ONLINE EVENT
14th - 18th SEPTEMBER 2020

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Joan Bladé
Hospital Clinic, Barcelona, Spain

ISA INTERNATIONAL SOCIETY OF AMYLOIDOSIS

www.isa2020.org
On behalf of the international Society of Amyloidosis (ISA) and the PETHEMA Foundation it is my pleasure to invite you to the XVII ISA Symposium. It will be held from September 14th to 18th, 2020 in an on-line format. It was planned for March 1-5 in Tarragona as a regular meeting, but it was postponed and later transformed at the virtual form because of the COVID-19 pandemic.

The Symposium will consist of a five days scientific event. All types of amyloidosis will be covered: TTR (hereditary and wild-type), light chain (AL), AA as well as less frequent forms. There will be key lectures, plenary sessions, industry sponsored symposia, oral presentations plus meet-the-expert sessions and poster exhibition.

This Symposium is attended by all the outstanding researchers in all the fields of amyloidosis plus about 800 delegates particularly interested in these diseases. The ISA 2020 is a unique opportunity to learn on basic biology, early diagnosis and recognition of unusual forms, best prognostication and response assessment, new drug targets and the role of novel agents, including immunotherapy, as well as the updated results from early phase and large randomized clinical trials. Please join us for this exciting conference on amyloidosis.

Welcome to ISA 2020!!!

Joan Bladé
President ISA Symposium 2020

Hematology Department, Hospital Clinic. Barcelona Spain
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HIGH RESOLUTION CRYO-EM STRUCTURE OF A TRANSTHYRETIN-DERIVED AMYLOID FIBRIL FROM A PATIENT WITH HEREDITARY VAL30MET ATTR AMYLOIDOSIS

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Introduction: ATTR amyloidosis is one of the worldwide most abundant forms of systemic amyloidosis. The disease is caused by the misfolding of transthyretin protein and the formation of amyloid deposits at different sites within the body. The most common associated mutation giving rise to hereditary ATTR amyloidosis is Val30Met. Two main fibril morphologies have been found in ATTR amyloid deposits. Type A fibrils are more common and adopted by wildtype TTR and the vast majority of analysed mutational variants from hereditary amyloidosis including V30M. They are relatively short and haphazardly arranged in ultrathin sections of amyloidotic tissue and consist of a mixture of N-terminally truncated as well as full-length TTR. Type B fibrils are more elongated and arranged into bundle-like assemblies that primarily consist of full-length TTR protein. All organs of a patient show either type A or type B fibrils, and the fibril type of a patient does not change over time.

Objectives: The goal was to obtain a high-resolution structure of ex vivo transthyretin-derived amyloid fibril (Type A) to create a molecular model. This allows to gather detailed structural information on ATTR amyloid fibrils and to help develop a fundamental molecular understanding of the mechanism of disease.

Methods: Fibrils from amyloidotic heart tissue from a patient with hereditary Val30Met ATTR amyloidosis using recently established protocol were purified and analyzed via cryo-electron microscopy. Collected date was reconstructed using single particle based helical reconstruction with RELION.

Results: After cryo-electron microscopy of the purified Type A fibrils from patient tissue a 2.97 Å structure could be reconstructed. The fibril reconstruction shows a clear ~4.8 Å spacing, is polar, left-handed, possesses C1 symmetry and consists of a single, twisted protofilament. The fibril is mixed in the sense that it contains two non-homologous polypeptide chains an N-terminal and a C-terminal fragment of transthyretin.

Conclusions: The structure provides insights into the mechanism of misfolding and implies the formation of an early fibril state from unfolded transthyretin molecules, which upon proteolysis converts into mature ATTR amyloid fibrils. Given that the structure of an in vitro formed TTR fibril has nativelike structural characteristics and differs in this starkly from the presently analyzed fibril, this data underscores the importance of investigating patient-derived amyloid fibrils when analyzing the molecular basis of amyloid diseases.

Keywords: ATTR fibril, high resolution structure, cryo-EM.
DEFINING THE CARDIAC AMYLOID PROTEOME AND ITS ASSOCIATION WITH PATIENT CLINICAL CHARACTERISTICS AND OUTCOMES

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Introduction: Many proteins co-deposit with light chains (AL) or transthyretin (ATTR) in tissue and can be identified using laser capture microdissection and mass spectroscopy (LMD-MS). Understanding the cardiac amyloid proteome (CAP) may provide insight into the pathogenesis of cardiac amyloidosis. A major limitation in its accurate characterization is that many of the identified proteins are also abundant in normal tissue.

Objectives: To define the CAP in AL/ATTR and its association with clinical outcomes.

Methods: We used LMD/MS to identify proteins over-expressed in amyloid plaques compared to control tissues. We included 292 ATTRs, 153 ALs, 5 normal controls and 14 disease controls (5 restrictive, 9 hypertensive cardiomyopathies) which were combined for analyses. Protein abundance (normalized spectral counts) in AL and ATTR was compared to controls and proteins were considered part of the CAP if their abundance increased by 50%, (FDR of <0.05). Disease severity was assessed with the British (ATTR) and Mayo 2012 (AL) clinical staging systems.

Results: The CAP proteins are shown in the table. Collagen proteins were more abundant in AL whereas complement proteins were almost exclusively present in ATTR (p<0.0001). Despite a lower abundance of the amyloidogenic proteins, AL was associated with higher abundance of signature proteins (p<0.0001). In ATTR, complement proteins were increased in older patients (<0.001) and complement effector (C9) proteins were increased whereas regulatory (CFHR1, CFHR5) proteins were decreased (p<0.05) in higher cardiac stage patients. In AL CAPs, lambda light chain abundance was higher than kappa (P=0.01) despite similar FLC serum levels. Hierarchical clustering was performed separately for AL and ATTR and revealed clusters of patients with unique proteomic compositions. In ATTR, samples were organized based on decreasing protein abundance except for MAGEL2 and PIK3C/GDP1. Patients in the high PIK3/GDP1 cluster were twice as likely to die within 1 year from diagnosis (p=0.04). Their survival (OS) was worse independent of cardiac stage or mutation status (p=0.01). AL samples formed less homogeneous clusters. A high PIK3C/GPD1 cluster was again noted and had worse OS independent of cardiac stage (p=0.004).

Conclusions: We define the CAP in AL and ATTR and show the co-deposition of several proteins with variable functions. Our findings suggest that AL may cause more cardiac fibrosis and that complement activation may be important in TTR pathogenesis. AL can more effectively attract signature proteins than ATTR and lambda light chains may seed amyloid plaques more effectively than kappa. Finally, the CAP is prognostic of survival independent of existing staging systems in both AL and ATTR. Increased PIK3C was common in high risk patient subsets in both types irrespective of stage. PIK3C is essential for autophagy and impaired autophagy has been implicated in amyloid cardiotoxicity. Autophagy could represent a common mechanism of damage in AL and ATTR.
Table. The AL and ATTR cardiac proteomes organized by major protein groups. Proteins in **bold** are common across the two amyloid types.

<table>
<thead>
<tr>
<th>TTR, 29 proteins</th>
<th>AL, 19 proteins</th>
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<tr>
<td><strong>Signature</strong></td>
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<tr>
<td>TTR, 29 proteins</td>
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<td>1. APOA4</td>
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<td>2. APOE</td>
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<td>3. APCS</td>
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<td>2. COL1A1</td>
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<td>3. COL1A2</td>
<td>Other</td>
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<tr>
<td>4. COL3A1</td>
<td>1. AMBP</td>
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<td>5. COL6A1</td>
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<td>6. COL6A2</td>
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<td>7. DCN</td>
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<td>8. FBLN1-C</td>
<td>5. QSOX1</td>
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<td>9. FBLN1-D</td>
<td>6. SERPINE2</td>
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<td>10. PCOLCE2</td>
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IMMUNOGENETIC PROFILE OF PURIFIED PATHOLOGICAL PLASMA CELLS OF PATIENTS WITH LIGHT CHAIN AMYLOIDOSIS


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13 Hospital Son Espases.
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17 Hospital de Galdakao.
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Background: High-throughput sequencing (NGS) studies have rendered seminal knowledge in hematological malignancies. Unfortunately, the low incidence of light-chain amyloidosis (AL) and its low tumor burden account for the limited information on its tumor cell biology. Thus, greater knowledge about the immunogenetic landscape is required since potential differences between the genomic profiles of AL and multiple myeloma (MM) could help identifying patients with monoclonal gammopathies at greater risk of developing AL and monitor pre-symptomatic organ damage.

Aims: To perform whole exome sequencing (WES) in a series of patients with AL, to compare mutational profiles and copy number abnormalities (CNA) in AL vs MM. Moreover, to analyze by NGS the immunoglobulin (Ig) repertoires.

Methods: A total of 27 patients with confirmed diagnosis of AL were included. WES was performed in 56 paired samples of FACSorted bone marrow tumor plasma cells and peripheral blood mononucleated cells. Tumor samples was captured in triplicate using Agilent’s SureSelect V6 + UTR kit and sequenced on the Illumina platform. Data was analyzed with Strelka and Varscan2 software. CNVKit was used to CNA detection. We used the MMRF CoMMpass IA13c dataset (930 patients) to compare the mutational landscape of MM vs AL. Ig libraries were generated with NEBNext Fast DNA Library Prep Set, sequenced on an Ion S5 sequencer and were analyzed using an in house method.

Results: We identified a total of 718 exonic, non-Ig, nonsynonymous mutations with a variant frequency greater than 5% (683 SNV and 35 indel). Only 37 out of 662 (5.5%) mutated genes were altered more than once.

The most frequently mutated genes were FAT4, IGLL5, MUC16 and SSH2. With a median of 18 (8 – 92) mutations per sample, patients with AL are closer to MGUS (median of 19) rather than MM (median of 38, p<0.0001) in terms of mutational load. By contrast, presence of CNA was more frequent in AL (90.5%) than MGUS (60.6% in Mikulasova et al.) and similar to MM patients (virtually 100%). Interestingly, MM-defined driver mutations were undetected in AL (e.g. NRAS, BRAF, TRAF) or observed only once (e.g., DIS3 and DUSP2), most of them being subclonal in AL. Gains in chromosomes 9 and 19 were associated with inferior SPF, whereas del(13q) was associated with higher NT-proBNP levels. Furthermore, patients with +1q also displayed greater risk of cardiac involvement.
According to Ig repertoire, the most frequent IGH gene involved in AL was *IGHV3-48* (recurrence of 10.3%) and was significantly associated with kidney involvement (p=0.025), whereas *IGHV3-30* was the most recurrent (12%) in MM.

**Conclusions:** This study confirms previous observations that AL cannot be defined by a singular or a set of well-defined genetic events, and locate AL in the crossroad between MGUS and MM also in genetic grounds. Our results also provide new immunophenotypic markers that could emerge as novel risk-markers for AL in patients with monoclonal gammopathies.
OP04

FROM PROTEIN-PROTEIN INTERACTION TO PROTEIN CO-EXPRESSİON NETWORKS: A SYSTEMS BIOLOGY-BASED PERSPECTİVE TO İNVESTİGATEAMYLOIDOSİS DİSEASES.

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Introduction: In systemic amyloidosis, accumulation of misfolded proteins as extracellular amyloid fibrils causes severe organ dysfunction, but the molecular events of tissue damage are still largely unknown. Mass spectrometry-based proteomic analysis of tissues is a powerful approach to investigate these diseases, both for diagnosis and to highlight the molecular mechanisms altered by amyloid deposition. In fact, this approach allows a wide characterization of the proteome analyzed, thus providing plenty of data useful to address the study of amyloidosis using system biology-based computational approaches.[1]

Objectives: In addition to identifying relevant molecules, in terms of differentially expressed and topologically relevant proteins, the aim of our work is to provide an alternative computational method, based on experimental proteomic data, statistics and protein-protein interaction (PPI) networks, to in silico characterize protein complexes and biological processes affected by amyloid deposition.

Methods: The protein profiles obtained by analyzing the subcutaneous adipose tissue of 14 Healthy Controls, 23 Amyloidosis κ and 23 Amyloidosis λ patients were used to reconstruct a PPI network, by STRINGdb [2], and a protein co-expression network, per group, by Weighted Gene Co-expression Network Analysis (WGCNA) (1). Both networks were processed at functional and topological level to identify relevant proteins in terms of modules and hubs (1). Moreover, protein correlation and PPIs were combined to select regulated and/or dysregulated protein complexes and biological processes.

Results: Following the combination of experimental proteomic data, WGCNA and human PPI network, we found that, in comparison to healthy controls, the levels of subunits of some protein complexes, including ATP synthase, tubulin and collagen, were less correlated in both Amyloidosis κ and λ patients. On the contrary, proteins involved in biological processes like carbohydrate metabolism or unfolded protein response increased their score of correlation. On the other hand, the topological analysis of the reconstructed PPI networks permitted the identification of several hub proteins, including HSP90AB1 for Amyloidosis λ. Of note, some subunits of the T-complex protein 1, which play a role in the folding of actin and tubulin, resulted hubs for Amyloidosis κ.

Conclusion: Based on this study, amyloid deposition seems to affect the correlation of proteins composing biological complexes and processes related to cytoskeleton, metabolism and unfolded protein response. In particular, the loss of correlation among proteins we know to physically interact could be an indication of their dysregulation. Of course, these hypotheses need further validations by means of independent approaches. However, the preliminary results are encouraging and the proposed procedure could represent an alternative method for investigating a wider range of diseases.

Keywords: MS-based Proteomics, Protein-Protein Interactions, Co-expression Network

TARGETING DEUBIQUITYLATING ENZYMES USP14 AND UCHL5 IN SYSTEMIC IMMUNOGLOBULIN LIGHT CHAIN (AL) AMYLOIDOSIS

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Background: In AL amyloidosis, the underlying bone marrow (BM) residing plasma cell (PC) clone is usually small and poorly proliferating, and peculiar biologic and molecular features set it apart from multiple myeloma (MM). Expression of AL light chains induces endoplasmic reticulum (ER) stress and renders AL PCs particularly dependent on the proteocatabolic activity of the ubiquitin proteasome system (UPS). The proteasome inhibitor bortezomib forms the cornerstone of therapy for transplant-ineligible patients. However, a significant proportion of AL patients fail to respond to bortezomib-based combinations, especially those cases that exhibit the t(11;14) translocation, which is seen in >40% of AL patients. Recent studies have shown that targeting deubiquitinating (DUB) enzymes upstream of the 20S proteasome can overcome bortezomib-resistance in preclinical models of MM. In particular, USP14 and UCHL5 DUB enzymes are associated with the 19S regulatory particle lid of the proteasome that removes ubiquitin from target proteins to facilitate protein degradation by downstream 20S proteasome core particle.

Objectives: To examine the role of USP14/UCHL5 DUBs in AL using biochemical and pharmacological approaches.

Methods: We examined the activity of a small molecule inhibitor of the USP14 and UCHL5 DUBs b-AP15 against the AL cell line ALMC-2, as well as primary CD138+ and CD138- BM-derived cells immunopurified from AL patients at diagnosis. Moreover, we examined t(11;14)-positive MM cell lines U266 and KMS-12. For co-culture experiments, mesenchymal stromal cells (MSCs) were obtained from healthy individuals BM. Drug-induced accumulation of polyubiquitinated proteins was determined by Western blotting. Cell metabolism and cell viability was analyzed using CellTiterGlo, trypan blue and/or Annexin V-propidium iodide assays, respectively.

Results: 1) b-AP15 treatment triggered a dose-dependent reduction in cell metabolism and cell viability in U266, KMS-12 and ALMC-2 cells (IC50 range:100 and 200 nM). 2) Treatment of ALMC-2 cells with b-AP15 induced a progressive accumulation of polyubiquitinated proteins, followed by induction of apoptosis in these cells. 3) Treatment of primary BM-derived CD138+ PCs from AL patients (n=9 patients: 4 with t(11;14)-positive clones) with nanomolar concentrations of b-AP15 for 24h significantly decreased their metabolic activity, without markedly affecting matched CD138- fractions and PBMCs from normal healthy donors. These data suggest specific anti-AL activity and a favorable therapeutic index for b-AP15 in AL. 4) Finally, the cytotoxic activity of b-AP15 against AL cells was retained even in the presence of primary BM-derived MSCs.

Conclusions: Our preclinical data showing efficacy of b-AP15 in AL disease models validate targeting DUBs upstream of the proteasome to overcome proteasome inhibitor resistance, and provides the framework for clinical evaluation of USP14/UCHL5 inhibitors to improve patient outcome in AL.
MEMBRANE AND SOLUBLE B-CELL MATURATION ANTIGEN (BCMA) IN SYSTEMIC LIGHT-CHAIN AMYLOIDOSIS

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Introduction: Membrane-bound B-cell maturation antigen (mBCMA), a transmembrane glycoprotein in the TNF-receptor superfamily, is expressed on plasma cells and cleaved from the plasma cell-surface by γ-secretase, resulting in soluble serum BCMA (sBCMA). mBCMA is an important therapeutic target in multiple myeloma (MM) and may become an important target in systemic light-chain amyloidosis (AL). Therefore, in IRB and IACUC approved studies, we investigated mBCMA and sBCMA in AL patient marrow and blood specimens, modulated mBCMA on CD138-selected AL plasma cells with the γ-secretase inhibitor (GSI) LY-411575, and studied sBCMA in the blood of smoldering multiple myeloma patients (SMM) and in a mouse xenograft model with the λ light-chain secreting ALMC-1 AL cell line.

Methods: Mononuclear cells (MNC) were isolated from patient marrow aspirates with anti-CD138 microbeads (Miltenyi Biotec, Auburn, CA), and mBCMA expression was analyzed by flow cytometry using APC conjugated anti-CD269 (BCMA) antibody (Biolegend, San Diego, CA, USA) and CD138 expression by PE-conjugated anti-CD138 antibody (Biolegend) with isotype controls. sBCMA and free light chains (FLC) were measured by ELISA (R&D Systems, Minneapolis, MN; Bethyl Lab, Montgomery, TX). CD138-selected cells were cultured for 48 hours with GSI (LY-411575, Sigma Aldrich, St. Louis, MO) and changes in mBCMA evaluated. We injected 10⁷ ALMC-1 cells in the flanks of NOD scid (NSG) mice to create a xenograft model of AL clonal plasma cell disease (Jackson Laboratories, Bar Harbor, ME). Statistical calculations were performed with MedCalc (Ostend, Belgium).

Results: Marrow and blood were obtained from 22 AL patients. Medians of marrow plasma cells, involved FLC levels (iFLC), mBCMA expression on CD138⁺ plasma cells, and sBCMA levels were 5% (Interquartile range (IQR), 3-10%), 62.8 mg/L (21.2-271.5 mg/L), 39% (7-61) and 31.7 ng/mL (6.6-109.7) respectively. Blood was obtained from 16 SMM patients who had median iFLC and sBCMA of 115 mg/L (26.3-1420) and 86.5 mg/ml (15-168). sBCMA levels in AL and in SMM patients correlated with iFLC. sBCMA levels in AL and SMM patients were significantly different (Mann-Whitney, P < 0.01). In culture with GSI, mBCMA on ALMC-1 cells increased from 85% to 100% and on AL patient CD138⁺ cells from 36% to 68% (Paired t-test, P < 0.01). The sBCMA levels in CD138⁺ cell culture supernatants decreased by over 50%. In NSG mice with ALMC-1 xenografts, λ FLC and sBCMA levels were 949.1 mg/L (868.8-23629.2) and 3.8 ng/ml (0.9-23.6) and were significantly correlated (r= 0.99, p<0.01).

Conclusions: mBCMA expression is present on AL plasma cells and sBCMA is present in all AL patients and correlates with iFLC. mBCMA can be significantly up-regulated on AL patient plasma cells with GSI. sBCMA may be useful as a marker of disease activity in patients with low iFLC. These results provide the basis for applying anti-BCMA immunotherapies in clinical trials in relapsed refractory AL patients.
SKIN BIOPSY IN HEREDITARY TRANSTHYRETIN AMYLOIDOSIS WITH POLYNEUROPATHY IN FRANCE

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Objective: Hereditary transthyretin amyloidosis with polyneuropathy (ATTRv-PN) is a systemic disease with predominant peripheral nervous system involvement due to transthyretin amyloid depositions. Herein, we assessed data from a large series of ATTRv-PN patients to define the usefulness of skin biopsy (SB), alone or in combination with labial minor salivary gland biopsy (LSGB), in disclosing amyloid depositions. Moreover, intraepidermal nerve fiber density (IENFD) at three sites (ankle-thigh-wrist), and its relationship with clinical and para-clinical parameters, were also evaluated.

Methods: We reviewed the clinical and histopathological data from 218 symptomatic ATTRv-PN patients (carrying 20 different mutations; 60% with Val30Met) who underwent SB and/or LSGB in our centre between January 2012 and February 2019. Of them, 128 underwent both procedures. Moreover, we reviewed data from 36 asymptomatic carriers who underwent SB.

Results: SB can detect amyloid depositions in 80.32% of symptomatic subject. Combining SB and LSGB leads to a sensibility of 88.28% in early onset Val30Met patients. Amyloid was found in only 13.9% of asymptomatic carrier. Distribution patterns of amyloid deposition are the same for all mutations excepted peri-annexial and muscle cells who are over-represented in early onset Val30Met. All symptomatic subjects and 34/36 asymptomatic carriers showed IENFD reduction at ankle. IENFD was progressively reduced at all site according with mPND. IENFD correlates with disease severity and duration.

Conclusions: Skin biopsy is a mini-invasive, reliable and sensitive tool in detecting amyloid depositions, a major event to help the decision to initiate a disease modifying therapy in ATTRv-PN patients.

Keywords: ATTRv, polyneuropathy, skin biopsy, labial minor salivary gland biopsy, amyloid deposition, early biomarkers
LONG-TERM SAFETY AND EFFICACY OF PATISIRAN: GLOBAL OPEN-LABEL EXTENSION 24-MONTH DATA IN PATIENTS WITH HEREDITARY TRANSTHYRETIN-MEDIATED AMYLOIDOSIS

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Introduction/Background: Hereditary transthyretin-mediated (hATTR) amyloidosis, also known as ATTRv amyloidosis, is a progressive, life-threatening disease; the majority of patients develop a mixed phenotype including polyneuropathy and cardiomyopathy. The efficacy and safety of patisiran has been demonstrated over 18 and 24 months in Phase 3 (APOLLO) and Phase 2 Open-Label extension (OLE) studies, respectively, in patients with hATTR amyloidosis with polyneuropathy. Efficacy and safety results from an analysis of the ongoing Global OLE study are described, with the potential to present a 24-month update at the congress.

Methods: Multicenter, international, OLE, safety and efficacy study (NCT02510261) in eligible patients who completed parent studies, including APOLLO patients randomized to placebo (APOLLO/placebo, n=49) or patisiran (APOLLO/patisiran, n=137) and Phase 2 OLE patients (n=25).

Results: 211 patients were enrolled into Global OLE; data from 24-month assessments are anticipated by the end of 2019. As of September 24, 2018, 189 patients had 12-month assessments. Baseline demographics include: median age 64 years, 74% male, and 46% V30M. Safety profile remained consistent with previous studies. After 12 months of additional patisiran treatment in the Global OLE, durable improvement was seen for mNIS+7 (mean change [SEM]) in APOLLO/patisiran (-4.0 [1.9]) and Phase 2 OLE (-4.7 [3.5]) groups compared to their parent study baselines. Norfolk QOL-DN (not measured in the Phase 2 OLE) also showed durable improvement in APOLLO/patisiran patients (-3.9 [2.1]) following additional 12-months treatment in OLE. In the Global OLE, APOLLO/placebo patients experienced halting of disease progression and quality of life (QOL) improvement compared to Global OLE baseline after 12 months of patisiran (mNIS+7: -1.4 [2.4], Norfolk QOL-DN: -4.5 [2.5]), although they had progressed relative to APOLLO baseline (mNIS+7: +24.0 [4.2], Norfolk QOL-DN: +15.0 [3.4]) given the progression while on placebo in APOLLO.

Conclusions: Overall, patients with long-term exposure to patisiran demonstrated durability of efficacy. Despite marked progression on placebo during the 18-month APOLLO study, previously untreated patients exhibited halting of disease progression and QOL improvement following 12 months of patisiran. However, delay in treatment resulted in these patients accumulating greater disease burden compared to patients who started patisiran treatment earlier, demonstrating the need for early treatment. It is anticipated that the 24-month Global OLE safety and efficacy data will continue to demonstrate a positive benefit:risk profile for patisiran.

Keywords: hATTR, patisiran, polyneuropathy
LONG-TERM IMPACT OF TAFAMIDIS IN PATIENTS WITH LATE-ONSET HEREDITARY TRANSTHYRETIN AMYLOIDOSIS WITH STAGE I POLYNEUROPATHY

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Background: Tafamidis meglumine is approved in Europe for the treatment of hereditary transthyretin amyloidosis in adult patients with FAP stage I symptomatic polyneuropathy to delay neurological impairment. There are limited long-term real-world data on the efficacy of tafamidis in patients from populations characterized by late-onset phenotype associated with a wide range of different mutations, as observed in Italy.

Objective: To investigate the long-term outcome of patients with late-onset, FAP stage I hereditary ATTR polyneuropathy treated with tafamidis at a single referral centre in Italy.

Methods: The cohort includes consecutive symptomatic adult patients with a diagnosis of hereditary ATTR amyloidosis with stage I polyneuropathy given prospective longitudinal follow-up. For comparative purposes three different populations were predefined as follows: 1) patients treated with tafamidis according to EMA (European Medicines Agency) indication regardless of whether they discontinued the drug (intention to treat approach); 2) liver transplanted patients (OLT); 3) untreated patients because of lack of therapeutic options at the time of diagnosis.

Results: 116 consecutive patients (82 males, 71%) with hereditary ATTR and stage I polyneuropathy (21 different mutations, Val30Met 22.4%) were included. 71 patients were treated with tafamidis, 20 patients by liver transplant and 25 were untreated. The demographic and clinical characteristics of the three groups are presented in the Table. Survival of patients treated with tafamidis was significantly longer (median 83 months) compared with untreated patients (median 41 months, p<0.0001) and did not significantly differ from transplanted patients (median 126 months, p=0.8279). When analysis was limited to patients with heart involvement, a significant survival advantage was observed in patients treated with tafamidis (median 83 months) compared to untreated patients (median 38 months, p<0.0001). Similarly, significant difference in survival between tafamidis treated (median 83 months) and untreated patients (median 38 months, p=0.0001) was observed in patients with PND 2 which reflects a more advanced neurological impairment at baseline. PND progression by 1 point was significantly less frequent over 36 months in patients treated with tafamidis compared to untreated patients, irrespective of baseline PND score (p=0.001).

Conclusions: Tafamidis delays progression of neurological and cardiac impairment and has a significant survival benefit in patients with late-onset hereditary ATTR amyloidosis and stage I polyneuropathy associated with a wide range of mutations.

<table>
<thead>
<tr>
<th></th>
<th>Tafamidis (n=71)</th>
<th>No therapy (n=25)</th>
<th>OLT (n=20)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age at diagnosis, years</td>
<td>65 (31-78)</td>
<td>62 (31-68)</td>
<td>51 (31-66)**</td>
</tr>
<tr>
<td>Age at onset, years</td>
<td>63 (28-77)</td>
<td>59 (53-64)</td>
<td>50 (28-60)**</td>
</tr>
<tr>
<td>Age at onset &gt; 50 years</td>
<td>56 (79%)</td>
<td>22 (88%)</td>
<td>10 (50%)*</td>
</tr>
<tr>
<td>Male, n (%)</td>
<td>50 (70.4%)</td>
<td>20 (80%)</td>
<td>12 (60%)</td>
</tr>
<tr>
<td>Disease duration, months</td>
<td>31 (1-106)</td>
<td>33 (6-65)</td>
<td>30 (4-90)</td>
</tr>
<tr>
<td>PND=2, n (%)</td>
<td>23 (32%)</td>
<td>17 (68%)**</td>
<td>8 (40%)</td>
</tr>
<tr>
<td>Heart involvement, n (%)</td>
<td>44 (62%)</td>
<td>21 (84%)*</td>
<td>8 (40%)</td>
</tr>
<tr>
<td>NT-proBNP (pg/ml)</td>
<td>419 (29-13726)</td>
<td>824 (30-16299)</td>
<td>391 (38-12539)</td>
</tr>
<tr>
<td>Orthostatic hypotension, n (%)</td>
<td>29 (41%)</td>
<td>12 (48%)</td>
<td>7 (35%)</td>
</tr>
<tr>
<td>mBMI</td>
<td>994 (715-1562)</td>
<td>981 (573-1440)</td>
<td>1054 (670-1371)</td>
</tr>
</tbody>
</table>

Comparison vs tafamidis * p < 0.05, ** p < 0.01
OP10

EXTERNAL VALIDATION OF THE NATIONAL AMYLOIDOSIS CENTRE SCORE IN AN INTERNATIONAL COHORT OF PATIENTS WITH TRANSTHYRETIN CARDIAC AMYLOIDOSIS.

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Introduction: Cardiac transthyretin (ATTR) amyloidosis is an increasingly recognized, progressive, and fatal cardiomyopathy in which the natural history remains unclear. Recently, Gillmore and colleagues from the National Amyloid Center (NAC) in London, UK, have proposed a new staging system to predict long-term survival in ATTR patients based on NT-proBNP and eGFR values.

Objectives: We sought to externally validate this prognostic staging system in patients with both wild-type ATTR (ATTRwt) and hereditary ATTR (ATTRh) amyloid cardiomyopathy.

Methods: Clinical characteristics and survival data from cardiac TTR amyloidosis patients with NT-proBNP and eGFR data available evaluated at Columbia University Hospital (New York, US), University of Bologna (Italy), University of Pavia (Italy) and Hospital Puerta de Hierro (Madrid, Spain) were retrospectively collected. Patients were classified in 3 groups: Stage I (NT-proBNP ≤3000 ng/L and eGFR ≥45 ml/min), Stage II (NT-proBNP ≥3000 ng/L or eGFR ≤45 ml/min) and Stage III (NT-proBNP ≥3000 ng/L and eGFR ≤45 ml/min). A Cox regression model adjusting for age, as main predictor of death was performed.

Results: 406 patients with cardiac ATTR amyloidosis (83% male, mean age 76±8.8 years, 77.8% ATTRwt and 22.2% ATTRh) were included in this analysis. During a median follow-up of 23 months (IQR: 10.3-38.7), 111 (27%) patients died. Median survival was 68 months among 221 (54.4%) Stage I patients, 46 months in 136 (33.5%) Stage II patients, and 28 months in 49 (12.1%) Stage III patients (P=0.002). After adjusting for age, compared with Stage I, the hazard ratio (HR) for death for Stage II patients was 1.73 [95% confidence interval (CI) 1.11-2.71, P=0.016] and for Stage III was 2.95 (95%CI 1.71-5.08, P<0.001). These results are very similar to the original survival values reported in the NAC staging system cohort (median survival was 69.2, 46.7 and 24.1 months in Stage I, II and III, respectively (P<0.0001)). HR for death for Stage II and Stage III compared with Stage I in the NAC cohort were 2.05 [95%CI: 1.54-2.72, P<0.001] and 3.80 (95%CI 2.73-5.28, P<0.001), respectively.

Conclusions: NAC staging score is highly reproducible and accurately predicts prognosis in an international external cohort of patients with ATTR cardiac amyloidosis.
EVALUATION OF PATISIRAN WITH CONCOMITANT OR PRIOR USE OF TRANSTHYRETIN STABILIZERS IN PATIENTS WITH HEREDITARY TRANSTHYRETIN-MEDIATED AMYLOIDOSIS

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Introduction/Background: Hereditary transthyretin-mediated (hATTR) amyloidosis, also known as ATTRv amyloidosis, is a life-threatening disease caused by a mutation in the transthyretin (TTR) gene resulting in misfolded TTR proteins accumulating as amyloid fibrils in multiple organs. Current pharmacologic treatments include TTR stabilizers (tafamidis, diflunisal), which stabilize the TTR protein complex, and TTR silencing therapies (patisiran, inotersen), which reduce production of mutant and wild-type TTR protein.

Objectives: To evaluate safety and pharmacodynamics (PD) of patisiran alone or with concomitant TTR stabilizers from the Phase 2 Open-Label Extension study (OLE; NCT01961921) and safety and efficacy of patisiran in patients with prior TTR stabilizer use from the Phase 3 APOLLO study (NCT01960348).

Methods: During the 24-month Phase 2 OLE study, concomitant TTR stabilizer was permitted if initiated prior to study entry; safety and PD were summarized across concomitant TTR stabilizer groups in a post hoc analysis. In the 18-month APOLLO study, prior TTR stabilizer use was a stratification factor at randomization; patients were required to discontinue TTR stabilizers ≥14 days before study entry. Post-hoc safety and efficacy analyses across prior TTR stabilizer use groups were evaluated.

Results: In the Phase 2 OLE (N=27), 7 patients (25.9%) did not take a TTR stabilizer (patisiran alone) and 13 (48.1%) and 7 (25.9%) patients took concomitant tafamidis or diflunisal, respectively. Median exposure to patisiran was 736 days, to patisiran with concomitant stabilizers 736 days (tafamidis) and 421 days (diflunisal). Overall safety profiles across concomitant TTR stabilizer groups were consistent with the safety profiles of the respective therapies as monotherapy. Mean (standard error of the mean [SEM]) serum TTR reduction over 24 months was similar, regardless of whether a patient received patisiran alone or with concomitant tafamidis or diflunisal (83.4% [3.5], 81.8% [1.4], 81.3% [3.0]). At APOLLO baseline (N=225), 74 patients (32.9%) reported prior tafamidis, 45 (20.0%) prior diflunisal. Safety profile was consistent, regardless of prior TTR stabilizer use. At 18 months, mean (SEM) improvement in mNIS+7 was seen across all patisiran-treated patients, regardless of prior stabilizer use (no prior use -3.4 [2.69], prior tafamidis -6.3 [2.41], prior diflunisal -2.8 [2.68]), while placebo-treated patients progressed on average.

Conclusions: Evaluation of safety and PD data from a small cohort suggests that the PD profile of patisiran is unaffected by concomitant TTR stabilizer use. Additionally, these data indicate that patients benefit from patisiran treatment regardless of concomitant or prior use of TTR stabilizer.

Keywords: hATTR, patisiran, stabilizers
OPEN-LABEL STUDY OF PATISIRAN IN PATIENTS WITH HEREDITARY TRANSTHYRETIN-MEDIATED AMYLOIDOSIS WITH POLYNEUROPATHY POST-ORTHO TOPIC LIVER TRANSPLANT

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Introduction/Background: Hereditary transthyretin-mediated (hATTR) amyloidosis, also known as ATTRv amyloidosis, is a progressive, life-threatening disease; majority of patients develop a mixed phenotype of both polyneuropathy and cardiomyopathy. Orthotopic liver transplant (OLT) has historically been used to slow disease progression in early stage hATTR amyloidosis. However, it has been associated with disease progression due to wild-type amyloid fibril deposition. Patisiran significantly suppresses liver production of both mutant and wild-type transthyretin (wt TTR) and has been shown to halt or reverse polyneuropathy and improve quality of life (QOL) in patients with hATTR amyloidosis in the APOLLO study. Patisiran is approved in certain countries globally for the treatment of hATTR amyloidosis with polyneuropathy.

Objectives: To describe the baseline demographics, reduction in serum TTR levels following 3 weeks of treatment with patisiran, and 3-month interim safety results of patients enrolled in this Phase 3b open-label study evaluating the safety, efficacy, and pharmacokinetics (PK) of patisiran in patients with hATTR amyloidosis with polyneuropathy with disease progression post-OLT (NCT03862807).

Methods: Enrolled patients will receive patisiran 0.3 mg/kg intravenously once every 3 weeks for 12 months. Key inclusion criteria: adults who underwent OLT for treatment of hATTR amyloidosis ≥12 months before informed consent and had documented increase in polyneuropathy disability (PND) score either compared to pre-OLT assessment or between 2 assessments post-OLT. Key exclusion criteria: previous use of inotersen or patisiran and liver allograft rejection episodes or abnormal liver function tests suggestive of possible allograft rejection in the 6 months prior to informed consent. The primary objective is to evaluate serum TTR reduction with patisiran treatment. Assessments to evaluate the effect of patisiran on neuropathy, QOL, autonomic symptoms and nutritional status will also be conducted. Safety will be assessed throughout the study and up to 4 weeks after the last dose.

Results: 23 patients enrolled and received patisiran in the study. Median age was 58.0 years, 13 (56.5%) were males, and 11 (47.8%) had Val30Met mutation. At baseline, 1 (4.3%) patient had polyneuropathy disability (PND) score I, 9 (39.1%) had PND II, and 13 (56.5%) had PND IIIA/B. Eleven patients (47.8%) had New York Heart Association (NYHA) classification I, 6 (26.1%) had NYHA II, and none had NYHA III or IV at study baseline. Updated baseline demographic and disease characteristic data as well as reduction in serum TTR levels following 3 weeks of treatment with patisiran and interim 3-month safety data will be presented.

Conclusions: This study will continue to investigate the efficacy, safety, and PK of patisiran with the potential to address an unmet need in hATTR amyloidosis with polyneuropathy patients with disease progression post-OLT.

Keywords: hATTR, patisiran, OLT
HIGH RESOLUTION NERVE ULTRASOUND AS A DIAGNOSTIC TOOL FOR DIFFERENTIAL DIAGNOSIS AND PROGRESSION RECOGNITION IN TTR-RELATED FAMILIAL AMYLOIDOSIS

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Introduction/ Background: Hereditary neuropathies comprise a broad variety of progressively disabling diseases. Out of these, the transthyretin-related familial amyloidosis (hATTR) is of special interest due to its therapeutic relevance. To maximize the benefit from therapy, an early identification of the precise moment of disease onset is essential. The current standard, nerve conduction velocity (NCV) studies, is of limited use because of its low specificity, its dependency on patients’ compliance and the investigator’s experience. High resolution ultrasound (HRUS) of peripheral nerves is a new and promising method that might fill this gap. Recent studies have suggested algorithms for the differentiation of polyneuropathies by using an ultrasound score, the Ultrasound Pattern Sum Score (UPSS) (Grimm et al., 2016).

Objectives: Characterization of the ultrasound patterns of TTR-related familial amyloid neuropathy in different disease stages in comparison to other hereditary and acquired neuropathies such as chronic inflammatory polyneuropathy (CIDP) to enable an earlier diagnostic distinction.

Methods: At two clinical centers, we analyzed HRUS data in a cohort of 145 patients with different, genetically confirmed hereditary neuropathies and compared them to clinical and paraclinical results such as NCV studies. Cross-sectional area (CSA) of peripheral nerves were obtained at predefined landmarks in accordance with the UPSS protocol. We additionally assessed 35 immune-mediated neuropathy cases for comparison.

Results: We included a total number of 180 patients (145 hereditary, 35 immune-mediated neuropathies), 118 prospectively and 62 retrospectively. Hereditary neuropathies: The most frequent mutation was the heterozygous PMP22 duplication accounting for 33.7% (n = 49), followed by mutations in TTR and MPZ (both n=13, 8.9%), GJB1 (n = 12, 8.1%) and GLA (n=7, 4.8%). Acquired neuropathies: We retrospectively assessed 35 patients with CIDP and its variants (total=35). The CMT1A cohort showed the significantly highest nerve enlargement (UPSS mean=15.7±9.19 vs. CIDP mean=7.7 ±4.8, p=0.0001; TTR mean=2.9±2.8, p=0.0001). In hATTR patients, the UPSS ranged on a comparable score level with CMTX and HNPP patients (mean=2.9±2.8 vs 4.5±4.6 vs 1.4±2.0) and differed significantly from CIDP patients (p=0.001). Compared to other axonal neuropathies (mean=1.6±1.7), the UPS seemed to be higher in tendency. Herein, Coutinho disease stage 1 UPS was significantly enlarged compared to stage 0 (mean=5.2±1.6 vs 1.2±1.6, p=0.005). In the overall cohort, UPS values correlated with clinical parameters such as foot elevation strength and vibration perception at ankles.

Discussion
HRUS is a useful diagnostic tool for the differentiation of hereditary and inflammatory polyneuropathies serving as a potential biomarker for early disease progression in hATTR patients.
OP14

ORIGIN OF VAL30MET IN FAMILIAL AMYLOID POLYNEUROPATHY (TTR-FAP) IN PORTUGAL: A WALK THROUGH THE MUTATIONAL PATH

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Introduction: Variability in clinical presentation is present across all major worldwide Transthyretin Val30Met Familial Amyloid Polyneuropathy (TTR-FAP Val30Met) clusters in which a remarkably wide variation in age-at-onset (AO) with early and late disease forms has been described. Shedding some light on Val30Met ancestral origins, three independent mutational events have been hypothesized by Becker, Coutinho, Ohmori and Zaros et al, who suggested its association with the population differences in AO observed between each cluster. In Portugal it is also reported AO variation between TTR-FAP families from the different Portuguese main disease clusters. Nevertheless, whether there is a common origin for Val30Met mutation in the Portuguese cluster remains an open question.

Objectives: The main aims of this work were 1) to distinguish between one or multiple mutational events among the Portuguese Val30Met TTR-FAP kindreds, through characterization of the genetic background surrounding the Val30Met locus; 2) to estimate the age of its ancestral mutational event(s) and to disentangle the hypotheses on its spreading routes within Portuguese regions.

Methods: The access to the largest Val30Met TTR-FAP database available worldwide (~3000 patients, belonging to over 700 kindreds) allowed us to carry out an extensive family-based haplotype study, using STRs and SNPs markers, in several Portuguese Val30Met families originated from different disease clusters (Northern Coast, Inland and Central Coast).

Results: In our sample, mean AO was significantly higher in Val30Met patients from Inland compared to carriers from Northern Coast region ($p < 0.001$). Haplotype sharing analysis revealed a common haplotype of different lengths shared by almost all Val30Met carriers, providing strong evidence of a major single-founder effect for Val30Met in the Portuguese population, as previous suggested by Soares et al. According to the age estimates analysis, we hypothesized that the Portuguese Val30Met mutational event took place in the mid-VI century (AD) (1450–1475 years ago) and its dispersion occurred from Póvoa de Varzim and Vila do Conde to Inland region, corroborating the hypothesis of migration related to fishing/ farmer activities, as described in the literature. Although AO variability observed among Val30Met Portuguese kindreds does not seem to be attributable to different mutational origins, striking haplotypic differences downstream STR D18S1133 were found between Northern Coast vs. Inland and Central Coast carriers.

Conclusions: Further investigations on this genomic region may provide new insights on AO variability, such as the identification of other genetic modifiers that could guide for novel therapeutic strategies for TTR-FAP patients. With this study we were able to clarify Val30Met ancestral origins and its mutational path within the Portuguese cluster, strengthening the need for more surveillance of TTR-FAP Val30Met carriers.

Keywords: Familial Amyloid Polyneuropathy; Origin of mutation; Age-at-onset variability
99MTC-DPD SCINTIGRAPHY PREDICTS AMYLOID FIBRIL TYPE IN HEREDITARY TRANSTHYRETIN AMYLOIDOSIS

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2 Dept of Radiation Sciences, Radiation Physics, Umeå University, Sweden.
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Background: The amyloid fibril type has been shown to be related to the phenotype and to the outcome of transthyretin (ATTR) amyloidosis. Briefly, type A (full-length and fragmented ATTR) fibrils are associated with later disease onset and amyloid cardiomyopathy, whereas type B (full-length ATTR) fibrils are associated with earlier onset and mainly neuropathic complications. Parallel to histopathological tissue analysis, DPD or PYP scintigraphy has lately become an important diagnostic tool for ATTR amyloidosis, and DPD scintigraphy has also been shown to be associated with the amyloid fibril type.

Objectives: To re-evaluate the correlation between the outcome of 99mTc-DPD scintigraphy and the amyloid fibril type in ATTR amyloidosis.

Methods: Data from ATTR amyloidosis patients who had undergone 99mTc-DPD scintigraphy and had got their amyloid fibril type established from abdominal fat pad biopsies at the Amyloidosis Centre, Umeå University Hospital from 2012 to 2018 were evaluated. The biopsies had been analysed at the Department of Clinical Pathology, Uppsala University Hospital using Congo red staining to determine presence of amyloid and Western blot analysis to determine the type of amyloidosis (i.e. precursor protein and amyloid fibril type). Late disease onset was defined as onset at ≥50 years of age. Cardiac septum hypertrophy was defined as a septal thickness of >12 mm on echocardiogram.

Results: Seventy-two patients (65 % males) were available for analysis. Median age at examination was 68 (30-83) years. All patients had hereditary ATTR (ATTRv) amyloidosis, mainly ATTRV30M amyloidosis (90 %), and 54 (76 %) were late-onset cases. DPD scintigraphy was positive in 42 (58 %) of the patients and, among them, significantly more (p <0.01) had type A fibrils (88 %), late-onset disease (93 %) and septal hypertrophy (93 %). Of those with negative DPD scintigraphy, 29 (97 %) had type B fibrils, 15 (52 %) had late onset and 6 (20 %) had septal hypertrophy. Only one patient (76 year old female with ATTRV30M amyloidosis and heart and kidney failure) with type A fibrils had a negative DPD scintigraphy, whereas five patients (80 % males) with type B fibrils had a positive DPD scintigraphy and all of them were late-onset ATTRV30M patients (age 60-82 years) with cardiac septum hypertrophy. Overall, no difference in the outcome of DPD scintigraphy was found between the sexes (p = 0.83). All seven patients with non-V30M mutations displayed type A fibrils, positive DPD scintigraphy and septal hypertrophy, whereas all three patients with positive DPD and normal septum thickness had ATTRV30M amyloidosis and type A fibrils.

Conclusions: 99mTc-DPD scintigraphy is a good non-invasive predictor of cardiac amyloid and of the amyloid fibril type in ATTRv amyloidosis, however, a negative DPD scintigraphy does not rule out cardiac involvement. Further, older (and mainly male) patients with ATTRV30M amyloidosis and type B fibrils also seem to be at risk for amyloid cardiomyopathy.
HEPATIC EXPRESSION OF MUTANT TRANSTHYRETIN REMODELS PROTEOSTASIS MACHINERY IN HEREDITARY ATTR AMYLOIDOSIS

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BACKGROUND: Systemic amyloidosis represents a class of disorders in which misfolded proteins are secreted by effector organs and deposited as proteotoxic aggregates at downstream target tissues. Despite being well-described clinically, the contribution of effector organs such as the liver to the pathogenesis of these diseases is poorly understood.

OBJECTIVES: Here, we utilize our laboratory’s patient-specific induced pluripotent stem cell (iPSC)-based model of hereditary transthyretin (TTR) amyloidosis (ATTR amyloidosis) in order to define the contributions of amyloidogenic TTR-secreting hepatic cells to distal proteotoxicity observed in patients.

METHODS: We implemented a gene correction strategy to generate isogenic, ATTR amyloidosis patient-specific iPSCs expressing either amyloidogenic or wild-type TTR. We then utilized these gene edited iPSC lines in parallel with single cell RNA sequencing (scRNAseq) in order to identify transcriptional changes in iPSC-derived hepatic cells due only to the presence of the mutant, destabilized TTR. To assess functional consequences of activating adaptive unfolded protein response (UPR)-associated signaling in ATTR amyloidosis hepatic cells, we generated a patient-specific iPSC line capable of inducible ATF6 activation.

RESULTS: We developed a singular gene editing strategy capable of ameliorating all TTR genetic lesions. By employing this technology in a TTR^L55P ATTR amyloidosis iPSC line, we demonstrated total elimination of the secretion of destabilized, disease-causing TTR while wild-type levels remained unperturbed. scRNAseq of iPSC-derived hepatic cells revealed that expression of the pathogenic TTR^L55P mutation alone resulted in distinct transcriptional changes constituting activation of adaptive unfolded protein response (UPR)-associated signaling as well as known and novel chaperone genes. By recapitulating adaptive UPR activation in a branch-specific manner, employing our inducible ATF6 patient-specific iPSC line, we demonstrated that hepatic activation of branch-specific ATF6 signaling resulted in a significant reduction in the secretion of destabilized TTR^L55P, thereby highlighting a potential novel therapeutic pathway.

CONCLUSION: Together, these results suggest the capacity of the liver to act as a chaperone at a distance and contribute to downstream target organ pathogenesis, overturning the long-held belief in the field that ATTR amyloidosis livers are not different from normal individuals. Moreover, our data demonstrate the potential for UPR modulating therapeutics for the treatment of this and other systemic amyloid diseases.

KEYWORDS: hereditary ATTR amyloidosis; gene editing; single cell transcriptomics
DIAGNOSTIC POTENTIAL OF A NOVEL RT-qPCR-BASED ASSAY TO MEASURE CCND1 mRNA EXPRESSION LEVELS IN BONE MARROW PLASMA CELLS FROM AL AMYLOIDOSIS PATIENTS

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Background: AL amyloidosis is a life-threatening plasma cell (PC) tumor. A significant proportion of AL amyloidosis patients do not respond to frontline anti-plasma cell drugs and die before being offered second-line therapies. Hence, predicting which patients are more likely to resist frontline drugs has the potential to change the natural history of this disease. Recent studies in AL amyloidosis or multiple myeloma showed that the presence of the t(11;14)(q13;q32) or the consequent overexpression of the proto-oncogene CCND1, encoding cyclin D1, is associated with lower response rates towards bortezomib, and higher rates of response towards melphalan.

Objectives: As t(11;14) and CCND1 expression levels are not invariably associated, with a non-negligible proportion of t(11;14)-negative clones which overexpress CCND1, we hypothesized that a molecular diagnostic assay for measuring CCND1 levels in bone marrow (BM)-derived PCs may complement cytogenetics to guide treatment choices.

Methods: We designed different TaqMan-based, multiplexed assays for reliable CCND1 expression level measurements in purified BM-PCs. CD138+ plasma cells were obtained from diagnostic leftovers of bone marrow aspirated from treatment-naïve AL patients at diagnosis and CD138 purity was verified by flow cytometry. The same enriched plasma cell fraction was used for both transcriptional studies and interphase FISH.

Results: ACTR3 and ALG9 were selected as normalizing genes based on meta-analyses of transcriptomic datasets and subsequent experimental validation. According to MIQE guidelines, the TaqMan-based multiplex assays were technically validated, studying PCR efficiency, dynamic range and intra- and inter-assay variation. Moreover, the assays were compatible with low input RNA (obtained from 104 immunopurified CD138+ cells). We then applied the assays to measure CCND1 expression levels in BM plasma cells of an initial cohort of patients with AL and known t(11;14) status at diagnosis. Updated results of a larger cohort of patients will be presented.

Conclusions: Our results show variable CCND1 expression levels in BM-derived PCs, which can be explained only in part by the t(11;14) translocation status, thus strengthening the rationale for the development of a complementary test to measure CCND1 levels in these tumors. Further studies will be needed to fully explore the potential clinical utility of a TaqMan-based assay for CCND1 measurements in AL, especially in terms of prognostication of response to novel agents and guidance for treatment choices, towards a precision medicine approach.

Keywords: Molecular cytogenetics, Cyclin D1, AL amyloidosis
MACHINE LEARNING PREDICTS IMMUNOGLOBULIN LIGHT CHAIN TOXICITY THROUGH SOMATIC MUTATIONS

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3 University of Lausanne.

Background: In light chain (AL) amyloidosis, pathogenic monoclonal light chains (LCs) form toxic species and deposit as amyloid fibrils in target organs leading to organ failure and death. A known risk factor for AL is a pre-existing monoclonal gammopathy of undetermined significance (MGUS), which can remain in a benign condition or evolve either into AL or other clonal disorders. However, despite the presence of a link between MGUS and AL, delayed diagnosis remains common and reliable diagnostic tests able to predict whether MGUS patients are likely to progress to AL, are currently missing. Indeed, predicting the onset of AL is highly challenging as each patient carries a different pathogenic LC sequence, which is composed by a unique rearrangement of variable (V) and joining (J) immunoglobulin genes, and by a unique set of somatic mutations (SMs) acquired during B cell affinity maturation. Consequently, the development of specific prediction tools would be a crucial step to anticipate AL diagnosis and improve patients’ prognosis.

Objectives: We sought to identify predictive features of pathological light chains in the context of AL starting from their protein sequences. We aim at classifying LCs as either toxic or non-toxic, based on their likelihood to form toxic species in AL.

Results: We collected a database of toxic and non-toxic LCs of lambda (λ) isotype, since this latter is more prevalent than kappa (κ) in AL patients. The LC sequences of the collected database were firstly compared to the corresponding germline sequences and investigated through statistical analysis, showing that toxic sequences have a statistically significant higher chance of exhibiting somatic mutations as compared to non-toxic ones. Based on this finding, we devised LICTOR (λ-Light-Chain TOxicity predictoR), a machine learning approach able to predict λ LC toxicity starting from their amino acid sequences. LICTOR uses somatic mutations, exploited in sequence and structural features, as predictor variables to predict LC toxicity based on the hypothesis that SMs are the main discriminating factor of LC toxicity. LICTOR achieves specificity and sensitivity of 0.82 and 0.76, respectively, with an AUC of 0.87, making it a valuable tool for early AL diagnosis. Moreover, our results show that the presence or the absence of specific mutations at specific positions of LC structure, are key features used by LICTOR to classify LC sequences, which further underlines the pivotal role of SMs in the development of LC toxicity in AL.

Conclusions: LICTOR represents the first method able to accurately predict LC toxicity in AL. Hence, LICTOR may allow a timely identification of high-risk patients, such as MGUS patients likely to progress to AL, paving the way for early treatment and higher survival rates.

Keywords: light chain amyloidosis, prediction of light chain toxicity, machine learning.
OP19

DROSOPHILA MELANOGASTER AS A MODEL ORGANISM FOR ATTR AMYLOIDOSIS

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Introduction: Transthyretin (TTR) is a 55 kDa homotetrameric protein composed of four identical monomers of 127 amino acid residues. More than a hundred different mutations have been described in TTR and several of those are associated with amyloid formation while others have been shown to prevent misfolding. In hereditary forms of ATTR amyloidosis, a varying clinical phenotype occurs including age of onset, tissue distribution, symptoms, disease penetrance and prognosis.

Objectives: We selected Drosophila melanogaster as an organ-specific monitoring system to investigate whether TTR mutation is behind tissue tropism.

Methods: The Gal4-UAS system was used to drive the expression of human TTR with single mutations (TTR-V30L, TTR-V30M, TTR-L55P, TTR-R104H, TTR-A109S, TTR-A109T, TTR-L111M and TTR-T119M) wild type (wt) TTR and fragments of TTRwt (wt 1-49 and TTRwt 50-127) for site-specific expression in Drosophila. Fatbody-Gal4 was used to express protein in the adipose tissue, Hand-C-Gal4 in the heart, GMR-Gal4 in the eyes and Nrv2-Gal4 in peripheral nerves.

Results: Expression of mRNA and protein was confirmed with rt-PCR and western blot, respectively. In western blot, TTR monomers, dimers and tetramers were present. Transmission electron microscopy showed that aggregates composed of fibrillar structures and highly ordered spheres formed when the expression was driven by fatbody-Gal4. In flies with TTR expression driven by Nrv2-Gal4 driver movements were recorded for 120 hours using the Drosophila Activity Monitoring System (DAMS). In 10 days old flies, expression of TTRV30L, TTRV30M, TTRL55P, TTRA109S and TTRwt 1-49 resulted in a reduced daytime activity compared to control flies. In 20 days old flies expressing TTRV30L, TTRL55P, TTRT119M and TTRwt 50-127 a shift in the circadian rhythm was observed. Semi-Automated Optical Heartbeat Analysis (SOHA) was used to monitor effects of TTR expression driven by Hand-C-Gal4. The technique enables studies of heart rate, periodicity and contractility. In 15 day old flies, expression of TTRL111M resulted in a significant arrhythmia (p <0.01). The fractional shortening, an indicator of cardiac contractility, was significantly reduced over 30 days in TTR transgenic flies, with the exception of TTRR104H and TTRA109T, compared to controls (P <0.001).

Conclusions: The result of site directed expression of the selected TTR mutants resulted in the development of different phenotypes. These phenotypes were not dependent on TTR expression levels and could therefore depend on local effects caused by the specific TTR mutant.

ELEVEN DIFFERENT AMYLOID TYPES IDENTIFIED IN CUTANEOUS AMYLOIDOSIS BY PROTEOMICS-BASED TYPING

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¹ Mayo Clinic.

Introduction: Amyloidosis is characterized by deposition of insoluble misfolded proteins that form beta-pleated sheets. Cutaneous amyloidosis can be either localized (primary cutaneous amyloidosis) or systemic (secondary amyloidosis). At present 36 human amyloid proteins have been described and accurate determination of amyloid type is essential for optimal patient management. However, antigen-antibody-based typing methods such as immunohistochemistry are potentially unreliable for amyloid typing due to poor sensitivity (bias towards common expected amyloid types) and poor specificity (lack of antigen-antibody specificity).

Objectives: To understand the proteomic and demographic spectrum of disease in cutaneous amyloidosis by utilizing the current clinical standard of mass spectrometry-based proteomics to type cutaneous amyloid deposits.

Methods: We queried our liquid chromatography and tandem mass spectrometry (LC-MS/MS) amyloid typing database (2009 – 2018) for skin tissue specimens. This database consists of internal and external specimens for which Congo red-stained amyloid deposits were laser microdissected and subjected to shotgun proteomics using a clinically validated proteomics method.

Results: We identified 754 cutaneous amyloid cases (16% internal; 84% external). Table 1 summarizes the frequency and demographic information of the 11 amyloid types in our cohort. There were 367 females and 378 males (9 unknown); median age of 68. AL, which may be localized or systemic, was the most common type (67% of total cases). Insulin amyloidosis (AIns: 13%) and KRT5-14 amyloidosis (11%) were common localized forms. Of the 40 specimens with ATTR amyloidosis (5% of total), 9 had amino acid abnormalities consistent with TTR mutations by proteomic analysis [p.V142I (n=7), p.P44S (n=1) and p.T80A (n=1)]. By differential laser microdissection, 10 cases contained two different amyloid types: KRT5-14-type amyloid in the papillary dermis and AL-type amyloid in deep dermal and perivascular regions.

Conclusions: Cutaneous amyloidosis encompasses at least 11 different amyloid types that may be localized (AIns, KRT5-14, AEnf), systemic (ATTR, AB2MG, AA, AGel, AApoA1, AApoAIV) or both (AL, AH). Accurate determination of amyloid type is essential for patient management. Mass spectrometry, which unambiguously detects all amyloid types in a single assay, is the optimal technique for this application. KRT5-14 amyloid should be under consideration as a new canonical amyloid type related to skin.

<table>
<thead>
<tr>
<th>Type</th>
<th>Number (%)</th>
<th>Median Age</th>
<th>Gender (F/M/U)¹</th>
</tr>
</thead>
<tbody>
<tr>
<td>AL</td>
<td>505 (66.98%)</td>
<td>68</td>
<td>265/236/4</td>
</tr>
<tr>
<td>AIns</td>
<td>102 (13.53%)</td>
<td>66</td>
<td>39/63/0</td>
</tr>
<tr>
<td>KRT5-14*</td>
<td>84 (11.14%)</td>
<td>68</td>
<td>47/36/1</td>
</tr>
<tr>
<td>ATTR</td>
<td>40 (5.31%)</td>
<td>79</td>
<td>5/34/1</td>
</tr>
<tr>
<td>AH</td>
<td>8 (1.06%)</td>
<td>65</td>
<td>3/5/0</td>
</tr>
<tr>
<td>AB2MG</td>
<td>5 (0.66%)</td>
<td>70</td>
<td>3/1/1</td>
</tr>
<tr>
<td>AA</td>
<td>4 (0.53%)</td>
<td>64</td>
<td>2/1/1</td>
</tr>
<tr>
<td>AEnf</td>
<td>2 (0.27%)</td>
<td>58</td>
<td>0/1/1</td>
</tr>
<tr>
<td>AGel</td>
<td>2 (0.27%)</td>
<td>64</td>
<td>1/1/0</td>
</tr>
<tr>
<td>AApoAI</td>
<td>1 (0.13%)</td>
<td>72</td>
<td>1/0/0</td>
</tr>
<tr>
<td>AApoAIV</td>
<td>1 (0.13%)</td>
<td>71</td>
<td>1/0/0</td>
</tr>
</tbody>
</table>

Table 1: Frequency of cutaneous amyloid types. #F = female, M = male and U = unknown. *KRT5-14 = amyloid type characterized by overexpression of high molecular weight keratins 5 and 14.
NEW ORGAN RESPONSE CRITERIA FOR LIGHT CHAIN AMYLOIDOSIS: AN INTERNATIONAL VALIDATION STUDY


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Introduction: With improved treatment options for AL amyloidosis, more patients achieve hematological response, leading to a higher rate and depth of organ response. A previous study proposed that graded organ response depth criteria can provide survival discrimination.

Objective: To validate organ response depth criteria in an international collaborative study.

Methods: AL amyloidosis patients who were diagnosed between January 2010 and December 2015 and achieved at least a partial hematologic response for any line of therapy commenced within 12 months from diagnosis were included (hematologic non responders were assumed not to achieve organ responses). All patients had an assessable organ for response (heart: NT-proBNP >650 pg/mL or BNP >150 pg/mL; kidneys: >0.5 gram proteinuria/24-h; liver: alkaline phosphatase >1.5 upper limit of normal (ULN). Complete response (CR) was defined as: heart, NT-proBNP ≤350 ng/L, BNP≤80 ng/L; kidneys, proteinuria ≤200 mg/24-h in the absence of a decrease in estimated glomerular filtration rate ≥25%; and liver, serum alkaline phosphatase ≤2X institutional lower limit of normal. Very good partial response (VGPR) was defined as >60% reduction in organ parameter not meeting organ CR, partial response (PR) as 31-60% reduction in organ parameter, and as no response (NR) ≤30% reduction in organ parameter. Survival curves were plotted with the above criteria for each organ using the Kaplan-Meier method.

Results: Data from 6 centers with 579 patients are included. The median age at diagnosis was 63 years; 336 patients (58%) were male. Sixty-five percent were alive at the end of follow-up at a median follow-up of surviving patients of 72 months. Eighty percent of patients received one line of therapy in the initial 12 months from diagnosis, the most common regimen bortezomib-containing (61%). Best hematological response was CR in 37%, VGPR response in 40% and PR in 23% of patients. Three hundred and fifty four patients were evaluable for cardiac response, with a median baseline NT-proBNP of 3402 pg/mL. Overall survival (OS) was significantly associated with depth of cardiac response (5-year OS 86%, 78%, 62% and 25% for cardiac CR, VGPR, PR and NR, respectively; p<0.001). Three hundred and seventy one patients were evaluable for renal response, with a median baseline proteinuria of 5.2 g/24-h. The depth of renal response correlated with survival (5-year OS 91%, 82%, 72% and 46% for renal CR, VGPR, PR and NR, respectively; p<0.001). Hepatic response was assessable in 75 patients, in whom survival increased with a deeper hepatic response (5-year OS 100%, 88%, 46%, 43% for hepatic CR, VGPR, PR and NR, respectively; p<0.001).

Conclusions: Depth of organ response among heart, kidney and liver involvement correlates with survival. This supports a change for organ response criteria from the current response/nonresponse to a graded response system, similar to the hematological response criteria.
THE QUEST FOR INDICATORS OF PROFOUND HEMATOLOGIC RESPONSE IN AL AMYLOIDOSIS: COMPLETE RESPONSE REMAINS THE OPTIMAL GOAL OF THERAPY

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Background: The ISA defines complete response as negative serum & urine immunofixation plus a normal FLC-ratio (FLCR). A new definition of “stringent-CR” (CR and dFLC<10 mg/L) was proposed.

Objective: We compared the outcome of patients who obtained a profound reduction of FLC but do not qualify for CR with those who reached CR and we checked whether very low FLC levels identify a subgroup with better outcome among CR patients.

Methods: Our dataset was searched for newly-diagnosed patients who reached at least one the following endpoints 6 months after treatment: 1) complete response (CR) or, in patients who do not qualify for CR, 2) normalization of involved-FLC concentration (normal-iFLC), dFLC <10 mg/L (dFLC10) and normalization of FLCR (normal-FLCR). We calculated overall survival (OS) and time to next line of therapy or death (TNTD) from the time of response assessment.

Results: 434 patients (Table) were included: CR (161), normal-iFLC (114), dFLC10 (144) and normal-FLCR (220). No differences were seen between the CR and all the others in baseline variables: age, sex, organ involvement, Mayo 2004/European staging, renal staging and type of treatment. The reasons for not qualifying for CR in the normal-iFLC group was abnormal FLCR only in 4 (3%), positive s&u-IFE only in 81 (72%), and both in 29 (25%). The reason for not qualifying for CR in the dFLC10 group was abnormal FLCR only in 4 (3%), positive s&u-IFE only in 104 (72%) and both in 36 (25%). Among patients in CR 103 (64%) had normal iFLC levels and 86 (53%) had a dFLC <10 mg/L. The median follow-up of living patients was 60 months. Patients who achieved CR enjoyed a significantly longer OS (median survival not reached) compared to patients who reached any of the other endpoints but did not qualify for CR. In particular, median survival in normal-iFLC was 91 months (P=0.033), in dFLC<10 85 months (P<0.001) and in normal-FLCR 79 months (P<0.001). Also, median TNTD was significantly longer in patients who obtained CR (75 months) compared to the normal-iFLC (33 months, P<0.001), the dFLC10 (39 months, P<0.001), and the normal-FLCR (24 months, P<0.001). We then evaluated whether achieving a normal iFLC or a dFLC <10 mg/L (stringent-CR) was associated with a better survival among patients who did qualify for CR. There was no significant difference in OS and TNTD between subgroups of patients in CR according to iFLC or dFLC response. In addition, no differences were seen in frequency of cardiac (51 vs. 47%, P=0.34) and renal (51 vs. 47%, P=0.34) responses amongst patients in CR with or without the additional factors.

Conclusion: CR is associated with best survival in AL amyloidosis and should be the goal of therapy if tolerability and patient frailty allow. Higher sensitivity tools to identify residual clonal disease (mass-spectrometry on serum and urine, next generation sequencing and next generation flow cytometry on bone marrow) are needed to detect patients in CR at higher risk of relapse.

Keywords: Light Chain, Response, Survival

Table. Patients characteristics

<table>
<thead>
<tr>
<th></th>
<th>Complete Response (N=161)</th>
<th>No CR &amp; normal-iFLC (N=114)</th>
<th>No CR &amp; dFLC&lt;10 mg/L (N=144)</th>
<th>No CR &amp; normal-FLCR (N=220)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, years</td>
<td>64 (57-69)</td>
<td>64 (55-70)</td>
<td>63 (56-68)</td>
<td>63 (55-68)</td>
</tr>
<tr>
<td>Male sex</td>
<td>87 (54)</td>
<td>58 (51)</td>
<td>83 (57)</td>
<td>133 (60)</td>
</tr>
<tr>
<td>Heart, kidney</td>
<td>110 (68) / 115 (71)</td>
<td>79 (69) / 76 (67)</td>
<td>107 (74) / 103 (71)</td>
<td>165 (75) / 158 (72)</td>
</tr>
<tr>
<td>Liver, soft tissue</td>
<td>15 (9) / 24 (15)</td>
<td>9 (8) / 14 (12)</td>
<td>20 (14) / 19 (13)</td>
<td>28 (13) / 29 (13)</td>
</tr>
<tr>
<td>PNS, ANS</td>
<td>17 (11) / 11 (7)</td>
<td>12 (10) / 5 (4)</td>
<td>8 (5) / 7 (4)</td>
<td>18 (8) / 20 (9)</td>
</tr>
<tr>
<td>Cardiac stage</td>
<td>32 (20) / 78 (49)</td>
<td>30 (26) / 49 (43)</td>
<td>28 (20) / 72 (49)</td>
<td>40 (19) / 108 (48)</td>
</tr>
<tr>
<td>Renal stage</td>
<td>76 (47) / 61 (38) / 24 (15)</td>
<td>53 (46) / 54 (47) / 7 (6)</td>
<td>56 (40) / 67 (45) / 21 (15)</td>
<td>82 (37) / 104 (48) / 34 (15)</td>
</tr>
<tr>
<td>eGFR &lt;30 mL/min</td>
<td>27 (17)</td>
<td>16 (14)</td>
<td>34 (23)</td>
<td>49 (22)</td>
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<tr>
<td>Treatment type</td>
<td>MDex 48, (30) Bortezomib 89 (55)</td>
<td>MDex 26 (21) Bortezomib 77 (67)</td>
<td>MDex 28 (20) Bortezomib 93 (64)</td>
<td>MDex 58 (26) Bortezomib 122 (55)</td>
</tr>
<tr>
<td>Cardiac response</td>
<td>50 (50)</td>
<td>29 (44)</td>
<td>30 (35)</td>
<td>39 (29)</td>
</tr>
<tr>
<td>Renal response</td>
<td>54 (53)</td>
<td>45 (47)</td>
<td>44 (47)</td>
<td>63 (42)</td>
</tr>
</tbody>
</table>
MINIMAL RESIDUAL DISEASE POSITIVITY BY MULTIPARAMETER FLOW CYTOMETRY HINDERS ORGAN INVOLVEMENT RECOVERY IN AL AMYLOIDOSIS PATIENTS IN COMPLETE RESPONSE

Introduction: In multiple myeloma, next generation flow cytometry (NGF) is used to detect minimal residual disease (MRD). This technique offers robust endpoints for clinical trials, as well as for optimizing individual patient treatment. The evaluation of the role of MRD in light chain (AL) amyloidosis is an emerging area of interest.

Objectives: we searched MRD by NGF in 108 patients with AL amyloidosis who were known to be in complete response (CR) and we assessed the prognostic impact of MRD status.

Methods: CR was defined as per current criteria (negative serum and urine immunofixation and normal free light chain ratio). Bone marrow samples were processed following the Euro Flow Bulk Lysis Standard Operating Protocol and stained with the EuroFlowIMF MM MRD panel. A median number of 5x10⁶ events were acquired (range 5×10⁶ – 7×10⁶) using a FACSCanto II instrument. Data were analyzed using the Infinicyt software by an operator blind to clinical data. A discreet population of clonal plasma cells comprising ≥50 events defined MRD positivity (0.001% limit of detection, i.e. 10⁻⁵ sensitivity).

Results: One hundred and eight patients were tested (16 were found to have relapsed at the time of MRD assessment with monoclonal components detectable and MRD+) and 92 satisfied current criteria for CR. Their clinical characteristics are reported in the Table. Forty-two patients (46%) had no detectable MRD. In the 50 patients with MRD, a median of 447 PCs with abnormal phenotype (range 54-3581), corresponding to 0.02% (range 0.01-0.3%), were detected. The LC restriction defined the PC clonality and corresponded to the expected pattern of clonal isotype expression based on the type of monoclonal protein in all cases. There was no significant difference in clinical variables measured at baseline in patients with and without detectable MRD except for lower dFLC at the time of CR achievement in the MRD negative group. We compared organ involvement data at the time CR was first recorded and at the time of MRD assessment to detect organ responses occurring after achievement of CR. Patients without detectable MRD were more likely to attain renal [92% (23/25 evaluable) vs. 57% (15/26 evaluable), P=0.005] and cardiac response [95% (18/19 evaluable) vs. 71% (20/28 evaluable), P=0.046]. After a median follow-up of living patients of 23 months from the time of MRD assessment, 3 patients, all with persistent MRD died. Time to hematologic progression was significantly longer in MRD-negative patients: only 1 patient without detectable MRD progressed, compared to 13 in MRD positive patients

Conclusion: In conclusion, NGF can effectively detect MRD in AL amyloidosis patients otherwise in CR and persistent MRD may explain persistent organ dysfunction in these patients. In subjects who attain CR, testing for MRD should be offered, especially if CR is not accompanied by organ response, and in case MRD is present, further chemotherapy could be considered, carefully balancing residual organ damage, patient frailty, and possible toxicity.

Keywords: Amyloidosis, Response, Prognosis
Table 1. Patients characteristics (total number of patients N=92)

<table>
<thead>
<tr>
<th>Variable</th>
<th>MRD positive (N=50) N (%) / median (IQR)</th>
<th>MRD negative (N=42) N (%) / median (IQR)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male sex</td>
<td>33 (66) / 55-66</td>
<td>21 (50) / 55-68</td>
<td>0.128</td>
</tr>
<tr>
<td>Age at diagnosis, years</td>
<td>59 (55-66)</td>
<td>61 (55-68)</td>
<td>0.280</td>
</tr>
<tr>
<td>Organ involvement at diagnosis</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>heart</td>
<td>36 (67) / 55-66</td>
<td>23 (54) / 55-68</td>
<td>0.093</td>
</tr>
<tr>
<td>kidney</td>
<td>30 (60) / 55-66</td>
<td>31 (73) / 55-68</td>
<td>0.172</td>
</tr>
<tr>
<td>liver</td>
<td>11 (22) / 55-66</td>
<td>4 (9) / 55-68</td>
<td>0.117</td>
</tr>
<tr>
<td>Cardiac stage at diagnosis</td>
<td>(available in N=45)</td>
<td>(available in N=40)</td>
<td></td>
</tr>
<tr>
<td>I</td>
<td>11 (24)</td>
<td>17 (43)</td>
<td>0.084</td>
</tr>
<tr>
<td>II</td>
<td>18 (40)</td>
<td>12 (30)</td>
<td>0.341</td>
</tr>
<tr>
<td>III</td>
<td>15 (34)</td>
<td>9 (22)</td>
<td>0.227</td>
</tr>
<tr>
<td>IIIb</td>
<td>1 (2)</td>
<td>2 (5)</td>
<td>0.582</td>
</tr>
<tr>
<td>Renal stage at diagnosis</td>
<td>(available in N=41)</td>
<td>(available in N=39)</td>
<td></td>
</tr>
<tr>
<td>I</td>
<td>20 (49)</td>
<td>18 (46)</td>
<td>0.818</td>
</tr>
<tr>
<td>II</td>
<td>18 (44)</td>
<td>18 (46)</td>
<td>0.843</td>
</tr>
<tr>
<td>III</td>
<td>3 (7)</td>
<td>3 (8)</td>
<td>0.951</td>
</tr>
<tr>
<td>eGFR, mL/min per 1.73 m²</td>
<td>86 (61-90)</td>
<td>76 (60-89)</td>
<td>0.158</td>
</tr>
<tr>
<td>BMPC at diagnosis (%)</td>
<td>8 (4-14)</td>
<td>9 (4-15)</td>
<td>0.865</td>
</tr>
<tr>
<td>dFLC at diagnosis, mg/L</td>
<td>141 (65-488)</td>
<td>112 (34-397)</td>
<td>0.303</td>
</tr>
<tr>
<td>dFLC at aCR first documentation, mg/L</td>
<td>6.4 (2-16)</td>
<td>1.4 (0-5)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Light chain only</td>
<td>30 (60)</td>
<td>19 (45)</td>
<td>0.166</td>
</tr>
<tr>
<td>Exposure to 2 lines of therapy before first aCR documentation</td>
<td>25 (50)</td>
<td>15 (36)</td>
<td>0.177</td>
</tr>
<tr>
<td>Time from diagnosis to first aCR documentation, months</td>
<td>9.6 (6-15)</td>
<td>11 (5-19)</td>
<td>0.823</td>
</tr>
<tr>
<td>Time from first aCR documentation to MRD assessment, months</td>
<td>13 (5-30)</td>
<td>12 (6-47)</td>
<td>0.368</td>
</tr>
<tr>
<td>Autologous stem cell transplant</td>
<td>19 (38)</td>
<td>16 (38)</td>
<td>0.991</td>
</tr>
<tr>
<td>Melphalan</td>
<td>23 (46)</td>
<td>14 (33)</td>
<td>0.227</td>
</tr>
<tr>
<td>Bortezomib</td>
<td>45 (90)</td>
<td>36 (86)</td>
<td>0.547</td>
</tr>
<tr>
<td>Lenalidomide</td>
<td>12 (24)</td>
<td>3 (7)</td>
<td>0.031</td>
</tr>
<tr>
<td>Cardiac response at the time aCR was documented</td>
<td>16/29 (55)</td>
<td>15/21 (71)</td>
<td>0.262</td>
</tr>
<tr>
<td>Renal response at the time aCR was documented</td>
<td>12/29 (52)</td>
<td>19/31 (61)</td>
<td>0.470</td>
</tr>
</tbody>
</table>
IN SYSTEMIC LIGHT-CHAIN AMYLOIDOSIS THE BEST HEMATOLOGIC RESPONSE FOR LONG-TERM SURVIVAL IS iFLC < 10mg/L

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Background: Hematologic response in light-chain amyloidosis (AL) is evaluated by use of free light chain (FLC) assay, employing the difference (dFLC) between involved (iFLC) and uninvolved FLC and the FLC ratio but not the depth of change in the pathologic iFLC; emerging data have controverted the definitions of both the amyloid complete response (aCR) and the very good partial response (VGPR) (AJH 2005;79:319; JCO 2012;30:4541). Combination therapy with daratumumab in newly diagnosed patients achieves dFLC levels <10mg/L in over 70% of cases and often makes the FLC ratio incalculable (Abs #S875 24th EHA Congress, 6/15/19). We therefore examined overall survival (OS) in patients achieving ≥ VGPR in relation to disease- and treatment-related variables.

Methods: Patients diagnosed with AL between 2005-2017 achieving ≥ VGPR after treatment were included in this IRB-approved retrospective study. Cox proportional-hazards regression analyses were used to assess the impact on OS of baseline variables (gender, age, iFLC, dFLC, cardiac and renal stage), and treatment-related variables (receiving stem cell transplant (SCT), aCR, VGPR, iFLC and dFLC at best response). iFLC categories at best response were defined as <10, 10-20 or >20mg/L. MedCalc was used for all statistical analyses.

Results: The 133 patients eligible had a median age of 60.5 years (range, 35-81); 78 were men. AL, λ-type, was present in 114 (86%). Median bone marrow plasma cells were 10% (1-50) and iFLC and dFLC were 135mg/L (29.4-9780) and 123mg/L (4-9770); 89 patients (66%) had cardiac (stage 2=49, 3=33) and 108 (81%) renal involvement (stage 2=52, 3=19). Of baseline variables, only age and cardiac stage 3 significantly predicted OS.

Ninety one patients (68%) had bortezomib-based initial therapy; 63 (47%) received SCT and consolidation. Overall 79 (59%) and 54 (41%) achieved an aCR or VGPR respectively. With a median follow-up of 63 months, iFLC best response <10mg/L had over 95% survival at 120 months, while those achieving iFLC best response 10-20mg/L or >20mg/L had median OS of 96 and 121 months (p<0.01). Patients in the iFLC response groups differed only in baseline iFLC, dFLC, cardiac and renal stage. With treatment they differed in the utilization of SCT and in aCR rates (Table 1). In the iFLC <10mg/L group, 25 patients achieved iFLC<10mg/L in a median of 4 months (1-9) and 17 in a median of 31 months (12-80).

Conclusions: Advanced age and stage 3 cardiac involvement negatively impacted OS in patients achieving ≥ VGPR to therapy. Achieving an iFLC <10mg/L predicted markedly improved OS. These data justify the practice of changing therapy early in those with sub-optimal hematologic responses and also indicate that further studies are needed to assess hematologic response and long-term survival in AL in the era of monoclonal antibody therapy.

Table 1. variables Affecting iFLC Response

<table>
<thead>
<tr>
<th>Variables</th>
<th>Post-rt iFLC &lt;10 (N=42)</th>
<th>Post-rt iFLC= 10-20 (N=37)</th>
<th>Post-rt iFLC &gt;20 (N=54)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline at diagnosis</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cardiac stage 0/1/2/3</td>
<td>24/1/10/7</td>
<td>7/5/15/10</td>
<td>13/1/24/16</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Cardiac stage 0/1/2/3</td>
<td>11/12/17/2</td>
<td>12/6/12/7</td>
<td>2/19/23/10</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>iFLC, median (range)</td>
<td>75 (21-4325)</td>
<td>198 (50-1650)</td>
<td>250 (39-9780)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>dFLC, median (range)</td>
<td>62 (6-4320)</td>
<td>182 (33-1670)</td>
<td>192 (4-9770)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Treatment</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Underwent SCT (%)</td>
<td>28 (67)</td>
<td>16 (43)</td>
<td>19 (35)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>dFLC&lt;10mg/L at best response (%)</td>
<td>41 (98)</td>
<td>28 (76)</td>
<td>11 (20)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>aCR rates (%)</td>
<td>34 (81)</td>
<td>25 (68)</td>
<td>20 (37)</td>
<td>&lt;0.01</td>
</tr>
</tbody>
</table>
COMPARISON OF MEASURES OF COMPLETE HEMATOLOGIC RESPONSE AFTER HIGH DOSE MELPHALAN AND AUTOLOGOUS STEM CELL TRANSPLANTATION FOR AL AMYLOIDOSIS

S. Sarosiek 1, J. M. Sloan 1, K. Quillen 1, D. Brauneis 1, V. Sanchorawala 1

1 Boston University.

Introduction: The criteria for hematologic response in AL amyloidosis currently defines a complete response (CR) as negative serum and urine immunofixation (IFE) and normal serum free light chain ratio (FLCR), very good partial response (VGPR) as a difference in involved to uninvolved serum free light chains (dFLC) <40mg/L, and partial response (PR) as a decrease in the dFLC by >50% compared to baseline. Recent literature suggests that a normal involved free light chain (iFLC), iFLC <20mg/L, or low difference in free light chains (dFLC) may more accurately predict outcomes in patients with AL amyloidosis.

Objective: To evaluate overall survival (OS) in patients achieving a hematologic CR (hemCR) after HDM/SCT compared to those who did not meet criteria for CR, but achieved a normal iFLC, an iFLC <20mg/L, a dFLC <10mg/L, or a normal free light chain ratio (FLCR).

METHODS: Patients who were treated with high dose melphalan and autologous stem cell transplantation (HDM/SCT) between January 30, 2003 and December 1, 2017 were included in this retrospective review. Kaplan-Meier survival curves were generated to compare OS between patients with hemCR by ISA criteria compared with those not achieving a CR but with a normal iFLC, iFLC <20mg/L, dFLC <10mg/L, or normal FLCR.

Results: During the specified 15 years, 327 patients with AL amyloidosis received HDM/SCT, of which 35 (11%) were excluded due to death prior to 6 month assessment after SCT or lack of follow-up. Of those evaluable at 6 months (n=292), the hematologic responses were as follows: 85 (29%) CR, 129 (44%) very good partial response, 33 (11%) partial response, and 45 (15%) with no response/progression of disease. Of the total cohort (n=292) the number of patients achieving stringent hematologic responses of dFLC <10mg/L, normal iFLC, iFLC <20mg/L, and normal FLCR were 126 (43%), 141 (48%), 114 (39%), and 168 (58%), respectively. Of those patients without a CR (n=207), 78 patients had a normal iFLC, 61 had an iFLC <20mg/L, 63 patients had a dFLC <10mg/L, and 83 had a normal FLCR. Those with normal iFLC, dFLC <10mg/L, and normal FLCR in the absence of a hemCR had worse OS compared with those in hemCR (p=0.0165, 0.0057, and 0.0004, respectively). Those with iFLC <20mg/L had OS not significantly different than those with hemCR (p=0.1533). Of patients with a hemCR, OS was not significantly improved when comparing those with dFLC <10 v. ≥10 (p=0.6826) or those with normal iFLC v. elevated iFLC (p=0.0793). OS was significantly improved when iFLC <20mg/L was achieved with a hemCR (p=0.0264).

Conclusions: Serum and urine IFE negativity remain an important part of the hematologic response criteria as evidenced by significantly worse survival of those patients achieving only dFLC<10mg/L, normal iFLC, or normal FLCR with continued IFE positivity. OS is similar in patients with hemCR and iFLC <20mg/L and those meeting both iFLC <20mg/L and hemCR criteria have superior OS.

Keywords: Complete Response (CR), Involved Free Light Chains (iFLC), Low Difference In Free Light

<table>
<thead>
<tr>
<th>Total cohort (n=292)</th>
<th>Not meeting ISA hemCR criteria (n=207)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total patients, n (%)</td>
<td>85 (29)</td>
</tr>
<tr>
<td>Female, n(%)</td>
<td>35 (41)</td>
</tr>
<tr>
<td>Median age at transplant, y (range)</td>
<td>56 (26-70)</td>
</tr>
<tr>
<td>dFLC prior at transplant, median (range)</td>
<td>60 (0.2-2282)</td>
</tr>
<tr>
<td>Renal involvement, n (%)</td>
<td>70 (82)</td>
</tr>
<tr>
<td>Baseline renal stage, median</td>
<td>2</td>
</tr>
<tr>
<td>Cardiac involvement, n (%)</td>
<td>44 (52)</td>
</tr>
<tr>
<td>Baseline cardiac stage, median</td>
<td>2</td>
</tr>
</tbody>
</table>
THE IMPACT AND IMPORTANCE OF POST-RENAL TRANSPLANTATION HAEMATOLOGICAL RESPONSE ASSESSMENT IN AL AMYLOIDOSIS

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Background and Methods: In AL amyloidosis, patients with renal involvement have a 19-42% risk of progression to end-stage renal failure (ESRF), which is associated with a polyclonal free light chain (FLC) rise making quantification of the monoclonal element of the involved free light chain (iFLC) difficult. When renal transplantation is considered, accurate assessment of haematological response (HR) is critical to minimise the risk of amyloid accumulation within the allograft.

We assessed FLCs in 40 patients who underwent renal transplantation at the UK National Amyloidosis Centre, 2004-2019.

Results: Baseline characteristics were: age median 53.5 years (range 38-69 years), 60% male, median ECOG 1 (0-3), 70% lambda restricted. Extra-renal organ involvement was: 40% cardiac, 52.5% liver, 7.5% peripheral nerve, 12.5% autonomic nerve, 7.5% soft tissue. Median time from haematological response to renal transplantation was 29 months (0-93 months). At transplantation, haematological responses were: complete response (CR) – 24 (60.0%), very good partial response (VGPR) – 6 (15.0%), partial response (PR) – 6 (15.0%), no response (NR) – 3 (7.5%) and 1 non-evaluable. Post-transplant, 7 patients (17.5%) had their haematological response re-classified of which 6/7 (85.7%) were assigned an improved response. A pre-transplant CR did result in an improved PFS (p=0.024) and OS (p=0.015). However, reclassification of HR after renal transplantation, once the polyclonal element of the light chain excess was largely removed, improved significance (p=0.0001). Furthermore, percentage and absolute difference between free light chains (dFLC) and iFLC were only predictive post-transplant. A dFLC 10mg/L (p=0.0001). Other than HR, only cardiac involvement predicted survival based on univariate analysis (p=0.01).

In patients with renal failure, only a CR is predictive of survival based on pre-renal transplant light chain assessment. However, some patients will be misclassified by this method and, as such, post-transplant FLC assessment is essential. Of note, a wide normal range (0.37-3.1) has been suggested for patients with ESRF, which if incorporated into HR criteria in this cohort, removed any reclassification of HR post-renal transplant. This method requires study in a larger group of patients in order to validate its use.

Conclusions: In summary, patient survival following renal transplantation is dictated by HR. The pretransplant assessment may be best assigned using a previously published new normal range for patients with ESRF but post-transplant assessment is critical for prognosis. Cardiac involvement negatively impacts survival and highlights the importance of careful patient selection.

Keywords: amyloidosis, transplantation, renal.
PREVALENCE AND SURVIVAL IMPACT OF ATRIAL FIBRILLATION IN PATIENTS WITH TRANSTHYRETIN CARDIAC AMYLOIDOSIS. ANALYSIS FROM A LARGE INTERNATIONAL COHORT

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¹ Hospital Universitario Puerta de Hierro (Madrid, Spain).
² National Amyloid Center (London, UK).
³ Columbia University Hospital (New York, US).
⁴ University of Bologna (Italy).
⁵ University of Pavia (Italy).

Background: Atrial fibrillation (AF) is the most common arrhythmia found in cardiac transthyretin (ATTR) amyloidosis. Prevalence and incidence data of AF in ATTR cardiac amyloidosis patients are scarce and derived mainly from small and mostly single-center cohorts. Moreover, it is unknown how the development of AF can modify prognosis in ATTR.

Objectives: We sought to describe prevalence and incidence of AF and to evaluate its influence in survival in a large multicenter international cohort of patients with both wild-type ATTR (ATTRwt) and hereditary ATTR (ATTRh) amyloid cardiomyopathy.

Methods: Clinical characteristics at initial evaluation and survival data from cardiac TTR amyloidosis patients evaluated at 5 international amyloid centers: Columbia University Hospital (New York, US), University of Bologna (Italy), University of Pavia (Italy), National Amyloid Center (London, UK) and Hospital Puerta de Hierro (Madrid, Spain) were retrospectively collected. Presence of AF at baseline and its appearance during follow-up was captured. We performed survival analysis and multivariant analysis to evaluate the impact of AF at baseline on prognosis and its interactions with other predictors associated with death.

Results: 1157 patients with cardiac ATTR amyloidosis (87.4% male, mean age 75.78.6 years, 83% ATTRwt and 17% ATTRh) were included in this analysis. Prevalence of atrial fibrillation (AF) at baseline was 49.6% (CI95% 46.7-52.5%). During a median follow-up of 23.2 months (IQR: 12.2-39.2), 187 patients (16.2%, 29.8% of those who initially were in sinus rhythm) developed AF and 379 (32.8%) remained in sinus rhythm. Median time from initial evaluation to AF appearance in those who developed AF was 17.4 months (IQR: 7.2-31.7). AF presence at baseline was associated with lower survival (HR 1.53; CI95%: 1.24-1.89). Survival rate was also lower among patients with AF than in patients who developed AF during follow-up (HR 0.56; CI 0.42-0.75), and those who remained in sinus rhythm (HR 0.69; CI 0.54-0.89). Survival rates of patients who developed AF during follow-up and those who maintained sinus rhythm were similar. In a multivariant analysis, including all variables associated to death at univariate analysis, only age, diabetes, Nt-proBNP, LVEF and renal function (eGFR) remained in the model. When adjusting for these variables, presence of AF at baseline evaluation was not statistically associated with lower survival (HR 1.23 CI95%: 0.94-1.63).

Conclusions: AF is very common in ATTR cardiac amyloidosis. Almost half of patients with ATTR cardiac amyloidosis show this arrhythmia at initial evaluation and one third of those on sinus rhythm develop it during follow-up. Presence of AF at initial evaluation is associated with decreased survival but its impact is lost when adjusted for other prognostic factors.
OP28

IMPACT ON SURVIVAL OF N-TERMINAL PRO-B-TYPE NATRIURETIC PEPTIDE (NT-proBNP) INCREASE AFTER DIAGNOSIS FOR CARDIAC TRANSTHYRETIN AMYLOIDOSIS

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1 Referral Center for Cardiac Amyloidosis, Mondor Amyloidosis Network, Henri Mondor Teaching Hospital, Créteil, France.
2 EXYSTAT, Malakoff, France.

Introduction/Background: Cardiac transthyretin (ATTR) amyloidosis is an increasingly recognized, progressive, and fatal cardiomyopathy, for which the natural history remains unclear.

Objectives: Aim1 was to assess the impact of NT-proBNP changes on mortality in amyloid cardiomyopathy untreated patients. Aim2 was to assess the NT-proBNP changes before and after treatment with Tafamidis.

Methods: Data from 648 consecutive patients included between 2007 and 2019 from an ongoing monocentric, longitudinal, observational registry on the natural history of ATTR at the Henri Mondor Teaching Hospital were used. For aim 1, patients with at least 2 NT-proBNP available values (N=401) were included. NT-proBNP initial values were categorized in four groups according to quartiles values. Patients were stratified according to the quartile increase (group A) or not increase (Group B). Event of interest was the first observed event between death, decompensation or heart transplant. Patients were censored if they started any ATTR treatment or if no event occurred at the end of the follow up or at the cut-off date (31OCT2019). Cox proportional hazards regression model was used to calculate the hazard-ratio (HR) and log-rank test to compare the two survival curves. Crude and adjusted analyses on NYHA Class, ATTR variant, NAC-ATTR stage and Strain baseline were performed.

For aim 2, 265 patients treated with Tafamidis were considered, having at least 2 NT-proBNP values available (before and under treatment) in order to document NT-proBNP evolution. Slopes before and under treatment were evaluated and compared using a regression model including time*treatment interaction.

Results: For aim 1, 400 patients were included, with a mean age of 77 years, 83% Male, and a median follow up of 12.8 Months [11.1; 14.4] and 158 (39% maturity) events were observed. Median time to event among cohort A (n=95) was 23.5 months [17.2; 28.8] and 31.6 [25.1; 38.4] months for cohort B (n=305) (HR: 0.65, 95% Confidence Interval (CI) [0.47; 0.91], P=0.01). In adjusted model, HR[95%CI] was 0.53 [0.35, 0.81], P<0.01. Median time to death among cohort A was 31.5 months [26.2; 39.8] and 52.3 [37.5; 71.1] months for cohort B (HR[95%CI] 0.65 [0.43; 0.98], P=0.04; 100 events; 25% maturity).

For aim 2, 265 patients were included and treated with Tafamidis, with a mean age of 74 years, 80% Male, and a median follow up of 20.5 Months [16.1; 23.9]. NT-proBNP increased before treatment, (slope positive p=0.005) and decreased after treatment (slope negative p<0.0001 and p<0.0001 for the 2 slopes comparison). The effect increased with time under treatment.

Conclusion: In this large database study of patients with ATTR, increase of NT-proBNP is an independent predictor of survival for untreated patients. For Tafamidis treated patients, this NT-proBNP increase was reversed under treatment.

Keywords: Amyloidosis • NT-proBNP • Tafamidis
DIAGNOSTIC VALUE OF SUBCUTANEOUS ABDOMINAL FAT TISSUE ASPIRATES IN CARDIAC AMYLOIDOSIS

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1 University Medical Center Groningen.

Background: Endomyocardial biopsy is the gold-standard for detection of cardiac amyloidosis (CA), however it has a potential risk of serious complications and requires specialist technical expertise and equipment. Abdominal subcutaneous fat tissue aspiration (ASFTA) is simple, safe, and inexpensive, though profoundly divergent sensitivities (14-90%) have been reported [1,2,3].

Objectives: To assess the diagnostic value of routinely performed ASFTA in patients with cardiac amyloidosis at our center and to reconcile between-center discrepancies.

Methods: The diagnostic value of ASFTA was assessed in 248 consecutive patients with CA, of whom 100 had amyloid light-chain (AL), 82 hereditary transthyretin (ATTRv), and 66 wild-type transthyretin (ATTRwt) amyloidosis. The diagnosis of CA was either established by the presence of both otherwise unexplained increased left (≥12 mm) or right (≥7 mm) ventricular wall thickness on transthoracic echocardiography or cardiac magnetic resonance imaging and histological confirmation of the presence and type of amyloid, or according to non-histological diagnostic criteria in ATTR amyloidosis [4]. All patients with ATTR amyloidosis underwent sequencing of the transthyretin (TTR) gene. Fat tissue aspirates in ATTRv were obtained within the same timeframe that CA was confirmed. Congo red-stained tissue was assessed in three smears by two independent observers and semi-quantitatively scored 0 through 4 according to our center’s standard procedure [3].

Results: Amyloid deposition in Congo red-stained ASFTAs was detected in 99/100 (99%) patients with AL, 75/82 (91%) with ATTRv, and 44/66 (67%) with ATTRwt amyloidosis. The number of fat smears of patients with AL, ATTRv, and ATTRwt semi-quantitatively judged 1+ (less than 1% of inspected area) were 7/100 (7%), 13/82 (16%), and 29/66 (44%), respectively, leaving 92/100 (92%), 62/82 (76%), and 15/66 (23%) smears graded 2+ to 4+, respectively. In patients with ATTRwt, no relation was found between the Congo red status of ASFTAs (negative/positive) and age, gender or cardiac bone tracer uptake (Perugini score).

Conclusion: Aspiration of abdominal subcutaneous fat tissue has a good diagnostic sensitivity in cardiac AL and ATTRv amyloidosis, and a reasonable sensitivity in ATTRwt. By considering semi-quantitatively judged 1+ fat smears as negative, we approximated the sensitivity reported by Fine et al., raising the suggestion that thorough assessment by two independent observers should become rule rather than exception in exposing elusive amyloid deposits, which may otherwise remain concealed.

<table>
<thead>
<tr>
<th>Table 1. Characteristics of the 248 patients with cardiac amyloidosis</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>No. of participants</strong></td>
</tr>
<tr>
<td>---------------------------------------------------------------</td>
</tr>
<tr>
<td>248</td>
</tr>
<tr>
<td><strong>Sociodemographics</strong></td>
</tr>
<tr>
<td>Age, years</td>
</tr>
<tr>
<td>Males</td>
</tr>
<tr>
<td><strong>Fat aspiration biopsy</strong></td>
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<tr>
<td>Positive</td>
</tr>
<tr>
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</tr>
<tr>
<td>0</td>
</tr>
<tr>
<td>1</td>
</tr>
<tr>
<td>2</td>
</tr>
<tr>
<td>3</td>
</tr>
<tr>
<td>4</td>
</tr>
</tbody>
</table>

Abbreviations: AL, amyloid light-chain. ATTRv, hereditary transthyretin. ATTRwt, wild-type transthyretin.
Data are mean (SD), number (percentage).
OP30

DESCRIBING THE ECHOCARDIOGRAPHIC PHENOTYPE OF TRANSTHYRETIN CARDIAC AMYLOIDOSIS- WHAT ARE THE PREDICTORS OF PROGNOSIS?

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2 Tuscan Regional Amyloid Center, Careggi University Hospital, Florence, Italy.
3 Heart Failure Unit, Cardiology University Department, IRCCS Policlinico San Donato, Milan, Italy.
4 Barts Heart Centre, St Bartholomew’s Hospital, London, UK.

Background: Transthyretin amyloidosis cardiomyopathy (ATTR-CM) is an increasingly recognised, progressive and ultimately fatal cause of heart failure.

Objectives: We sought to characterize the structural and functional echocardiographic phenotype and their independent association with prognosis across the spectrum of wild-type (wt-ATTR-CM) and hereditary transthyretin cardiomyopathy (hATTR-CM).

Methods: Patients referred to the National Amyloidosis Centre, United Kingdom between 2000 and 2019 in whom a diagnosis of ATTR-CM was confirmed were invited to participate in a prospective clinical follow up program evaluating cardiac parameters and survival. The diagnosis of ATTR-CM was established on the basis of heart failure symptoms together with a characteristic amyloid echocardiogram and either direct endomyocardial biopsy proof of ATTR amyloid, or presence of ATTR amyloid in an extra-cardiac biopsy along with cardiac uptake on 99mTc-DPD scintigraphy, or Perugini grade 2 or 3 cardiac uptake on 99mTc-DPD scintigraphy in the absence of an abnormal serum free light chain ratio and monoclonal immunoglobulin in the serum and urine by immunofixation.

Results: We studied 1240 patients with ATTR-CM comprising 766 with wtATTR-CM and 474 with hATTR-CM, of whom 314 had the V122I variant and 127 the T60A variant. At diagnosis, patients with V122I-hATTR-CM had the most severe degree of systolic and diastolic dysfunction across all echocardiographic parameters and patients with T60A-hATTR-CM the least; patients with wtATTR-CM had intermediate features. Stroke volume index, right atrial area index, longitudinal strain, and E/e’ were all independently associated with mortality (p<0.05 for all). Severe aortic stenosis was also independently associated with prognosis, conferring a significantly shorter survival (median survival 22 versus 53 months, p=0.001).

Conclusions: Our findings suggest that ATTR-CM, traditionally considered to be predominantly a disease of diastolic heart failure is characterised by a complex pathophysiology as demonstrated by echocardiography, with varying degrees of severity across the spectrum of ATTR-CM.

Keywords: Amyloidosis, Transthyretin, Echocardiography
CARDIAC TRANSTHYRETIN WYLD TYPE AMYLOIDOSIS (ATTRwt): A PROSPECTIVE STUDY OF 400 PATIENTS FOLLOWED AT THE ITALIAN REFERRAL CENTER

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**Background:** Cardiac wild type transthyretin (ATTRwt) amyloidosis, formerly known as senile systemic amyloidosis, is an increasingly recognized, progressive, and fatal cardiomyopathy. Two biomarkers staging systems were proposed based on NT-proBNP (in both cases) and cardiac troponin or estimated glomerular filtration rate that are able to predict survival in this population. The availability of novel effective treatments requires large studies to describe the natural history of the disease in different populations.

**Objective:** To describe the natural history of the disease in a large, prospective, national series.

**Methods.** Starting in 2007, we protocolized data collection in all the patients diagnosed at our center (n=400 up to 7/2019).

**Results:** The referrals to our center increased over time: 5 cases (1%) between 2007-2009, 33 (9%) in 2010-2012, 90 (22%) in 2013-2015 and 272 (68%) in 2016-2019. Median age was 76 years [interquartile range (IQR): 71-80 years] and 372 patients (93%) were males. One hundred and seventy-three (43%) had atrial fibrillation, 63 (15%) had a history of ischemic cardiomyopathy and 64 (15%) underwent pacemaker or ICD implantation. NYHA class was I in 58 subjects (16%), II in 225 (63%) and III in 74 (21%). Median NT-proBNP was 3064 ng/L (IQR: 1817-5579 ng/L), troponin I 0.096 ng/mL (IQR: 0.063-0.158 ng/mL), eGFR 62 mL/min (IQR: 50-78 mL/min). Median IVS was 17 mm (IQR: 15-19 mm), PW 16 mm (IQR: 14-18 mm) and EF 53% (IQR: 45-57%). One-hundred and forty-eight subjects (37%) had a concomitant monoclonal component in serum and/or urine and/or an abnormal free light chain ratio. In these patients, the diagnosis was confirmed by immunoelectron microscopy or mass spectrometry. In 252 (63%) the diagnosis was based on bone scintigraphy. DNA analysis for amyloidogenic mutations in transthyretin and apolipoprotein A-I genes was negative in all subjects. The median survival of the whole cohort was 59 months. The Mayo Clinic staging based on NT-proBNP (cutoff: 3000 ng/L) and troponin I (cutoff: 0.1 ng/mL) discriminated 3 different groups [stage I: 131 (35%), stage II: 123 (32%) and stage III: 127 (33%)] with different survival between stage I and II (median 86 vs. 81 months, P=0.04) and between stage II and III (median 81 vs. 62 months, P<0.001). The UK staging system (NT-proBNP 3000 ng/L and eGFR 45 mL/min), discriminated three groups [stage I: 170 (45%), stage II: 165 (43%) and stage III: 45 (12%)] with a significant difference in survival: between stage I and stage II (86 vs. 52 months, P<0.001) and between stage II and stage III (median survival 52 vs. 33 months, P=0.045).

**Conclusions:** This is one of the largest series of patients with cardiac ATTRwt reported so far. The diagnoses increased exponentially in recent years, One-third of patients has a concomitant monoclonal gammopathy and needed tissue typing. Both the current staging systems offered good discrimination of staging and were both validated in our independent cohort.

**Keywords:** Transthyretin, Survival, Biomarkers
REGIONAL CARDIAC UPTAKE OF 99-TC-DPD IS A NOVEL POWERFUL AND INDEPENDENT PROGNOSTIC MARKER IN CARDIAC ATTR WILD TYPE AMYLOIDOSIS

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2 Nuclear Imaging Unit, IRCCS Policlinico San Matteo, Pavia, Italy.
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Background: Skeletal scintigraphy with bone tracers is a key tool for cardiac ATTR diagnosis. However, the prognostic value of this method has not been systematically assessed.

Objectives: We evaluated the prognostic relevance of a quantitative method to assess regional 99mTc-DPD uptake by SPECT in the heart of patients with ATTRwt.

Methods: all ATTRwt patients (n=229) who underwent clinical assessment and bone scintigraphy at our center (from 2012 to 2019). Patients received approximately 700 MBq of 99mTc-DPD. Planar whole body acquisition 10’ after the injection followed by cardiac SPECT after 3 hours was performed. SPECT data were reconstructed into 64x64 matrices with an ordered-subset expectation maximization algorithm on a GE Xeleris workstation. For each wall and for the apex, a circular region of interest (ROI) of twenty pixels each was manually drawn and a value equating to the number of counts contained in the ROI was obtained. Partial correlation of ln-transformed ROI and biomarkers was retrieved from a multivariable regression model, while controlling for cardiac wall. Multivariable Cox regression was used to assess the prognostic role of lnROI while adjusting for wall, NT-proBNP, cTnI and eGFR. Hazard ratios and 95% confidence intervals (HR, 95%CI) were computed. The Harrell’s c statistic was reported for model discrimination. The interaction of biomarker and wall on survival was assessed; also, to account for intra-subject correlation of measures, within subject robust standard errors were computed.

Results: Median follow-up was 21 months (IQR 11, 40) and 39 (17%) patients died. Median age was 76 years (IQR, 72-80), NT-proBNP 2944 ng/L (IQR, 1815-5319), cTnI 0.095 ng/L (IQR, 0.062-0.144) and eGFR 62 mL/min (IQR, 51-77). ROI did not correlate with any of NT-proBNP, eGFR, age, cTnI or mLVWT (R<1% in all cases). All analyses were adjusted for cardiac wall. At the multivariable Cox regression, there was a linear increase in the risk of death associated with lnROI, which was independent of cardiac wall, NT-proBNP, cTnI and eGFR. Only cTnI maintained a significant prognostic value (Table). The association of lnROI and mortality was not modified by the site of measurement (test for interaction with cardiac wall p=0.818). At the predefined subgroup analysis, the risk of death was similar for all walls; we computed the optimal cut-off for 12 months survival at the apex (a region usually lately involved) to 4193 (AUC: 0.68, sensitivity 80%, specificity 68%). At the multivariable Cox regression, apex ROI>4193 was an independent predictor of death (HR 3.60, 95%CI 1.45-8.93, p=0.006) and outperformed all the biomarkers tested.

Conclusions: The quantitative assessment of ROI uptake at cardiac SPECT is a powerful predictor of survival in ATTRwt patients, independent of and outperforming the other known prognostic factors. This observation warrants validation with prolonged follow-up and in independent series.

Keywords: Transthyretin, Prognosis, Scintigraphy

Table. Cox proportional hazard multivariate model.

<table>
<thead>
<tr>
<th>Variable</th>
<th>HR (95%, CI)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>lnROI</td>
<td>2.14 (1.16-3.94)</td>
<td>0.014</td>
</tr>
<tr>
<td>NT-proBNP&gt;3000 ng/L</td>
<td>1.72 (0.90-3.27)</td>
<td>0.101</td>
</tr>
<tr>
<td>eGFR &lt;45 mL/min</td>
<td>1.11 (0.46-2.72)</td>
<td>0.813</td>
</tr>
<tr>
<td>cTnI &gt;0.1 ng/L</td>
<td>1.98 (1.03-3.78)</td>
<td>0.040</td>
</tr>
</tbody>
</table>
FINNISH GELSONI AMYLOIDOSIS CAUSES SIGNIFICANT DISEASE BURDEN BUT DOES NOT AFFECT SURVIVAL

S. Eeva-Kaisa ¹, M. Tuuli ¹, K. Sari ¹, K. Tero ², A. Sari ³

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Introduction: Hereditary gelsolin (AGel) amyloidosis is an autosomal dominantly inherited systemic amyloidosis that manifests with the characteristic triad of progressive ophthalmological, neurological and dermatological sign and symptoms. The National Finnish Gelsolin Amyloidosis Registry (FINGAR phase I) was founded in 2013 to collect clinical data on patients with AGel amyloidosis, including altogether approximately one third of the Finnish patients.

Objects: We aim to deepen knowledge on the disease burden and its progression as well as life span of the patients using data from the FIN-GAR phase II registry, updated five years after the original FIN-GAR phase I registry.

Methods: We sent an updated questionnaire concerning the symptoms and signs, symptomatic treatments and subjective perception on disease progression to 240 members of the Finnish Gelsolin Amyloidosis Association (SAMY). We also analyzed the lifespan of 478 patients using the relative survival (RS) framework.

Results: The updated FIN-GAR phase II registry includes 261 patients. Follow-up time after FINGAR phase I was 4.9 years (range 4.1-5.6). Symptoms and signs corresponding to the classical triad of ophthalmological (dry eyes in 93%; corneal lattice amyloidosis in 89%), neurological (numbness, tingling and other paresthesias in 75%; facial paresis in 67%), and dermatological (drooping eyelids in 86%; cutis laxa in 84%) manifestations were highly prevalent. A total of 74%, 53% and 72% of the patients reported that their ophthalmologic, neurologic and dermatologic symptoms, respectively, have got worse during the follow-up. Cardiac arrhythmias were reported by 15% of the patients and 5% had a cardiac pacemaker installed. Proteinuria was reported by 13% and renal failure by 5% of the patients. A total of 65% of the patients had undergone a skin or soft tissue surgery, 26% carpal tunnel surgery and 24% at least unilateral cataract surgery. As regards life span, relative survival estimates exceeded 1 for males and females until the age group of 70-74 years, for which it was 0.96.

Conclusions: AGel amyloidosis causes a wide variety of ophthalmological, cutaneous and neurological symptoms that together with repeated surgeries cause a clinically significant disease burden. Renal and cardiac manifestations are rare as compared to other systemic amyloidoses, explaining in part the finding that AGel amyloidosis does not shorten the life span of the patients at least for the first 75 years.
EXCELLENT OUTCOMES OF ISOLATED RENAL TRANSPLANTATION FOR HEREDITARY FIBRINOGEN (AFIB) AMYLOIDOSIS

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2 Waikato DHB, Hamilton, NZ.

Introduction: Mutations in the fibrinogen Aα chain are the commonest cause of hereditary renal amyloidosis in the United Kingdom. The phenotype of AFib amyloidosis is predominantly renal, presenting with nephrotic syndrome in the 50s or 60s and typically progressing Chronic Kidney Disease (CKD) to end-stage renal disease (ESRD). Previously we reported (Pinney et al, Am J Transpl 2013;13:433) the outcome of renal transplantation in 11 patients receiving isolated kidney transplants for AFib with a median graft survival 7.3 years.

Objectives: Review and update outcomes of renal transplantation for AFib amyloidosis.

Methods: The database of the UK National Amyloidosis Centre (NAC) was used to identify all patients who had received an isolated renal transplant for AFib amyloidosis; combined liver/kidney transplants were excluded. Patients were reviewed annually at the NAC including renal function (by measured creatinine clearance (CrCl)), proteinuria and SAP scintigraphy.

Results: Among 153 patients with AFib reviewed at NAC, 36 patients (22 males) received 40 isolated renal transplants (30 cadaveric, 10 live unrelated donor). Median age at transplantation was 60.1 years. Median follow-up from transplantation was 66 months. Four transplants (10%) failed immediately for technical reasons and were not included in analyses of graft survival. Median death censored graft survival of the remaining 36 transplants was not reached; Kaplan-Meier estimates of renal allograft survival were 100% at 5 years and 68% at 10 years. Six grafts failed at a median of 81 months (range 70-131) and although amyloid deposits were identified in 4/4 allograft biopsies undertaken to investigate a rising creatinine, in no case was amyloid considered to be the sole cause of allograft failure. Serial SAP scintigraphy showed amyloid deposition in the renal allograft at a median of 7 years post-transplant (range 4-10 years). Proteinuria rose from a median of 0.1g/day in years 1-6 to 0.3g/day in years 7-8 and 0.5g/day in years 9-10 post-transplant. Median CrCl was 55mL/min in years 1-6, 48mL/min in years 7-8 and 41mL/min in years 9-10.

Conclusions: Renal allograft survival is excellent (68% at 10 years) in AFib amyloidosis. Amyloid deposition occurs in the transplanted kidney, as shown by SAP scintigraphy, at a median of 7 years and is typically accompanied by onset of gradually progressive proteinuric CKD.

Keywords: Renal Transplantation / Fibrinogen
A PHASE II STUDY OF ISATUXIMAB (SAR650984) (NSC-795145) FOR PATIENTS WITH PREVIOUSLY TREATED AL AMYLOIDOSIS (SWOG S1702; NCT#03499808)

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2 SWOG Statistics and Data Management Center.
3 Boston University Medical Center.
4 Memorial Sloan Kettering Cancer Center.
5 University of Michigan Comprehensive Cancer Center.
6 Mayo Clinic.
7 Loyola University Medical Center.
8 Oregon Health and Science University.
9 The University of Texas MD Anderson Cancer Center.

Background: Isatuximab (SAR650984) is an IgG1k monoclonal antibody that binds with high affinity to CD38 expressed on plasma cells in AL amyloidosis. It has been shown to be efficacious and well tolerated in relapsed and refractory multiple myeloma as a single agent and in combination. Here we report the patient characteristics and preliminary safety results of a prospective multi-center, phase II study of isatuximab in previously treated patients with AL amyloidosis (NCT03499808).

Materials/Methods: Eligibility included age ≥ 18 years, relapsed or refractory systemic AL amyloidosis, ≥ 1 prior line of therapy, measurable disease, at least one organ involved, not refractory to daratumumab, ECOG performance status ≤ 2, creatinine clearance ≥25 mL/min, and NT-proBNP ≤8500 pg/mL. Patients received isatuximab intravenously 20 mg/kg weekly during the first 28 day cycle and every other week during cycles 2 through 24 for a maximum of 24 cycles. The primary objective was hematologic response with secondary objectives of organ response, safety, progression free survival, and overall survival.

Results: At data cut-off (November 15, 2019), 43 patients were enrolled from March 08, 2018 to September 30, 2019 at 14 institutions. Thirty eight patients were eligible with 37 being evaluable for toxicity. The median age was 69.8 years (range, 40.1-83), with 22 patients (58%) being female. Two patients (5.3%) had prior daratumumab exposure. Twenty one patients (55%) had only one organ system involved while 17 patients (44.7%) had multi-system involvement. Twenty six patients (68%) had cardiac involvement and 16 patients (42%) had renal involvement. Twenty six patients remain on treatment with 12 patients discontinuing treatment prior to 24 cycles due to adverse events (AEs) (n=4), other-not protocol specified (n=2), disease progression (n=2), refusal unrelated to an AE (n=2), and death unrelated to treatment (n=1). Ten patients (27%) experienced at least one grade ≥ 3 AE (any cause). The most common ≥ grade 3 AEs were infusion related reactions (n=3), lymphopenia (n=3), and lung infection (n=2). The most common drug-related AEs were infusion related reactions in 18 patients (48.6%) followed by infection (any cause) in 10 patients (27%), lymphopenia in 8 patients (22%), and anemia in 7 patients (18.9%). The majority of infusion related reactions were grade 1 or 2 (n=15).

Conclusions: The administration of isatuximab in previously treated patients with AL amyloidosis is associated with a good safety profile. Similar to other monoclonal antibodies against CD38, the main toxicities were related to infusion reactions and infections. The data will be updated at the meeting.

Support: NIH/NCI/NCTN grant awards U10CA180888, U10CA180819, CA180820, and CA180821; and in part by Sanofi-Aventis (Sanofi US).
IXAZOMIB-DEXAMETHASONE VERSUS PHYSICIAN’S CHOICE IN RELAPSED/REFRACTORY SYSTEMIC AL AMYLOIDOSIS: RESULTS FROM THE PHASE 3 TOURMALINE-AL1 TRIAL


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3 National Amyloidosis Centre, the Royal Free London NHS Foundation Trust, University College London, London, UK.
4 Department of Medicine V (Hematology, Oncology and Rheumatology), Amyloidosis Center Heidelberg, Heidelberg University Hospital, Heidelberg, Germany.
5 Division of Hematology/Oncology, Department of Medicine, Samsung Medical Center, Sungkyunkwan University School of Medicine, Seoul, Korea.
6 Amyloidosis Center, Boston University School of Medicine, Boston Medical Center, Boston, MA, USA.
7 Memorial Sloan Kettering Cancer Center, New York, NY, USA.
8 Westmead Amyloidosis Service, Westmead Hospital, Sydney, on behalf of the Australian Amyloidosis Network, Australia.
9 Department of Hematology, Japanese Red Cross Medical Center, Tokyo, Japan.
10 John C. Davis Myeloma and Amyloid Program, Tufts Medical Center, Boston, MA, USA.
11 Millennium Pharmaceuticals, Inc., Cambridge, MA, USA, a wholly owned subsidiary of Takeda Pharmaceutical Company Limited.
12 Division of Hematology, Mayo Clinic, Rochester, MN, USA.

Background: There are currently no approved treatments for AL amyloidosis; active, tolerable treatments that elicit a deep haematological response and improve organ function are needed. The oral proteasome inhibitor ixazomib (I) is approved with lenalidomide (L)-dexamethasone (Dex) for the treatment of multiple myeloma patients (pts) who have received ≥1 prior therapy.

Methods: Relapsed/refractory AL (RRAL) pts were randomised to I (4.0 mg, d 1, 8, 15) plus Dex (20 mg, d 1, 8, 15, 22) or physician’s (phys) choice (Dex alone or plus melphalan [M], cyclophosphamide [C], thalidomide [T], or L) in 28-d cycles until disease progression or unacceptable toxicity. Primary endpoints were 1) overall haematologic response rate, and 2) 2-yr vital organ deterioration or death.

Results: 168 pts were randomised; 85 to IDex and 83 to phys choice (47 LDex, 24 MDex, 10 CDex, 2 TDex); median age was 65 (range 38–84) vs 66 (33–82) yrs, 56% vs 63% had cardiac and 66% vs 58% had renal involvement (33% vs 23% had both) at diagnosis. Haematological responses were seen in 53% (IDex) vs 51% (phys choice; odds ratio [OR] 1.10, p=0.762). Higher complete response (CR) rates were seen with IDex (26%) vs phys choice (18%). Time-to-event data favoured pts treated with IDex vs phys choice, including time to vital organ deterioration/death (34.8 vs 26.1 mos, Table). Vital organ response was 36% with IDex vs 11% with phys choice (cardiac/renal response rates: 18%/28% vs 5%/7%). At data cut-off, median treatment duration was 11.7 (IDex) vs 5.0 mos (phys choice); 21% vs 6% of pts remained on treatment. Grade ≥3 adverse event (AE) rates were 62% vs 56%, 34% vs 41% of pts had drug-related grade ≥3 AEs, 26% vs 20% of pts had AEs resulting in discontinuation, and there were 6% vs 5% on-study deaths. Common grade ≥3 AEs were fatigue (9% vs 9%), anaemia (2% vs 11%), cardiac failure and dyspnoea (each 6% vs 4%), peripheral oedema, and pneumonia (each 5% vs 5%).

Conclusions: IDex resulted in an improved CR rate and duration of response and, although the primary endpoint was not met, all clinically relevant time-to-event endpoint data favoured IDex vs phys choice. IDex was generally well tolerated and associated with a doubling of treatment duration vs phys choice. TOURMALINE-AL1 is the first phase 3 trial in RRAL and shows significant outcome improvements for IDex vs phys choice.

Keywords: ixazomib, relapsed/refractory.
<table>
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<tr>
<th></th>
<th>IDex n=85</th>
<th>Phys choice n=83</th>
<th>HR; p-value* (unless stated)</th>
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</thead>
<tbody>
<tr>
<td>Time to vital organ deterioration/death†</td>
<td>34.8</td>
<td>26.1</td>
<td>0.525; p=0.012</td>
</tr>
<tr>
<td>2-yr vital organ deterioration or death rate‡, %</td>
<td>40</td>
<td>45</td>
<td>OR 0.83; p=0.608</td>
</tr>
<tr>
<td>OS</td>
<td>Not estimable</td>
<td>40.8</td>
<td>0.837; p=0.478</td>
</tr>
<tr>
<td>Time to subsequent therapy</td>
<td>26.5</td>
<td>12.5</td>
<td>0.615; p=0.027</td>
</tr>
<tr>
<td>Overall PFS†</td>
<td>11.2</td>
<td>7.4</td>
<td>0.673; p=0.043</td>
</tr>
<tr>
<td>Haematological PFS†</td>
<td>20.1</td>
<td>16.7</td>
<td>0.790; p=0.283</td>
</tr>
<tr>
<td>Vital organ PFS†</td>
<td>18.0</td>
<td>11.0</td>
<td>0.62; p=0.019</td>
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<tr>
<td>Time to treatment failure†</td>
<td>10.1</td>
<td>5.2</td>
<td>0.600; p=0.005</td>
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</tbody>
</table>

*HR/OR<1 indicates an effect favouring IDex
†Per physician assessment; ‡Per adjudication committee.
This study was sponsored by Millennium Pharmaceuticals Inc., a wholly owned subsidiary of Takeda Pharmaceutical Company Limited.
OP37

SUBCUTANEOUS DARATUMUMAB + CLOPHOSPHAMIDE/BORTEZOMIB/DEXAMETHASONE IN NEWLY DIAGNOSED AL AMYLOIDOSIS: UPDATED SAFETY RUN-IN RESULTS OF ANDROMEDA


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8 Department of Lymphoma and Myeloma, Division of Cancer Medicine, The University of Texas MD Anderson Cancer Center, Houston, TX, USA.
9 Division of Hematology, The Ohio State University Comprehensive Cancer Center, Columbus, OH, USA.
10 Winship Cancer Institute, Emory University, Atlanta, GA, USA.
11 School of Medicine, Oregon Health & Science University, Portland, OR, USA.
12 Division of Hematology and Hematologic Malignancies, University of Utah School of Medicine, and the Huntsman Cancer Institute, Salt Lake City, UT, USA.
13 Department of Hematology and Hematopoietic Cell Transplantation, Judy and Bernard Briskin Center for Multiple Myeloma Research, City of Hope, Duarte, CA, USA.
14 Department of Medicine, Boston University School of Public Health, Boston, MA, USA and Amyloidosis Center, Boston University School of Medicine and Boston Medical Center, Boston, MA, USA.
15 Janssen Research & Development, LLC, Spring House, PA, USA.
17 Amyloidosis Research and Treatment Center, Fondazione Istituto di Ricoero e CurA a Carattere Scientifico (IRCCS) Policlinico San Matteo, and Department of Molecular Medicine, University of Pavia, Pavia, Italy.

Background: Achievement of deep and sustained hematologic response in AL amyloidosis is needed to improve organ function and overall survival. In the safety run-in cohort of ANDROMEDA (NCT03201965), a phase 3 study of subcutaneous daratumumab (DARA SC) with cyclophosphamide, bortezomib, and dexamethasone (CyBorD) in patients with newly diagnosed AL amyloidosis, deep hematologic responses and a tolerable safety profile were observed.

Objectives: We present updated safety and efficacy data for the 28 patients enrolled in the safety run-in cohort of ANDROMEDA.

Methods: Eligible patients had ≥1 involved organs, Eastern Cooperative Oncology Group (ECOG) score ≤2, estimated glomerular filtration rate ≥20 mL/min/1.73 m², and NT-ProBNP ≤8,500 ng/L. In the safety run-in, patients received DARA (1,800 mg in 15 mL) co-formulated with recombinant human hyaluronidase PH20 (rHuPH20; ENHANZE® drug delivery technology, Halozyme, Inc.) administered subcutaneously (SC) QW Cycles 1-2, Q2W Cycles 3-6, and Q4W thereafter for ≤2 y. Cy 300 mg/m² PO or IV, Bor 1.3 mg/m² SC, and D 40 mg were administered QW ≤6 cycles. Hematologic response evaluations were Q4W cycles 1-6 and every other month thereafter. Amyloidosis complete response, based on consensus criteria, required normalization of FLC levels and ratio, and negative serum and urine immunofixation (IFE). Patients with negative serum and urine IFE and normalized involved FLC levels without normalized FLC ratio (due to suppression of uninvolved FLC below LLN) were classified as having a modified complete response.

Results: Patients (N = 28) had a median (range) age of 67.5 (35-83) years and a median of 2 (1-4) involved organs. Heart and kidney involvement affected 60.7% and 67.9% of patients, respectively; 17.9% were Mayo cardiac stage IIIA at screening. Median follow-up was 17.6 (1.3-20.4) months. Median treatment duration was 15.1 (0.2-20.1) months. Patients received a median of 16 (1-23) treatment cycles; 22 (78.6%) patients have received DARA SC maintenance (>6 cycles). The overall hematologic response rate was 96.4% and the rate of very good partial response (VGPR) or better was 82.1%. A total of 15 (53.6%) patients achieved CR; 5 of these 15 patients achieved CR with all criteria except normalization of the FLC ratio. The proportions of patients achieving stringent hematologic responses of dFLC levels <10 mg/L and iFLC levels ≤20 mg/L were 67.9% and 71.4%, respectively. Cardiac, renal and hepatic responses were seen in...
52.9%, 83.3% and 50.0% of evaluable patients, respectively. No new safety signals were identified. Updated efficacy and safety data will be presented at the meeting.

**Conclusions:** DARA SC with CyBorD achieves high rates of deep hematologic responses and encouraging organ responses with no new safety concerns in patients with newly diagnosed AL amyloidosis.

**Suggested keywords (3):** Daratumumab, CyBorD, AL amyloidosis,
ASSESSMENT OF MINIMAL RESIDUAL DISEASE USING MULTIPARAMETRIC FLOW CYTOMETRY IN TREATED PATIENTS WITH AL AMYLOIDOSIS

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Background: Despite achieving a hematologic complete response (hemCR), ~20% of patients with AL amyloidosis do not attain organ response and ~30% experience hematologic relapse. The assessment of minimal residual disease (MRD) by multiparametric flow cytometry (MFC) as a more stringent treatment response category is of interest in AL amyloidosis.

Objective: To compare MRD-negative vs. MRD-positive treated patients in hemCR and evaluate the association of MRD-status with organ response.

Methods: Bone marrow aspirates were assessed with a cross-validated 2-tube 10-color antibody panel. A target of $2 \times 10^6$ events were measured using a Beckman Coulter Navios flow cytometer and analyzed using Kaluza software (sensitivity level: 1 in $10^5$ nucleated cells or higher). MRD-positivity was defined as a clonal plasma cell population of ≥20 events with ≥2 aberrancies. Statistical differences were estimated by the $\chi^2$ and Mann-Whitney U tests.

Results: Of 65 patients in hemCR, 52 (80%) had renal and 29 (45%) had cardiac involvement. Final treatment achieving hemCR was high-dose melphalan and stem cell transplantation for 32 (49%), bortezomib-based for 16 (25%) and an anti-CD38 monoclonal antibody for 14 (22%) patients. Three were still receiving daratumumab therapy at the time of hemCR achievement and MRD assessment. For 24 (37%) patients, ≥2 lines of therapy were required. Median time from hemCR achievement to MRD analysis was 38 months (range 0–260). 29 (45%) patients were MRD-negative and 36 (55%) were MRD-positive. Comparing the MRD-negative and MRD-positive groups, renal response was observed in 86% vs. 64% ($P = .05$), cardiac response in 75% vs. 59% ($P = .37$) and any organ response in 90% vs. 75% ($P = .13$) of patients. Depth of organ response as measured by percentage improvement in proteinuria (96% vs. 91%, $P = .16$) and BNP (55% vs. 46%, $P = .66$) was not significantly higher among those with MRD-negativity.

Conclusions: For patients with AL amyloidosis in hemCR, MRD-negativity by MFC was associated with statistically higher probability of renal response, but not cardiac or any organ response, nor depth of organ improvement. Results may be confounded by higher baseline dFLC and MRD testing occurring at a later time point after hemCR achievement in the MRD-negative cohort (71 vs. 32 months, $P = .27$), thereby allowing more time for organ recovery.

### TABLE 1: Clinical characteristics according to MRD group

<table>
<thead>
<tr>
<th>MRD-negative (N = 29)</th>
<th>MRD-positive (N = 36)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Median age (range)</td>
<td>60 (32–76)</td>
<td>61 (30–74)</td>
</tr>
<tr>
<td>% female</td>
<td>48%</td>
<td>28%</td>
</tr>
<tr>
<td>Light chain type λ/κ</td>
<td>24/5</td>
<td>29/7</td>
</tr>
<tr>
<td>Median dFLC mg/L (range)</td>
<td>129 (34–3120)</td>
<td>70 (6–462)</td>
</tr>
<tr>
<td>Bone marrow PC % (range)</td>
<td>10% (3–30)</td>
<td>10% (1–25)</td>
</tr>
<tr>
<td>t(11;14)</td>
<td>6/15 (40%)</td>
<td>8/21 (38%)</td>
</tr>
</tbody>
</table>

**Final treatment achieving hemCR**

- HDM/SCT: 15 (52%) vs. 17 (47%) ($P = .72$)
- Bortezomib-based: 6 (21%) vs. 10 (28%) ($P = .51$)
- Anti-CD38 monoclonal Antibody: 8 (28%) vs. 6 (17%) ($P = .29$)
- Immunomodulatory drug: 0 (0%) vs. 3 (8%) ($P = .29$)
- # requiring >1 treatment regimen to achieve hemCR: 11 (38%) vs. 13 (36%) ($P = .78$)
- Median # of months from hemCR to MRD testing: 71 (0–238) vs. 32 (0–260) ($P = .27$)

**Organ response at the time of MRD assessment**

- Any organ: 26/29 (90%) vs. 27/36 (75%) ($P = .13$)
- Renal: 21/24 (88%) vs. 18/28 (64%) ($P = .05$)
- Cardiac: 9/12 (75%) vs. 10/17 (59%) ($P = .37$)
- Hepatic: 3/3 (100%) vs. 7/7 (100%)

Keywords: AL amyloidosis; minimal residual disease; multiparametric flow
OP39

ONE-YEAR EVALUATION OF THE INCIDENCE AND DISTRIBUTION OF AMYLOIDOSIS DISEASES IN GERMANY: NATIONAL CLINICAL AMYLOIDOSIS REGISTRY

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2 Medical Department V, Amyloidosis Center.
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4 DKFZ Heidelberg, Biostatistics.

Background: Currently there are no data on the epidemiology of systemic amyloidosis in Germany. Previous studies in other regions estimated an incidence of 8 to 14 cases per million population per year.

Objectives: Our aim was to collect epidemiological data on amyloidosis diseases in Germany. These data were used to determine incidence and distribution of amyloidosis types. Also, data on diagnosis, prognosis and survival were collected.

Methods: The registry population consist of all reported cases of newly diagnosed amyloidosis patients between 4.1.18 - 8.1.19. Data cutoff of the analysis was May 31, 2019. Patients were included in the registry by two main ways: personal presentation at the amyloidosis outpatient clinic or phone contact of the attending physician to the Amyloidosis Center. Inclusion criteria were either a Congo Red positive tissue sample or unequivocal findings in bone scintigraphy (ATTR amyloidosis). Patient were contacted every 6 months to receive QoL data using EORTC QLQ-C30, EQ-5D-5L and SF36-v2 questionnaires.

Results: During the first year of the registry 441 patients were included, mainly through the amyloidosis outpatient clinic (n=268, 60.8%). Seventy-one percent of cases were male (n=311). The median age at diagnosis was 71 years. The subgroup distribution was as follows: 46% AL amyloidosis, 41.5% ATTR amyloidosis, 6.8% local amyloidosis, 4.1% AA amyloidosis, 0.5% others and 1.1% type unknown. Diagnosis was confirmed by biopsy in 88% and in 12% by bone scintigraphy. The most common involved organs were heart (76%) and kidney (33%). Cardiac staging systems were used for AL (Kumar et al., 2014) and ATTR (Gillmore et al., 2018). Data on survival were available in 399 cases (median follow-up of 10.5 months). During the first year 60 patients died (9 ATTRwt and 42 AL, 9 other). The most frequent cause of death was amyloidosis itself (n=27, 45%). Five percent of patients died due to therapeutic complications. Compared to ATTRwt patients with AL amyloidosis had a higher risk to die, 1 year survival was 92% and 76%, respectively. In multivariate analysis (including age, cardiac staging systems and time from symptoms to diagnosis), factors associated with worse overall survival were increasing age (HR 1.04 for AL and 1.1 for ATTRwt) and advanced heart involvement (HR 3.98 for AL and 9.23 for ATTRwt). The heart stage III had the most significant influence on survival (AL: HR 3.98, p = 0.026; ATTRwt: HR 9.23; p=0.01).

Summary: First results of the registry show a high number of ATTRwt patients in the German population. Patients with AL amyloidosis have a worse overall survival compared to ATTRwt.

Conclusion: The first year of the German registry was successful to recruit clinical data of all types of amyloidosis of patients newly diagnosed in 2018. We will start an extended registry in 2020 including biopsy reports of reference pathologists.

Keywords: German registry, survival.
LOCALISED LARYNGEAL AMYLOID – A SERIES OF 100 CASES


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Introduction: Localised laryngeal amyloidosis, accounts for 0.2 to 1.2 per cent of all benign laryngeal tumours. We here describe a large national case series.

Methods: A retrospective review was performed to identify all patients diagnosed with localised laryngeal amyloid between 2000 and 2017. Patient demographics and disease profile was collated, including age and sex, presenting symptoms and signs, exact location of amyloid deposit, treatments and progression of disease, if any.

Results: One hundred patients were identified from the database for review (54 female, mean age at diagnosis 54 (range 14 – 83) years). Dysphonia was the most common primary complaint (83%) other symptoms included cough, shortness of breath, foreign body sensation and wheeze. Four patients were diagnosed as an incidental finding. The glottis involved in 53 patients, the supraglottis in 44, the subglottis 26 and 11 had amyloid extending into the tracheobronchial tree. 8% had amyloid outside the larynx – 3 nasal cavity, 1 palate, 2 paranasal sinus and 2 in the postnasal space. Of the 100 patients, two thirds (66) had amyloid localised to one laryngeal subsite with 34 patients having more than one site involved. Most amyloid deposits (56/100) were single discrete lesions. Of the 66 patients who had amyloid in one subsite, 10 had multifocal disease or more than one deposit. None of the patients who were diagnosed with laryngeal amyloid progressed to systemic amyloidosis. Many patients did however, progress locally with further lesions within the same laryngeal subsite (26) or into other subsites (17). 32 patients required only one treatment while 34 had a more progressive disease that required multiple treatments over a long period of time. For one patient, the period between treatments was 14 years. Three patients radiotherapy.

Discussion: We describe the largest reported cohort of patients with localised laryngeal amyloidosis. Similar to case series already in the literature, we found no clear sex predominance. Localised laryngeal amyloid can present at any age with our patient cohort ranging from 14 to 83 years of age. Although the data suggests that the glottis and supraglottis where the more common subsites (53% and 44% respectively), we suggest the more important message is the multifocal nature of the localised laryngeal amyloidosis. Over a third of the patients (34%) had amyloid deposits in more than one laryngeal subsite, and 19% of patients had amyloid deposits outside the larynx (8% superiorly in the upper airway and 11% inferiorly into the tracheobronchial tree). This highlights the need for comprehensive examination of the entire airway looking for multiple amyloid deposition.
VALIDATION OF A NOVEL MODEL FOR THE COMPARATIVE ANALYSIS OF AMYLOID-REACTIVE BIOLOGICALS USING A SINGLE MOUSE

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**Introduction:** Synthetic bifunctional peptopes, which combine a multi-amyloid binding motif and an epitope sequence for an opsonizing antibody, have been developed for use in pre-targeting immunotherapy of amyloidosis. Binding of peptopes to amyloid in vivo depends on various factors including amyloid binding affinity, accessibility of the amyloid to the circulation, elimination half-life and secondary pharmacology. Modifications to peptope structure may alter accumulation within tissue amyloid. Therefore, assessing the effects of structural changes on amyloid binding of peptopes is best achieved using a murine model of amyloidosis. Previous studies have used between-subjects (BS) designs to assess the comparative uptake of 125I-labeled peptides in independent cohorts of AA-amyloid mice. However, differences in amyloid load in respective cohorts can reduce the precision of comparison. We hypothesized that a single mouse within-subjects (WS) study, using 125I and 124I-labeled reagents, from which multiple samples of tissue are collected and quantified, can reduce variability and serve as a more precise method of comparing relative amyloid accumulation.

**Objectives:** The goal of this study was to statistically compare the effects of BS and WS study designs when analyzing amyloid accumulation of 125I- and 124I-labeled peptopes.

**Methods:** Peptopes were synthesized (Genscript, NJ) and purified by reverse phase HPLC. Radiolabeling of p93 used 124I (~600 keV), and p98 was labeled with 125I (~30 keV emission). AA amyloid was induced in H2/hIL-6 transgenic mice by IV injection of AEF. Mice were injected with a mixture of 125I-p98 and 124I-p93. In the BS study, two cohorts of n = 3 mice were injected, and a single sample of muscle, liver, pancreas, spleen, kidney, stomach, upper/lower intestine was collected. For the WS study, one mouse was injected and three independent samples of each tissue harvested. Organ uptake of peptopes was quantified by gamma counting and the study designs compared with respect to differences in variances, means and mean differences.

**Results:** When comparing the variances of peptope 124I - p93, there were no differences in the liver, pancreas, spleen, stomach or lower intestine. For peptope 125I-p98, there were no differences in variance for the muscle, liver, pancreas, spleen, stomach, upper intestine or lower intestine. Similarly, no difference on the means for p93 or p98 in the BS and WS designs were detected for any organ. For the mean differences (p93 vs. p98) comparison of BS and WS designs, no differences were detected for any organ.

**Conclusions:** The ability to perform comparative analyses of amyloid-reactive biologicals in individual mice (n = 1) using dual-energy biodistribution measurements and a WS design increases the precision of comparison and reduces the use of animals to generate meaningful data. This creative study design might improve iterative development of next generation biological imaging agents and therapeutics for amyloidosis.
CURCUMIN PROMOTES PROGRESSION OF AApoAII AMYLOIDOSIS IN MICE BY ACTIVATING THE PPARα SIGNALING PATHWAY

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2 Department of Aging Biology, Institute of Pathogenesis and Disease Prevention, Shinshu University Graduate School of Medicine.
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4 Department of Neuro-health Innovation, Institute for Biomedical Sciences, Interdisciplinary Cluster for Cutting Edge Research, Shinshu University, Matsumoto Japan.

Introduction: In mouse AApoAII amyloidosis, apolipoprotein A-II (ApoA-II) polymerizes to form amyloid fibrils (AApoAII) that are associated with aging. We have demonstrated that administration of oxidative stress inhibitors (e.g., tempol and apocynin) suppresses AApoAII amyloidosis progression in mice [1]. Curcumin, a polyphenol compound, is considered to have beneficial biological effects such as antioxidant and anti-inflammatory activity, and also exhibits anti-amyloid effects in vitro and in vivo [2,3]. To elucidate the mechanisms by which curcumin affects systemic amyloidosis, we investigated amyloid deposition and molecular changes in a mouse model of AApoAII amyloidosis when the mice were fed a diet supplemented with curcumin.

Materials & Methods: Eight-week-old female R1.P1-Apoa2c mice were injected with AApoAII amyloid fibrils (1µg) to induce amyloidosis and were fed a normal diet (A-NT group) or a diet supplemented with 0.5%w/w or 2%w/w curcumin (A-Cur group). As a control, mice were also fed a normal (Con group) or curcumin-supplemented (Cur group) diet without amyloid induction to analyze the effects of curcumin intake. At 8 or 12 weeks after beginning the diet, the mice were sacrificed for histochemical and biochemical analysis. AApoAII amyloid deposition was evaluated in Congo red-stained sections using polarizing microscopy and the plasma concentrations of ApoA-II and high-density lipoprotein (HDL) were measured. We also performed a comprehensive analysis of mRNA expression in the liver using RNA-Seq to investigate the molecular mechanisms associated with AApoAII amyloidosis that are affected by curcumin.

Results: Unexpectedly, we found that dietary supplementation with curcumin (A-Cur) for 12 weeks significantly increased the degree of AApoAII amyloid deposition relative to those in the control mice (A-NT) without supplementation, especially in the liver and spleen. The liver weight, plasma ApoA-II and HDL concentration were also significantly elevated in the curcumin-supplemented groups. RNA-Seq analysis in liver tissue revealed that curcumin intake affected liver lipid metabolism via the peroxisome proliferator-activated receptor (PPAR) pathway, resulting in increased Apoa2 mRNA expression. Furthermore, the observed increase in liver weight could also be due to activation of Pparα gene expression and peroxisome proliferation.

Discussion & Conclusions: These findings suggest that curcumin activates the PPARα signaling pathway to promote increases in ApoA-II levels that in turn result in increased amyloidosis. Such reverse regulation of PPARα signaling pathways could represent new therapeutic strategies against AApoAII amyloidosis.

Key words: AApoAII amyloidosis, PPARα, curcumin

References:
IN VITRO ASSAY SYSTEM DEMONSTRATING TOXIC EFFECTS OF A PATIENT-DERIVED AMYLOIDOGENIC LIGHT CHAIN IN PRIMARY HEART AND KIDNEY CELLS

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Introduction: Amyloidogenic light chain (AL) amyloidosis is a systemic protein misfolding and deposition disease, which most commonly affects the heart, the kidney or both. This organ tropism seems to depend on the synergistic effects of light chain (LC) sequence alterations, posttranslational modifications and organ environment. Yet, the exact mechanisms are widely unclear.

Objectives: With the help of our research cooperation partners, we aim to replicate and elucidate the kidney and heart tropism of AL amyloidosis patients in vitro and characterise interactions of kidney and heart cells with patient-derived and recombinantly expressed LCs and variable domains (VLs). Here, a combination of cellular assays was used to determine the effects of LCs and VLs on proliferation, cell viability and overall cell survival.

Methods: In detail, three different primary rat cell types from kidney (mesangial cells, proximal tubular cells) and heart (cardiomyocytes) were incubated with 15 µg/ml recombinant patient LC or VL and subsequently employed in live / dead fluorescence staining and evaluated by flow cytometry. We analysed immediate effects of LC and VL treatment for 8 h on kidney cell proliferation in BrdU labelling assays. Metabolic activity as an indicator of cell viability was measured after 24 h in mesangial and tubular cells by a resazurin reduction assay.

Results: So far, LC and VL from one patient with cardiac symptoms and the corresponding germline lambda LC and VL were tested. In accordance with the reported phenotype, rat cardiomyocytes showed approximately 1.4 times increased fraction of dead cells after 48 h co-incubation with germline LC, with a 10 % increased toxic effect in patient LC treated cells. Preliminary data further indicate a cytotoxic effect of the amyloidogenic LC in mesangial and tubular cells (approximately +20 % and +10 % dead cell fraction compared to medium control, respectively). Notwithstanding, kidney cells displayed a small increase in proliferative activity immediately after addition of LC or VL (=10% in mesangial cells, ≈20 % in tubule cells). Finally, no differences in metabolic activity were observed in kidney cells after 24 h co-incubation, possibly due to low LC and VL concentrations obliterating differences between germline and amyloidogenic sequences.

Conclusions: In conclusion, we were able to establish a set of assays to replicate organ tropism of patient-derived LCs and VLs, which revealed both cell type-specific differences in the toxicity. In a next step, we will investigate species-dependency by subjecting human primary cells to the same proliferation, viability and toxicity assays. Already, these cellular assays can be used to compare the toxicity of distinct patient LCs and determine crucial LC sequence variations and modifications.

Keywords: amyloidogenic light chains, organ tropism, cellular assays.
DEVELOPING A HUMANIZED TRANSTHYRETIN MOUSE MODEL FOR THERAPEUTIC VALIDATION

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2 Intellia Therapeutics.

Transthyretin amyloidosis (ATTR) is a rare disorder arising from misfolded transthyretin (TTR) protein which aggregates and deposits as amyloid deposits in target organs. The familial form involves genetic mutations which usually destabilize circulating tetramers and leads to amyloid deposition in numerous sites in the body, including peripheral nerves, heart, and kidneys. It is becoming clear that wild-type (WT) ATTR, in which un-mutated monomers fibrillize and aggregate, is a significantly under-diagnosed cause of heart failure in certain populations.

Regeneron utilizes a high-throughput pipeline to create genetically-modified mouse and rat models, in order to enable in vivo evaluation of our targets portfolio for our Therapeutic Focus Areas. A key tool in our toolbox is humanizations, in which we replace the endogenous rodent gene with the human counterpart such that the human gene is under control of native regulatory elements. Humanizations are an important part of Regeneron’s collaboration with Intellia Therapeutics as we evaluate CRISPR/Cas9 lipid nanoparticles as a modality to treat diseases such as ATTR.

Here we describe the design and generation of a series of human transthyretin alleles precisely replacing the murine gene. In order to take full advantage of the coding exons for gRNA design, we first replaced the murine gene (from the start to the stop codon) with the human gene (start codon to past the polyadenylation signal and 3'-UTR). These humanized TTR mice carry either unmutated (WT) or human disease-relevant mutant alleles. We describe a second generation of humanization which should improve human protein expression. Finally, we demonstrate the utility of rationally designed mouse models to validate transthyretin as a potential target for CRISPR/Cas9 therapeutics.

Key words: Transthyretin amyloidosis, mouse models, human TTR
NEW GENETIC TOOL TO ASSAY TRANSTHYRETIN AGGREGATION IN A YEAST MODEL SYSTEM.

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Background: Transthyretin amyloidosis (ATTR) is a protein misfolding disorder in which monomeric transthyretin protein (TTR) misfolds, assembles into amyloid, and infiltrates systemically. Internalization and cytoplasmic accumulation of TTR aggregates is associated with changes to the proteostasis system, such as chaperone and autophagy activity. Since proteostasis becomes less efficient with age, a lower intracellular proteostasis response may contribute to age-associated cytotoxicity.

Objective: Develop a tractable system to study intracellular TTR aggregation in vivo that lends itself to studying the impact of TTR aggregation on proteostasis and provides a means for high-throughput screening.

Methods: The yTRAP (yeast transcriptional reporting of aggregating proteins) system was recently developed to detect the aggregation-state of proteins associated with yeast prions. Here, we have adapted the yTRAP system for the study of TTR. Wildtype TTR protein, as well as several mutant derivatives, are fused to a synthetic transcriptional activator (synTA). When TTR fusion protein is soluble, the transcriptional activator binds to a synTA responsive promoter that drives expression of a mNeonGreen reporter gene. When TTR is aggregated, the fused transcriptional activator is no longer able to bind to the promoter and mNeonGreen is not expressed. Therefore, mNeonGreen intensity is a direct measure of the amount of soluble TTR within the cells, and an indirect measure of protein aggregation. Aggregation states associated with wildtype, V30M, L55P, L110Q, and V122I alleles in TTR were assessed using fluorescent microscopy and flow cytometry.

Results: TTR expression leads to the formation of intracellular cytoplasmic aggregates that are SDS-resistant and thermostolerant in yeast. Wildtype TTR shows dramatically lower mNeonGreen intensity compared to control strains containing the synTA protein only, indicating that the TTR protein forms intracellular aggregates in the yTRAP system. Wildtype and L110Q alleles show overlapping yTRAP flow cytometry profiles, whereas L55P and V122I exhibit mNeonGreen intensity profiles that are decreased compared to wildtype TTR alleles. This data suggests that there is a higher proportion of the L55P and V122I protein exists in the aggregated state. Surprisingly, V30M alleles shows a substantial decrease in mNeonGreen intensity, indicating that most of the V30M protein is aggregated within the yeast system.

Conclusions: We have established a genetic tool that can assess the TTR aggregation state in yeast cell populations. This yTRAP system allows us to understand protein aggregation associated with different TTR alleles in vivo, and provides a means to study how intracellular TTR aggregation influences cellular proteostasis. This system also provides a platform to pursue high-throughput studies to identify agents that enhance TTR aggregate solubilization.
GENETICS KNOWLEDGE OF PATIENTS WITH HEREDITARY TTR AMYLOIDOSIS

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Introduction: When someone is diagnosed with a hereditary autosomal dominant condition such as hereditary transthyretin amyloidosis (haTTR), the diagnosis has implications beyond the individual as all first degree relatives have a 50% risk to also carry the pathogenic variant. Given the recurrence risk it is important for individuals to share this information with relatives, and often the first step to sharing this information begins with comprehension. Previous research on patient understanding of the genetics aspects of hereditary conditions has focused in the prenatal and oncology settings. There has not yet been an exploration of patient understanding of the genetics of haTTR.

Objectives: Assess patient and family member knowledge of genetic concepts related to haTTR.

Methods: A 13 question true/false survey to assess the genetic knowledge of hereditary amyloidosis was created based off Erblich et al. (2005) validated survey to assess genetic knowledge in the breast cancer setting. Survey content was then reviewed and revised by four experts in the field. The survey was sent to the members of the Amyloidosis Support Group email list serve and posted on their Support Group Facebook page.

Results: One hundred and seventeen individuals participated in the survey, with 114 individuals completing it. The majority of participants had previously been diagnosed with haTTR (82%), and the remaining 18% were family members. Participant demographics were equally split between male and female (male =54%). The majority of participants identified as Caucasian (83%), had a household income above $60,000 annually (62.3%), and had received at least a bachelor’s degree (65%). Over half of the participants had previously attended a support group meeting (54%), and almost half had previously seen a genetic counselor (42%).

On average, participants correctly answered 82% of the questions. Almost 90% of individuals answered the questions correctly regarding recurrence risk for family members. The question most commonly answered incorrectly (31%) pertained to testing minors for amyloidosis. There was a trend that patients who had either seen a genetic counselor and/or attended a support group meeting answered more questions correctly. Individuals who answered less than 50% of the questions correctly had never seen a genetic counselor.

Conclusions: The data suggests that many patients do have an accurate understanding of the genetics and hereditary nature of haTTR and it seems that genetic counseling or attending a support group meeting may help improve knowledge in this area. More education may be needed to discuss the concerns regarding genetic testing of minors for adult onset conditions. Of note, there are limitations to the generalizability of our results given the high socio-economic status of the majority of the participants. Future research should focus on determining whether patients are subsequently informing family members of the risk and if there are barriers to cascade screening.
SERPINA1 MODULATES EXPRESSION OF AMYLOIDOGENIC TTR

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In hereditary transthyretin amyloidosis (ATTR amyloidosis), mutations of the transthyretin (TTR) gene result in deposition of TTR in various tissues followed by disease, commonly manifested by polyneuropathy and/or cardiomyopathy. Using induced pluripotent stem cells (iPSCs) we recently could identify alpha-1-antitrypsin (SERPINA1, AAT1) as being related to ATTR [1]. SERPINA1 and TTR are both proteins of the acute phase response. During inflammatory processes, SERPINA1 expression is induced, while TTR levels are decreased. Yet, the exact mechanisms and functions of TTR downregulation are not understood and a direct link between SERPINA1 and TTR was not previously mentioned.

The aim of this study was to evaluate the inverse expression of SERPINA1 and TTR by modulation of SERPINA1 expression in the context of ATTR amyloidosis.

The human hepatoma cell line HepG2 was used for SERPINA1 knockdown and Oncostatin-M (OSM) treatment. TTR V30M mice were treated with OSM and antisense oligonucleotide (ASO) directed against mSERPINA1. TTR deposition in tissues was evaluated by immunohistochemical staining. SERPINA1 and TTR were examined by RT-qPCR and ELISA.

In HepG2 cells, knockdown of SERPINA1 mRNA was followed by induction of TTR mRNA and protein (factor 2.2 ± 0.4 and 3.0 ± 0.4, respectively). In contrast OSM, a known inducer of SERPINA1, resulted in downregulation of TTR in a dose-dependent manner. At 100 µM OSM, a 2.0 ± 0.9-fold and 6.0 ± 0.1-fold downregulation of TTR mRNA and protein, respectively, was observed. Inspection of sera derived from TTR knockout mice revealed significantly higher SERPINA1 levels (4.7 ± 0.3 mg/ml) in comparison to wildtype mice (3.2 ± 0.6 mg/ml). In addition, V30M mice also displayed lower SERPINA1 levels (2.9 ± 0.4 mg/ml). OSM treatment of V30M mice for 24 h could induce SERPINA1 by a factor of 1.5 ± 0.1 and 1.9 ± 0.1, while TTR was concomitantly downregulated by 1.6 ± 0.1 and 2.3 ± 0.2 (mRNA and protein, respectively). Downregulation of mSERPINA1 following ASO treatment resulted in an increased TTR deposition in V30M mice (factor 2.46 ± 0.38) in various tissues, including dorsal root ganglion (DRG).

SERPINA1 knockdown, OSM stimulation and ATTR V30M mice suggest a previously unrecognized direct correlation of SERPINA1/TTR expression at the transcriptional and protein level. The molecular mechanism of the tight SERPINA1/TTR regulation in the liver is however not known. Ongoing experiments will reveal whether SERPINA1 represents a novel player to modulate disease in ATTR.

THE CHALLENGE IN DEVELOPING AN ANIMAL MODEL FOR AL AMYLOIDOSIS

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Background
Several proteins can cause amyloidosis but the most common form of systemic amyloidosis is the AL Amyloidosis. It is related to the deposit of a monoclonal immunoglobulin (Ig) free Light Chain (LC) produced by a proliferating plasma cell (PC) clone (from Monoclonal Gammapathy of Undetermined Significance to Multiple Myeloma) in different organs such as heart, kidney, liver, spleen and the digestive system. As the amyloid builds up, it gradually affects organ function. Due to the inherent variability of Igs, which differs in sequence from one patient to the other, and the wide spectrum of the pathology, reliable experimental models are challenging to obtain.

Methods
We developed a good strategy to establish mouse lines with high levels of serum pathogenic Igs isolated from patients1. This method proved to be efficient in mimicking several Ig-related human deposition diseases2,3,4 but failed to reproduce AL amyloidosis on its own. By different approaches, we tried triggering AL amyloidosis in our mouse line producing a free λ-LC. For this, we injected amyloid-enhancing factors (AEF), triggered another form amyloidosis (AA Amyloidosis) and induced systemic acidosis. Furthermore, we tried stressing mice PCs by low-dose Bortezomib (Bz) treatment, hoping to generate β- structured aggregates necessary for fibril formation. We have also created a rat model expressing a κ-LC using a similar transgenic strategy as for the mouse. In vitro fibril formation for both LC was performed to confirm their capacity of fibrillation.

Results
In the mouse line (with serum free LCs 494±90 μg/ml), LCs are highly detectable, especially in spleen and kidneys, but there is no Congo Red (CR) positive staining, even after AEF, AA Amyloidosis induction or systemic acidosis. Mice treated with Bz recovered completely, but once again, no deposits were found. Additionally, no intrinsic toxicity of the LC was observed as they have a normal lifespan (around 2 years). The rat line has lower serum free LC levels (26±8 μg/ml) as they still carry the murine Heavy Chain, thus generating complete Igs. In this model, the LC can be detected in lower quantities in kidney and spleen. Their characterization is still ongoing. Fibril formation in vitro using purified variable domain (VL) of the λ-LC and κ-LC was successful (confirmed by Thioflavin T and atomic force microscopy) and seeding with short fragments of pre-formed fibrils rapidly induced fibril formation. However, the attempt to form fibrils using the full-length LC in mice serum was unsuccessful up to now, even in the presence of VL seeds.

Conclusion
Despite the high levels of serum free LCs in the mouse line we seem to be unable to obtain CR positive deposits in vivo. We believe that the negative results could provide invaluable information for AL amyloidosis: understanding why these animals can cope with the pathogenic LCs might give us a new insight into the disease and to develop new therapies.
PM009

A FIRST PENETRANCE ESTIMATE IN VAL30MET HEREDITARY TRANSTHYRETIN AMYLOIDOSIS (HATTR) FROM MALLORCA.

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Background
Mallorca is the third most important focus of hATTR Val30Met (pVal50Met) in Europe. Hither, previous epidemiological studies estimated the disease prevalence at 5/100,000 inhabitants, including patients with early (< 50 years) and late age of onset (≥ 50 years). The usual phenotypic presentation is a progressive axonal sensorimotor polyneuropathy, although some late onset patients present an associated cardiomyopathy in a so called mixed phenotype (Réines JB et al 2016). The reason for such phenotypic variation between patients, eventually within the same family, remains non elucidated. Acknowledge the risk of being affected (penetrance) for gene carriers is important to guide their management across time, allowing early diagnosis and therapeutic initiation timely.

Aim
Estimate Penetrance in a large sample of Val30Met hATTR families from Mallorca.

Materials and methods
Relevant genealogical data were collected, between 2002 and 2018, from Val30Met hATTR families monitored at the Hospital Universitario Son Llàtzer, referral center for hATTR in Mallorca.
The penetrance function was estimated using the non-parametric method based on a survival analysis (NPSE). In addition, risk estimates were analysed according to gender and to the parent of origin (POO) computed as covariates in a Cox model.

Results
Among, 51 unrelated Val30Met hATTR families initially investigated, 48 (928 subjects) were retained for the analysis, including 147 patients (86 men), 123 asymptomatic carriers (53 men) and 20 obligate carriers (10 men). In total, the rate of genotyped individuals in the families was 42%.
Mean age at onset (AO) (± Standard Deviation) was 49 (±15) years, with a wide range from 22 to 79 years and no difference of AO according gender.
Overall, the disease risk was below 7% [Confidence interval: 3-10] before 30 years-old, increasing progressively i.e. 25% [19-31] at age 50 years then more rapidly, up to 74% [61-83] at 90 years.
Penetrance was similar in both gender. Also, we found similar risks in case of maternal versus paternal inheritance. These latter findings contrast with the POO effect previously reported in the Portuguese and Swedish Val30Met-hATTR families.

Conclusion
The present study gives first insights on penetrance estimates in a representative sample of Val30Met hATTR families from Mallorca. Herein, our analysis showed intermediate risks increasing from the age 30 years, reflecting the mixed clinical picture, including patients with early and late AO. Penetrance remained incomplete at age 90 years. No gender effects could be detected in this sample. Our data are of importance to guide genetic counselling and monitoring of asymptomatic gene carriers in this area of high disease prevalence.
PM010

ANALYSIS OF PLASMA CELL CHARACTERISTICS IN PATIENTS WITH SYSTEMIC LIGHT-CHAIN AMYLOIDOSIS

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Objective: To investigate the number and immunophenotypic characteristics of malignant plasma cells in bone marrow and their relationship with disease risk stratification in patients with primary systemic light chain amyloidosis (AL).

Methodology: Using multiparametric flow cytometry (MFC) to measure the expression of CD19, CD38, CD138, CD56, CD117 and κ/λ light chain restriction of plasma cells in all patients. Patients with monoclonal gammopathy of renal significance (MGRS) and multiple myeloma (MM) were selected in control group. Laboratory examination results were collected to observe the clinical characteristics of AL patients.

Results: A total of 119 AL patients and a control group consist of 49 patients with non-AL MGRS (27 cases of Proliferative glomerulonephritis with monoclonal Ig deposits, 16 cases of monoclonal immunoglobulin deposition disease, 3 cases of light chain deposition disease, 2 cases of heavy chain deposition disease, 1 case of heavy and light chains amyloidosis) and 19 patients with MM are enrolled in this study. Among all AL patients 96(80.7%) cases were found out light chain restriction, the positive expression rate of CD19, CD56, CD117 on plasma cell surface was 10.4% 71.9% 46.9%, respectively. The number of bone marrow plasma cells in AL patients was higher than which in non-AL MGRS patients($P<0.001$) and lower than that in MM patients($P<0.001$), compared with the latter two, the positive rate of CD117 of monoclonal plasma cells in AL patients was higher ($P=0.001, P<0.001$). The number of plasma cells in AL patients with CD19 negative on monoclonal plasma cells’ surface was higher than that in patients with CD19-positive monoclonal plasma cells($P=0.013$). Furthermore, based on Mayo staging system, the highest proportion of monoclonal plasma cells in AL patients was 0.98% (0.6%, 1.37%) in stage IV, and the lowest was 0.53% (0.29%, 1.03%) in stage II. The difference between the two groups was statistically significant ($P=0.033$). The positive rate of CD117 was higher in stage IV patients than in stage II and III patients ($P=0.032, P=0.021), and higher in stage I patients than in stage II patients ($P=0.029$).

Conclusion: The number and immunophenotypic characteristics of monoclonal plasma cells in AL patients are different with those in non-AL MGRS and MM patients. Besides, patients with high Mayo stage in AL have higher clonal plasma cell load and CD117 positive rate. Altogether, using multiparametric flow cytometry to detect the number of bone marrow plasma cells and their immunophenotypic characteristics can helping diagnosis and risk assessment of AL.

Key words systemic light-chain amyloidosis multiparametric flow cytometry monoclonal plasma cell.
EUROPEAN PROTEOMICS AMYLOID NETWORK (EPAN), A MULTICENTER COLLABORATION TO STANDARDIZE METHODS AND IMPROVE KNOWLEDGE ON DISEASE MECHANISMS.

P. Mauri1

1 EPAN.

Introduction: Amyloidosis is a protein misfolding disease where the pathogenesis, diagnosis and treatment is dependent on the type of protein deposited in affected tissues. Proteomics is now used in a number of centers for fundamental research into the mechanisms of amyloidosis, studying the events leading to amyloid-related tissue damage and for evaluating pharmacological therapies. Identification of the deposited amyloidogenic protein is central to clinical diagnosis, and proteomics, pioneered at the Mayo Clinic, is also now being applied for amyloid typing; indeed, proteomics has been suggested as the new gold standard for characterizing amyloid deposits to replace immunological approaches such as IHC and immunofluorescence. There are a number of issues with amyloid proteomics that need to be considered and agreed before it can be fully accepted as an accredited clinical diagnostic test.

Objectives: The European Proteomics Amyloid Network (EPAN) was inaugurated in December 2017 at a meeting in London as a forum to discuss best practice in the application of amyloid proteomics and allied techniques both for clinical diagnosis and to understand the natural history of the disease.

Results: There have been two meetings of EPAN: in London 2017 and Uppsala 2019. These involved representatives from France, Germany, Italy, Netherlands, Spain, Sweden, Switzerland and the UK. Research updates were received from each center and a number of issues were discussed including data sharing, sample exchange, databases, quantitation, protocol standardization and accreditation. Some of the common challenges in applying proteomics for clinical diagnosis were covered. Three main areas have been selected to initiate inter-center studies: Data exchange: the initial focus of this group was to exchange LC-MS/MS data between groups and evaluate performance of different software platforms (Mascot, Proteome Discoverer, MaxQuant, Peaks) for identifying amyloid. MS data was shared between the London and Milan centers and showed good reproducibility in terms of amyloid identification. Sample exchange: it was agreed that different tissue preparations, mainly, fat and dissectates from FFPE would be analyzed at different centers. This would act as a basis for an inter-laboratory validation scheme. Database preparation: a global database will be prepared focusing on the immunoglobulin light chain sequences. Preliminary results will be reported.

Conclusion: As proteomics becomes more commonly applied, particularly in the absence of supporting information such as immunohistochemistry and genotyping, it will become important to understand the underlying issues to share good practice and agree common European guidelines on using the technique in clinical diagnosis. EPAN was set up to forward these aims. The next meeting will be in Kiel in 2020/2021 where we welcome all those interested in amyloid proteomics to participate.

Keywords European Amyloid Proteomics Network, MS-based proteomics, databases, computation
INCREASED STRUCTURAL FLEXIBILITY OF APOA-I AMYLOIDOGENIC VARIANTS IN SMALLER HDL PARTICLES IMPROVES THE CHOLESTEROL EFFLUX ABILITY

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Apolipoprotein A-I (ApoA-I) of high-density lipoprotein (HDL) is a protein endowed with a plethora of health promoting effects, such as anti-inflammatory, antioxidant and atheroprotective activities. However, specific ApoA-I mutations are known to cause a hereditary, autosomal dominant, late onset amyloidosis. Carriers of such ApoA-I amyloidogenic mutations do not show higher risk of developing cardiovascular diseases although their ApoA-I and HDL-cholesterol levels are lower compared to healthy subjects [1-3]. The molecular and structural basis for the improved catalytic function of the ApoA-I amyloidogenic variants is not yet understood.

Here we aimed at providing an explanation to the paradox observed in the clinical phenotype by correlating the structural properties of ApoA-I amyloidogenic variants in HDL particles with their ability to mediate cholesterol mobilization.

Serum samples from Leu75Pro (L75P) and Leu174Ser (L174S) carriers, as well as from healthy donors, were analyzed by western blot and tested for their ability to stimulate cholesterol efflux from macrophages. Reconstituted 8.4 and 9.6 nm HDL nanoparticles (rHDL) containing ApoA-I variants were also used for cholesterol efflux experiments and were structurally characterized by synchrotron radiation circular dichroism and hydrogen-deuterium exchange followed by mass spectrometric analysis. Finally, conventional circular dichroism was employed to measure rHDL thermal stability.

We found that carriers of ApoA-I amyloidogenic variants were characterized by a peculiar HDL particle profile, showing a higher relative abundance of the 8.4 nm vs 9.6 nm particles, compared to the healthy donors. Moreover, serum from L75P and L174S-ApoA-I carriers showed a higher capacity to stimulate cholesterol efflux from macrophages, with respect to serum from control subjects. Finally, low-resolution structural analysis of the rHDL particles revealed that L75P- and L174S-ApoA-I proteins in 8.4 nm rHDL are characterized by an altered secondary structure composition and a more relaxed binding to lipids in the region close to the mutation site, as compared to their native counterpart. Importantly, these structural differences and increased flexibility did not affect particle stability.

These results prove that the unfavourable lipid profile of carriers of ApoA-I amyloidogenic variants is counterbalanced by a higher amount of smaller HDL particles, which are particularly efficient at mediating cholesterol efflux from macrophages. This improved functionality appears to be due to an overall increased higher structure plasticity.

VAL122ILE ATTR ITALIAN FOUNDER POPULATION GENETIC ANCESTRY ANALYSIS, NON ONLY AN AFRO-AMERICAN PATHOGENETIC VARIANT

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Background: Transthyretin amyloidosis is a life-threatening disorder caused by the deposition of TTR amyloid in various tissues and organs. The most common worldwide pathogenic variant with almost exclusive cardiac involvement is Val122Ile (rs76992529), with an allele frequency of 3.5% in the U.S. African-American population, but rare in Caucasians. Unexpectedly, we identified 23 Caucasian individuals with Val122Ile in our amyloidosis referral center (9 affected patients, 14 carriers), belonging to 9 unrelated families.

Objective: To determine the ancestral origin of the Tuscan founder population of TTR Val122Ile carriers.

Methods: A total of 24 individuals were included in the analysis (our 23 probands and relatives from Val122Ile families and the Caucasian reference sample NA10851 (CEU – Utah resident with European ancestry). All samples were genotyped using the EUROFORGEN Global AIM-SNP array1, inclusive of 127 highly informative SNPs to infer genetic ancestry. We have performed a principal component analysis (PCA) of the 9 unrelated probands and NA10851, compared with the Phase 3 of the 1000 Genomes Project data, comprising 2504 unrelated individuals from >20 distinct populations.

Results: all our samples but one (from Argentina) cluster very close to the super-cluster of European populations, and distant from the populations of African ancestry. The proband from Argentina and the Caucasian reference sample NA10851 cluster close to Mexicans and Peruvians, and the super-cluster of European populations, respectively, confirming the robustness of the analysis.

Conclusions: Based on this result, we can confidently conclude that our samples from Tuscan families in which the TTR Val122Ile variant segregates are of ancestral European origin, with no mixed African ancestry, implying that the same variant originated in Africans and Europeans independently and not as result of genetic admixture. These findings suggest the presence of a mutational hot spots in TTR, with potential impact on the epidemiology of amyloidosis worldwide.
PM014

IDENTIFICATION AND CRYO-EM STRUCTURAL CHARACTERISATION OF CARDIAC AMYLOID FIBRILS FROM AN AL PATIENT

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Introduction: Systemic light chain amyloidosis (AL) is a life-threatening disease caused by aggregation and deposition of monoclonal immunoglobulin light chains (LC) in target organs. Severity of heart involvement is the most important factor determining prognosis. The abundant accumulation of amyloid deposits in the heart of patients is the hallmark of cardiac AL and it leads to abnormal thickening of heart walls and to poor heart performance. Thus, characterising the architecture of AL amyloid fibrils is of paramount relevance to understand the molecular bases of AL amyloidosis. Moreover, LC molecules present in amyloid deposits are mostly proteolysed, however, the molecular bases of such observation are poorly characterised.

Objectives: Consequently, we aim to determine the fibrillar architecture of amyloid deposits from a relevant ex vivo sample such as the heart of an AL patient.

Methods: Amyloid fibrils were isolated from left ventricle specimens acquired during autopsy. The monoclonal amyloidogenic LC responsible for such deposits, labelled AL55, was sequenced from its coding mRNA from bone marrow plasma cells. The fibrillar sample was analysed using a proteomic approach in order to determine the LC fragments present in the amyloid deposits. Finally, fibrils were analysed by single molecule cryo-electron microscopy (Cryo-EM) in order to determine a high-resolution structure of the amyloid fibrils.

Results: The LC responsible for amyloid deposits has been identified as belonging to λ6 isotype which is markedly overrepresented in AL patients. Mass spectrometry analysis of solubilised fibrils indicates that a sizable minority of LC molecules from the deposits are full length however most of the molecules are shorter fragments. The most common cleavage sites were found around residue 129 and 150, both positions are located in LC constant domain (C). Then, the 4.0 Å resolution Cryo-EM of amyloid fibrils was then elucidated. The helical fibrils are composed of a single protofilament, showing typical 4.9 Å stacking and cross-β architecture. Two distinct polypeptide stretches (residues 1-37 and 66-105) both belonging to the LC variable domain (Vl) fit the fibril density. Despite Vl high sequence variability, residues stabilizing the fibril core are conserved through different cardiotoxic Vl, highlighting structural motifs that may be common to other misfolding-prone LCs. The whole constant domain is invisible in the structure fibrillar core, suggesting that it is not relevant for fibril assembly and that it likely lies without a rigid conformation on the surface of fibrils.

Conclusions: Our data shed light on the architecture of LC amyloids, correlate amino acid sequences with fibril assembly, providing the grounds for development of innovative medicines.

Keywords: Light Chain Amyloidosis, amyloid fibril, Cryo-EM structure
UNDERSTANDING THE MOLECULAR BASES OF AMYLOIDOGENIC LIGHT CHAINS SOLUBLE TOXICITY

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Introduction: In light chain amyloidosis (AL), fibrillar deposition of monoclonal immunoglobulin light chains (LCs) in vital organs, such as heart, is associated with severe dysfunction. In addition to the cellular damage caused by fibril deposition, direct toxicity of soluble prefibrillar amyloidogenic proteins has been reported, in particular for cardiotoxicity. Clinical data indicate that lowering circulating level of soluble LCs ameliorates heart performance without reducing the amyloid load. However, the molecular bases of proteotoxicity by soluble LCs have not been clarified.

Objectives: Thus, we aim to shed light on the biochemical and biophysical properties typical of amyloidogenic LCs which in parallel to amyloid aggregation also display cardiotoxicity as soluble species.

Methods: Here, to address this issue, we rationally engineered the amino acid sequence of the highly cardiotoxic LC H6 by introducing three residue mutations, designed to reduce the dynamics of its native state. The resulting mutant (mH6) has been tested first to assess its toxicity in vivo and in vitro compared to its parent protein (H6). Then a thorough biophysical characterisation focused into determining H6 and mH6 fold and kinetic stability and protein flexibility have been performed. Moreover, the crystal structure of mH6 allowed the high-resolution structural characterisation and comparison to dissect differences and similarities between the two LC variants.

Results: We observed that mH6 is less toxic than its parent H6 to human cardiac fibroblasts and C. elegans. Worms incubated with H6 displays lower pharyngeal pumping and oxidative stress while H6 incubation results in vitality reduction of human cardiac fibroblasts. Conversely, the toxicity observed for mH6 is very comparable to the one of non-toxic and non-amyloidogenic LC. The high sequence and structural similarity, together with the different toxicity, make H6 and its non-toxic designed variant mH6 a test case to shed light on the molecular properties underlying soluble toxicity. Spectroscopic data and limited proteolysis indicate that H6 displays poorly cooperative fold, higher flexibility and kinetic instability, and a higher dynamic state in its native fold. The comparison of the crystal structures of H6 and of mH6 indicate that the mutations introduced in mH6 do not significantly alter the LC fold ruling out that the observed effects are due to poorly or differently folded protein.

Conclusions: Taken together, the results of this study indicate the existence of a strong correlation between the overall conformational properties of the native fold and the proteotoxicity observed in AL patients with cardiac involvement.

Keywords: Light Chain Amyloidosis, soluble toxicity, protein stability and dynamics
AMYLOIDOGENIC LIGHT CHAIN ENDOCYTOSIS IN RENAL PROXIMAL TUBULAR CELLS LEADS TO IMPAIRED AUTOPHAGY

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Key words: AL amyloidosis, proximal tubulopathy, autophagy

Topic: Genetics and basic science in amyloidosis

Introduction: AL amyloidosis is the result of clonal production of amyloidogenic immunoglobulin light chain (LC) proteins, often resulting in renal failure. Although amyloid fibril deposition of LC proteins is known to cause of renal damage in AL amyloidosis, amyloid precursor proteins might also directly impair renal tubular function at the cellular level, independent of fibril formation. Light chains are actively reabsorbed in the proximal tubular epithelial cells (PTECs) by endocytosis and degraded in lysosomes. Lysosomes are also essential for functional autophagy, a process responsible for the removal of damaged mitochondria (mitophagy) and denatured proteins.

Objectives: Using urine-derived LC (Bence-Jones protein; BJP), we tested the hypothesis that endocytosis of amyloidogenic LC causes PTEC injury by inhibiting tubular cell injury. We also investigated whether the level of autophagic inhibition correlated to BJP stability.

Methods: Cultured primary PTECs extracted from kidneys of Balb/C mice were exposed to 6 separate BJPs, each purified from patient urine. BJPs were derived either from patients with AL amyloidosis featuring associated nephropathy or from a non-amyloid myeloma patient, as control. The apparent molecular weights of the BJPs were assessed by gel electrophoresis and western blotting, which revealed a range of molecular species including monomers, disulfide-linked and non-covalent dimers, fragments and larger species. Autophagic flux was estimated by immunoblot using the autophagy marker LC3-II, in both bafilomycin treated and untreated cells. Autophagosomes were quantified in live cells using fluorescent microscopy, as well as a microplate reader. Mitochondrial respiration and reactive oxygen species production were measured in live cells. Mitochondrial morphology was also assessed using confocal microscopy. The stability of each BJP was assessed using limited proteolysis by a broad spectrum protease, proteinase K, at neutral pH. The extent of protein degradation after two hours of protease treatment was measured by western blotting.

Results: AL patient-derived BJPs caused inhibition of autophagy at various levels while control BJPs did not lead to significant changes in autophagy. BJP-exposed cells accumulated damaged mitochondria with altered mitochondrial respiration and morphology leading to increased ROS production. There was no direct relationship between the apparent stability of each full-length LC and the level of autophagy inhibition observed in the various samples.

Conclusions: Dysfunctional autophagy and mitophagy caused by direct cellular toxicity of BJPs likely contribute to tubular cell toxicity in AL amyloidosis but the extent of inhibition does not seem to correlate with in vitro stability of BJPs assessed by limited proteolysis at neutral pH.
DISENTANGLING LARGE ANTICIPATION IN FAMILIAL AMYLOID POLYNEUROPATHY VAL30MET FAMILIES: A WHOLE-GENOME SCREENING

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Introduction: Familial Amyloid Polyneuropathy (FAP) is an autosomal dominant systemic amyloidosis, showing sensorimotor symptoms and progressive incapacity, leading to death 10-15 years after onset. FAP occurs as a result of an inherited point variant in the transthyretin (TTR) gene, Val30Met being the commonest. Current consensus is in line with the notion that while each disease-causing variant sensitizes the genome to a primary clinical manifestation, additional variants in the genetic background modulate the phenotype expression. This disorder shows anticipation of age-at-onset (AO) - an earlier-onset in the offspring when compared with the affected late-onset parent. This variation suggests that genetic modifiers, either conferring protection or increasing risk, contribute to clinical manifestation in TTR-FAP. A single etiological mechanism is insufficient to explain the causal origins of phenotypic heterogeneity observed in AO, which acts as a complex trait with diverse factors involved.

Objectives: The aim of this study was to explore other genes and mechanisms that may act as AO modifiers in TTR-FAP.

Methods: To accomplish this, we collected DNA samples of TTR-FAP Val30Met families with large anticipation (comprising 3 generations) and performed a whole-genome sequencing. In this extreme phenotypes' design, we compared the genetic profile between extreme AO (early- and late-onset).

Results: Anticipation was obvious in the selected families with a mean decrease in AO of 20 years in each generation, resulting for both families, in a difference of ≈ 40 years in AO between the grandmother and the grandchild. Then, we analyzed an intersection of genetic variants between the families’ core, allowing us to isolate the variants that overlap between families and which will in part constitute the complex genetic architecture of large anticipation. We found 195,850 variant calls with 63% of them being located in intron region. Our analyses are now focused in exonic regions (1.3%) and allowed us to discover new FAP-risk-genes and through a gene-set enrichment analysis, we also identified potential novel pathways as well as significant protein-protein interaction (PPI) network associated with AO of TTR-FAP.

Conclusions: Familial-based deep sequencing provides the most robust approach for definition of the genetic determinants of anticipation that can help us to find new mechanisms related with AO variability in TTR-FAP.

Keywords: Genome; Anticipation; TTR
MELFLUFEN IS A NOVEL ALKYLATING AGENT DEMONSTRATING TOXICITY TO AMYLOIDOGENIC PLASMA CELLS

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Introduction: Plasma cell directed therapeutics for immunoglobulin light-chain (AL) amyloidosis, which are aimed at preferentially eliminating the clonal population of amyloidogenic cells in bone marrow, are expected to reduce production of toxic light chain and alleviate deposition of pathogenic amyloid fibrils, thereby restoring organ function.

Melphalan fluifanamide ethyl ester, melflufen, is a highly lipophilic, peptidase potentiated alkylating agent with rapid cellular uptake and potent toxicity in myeloma cells. Previously it was demonstrated that multiple myeloma plasma cells are sensitive to melflufen, suggesting that the drug might be useful to directly eliminate amyloidogenic plasma cells, thereby reducing the amyloid load in patients.

Objectives: To assess potential efficacy in amyloidosis patients and to explore the mechanism of action, we examined effects of melflufen on amyloidogenic plasma cell lines as well as primary patient samples.

Methods: The degree and mechanism of cellular toxicity and effects on the unfolded protein response (UPR) were measured in response to either melflufen or melphalan in multiple light chain secreting malignant human plasma cell lines by western blot and qRT-PCR for UPR signature genes. Primary CD138+ plasma cells from bone marrow of amyloidosis patients were analyzed by single cell sequencing to examine gene signatures associated with melflufen sensitivity in amyloidogenic plasma cells.

Results: Melflufen demonstrated increased potency in multiple myeloma cell lines compared to melphalan, inducing malignant plasma cell death on established light chain secreting plasma cell lines. ALMC-1 and ALMC-2 cells displayed resistance to melphalan, but demonstrated an IC50 of approximately 1 µM to melflufen, with a significant reduction in light chain production, while JJN-3 cells were sensitive to melphalan, with an approximate 5 fold increase in sensitivity to melflufen. Melflufen sensitivity was associated with enhanced plasma cell apoptosis, with differing effects on the UPR pathway compared to melphalan. Analysis of ex vivo primary CD138+ bone marrow plasma cells from 10 individual patients with amyloidosis demonstrated similar superior melflufen sensitivity, and single cell sequencing of these samples explored gene signatures specific to sensitive samples, including genes associated with melflufen processing and mechanism of action.

Conclusions: These findings provide evidence that melflufen mediated toxicity, previously described in myeloma cells, extends to amyloidogenic plasma cells and elucidates the mechanism of action, supporting the rationale for the evaluation of melflufen in patients with AL amyloidosis.
DIVERSE CHEMOTYPES OF SMALL MOLECULES THAT STABILIZE AMYLOIDOGENIC IMMUNOGLOBULIN LIGHT CHAINS

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Background: Light chain amyloidosis (AL) is caused by the misfolding and aggregation of monoclonal light chains (LCs), a process which leads to organ damage. No treatments are specifically approved for AL, but the current standard of care is the use of cytotoxic chemotherapy designed to eradicate the monoclonal plasma cell expansion that secretes LCs. However, many patients, especially those with cardiac involvement, are too sick to tolerate these regimens, and others relapse after hematologic response. Thus, new efficacious therapies for AL are needed.

Objectives: Aggregation-prone LCs have low kinetic stability, i.e. high rates of transient unfolding from the natively folded, nontoxic LC into aggregation-competent conformations. We hypothesize that small molecule kinetic stabilization of unstable, amyloidogenic LCs would prevent the misfolding and aggregation that causes organ damage in AL patients. This novel strategy would be complementary to current and emerging treatments for AL. The analogous kinetic stabilizer drug tafamidis has been clinically successful in treating the transthyretin amyloidoses.

Methods: We developed a high-throughput screening assay and used it to identify small molecules that protect an amyloidogenic LC from aberrant proteolysis enabled by conformation excursions, a measure of kinetic stability. We characterized the hits from the screen using X-ray crystallography, nuclear magnetic resonance (NMR), and measurement of dissociation constants.

Results: We identified five chemotypes of small molecules that stabilize LCs from aberrant proteolysis in vitro. The screening hits have modest affinity for LCs (low micromolar dissociation constants). X-ray crystallography and NMR data show that the molecules bind the variable domain dimerization interface and interact with residues that are highly conserved among amyloidogenic LC sequences. We identified two small molecule binding pockets: a deep cavity at the core of the dimer interface, and a cavity near the complementary-determining regions. The latter cavity is unoccupied for some hits or only partially occupied for others, and thus could represent a starting point for medicinal chemistry-based optimization of the kinetic stabilizers.

Conclusions: We have identified small molecules of diverse chemotypes that kinetically stabilize amyloidogenic LCs. We will utilize conserved residues in the binding site and a substructure combination strategy to develop highly potent and specific kinetic stabilizers that could become drug candidates for AL.

Keywords: AL amyloidosis; drug development
Category: Genetics and basic science in amyloidosis
DEFINING MOLECULAR SIGNATURES OF MUTATION-SPECIFIC ORGAN TROPISM IN HEREDITARY TRANSTHYRETIN AMYLOIDOSIS

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Background: Hereditary ATTR amyloidosis is a devastating, multi-system protein folding disorder resulting from >100 described mutations in the transthyretin (TTR) gene. Complicating the development of broadly efficacious therapeutics, patients with the disorder exhibit extreme mutation-specific phenotypic variation in disease pathology. Highlighting this point, some mutations (e.g. TTRV122I) typically manifest in amyloid deposits in cardiac tissue with resultant cardiomyopathy, while others (e.g. TTR255P) produce neuronal-directed amyloid deposits throughout the peripheral nerves.

Objectives: In these studies, we sought to identify molecular signatures resulting from exposure to pathologically-diverse TTRs. In doing so, we investigated 1) how target cells respond when exposed to pathologic TTRs in the presence and absence of the kinetic stabilizer tafamidis and 2) how this response changes across diverse TTR variants and target cell types.

Methods: Neuronal (SH-SY5Y), cardiac (AC16), and hepatic (HepG2) cell lines were exposed to wild-type, neuropathy-, and cardiomyopathy-associated TTRs in the presence and absence of tafamidis. Bulk RNA sequencing (RNAseq) was performed to identify distinct transcriptional signatures resulting from exposure to each TTR. In parallel, following TTR dosing, changes in chromatin architecture, known to contribute to the regulation of gene expression, was assessed via ATACseq. As positive controls for cellular stress, cells were subjected to heat shock and exposed to the global ER stressor, thapsigargin.

Results: Transcriptomic profiling revealed molecular signatures resulting from exposure to each independent TTR. Interestingly, when cardiac- or neuronal-specific TTRs were exposed to their clinically-associated cell type (e.g. cardiomyopathy-associated TTRV122I on cardiac cells), distinct transcriptional signatures emerged which were not seen in mismatched pairings (e.g. TTRV122I on neuronal cells). These tissue-specific signatures were eliminated in the presence of tafamidis. Moreover, exposure to diverse TTRs also resulted in distinct alterations in chromatin architecture. Inducing global ER stress via addition of thapsigargin and heat shock similarly resulted in gross epigenetic remodeling.

Conclusions: Using next generation genomic approaches, we identified distinct molecular changes as a result of exposure to diverse, misfolded TTRs. Moreover, different TTRs elicit different responses in the same cell type, suggesting that the well-documented, mutation-specific organ tropism phenomenon could result from differences in how each target cell type responds to its associated pathogenic TTR. Furthermore, target cell types involved in ATTR amyloidosis pathogenesis respond to the presence of TTR differently at the transcriptional and epigenetic levels. Lastly, we demonstrate alleviation of novel stress signatures via addition of kinetic stabilizer tafamidis.

Keywords: hereditary ATTR amyloidosis; organ tropism; genomics
INVESTIGATION OF ADIPOCYTE DYSFUNCTION IN AL AMYLOIDOSIS PATIENTS WITH AMYLOID DEPOSITION IN SUBCUTANEOUS ABDOMINAL FAT


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Introduction: Amyloid deposits frequently infiltrate white adipose tissue in systemic amyloidosis; abdominal subcutaneous fat is indeed the tissue electively acquired for demonstrating amyloid fibrils. White fat is a crucial organ for energy storage and mobilization, with important endocrine activity. Weight loss and malnutrition are typical features of amyloidosis and increase treatment toxicity; however, how amyloid deposition could affect the function of adipocytes is understudied. Adipokines, i.e. adipocytes-secreted hormones, are involved in a variety of paracrine and endocrine activities. To date, the only quantitative parameter used to assess nutritional status has been body mass index (BMI).

Objectives: In this pilot study, we aimed to explore adipocyte dysfunction in systemic AL amyloidosis patients with fat tissue involvement, through: 1) measurement of circulating levels of adiponectin and leptin, and evaluation of their correlation with modified BMI (mBMI); 2) assessment of adipocyte size and morphology in fat tissue samples.

Methods: Stored frozen plasma samples from consecutive unselected patients with newly diagnosed AL amyloidosis and from controls, with available baseline samples and complete clinical characterization, were analyzed. Levels of adiponectin and leptin in plasma were measured by commercial ELISA assays. Size and morphology of adipocytes in fat aspirates was histologically evaluated.

Results: Plasma samples from 50 AL patients (42 ALλ, 8 ALκ) and 44 individuals without systemic amyloidosis were analyzed. Median plasma adiponectin levels are significantly higher in patients compared to controls (13.5 µg/ml vs 7.6 µg/ml, p<0.05), whereas plasma leptin is lower in patients (median 4 ng/ml vs 14.1 µg/ml, p<0.05). The adiponectin/leptin (A/L) ratio was also significantly higher in patients (median 3.26, interquartile range 1.7-10.2) than in controls (median 0.54, interquartile range 0.27-0.92). The A/L ratio was inversely correlated with mBMI both in patients and controls. In patients, cell size is smaller, and cell morphology is altered, with loss of the normal cell-to-cell contacts and deformation of the adipocytes’ balloon-like shape.

Conclusions: This proof-of-concept study suggests that: 1) the adipokine status of AL patients with amyloid deposition in fat differs from that of individuals without systemic amyloidosis; 2) these hormones are informative indicators of the nutritional status of amyloidosis patients and could complement mBMI; 3) adipocyte function may be altered in presence of amyloid deposits. This information, through larger ongoing studies, could cast new light on the pathogenesis of metabolic alterations in amyloidosis, and help improve patients’ care and therapeutic management.

Keywords: adipocytes, adipokines, nutritional status
EVIDENCE FOR AN ANCIENT ORIGIN OF THE FGA p.Glu545Val (E526V) AMYLOIDOSIS-CAUSING MUTATION ENDEMIC IN NORTHERN PORTUGAL

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Fibrinogen alpha chain amyloidosis is widely spread throughout the world, and is most frequently associated with the FGA p.Glu545Val (E526V) mutation, particularly in European countries, with endemic foci of the disease identified in the UK and in northern Portugal. All identified Portuguese patients are from the same region, and a preliminary attempt to characterize the disease-associated haplotype hinted at a common ancestor, but whether this is true and how far back in time the founding event would have taken place is still much an open question.

In order to address these questions, we studied all available Portuguese patients and relatives, 56 individuals in total, 33 of which were mutation carriers, belonging to 12 extended families. Thirteen polymorphic short tandem-repeats spanning 5.8Mbp over the FGA gene in chromosome 4 were genotyped. A control population of 67 unrelated individuals was also genotyped for the same polymorphisms.

Haplotype phasing was carried out using an empirical linkage disequilibrium-based method implemented in the Beagle 4.1 computer program, with some manual adjustments to take into account pedigree constraints and to preserve parsimony. In total 7 different but closely related disease-associated haplotypes were identified, the most frequent of which (I), represented in 5 families, was presumed to be the ancestral haplotype (Table 1).

Table 1

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The age of the E526V mutation in this population was estimated by fitting a multipoint LD model, as implemented in the DMLE+ program. While this model is somewhat sensitive to estimates of population growth and other parameters, it consistently predicted a mutation age above 100 generations (2500 years). These results point to a relatively ancient mutation, which could explain, at least in part, its wide dissemination throughout the world. It would be interesting to extend this study to other populations, to see if there is evidence for a common ancestor, and to try to establish a pattern of mutation dissemination.

Keywords: Fibrinogen Amyloidosis; Haplotype; Mutation Age.
A STRATEGY FOR THE SELECTION OF qPCR REFERENCE GENES TO STUDY BONE MARROW DERIVED PLASMA CELLS FROM PATIENTS WITH AL AMYLOIDOSIS

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Background: In the next generation sequence era, quantitative PCR (qPCR) is still widely employed for both basic research and molecular diagnostics due to its ease of use, versatility and limited costs. Measurement of transcriptional levels through qPCR critically depends on reference genes used for normalization purposes, whose selection has to be specific for each clinical setting. To date, a systematic screening for reference genes in bone marrow (BM)-derived CD138+ plasma cells (PCs) from AL amyloidosis patients is missing.

Objectives: Here, we devised a strategy to identify and validate reference genes for transcriptional studies of BM-derived CD138+PCs from patients with AL amyloidosis or control subjects.

Methods: We performed a systematic review of published literature to compile a list of 150 candidate reference genes for qPCR experiments in human studies. Next, we interrogated the Gene Expression Omnibus (GEO) data repository to assess expression levels of these genes in two published transcriptomic studies on BM-derived CD138+PCs from patients with different plasma cell dyscrasias, including AL. For the top 14 most stable genes, we iteratively used Primer Blast, Ensembl, dbSNP and OligoAnalyzer to identify primers devoid of SNPs and lacking thermodynamically significant secondary structures and homo- and heterodimer formation. We used cDNA prepared from the AL plasma cell line ALMC-2 to perform technical validation analyses in accordance to MIQE guidelines. Genes were ranked according to their stability values using the geNorm software to identify the most stably expressed genes in AL patients and healthy controls.

Results: We designed primers for each of the 14 candidate genes. Technical validation experiments identified 6 genes out of 14 (ACTR3, ALG9, ESD, HSPA5, POLR2B, SDHA) with a dynamic range spanning 5 orders of magnitude of cDNA dilution, with PCR efficiency between 91 and 109%, intra- and inter-assay variation coefficient between 0.3 and 1.3% and 0.1 and 0.6%, respectively. Next, we analysed expression profiles of these 6 candidate genes in CD138+PCs isolated through MACS-sorting from diagnostic leftovers of 6 BM samples of AL amyloidosis patients and 3 healthy BM donors. Use of geNorm identified ALG9, ACTR3 and ESD as the most stable reference genes in this clinical setting. Results were confirmed even when their expression stability was compared with that of 5 commonly used reference genes (ACTB, B2M, GAPD, HMBS, HPRT1).

Conclusions: In this study we demonstrate that ALG9, ACTR3 and ESD are suitable reference genes for transcriptional analysis of BM CD138+PCs from patients with AL amyloidosis or healthy subjects. These genes may be instrumental to perform transcriptional studies on primary AL plasma cells, both for diagnostic and research purposes. The strategy presented here may be applicable to other clinical and experimental settings for which publicly available transcriptomic datasets are available.

Keywords: Transcriptional studies, qPCR, AL amyloidosis
AIDA: THE AUSTRIAN INTERDISCIPLINARY AMYLOIDOSIS REGISTRY

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Introduction: Amyloidosis is a severe rare disease that is typically characterized by an interdisciplinary character of both diagnosis, as well as treatment. Cardiological, renal, neurological and hematological manifestations are the most common of many.

As new treatments have become available higher awareness of amyloidosis and efficient diagnostic and therapeutic pathways are urgently warranted. To optimize our understanding of Amyloidosis high-quality capturing of deep “real world evidence” (RWE) data and corresponding bio-banks are essential. Facilitating these aims AIDA was established in 2019.

Methods: AIDA is a web-based clinical database (based on the Software CHES) designed to collect interdisciplinary clinical data concerning diagnosis, treatment, outcomes and survival of patients with the AL, AA and ATTR amyloidosis. Parametrization was carried out in respective interdisciplinary meetings with intensive IT support in a consensual way to define a minimum essential data set plus parameters of scientific interest. Data fields describe diagnostic features (histology, imaging, laboratory findings, etc.) as well as applied treatments (systemic therapy, transplantations, etc.), and supportive measures. Furthermore, the regular collection of electronic patient reported outcomes (ePROs) is planned and technically provided for.

With this poster we want to provide further insight into the development of the Austrian Interdisciplinary Amyloidosis Registry. The roadmap and an overview of the current data and number of amyloidosis patients will be demonstrated at the conference. Furthermore, data fields and administrative structure of AIDA as well as future analytical scenarios will be elucidated.

Data: Currently two Austrian centers contribute to the registry (Departments of Cardiology, Nephrology, Oncology and Hematology of the Medical Universities in Innsbruck and Vienna). Further roll-out is planned. By now 32 amyloidosis patients could be documented up to the abstract deadline. In the course of a systematic search, a backlog of over 250 amyloidosis patients (AL and ATTR) was identified, which are now being processed and documented in AIDA (update on basic data and patient characteristics to be presented at ISA2020) We assume that at least two to four additional centers can be acquired for the registry before the conference. Due to the web-based nature of the bi-lingual (German & English) registry, the inclusion of further centers is possible at any time and does not require much effort.

Conclusions: AIDA represents a unique database that allows monitoring the epidemiologic development and clinical course in patients with amyloidosis on a population level in Austria in an interdisciplinary context. AIDA will help physicians to better understand the nature of amyloidosis and contributes new information on this disease with the collected Real-World Evidence (RWE) and ePRO data.

Keywords: real world evidence (RWE), clinical registry & database, patient reported outcome (ePRO)
IN VITRO ALTERATIONS OF HUMAN CARDIAC CELLS EXPOSED TO AMYLOIDOGENIC CARDIOTROPIC LIGHT CHAINS: FOCUS ON THE CELL MEMBRANE

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Background: In light chain (AL) amyloidosis, heart involvement is common and dictates the prognosis. Established evidence indicates that cell and organ dysfunction is not only due to the effects of deposited fibrils, but also to direct damage by pre-fibrillar amyloidogenic light chains (LCs). Various experimental models to study cardiac LC proteotoxicity have been described (1-3); in vitro, exogenous cardiotropic LCs are internalized by primary human cardiac fibroblasts (hCF) and undergo intracellular trafficking (2,3).

Objectives: We are exploring the alterations occurring in hCF incubated with exogenous cardiotropic LCs, focusing especially on the cell membrane, port of entry of these toxic molecules and whose biochemical/biophysical properties are known to be altered by several misfolding-prone proteins (4).

Methods: Monoclonal amyloidogenic cardiotropic LCs (CT-LCs), and non-amyloidogenic myeloma LCs (MM-LCs) as controls, were purified, by ion-exchange chromatography, from urines of patients, or produced as recombinant proteins in E. coli (5). All patients were fully clinically characterized. Commercial primary human cardiac fibroblasts (hCF) were cultured in serum-free medium and incubated with CT-LCs or MM-LCs; cell damage was evaluated by MTT assay; dose-response curves experiments were performed (LCs concentration range: 1-50 μM). Cell membrane fluidity was investigated using a fluorescent lipophilic probe (pyrenedecanoic acid)-based assay. The effects of representative CT-LCs and MM-LCs on cell membranes composition are being investigated in parallel by FTIR microspectroscopy.

Results: 24 hours incubation with CT-LC significantly decreases MTT reduction by hCF; the extent of damage is linear for CT-LCs (3 distinct CLs) concentrations ranging from 3 to 20 μM, while incubation with MM-LCs (2 distinct CLs) showed no significant effects on cell viability for all concentrations tested. MicroFTIR-based ongoing analyses suggest that the composition of cell membrane lipids may be altered since early incubation time points (30 min) in hCF exposed to CT-LCs. Quantification of pyrenedecanoic acid incorporation by cells incubated with 3 distinct CT-LCs showed a significantly lower intrinsic signal compared to the non-amyloidogenic controls (2 distinct MM-LCs), suggesting that CT-LCs have a negative impact on membrane fluidity.

Conclusions: Our data suggest that exposure to CT-LC significantly affects hCF cellular properties, and in particular the properties of the cell membrane. Further investigation of this aspect will be instrumental to understand the role of this compartment in the pathogenesis of damage.


Keywords: Proteotoxicity; Human cardiac cells; Cell membrane
THE ROLE OF MICRORNAS IN THE PATHOGENESIS OF AL AMYLOIDOSIS

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Background: Systemic light chain (AL) amyloidosis is a clonal plasma cell disorder characterized by the deposition of misfolded immunoglobulin light chain products in vital organs, causing their dysfunction. MicroRNAs (miRNAs) are short, non-coding RNAs that regulate gene expression and have a role in cancer development and progression. MiRNAs can be purified from various biological specimens such as serum, plasma and bone marrow (BM) and may be used as biomarkers to distinguish between cancer patients and healthy individuals. Moreover, miRNA-mRNA interactions may determine the molecular mechanism by which miRNAs and their target genes are involved in AL amyloidosis and may suggest novel therapeutic options for affected patients. To date, knowledge about circulating or BM miRNAs in AL amyloidosis is lacking.

Aims: 1) To decipher specific miRNA signatures in AL amyloidosis patients compared to multiple myeloma (MM) and healthy controls. 2) To examine how miRNAs are involved in AL amyloidosis pathogenesis.

Methods: Plasma samples from peripheral blood were collected from healthy volunteers (n=10) and from newly diagnosed AL amyloidosis (n=15) and MM patients (n=10). BM samples (CD138+ cells) were collected from newly diagnosed AL amyloidosis (n=9), MM patients (n=7) and from healthy volunteers undergoing hip replacement (n=2). MiRNAs and mRNAs expression profiles were determined using the nCounter miRNA assay and RNA-seq respectively, and validated using quantitative RT-PCR. The bioinformatics software Ingenuity Pathway Analysis was used for integrative miRNA-mRNA analysis.

Results: Ten miRNAs were significantly differently expressed in BM samples from newly diagnosed AL amyloidosis patients compared to MM patients (Table 1). In plasma samples, 10 miRNAs were significantly differently expressed between AL amyloidosis and MM patients. Differential miRNA expression was found between healthy controls and AL amyloidosis or MM patients (Table 2).

Analysis of the mRNA expression profile of BM cells from AL amyloidosis and healthy controls showed differential expression of >2000 genes. Crossing the miRNAs’ binding site sequences (seed region) with their mRNA target sequence, revealed a potential involvement of miRNAs in regulating key signaling pathways related to cell cycle regulation and anti-apoptosis mechanisms including interleukin-4 signaling, interleukin-13 signaling, activation of MAPK and PI3K/AKT pathways and over-expression of BCL2.

Conclusions: We provide preliminary insight into the molecular mechanisms mediated by miRNAs and the aberrant expression of oncogenic/tumor suppressor genes. The differential expression of miRNAs in AL amyloidosis and MM, compared to healthy controls, may be used to understand disease pathogenesis and predict risk of progression to AL amyloidosis among patients with known plasma cell disorders. Additionally, the signaling pathways found to be involved in AL amyloidosis may assist in tailoring more specific treatments.

Keywords: Microrna, AL Amyloidosis
**Table 1.** Comparison of down- and upregulated miRNA expression in BM samples from AL amyloidosis and MM patients

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**Table 2.** Comparison of down- and upregulated miRNA expression in plasma samples from ALA and MM patients and healthy controls

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**Table 3.** Comparison of down- and upregulated miRNA expression in plasma samples - downregulated miRNAs

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Background: AL Amyloidosis is a multisystem disorder of clonal plasma cells (PCs) that produce an abnormal light chain, which misfolds and deposits in organs causing cellular stress, organ dysfunction, and eventually, death. The amyloidogenic PCs reside within the bone marrow microenvironment in close proximity to the cells of the innate and adaptive immune system. Changes in the composition and increased expressed of T cell inhibitory molecules is observed with the evolution of other related plasma cell proliferative disorders including the transition from Monoclonal Gammopathy of Undetermined Significance (MGUS) to Smoldering Multiple Myeloma (SMM) to MM thereby playing a role in disease progression. Immunosuppressive cells are also increased within the MM microenvironment and play an important role in tumor progression primarily through their capability to promote immune-escape, angiogenesis, drug resistance, and metastasis. Plasma cell disorders such as MGUS, SMM and MM and AL amyloidosis share similarities but they also demonstrate distinct differences. For example, clonal PCs in AL amyloidosis have similar phenotypic and copy number alteration profiles as those in MM, but their transcriptome is similar to that of normal PCs. Furthermore, the mutational pattern of AL amyloidosis is intermediate between those of MGUS and MM when studied using next-generation sequencing approaches. Given the presence of genomic instability within the amyloidogenic PCs, the immune system may have the capacity to recognize these lesions. The immune landscape of AL amyloidosis is not well characterized.

Methods: We performed flow-based longitudinal assessment of T cell inhibitory markers CCR7 and CD45RA as well as CD8 and PDL1 among three patients with different response to treatment as a pilot study.

Results: Patient A with persistent disease had the highest amount of PDL1 expression on CD8+ cytotoxic T cells (Figure 2) and had least numbers of CD45RA+CCR7+ CD8 effector T cells (Figure 1). Patient C, who attained a CR to therapy, had the lowest amount of PDL1 expression on CD8+ cytotoxic T (Figure 2) cells and had the most increase in CD45RA+CCR7+ CD8 effector T cells (Figure 1). Patient B who attained VGPR to therapy, had an intermediate amount of PDL1 expression on CD8+ cytotoxic T cells (Figure 2) and CD45RA+CCR7+ CD8 effector T cells (Figure 1).

Conclusion: This data would suggest a signal of impaired immunity within the AL amyloidosis patients who respond poorly to therapy and remain at risk of persistent/progressive disease. We are currently evaluating the immune profile of AL amyloidosis in a larger cohort to validate and better understand the immune landscape of light chain amyloidosis.
BCMA EXPRESSION IN AL AMYLOIDOSIS

S. Bal 1, A. Sigler 1, A. Chan 1, D. Chung 1, H. Hassoun 1, A. Dogan 1, S. Giralt 1, H. Landau 1
1 University of Alabama at Birmingham.

Introduction: AL Amyloidosis is a multisystem disorder of clonal plasma cells (PCs) that produce an abnormal light chain which misfolds and deposits in organs causing cellular stress leading to organ dysfunction and eventually death. Available therapies target plasma cells to stop the production of amyloidogenic light chains. The burden of clonal plasma cells in Bone Marrow (BM) is typically smaller than what is seen with Multiple Myeloma (MM) and potentially advantageous for immunotherapeutic strategies. B-Cell Maturation Antigen (BCMA) is a transmembrane protein that is involved in the regulation of B cell proliferation and survival as well as maturation/differentiation into PCs. Given the efficacy of therapies targeting BCMA in MM, we evaluated the expression of BCMA on the surface of clonal plasma cells in AL amyloidosis, which has not been studied thus far.

Methods: We identified all patients with AL amyloidosis who had available unstained bone marrow specimens at Memorial Sloan Kettering Cancer Center between 2012 and 2018. These specimens were stained for BCMA expression using immunohistochemistry (IHC; clone: D6, catalog: sc-390147, company: Santa-Cruz, monoclonal, dilution 1:400) with a clinical grade assay performed in a CLIA compliant setting. The samples were scored for expression, intensity, and site of staining.

Results: We identified twenty-eight diagnostic and six relapsed samples available for staining. The median age of the population was 63 years (range 41-73 years). There were 64% males and 36% females in the cohort. Lambda typic PCs were seen in 75% patients. By fluorescence in situ hybridization (FISH), t (11;14) was present in 36% patients, and gain of chromosome 1q and del 13q were each seen in 32% of patients. No patients in the cohort had t (4;14) or del 17p. At diagnosis, the median clonal PC percentage in the bone marrow was 10% (range 2-80%) with 64% having less than/equal to 10% plasma cells. The median BCMA expression was 80% (range 20-100%). The staining was predominantly membranous in 82% of patients and had a Golgi pattern in 11% with the median staining intensity of 2 (range 1-3). Six patients with diagnostic tissue also had relapsed bone marrow samples available. Among these patients, the median plasma cell burden at diagnosis was 35% (range 10-80), with majority of patients (83%) having greater than 10% clonal PCs. The median BCMA expression was 65% (range 50-80) at the time of diagnosis with 50% membranous staining, 17% had Golgi staining pattern, and 33% had Golgi-membranous staining. The median staining intensity was 1 (range 1-2). At the time of relapse, the median clonal PC burden was 13% (range 5-30). Median BCMA expression was 75% (range 50-100) with predominantly membranous staining (83%) and the median intensity of staining 1.

Conclusions: Our study represents the first description of BCMA expression on the surface of amyloidogenic PCs to our knowledge. BCMA is uniformly expressed by pathologic plasma cells in AL amyloidosis and remains present at the time of relapse. BCMA directed approaches may represent an effective therapeutic option in AL Amyloidosis and should be explored in the future.
**BLINDED POTENCY COMPARISON OF TRANSTHYRETIN KINETIC STABILIZERS IN HUMAN PLASMA**


**Background:** Wild-type (WT) transthyretin (TTR) misfolding and aggregation, manifesting in cardiomyopathy, is the most common TTR amyloid disease. Tafamidis is regulatory agency approved for use in slowing the progression of mutant and WT TTR cardiomyopathy by way of kinetic stabilization of the native, non-amyloidogenic TTR tetramer. Claims have been made that other kinetic stabilizers are superior to tafamidis.

**Objectives:** We performed a blinded potency comparison of TTR kinetic stabilizers added to healthy human plasma at several carefully determined, fixed, pharmacologically relevant concentrations.

**Methods:** We employed a subunit exchange assay conducted in pooled human plasma (derived from 5 healthy WT TTR donors) to compare the kinetic stabilizers AG10, tolcapone, tafamidis and diflunisal at 1, 5, 10 and 20 µM concentrations. This experiment facilitates rigorous quantification of pharmacologic tetramer kinetic stabilization (rate of tetramer dissociation) under physiological conditions in plasma wherein transthyretin likely aggregates in humans. Notably, tetramer dissociation is the rate-limiting step for TTR subunit exchange and TTR aggregation. The plasma concentration of the drug candidate and drugs was assessed by two independent methods to be sure that accurate kinetic stabilizer concentrations were used. One of the concentration measurements was performed by the core mass spectrometry laboratory at Scripps.

**Results:** In our poster, we will reveal triplicate head-to-head comparisons of the drug candidate AG10, and the drugs tolcapone, tafamidis and diflunisal in pooled healthy human plasma at the above mentioned, pharmacologically relevant small molecule kinetic stabilizer plasma concentrations. The extent of kinetic stabilization in human plasma will be correlated with the oral doses necessary to achieve said stabilization.

**Conclusion:** Our poster will report the relative potencies of the drug candidate AG10, and the drugs tafamidis, tolcapone and diflunisal towards WT TTR kinetic stabilization in human plasma. Other important pharmacologic considerations to further distinguish these small molecules will also be presented to provide perspective. Among these attributes are: safety (very important for daily kinetic stabilizer treatment over a decade or more), daily oral doses required to achieve complete kinetic stabilization of WT TTR in blood, pharmacokinetic properties, metabolites produced, and eye as well as blood-brain barrier permeability.

**Keywords:** TTR amyloidosis; cardiomyopathy
COLLAGEN ADDITION TO SYNTHETIC AMYLOID FIBRILS PRESENTS A “DON’T EAT ME” SIGNAL THAT PREVENTS MACROPHAGE PHAGOCYTOSIS

J. S. Foster 1, T. Richey 1, A. D. Williams 1, D. C. Wooliver 1, S. Macy 1, E. B. Martin 1, R. E. Heidel 2, S. Kania 3, S. J. Kennel 1, J. S. Wall 1

1 Department of Medicine, University of Tennessee Medical Center.
2 Department of Surgery, University of Tennessee Medical Center.
3 Department of Biomedical and Diagnostic Sciences, University of Tennessee College of Veterinary Medicine.

Introduction: Light chain-associated amyloidosis (AL) is a complex, compositionally heterogeneous, acellular material composed of immunoglobulin light chain fibrils in association with extracellular matrix components and more than 20 serum proteins, notably serum amyloid P, apolipoproteins and perlecan. Despite the non-native structure of AL fibrils, phagocytic cells of the innate immune system do not effectively eliminate this material in patients. Consequently, amyloid accumulation is generally progressive, leading to severe organ dysfunction and failure. We have recently shown that synthetic amyloid fibrils composed of a recombinant human $\lambda_6$ variable domain (rV$\lambda_6$Wil) are rapidly phagocytosed by macrophages when injected subcutaneously into nude (NU/NU) mice. In contrast, human AL$\lambda$ amyloid extract was resolved less quickly in the same model system. We, therefore, hypothesized that accessory molecules in the amyloid extract hindered the uptake and clearance of the material by macrophages.

Objectives: The goal of this study was to investigate the importance of amyloid-associated accessory molecules on macrophage-mediated phagocytosis in vitro.

Methods: Synthetic rV$\lambda_6$Wil fibrils were prepared by shaking in phosphate buffered saline at pH 7.4 and labeled with the pH-sensitive fluorophore, pHrodo red, which becomes fluorescent when taken up in acidic lysosomes. Uptake of fibrils, presented either in suspension or surface bound, to murine RAW264.7 or human THP-1 cells was monitored by measuring fluorescence at ~600 nm in a plate reader or by fluorescence microscopy. Fluorophore-labeled fibrils (20 $\mu$g) were mixed with rat or human collagen 1 (0.2 $\mu$g – 20 $\mu$g) and phagocytosis quantified. The effect of opsonization using murine 11-1F4 antibody (mAb) was assessed on collagen-treated fibrils.

Results: The addition of collagen 1, but not human serum amyloid P component, to rV$\lambda_6$Wil fibrils (20 $\mu$g) resulted in a dose-dependent inhibition of macrophage phagocytosis, with 80% inhibition achieved with 20 $\mu$g of collagen. The rate of fibril uptake by RAW264.7 cells over a 24 h incubation was similarly decreased in an exponential fashion by the presence of collagen. Fibrils coated with human collagen 1 were also resistant to phagocytosis by M0, M1 and M2-polarized THP-1 cells. The addition of the opsonizing IgG1κ m11-1F4 mAb to collagen 1-coated fibrils overcame the phagocytosis block in a dose-dependent manner but only significantly when the collagen content was 10% (w/w; i.e., 2 $\mu$g) or less.

Conclusions: Inexplicably, macrophages do not routinely clear amyloid deposits in patients with AL. We have shown that synthetic AL-like fibrils, which are efficiently taken up by macrophages in vitro, can be protected from phagocytosis by the addition of collagen 1. The addition of collagen 1 to tissue amyloid, by adjacent cells, may serve as a don’t eat me signal preventing cell-mediated clearance. This finding may lead to novel approaches to remove tissue amyloid.
AMYLOID BINDING AND OPSONIZATION PROPERTIES OF A NOVEL PEPTOPE-ANTIBODY COMPLEX

S. J. Kennel¹, J. S. Foster¹, A. D. Williams¹, D. C. Wooliver¹, S. Macy ¹, E. B1. Martin, J. S. Wall¹

¹ Department of Medicine, University of Tennessee Medical Center.

Introduction: Clearance of tissue amyloid remains an important goal in the treatment of patients with systemic amyloidosis. The persistent accumulation of amyloid results in organ dysfunction and severe morbidity. Amyloid clearance may result in restoration of function and improved patient outcomes. The prevailing mechanism for achieving this is antibody (mAb) binding and Fc-mediated phagocytosis by macrophages. We have developed a peptope construct comprising a pan-amyloid binding peptide and high-affinity epitope recognized by mAb 11-1F4 as part of a two-step mechanism for amyloid opsonization. We now demonstrate that premixing mAb 11-1F4 with a bifunctional peptope, designated p105, produces a complex which can recognize multiple types of amyloid and serve as an opsonin. We have shown that the p105-11-1F4 mixture: 1) Forms a stable, functional complex; 2) Binds diverse amyloids; 3) Specifically targets amyloid in a murine model of AA amyloidosis, and; 4) Enhances the phagocytosis of amyloid in vitro.

Objectives: The aim of this study was to assess the ability of a novel peptope-mAb complex to bind amyloid and induce its phagocytosis by macrophages. Such a reagent may serve as a novel amyloid-cleaving immunotherapeutic.

Methods: Peptope p105 was chemically synthesized (Genscript, NJ), and murine 11-1F4 was prepared by the National Cancer Institute. The complex was formed by incubation of p105 with 11-1F4 at a molar ratio of 2.5:1. For certain assays, 11-1F4 was labeled with iodine-125 before addition to p105. After characterization of the complex, binding to diverse amyloids was assessed using synthetic fibrils and human amyloid extracts in a pulldown assay. Reactivity with amyloid in vivo was evaluated in AA mice using small animal SPECT/CT and autoradiography. Complex-mediated phagocytosis was studied in vitro using pHrodo red-coupled amyloid extracts.

Results: Incubation of p105 and 11-1F4 resulted in ~95% of the mAb complexed with peptope. In a pulldown assay, p105-11-1F4 bound synthetic fibrils (84% bound vs. 2% for 11-1F4 alone) and human AL amyloid extracts consistent with the amyloid binding characteristics of the peptope. When injected into AA mice, 125I-11-1F4-p105 was retained in the spleen (11.1 % injected dose/g tissue), liver (23.2 %ID/g) and pancreas (12 %ID/g). In amyloid-free mice, liver and splenic uptake was 0.4 and 1.6 %ID/g, respectively. The complex was shown to specifically co-localize with amyloid in these tissues by autoradiography. When incubated with pHrodo red-coupled human AL amyloid extract, the complex caused a 3-fold increase in phagocytosis of the amyloid by RAW264.7 murine macrophages.

Conclusions: The p105-11-1F4 complex serves as an opsonizing agent for amyloid with the added benefit of enhanced reactivity to amyloid deposits not normally recognized by 11-1F4. The preformed complex could be used in a single injection to mediate clearance of diverse forms of amyloid through macrophage phagocytosis.
IMMUNOGLOBULIN LIGHT CHAIN PROTEOLYSIS AND AGGREGATION

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\textbf{Introduction:} A pilot proteomic study using multiple reaction monitoring mass spectrometry allowed us to obtain data on the concentrations of 153 target proteins present in sera from patients with AL amyloidosis, as well as a control group of healthy individuals. Results from this study showed a differential representation of 80.4\% (123/153) of the measured proteins in AL sera compared to matched controls; 39\% of 123 differentially represented proteins in the AL group were associated with catalytic activity, i.e. differing concentrations of either proteins that function as proteases or protease inhibitors. These data, together with reports on the presence of immunoglobulin (Ig) light chain (LC) fragments in AL serum, urine, and tissue deposits, as well as studies demonstrating enzymatic activity from neutrophil elastase and cathepsin G present on ex vivo AL amyloid fibrils, suggested that proteolysis caused by an imbalance of proteases and protease inhibitors might play a role in the pathobiology of AL amyloidosis.

\textbf{Materials and Methods:} Recombinant human Ig LC proteins were obtained using an E. coli expression system. Protein structure and stability were analyzed by far- and near-UV circular dichroism and fluorescence spectroscopy. LC fragments produced by limited proteolysis and LC peptides forming amyloid fibrils were identified by mass spectrometry. Amyloid fibril formation was monitored by thioflavin T binding, and fibrils were visualized by electron microscopy (EM).

\textbf{Results:} We compared the kinetics of proteolysis exhibited by several LC proteins containing amino acid sequences identical to patient cases including amyloid (AL LC), multiple myeloma (MM LC), or the corresponding k1 germline LC. Among these proteins, AL LC was the most thermodynamically unstable, while germline LC showed the greatest stability. Electrophoretic testing of the LC proteins, digested with trypsin or human neutrophil elastase (HNE) for discrete time intervals, showed the gradual disappearance of full-length LC upon proteolytic degradation in all reactions; however, AL LC demonstrated a lower resistance to proteolysis by trypsin compared to the MM and germline LC proteins. In the HNE reactions, rapid digestions of all studied LCs were indistinguishable. Mass spectral analysis of digest samples identified 13 major fragments of AL LC from reaction with trypsin and 19 shorter fragments from digestion with HNE. Moreover, peptide fragments appearing upon treatment of LC proteins with trypsin (but not with HNE) were able to form amyloid fibrils as demonstrated by ThT fluorescence and confirmed by EM measurements. Mass spectral analysis revealed that fibrils were formed by N-terminal fragments of the LC V-region.

\textbf{Discussion and Conclusions:} AL serum proteomic data suggesting an imbalance in circulating levels of proteases/protease inhibitors and electrophoretic analysis providing evidence of protein fragmentation in AL patient sera indicated that some abnormality in proteolytic processing of LCs may be important in AL amyloidosis. Faster fragmentation and fibril formation by AL LC compared to non-AL LCs revealed a correlation between LC stability, susceptibility to proteolysis, and propensity for aggregation and fibrillogenesis. AL-specific protease recognition sites introduced through somatic mutations are predominantly located within inter-strand loops and in the J-regions of the LC where they become easily accessible for cleavage by proteases. Partial proteolysis promotes unfolding of the proteins; however, intra-domain disulfide bonds within variable and constant regions of the LC prevent complete protein unfolding and degradation. Based on our findings, we propose that proteolytic cleavage of the AL LC protein increases solvent accessibility of exposed amyloidogenic “hot spots” leading to an increased aggregation propensity of the partially unfolded proteins.
SMALL MOLECULE STABILIZERS OF AMYLOIDOGENIC ANTIBODY LIGHT CHAINS

G. Morgan 1, N. Yan 2, E. Rennella 3, L. Kay 1, J. Kelly 2

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2 Scripps Research.
3 University of Toronto.

Background: In systemic AL amyloidosis, monoclonal light chains (LCs) or their fragments can misfold and aggregate to form insoluble, non-native amyloid fibrils that cause organ damage and eventually death. No drugs are approved specifically for AL amyloidosis, but off-label use of cytotoxic regimens approved for multiple myeloma can be effective. However, many patients are too sick at diagnosis to tolerate chemotherapy, especially those with significant cardiomyopathy. Treatments for frail AL amyloidosis patients are therefore urgently needed.

Objectives: Amyloidosis-associated LCs have been shown to be less stable than other LCs. Therefore, we hypothesize that stabilization of LCs by binding of a small molecule drug could reduce aggregation and benefit patients. This strategy is orthogonal and complementary to existing and emerging treatments for AL amyloidosis. A similar strategy has shown clinical benefit in ATTR amyloidosis. We therefore investigated the molecular mechanisms by which LCs aggregate and used high throughput screening to identify molecules that can stabilize LCs.

Methods: We used in vitro measurements of stability, dynamics and aggregation on recombinant LCs to investigate the molecular mechanisms of aggregation. We developed a high-throughput assay for measuring protein stability in microwell plates and used this assay to screen a large library of compounds. We validated hits from this screen using orthogonal assays for LC binding and stabilization.

Results: LC variable domains readily aggregate to form amyloid fibrils in vitro. Destabilization of variable domains by mutation increases the rate of aggregation, consistent with a model where amyloid is formed via self-association of the unfolded state. The LC constant domain stabilizes the structure of the full-length LC and there is a strong allosteric coupling between folding of the individual domains and self-association of two protein chains into a dimer. The presence of the constant domain also inhibits aggregation, even when the dimer interface is disrupted and the variable domain is destabilized. Our screen identified five structural classes of small molecules that stabilize the native dimeric structure of full-length LCs. Structural data demonstrate that at these molecules bind at dimerization interface within full-length light chains, utilizing variable domain residues that are highly conserved in most AL patients. The small molecule stabilizers identified bind to conserved residues at the variable domain–variable domain interface in the native dimer, stabilizing this putative non-toxic structure.

Conclusion: We have identified small molecules that stabilize amyloidogenic antibody light chains. We aim to develop small molecules with increased binding affinity that could become drug candidates for AL amyloidosis.

Keywords: AL amyloidosis; drug development; Amyloid fibrils
EXERCISE SUPPRESSES SYSTEMATIC AApoAII AMYLOIDOSIS PROGRESSION IN MICE

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1 Shinshu university.

Introduction: Apolipoprotein A-II (ApoA-II), the second most abundant protein in plasma high-density lipoprotein (HDL), is deposited as amyloid fibrils (AApoAII) in systemic organs, except for the brain, in association with aging in mice. Distinct exercise regimens are useful for preventing and treating various pathologies, and exercise has been reported to prevent amyloid beta deposition and improve memory impairment in Alzheimer’s disease (Lourenco MV et al. Nat Med, 2019). However, the cellular mechanisms underlying suppression of amyloidosis by exercise are not fully understood. In this study, we aimed to determine the mechanism by which exercise modulates systematic amyloidosis progression using a mouse model of AApoAII amyloidosis.

Materials and methods: AApoAII fibrils (1 μg/mouse) were administrated to 2-month-old R1.P1-Apoa2-/- female mice to induce amyloidosis. The mice were then subjected to 16 weeks of constant-speed running (30 minutes at 50% of their maximum running speed, 4 times/week) or interval running (the cycles of 3 minutes, 70% and 3 minutes, 30% of their maximum running speed repeated for 30 minutes, 4 times/week) on a treadmill. AApoAII amyloid deposition was then evaluated in Congo red-stained sections using polarizing microscopy.

Results: After exercise, AApoAII amyloid deposition in the mice was suppressed; especially in the liver and spleen, amyloid area were dramatically reduced compared with sedentary group (Table 1). In the interval-running group, the serum triglyceride level and white adipose tissue weight showed decreasing tendencies, and the glucose tolerance impaired by aging was improved. There was no change in the level of HDL cholesterol or Apoa1, Apoa2 expression in the liver, but the serum levels of both ApoA-I and ApoA-II were increased in the interval-running group. Additionally, AApoAII amyloid deposition improved glucose tolerance in an intraperitoneal glucose tolerance test.

Discussion and conclusions: We showed that exercise can prevent or slow down systemic AApoAII amyloidosis progression. Despite our attempts to determine the beneficial effects of exercise on amyloidosis, including anti-oxidation, autophagy activation, and suppression of endoplasmic reticulum stress, the mechanism of amyloidosis suppression by exercise is still not clear. For comprehensive analysis of the effects of exercise on amyloid deposition, we are planning next-generation RNA sequencing analyses using mouse liver and muscle samples.

Table 1. Exercise suppresses amyloid deposition

<table>
<thead>
<tr>
<th>Amyloid area (%)</th>
<th>N</th>
<th>AI</th>
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<th>Spleen</th>
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<td>2.8a</td>
<td>3.7c</td>
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<tr>
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<td>0.5d</td>
<td>0.6f</td>
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<tr>
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<td>2.0b</td>
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AI: The degree of amyloid deposition was defined as the average of the grade of amyloid deposition in seven organs (heart, liver, spleen, skin, tongue, small intestine, and stomach).
a vs. b: p<0.01; c vs. d: p<0.001; e vs. f: p<0.05
CRYO-ELECTRON MICROSCOPY STRUCTURES OF AMYLOID FIBRILS

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Introduction: Amyloid fibrils are associated with a range of debilitating diseases in humans and animals but the exact molecular structures of these pathogenic agents have recently remained elusive.

Objectives: To determine the structure of amyloid fibrils from patient tissue and to learn about the mechanism of fibril formation.

Methods: Cryo-electron microscopy (cryo-EM) and other methods

Results: We obtained cryo-EM structure of amyloid fibrils from three different forms of systemic amyloidosis: AA (Liberta F, Loerch S, Rennegarbe M et al., Nature Comm. 10, 1104, 2019), AL (Radamaker et al., Nature Comm. 10, 1103, 2019) and ATTR (Schmidt M et al. Nature Comm. 10, 5008, 2019). In addition, we obtained the structure of Aβ amyloid fibrils from Alzheimer’s disease/cerebral amyloid angiopathy (Kollmer M et al. Nature Comm. 10, 4760, 2019). Our data reveal the present of right-hand twisted cross-β sheets and provide, in specific cases, evidence about the order of events during fibril formation, such as whether proteolysis precedes fibril formation or vice versa.

Conclusions: Amyloid fibrils from patient tissue are structurally different from known amyloid-like fibrils formed in vitro. Our findings illuminate the structural repertoire of protein aggregates, provide insights into the mechanism of fibril formation in vivo, and inform about potential therapeutic strategies.

Keywords: Structure, Fibril, Mechanism.
A CELLULAR MODEL OF AMYLOID DEGRADATION USING MACROPHAGES CULTURED WITH MOUSE AAPOAII AMYLOID FIBRILS

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Introduction: Recent studies on amyloidosis have reported that macrophages are involved in both the formation and clearance of amyloid deposits [1] [2]. Cellular models of amyloid formation using macrophages have been developed to evaluate several types of amyloidosis, revealing the pathogenic roles of intracellular functions in amyloid deposition. However, the mechanism of amyloid fibril clearance by cells is less understood. In this study, we used a cell-based assay and mouse-derived apolipoprotein A-II amyloid fibrils (AApoAII) to investigate the roles of macrophages in amyloid-induced cellular degradation and cytotoxicity.

Methods: J774A.1 mouse macrophage cultures were treated with AApoAII for 3, 6, 12 or 24 h. The culture media collected from the cells at each time point were centrifuged at 20,000 rpm for 20 min to precipitate the amyloid fibrils. The remaining amyloid fibrils in the culture media were quantified by the thioflavin T fluorescence assay. The cytotoxicity induced by AApoAII fibrils was measured by the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay, and the gene expression levels of apoptosis-related genes were measured by qPCR. The macrophages were incubated with AApoAII fibrils for 24 h, washed with PBS twice, and incubated in AApoAII-free culture medium for 3, 6, 12 or 24 h to assess the cellular degradation of amyloid measured by the thioflavin T fluorescence.

Results: To determine whether macrophages degrade amyloid fibrils, J774A.1 macrophages were treated with AApoAII fibrils. The amount of AApoAII fibrils in the culture medium decreased in a time-dependent manner, and this decrease was prevented by the endosomal trafficking inhibitor VPS34-IN1 or the absence of living cells. AApoAII were associated with the cell surface after 24 h of co-incubation with macrophages. Incubation with AApoAII fibrils significantly reduced cell viability, as measured by the MTT assay, and decreased gene expression of the anti-apoptosis marker Bcl2. After washing the cells with PBS and incubating in AApoAII-free medium, the amount of AApoAII associated with the cell surface decreased in a time-dependent manner. Furthermore, the thioflavin T fluorescence was co-localized with lysosomes, suggesting the possibility that J774A.1 macrophages degrade AApoAII via the lysosomal pathway.

Conclusions: Recent studies have reported that amyloidosis results directly from an imbalance between amyloid formation and clearance [3]. Our results suggested that J774A.1 mouse macrophages degrade AApoAII fibrils via the endosomal–lysosomal pathway; thus, increasing endocytosis or lysosomal activity may contribute to the clearance of amyloid deposits. We suggest that further studies are needed to determine the mechanisms of amyloid-induced cytotoxicity and amyloid degradation by cells, necessary for maintaining amyloid clearance activity to suppress amyloidosis.

References:
DO PLANTS BECOME A GOOD TOOL FOR AMYLOID FIBRIL FORMATION STUDY?

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Introduction: One of the biggest problems in amyloidosis researches is to develop animal models for amyloidosis because most of animal models do not develop any amyloid fibrils in tissues or it takes much time even if the models have amyloid fibrils in tissues. We speculated plant might become a good tool for amyloid formation study because structure of plant is simple, and effects of amyloid precursor proteins and their amyloid fibrils on viability of plant are easy to be examined.

Methods: MicroTom (tomato) was subjected to in vitro amyloid formation study. One ml of human Aβ42 (10 μM) or transthyretin wild type TTRwt (10 μM) in PBS, pH 7.4 or PBS was given to the root of MicroTom, and appearance of leaves was precisely compared with those given PBS alone. Such experiments were also performed in the presence and absence of protease inhibitor cocktails. Each protein was extracted from the leaves in 6 and 24 hours after starting the experiments, and analyzed by Western blotting. Amyloid formation of proteins was evaluated by Western blotting and thioflavin T test.

Results: In Aβ42 and TTRwt administered groups, activity of plants went down and the diameters of stem become thinner than those in PBS group in a time dependent manner. In addition, Aβ and TTRwt oligomers were weaker detected in plants in 6 hours after starting experiments. The band became weaker which could be partially prevented by the administration of protease inhibitors. This suggest that intrinsic plant proteases might digest the precursor proteins. Aβ42 formed oligomer when measured by the thioflavin T test after 24 hours in PBS.

Conclusions: Plants can become a good tool for elucidating amyloid formation mechanism and may be used for screening candidate drugs for amyloidosis.
AMYLOID FORMATION IS SUPPRESSED IN MICROGRAVITY

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**Introduction/Background:** In the future, humans may live in space. However, it is unclear whether the effect of gravity on amyloid formation remains to the elucidated. In space, gravity in much has than that on the earth. In zero gravity, convection dose not occur, which may change amyloidigenicity of proteins. In this study, we analyzed the effect of microgravity on amyloid formation in several amyloid genic proteins.

**Methods:** The microgravity was produced using a gravity controller Gravite®. Insulin was incubated at 2 μM seed, pH 3.0, 37°C in a microgravity controlled by Gravite®. Human Aβ42 was incubated at 2 μM, pH 7.0, 37°C in the machine. Human TTR was incubated at 10 μM, pH 5.0, 37°C in the machine. These samples were incubated and measured for fluorescence intensity using the Thioflavin T method. Presence or absence of amyloid fibrils was evaluated by an electronic microscope.

**Results:** Insulin formed less amyloidosis in the microgravity compared to that on the ground. Furthermore, Aβ42 and TTR formed less amyloid fibrils in the same condition. However, amyloid fibrils were not morphologically different by an electronic microscopic analysis.

**Conclusions:** Gravity may be an accelerating factor on amyloid fibril formation.
Amyloid Fibril Composition Type is Consistent Over Time in Patients with Transthyretin Amyloidosis

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Background
Fibril composition in transthyretin (TTR) amyloidosis can be of two distinct types. In some patients, the amyloid deposits consist of a mixture of full-length TTR and large amounts of C-terminal TTR fragments (fibril type A) while in other patients the deposits consist of full-length TTR (fibril type B). The fibril conformation type is strongly correlated to the two clinical phenotypes seen among ATTRV30M amyloidosis patients, thereby offering a plausible explanation for the presence of the two phenotypic groups. Cardiac enlargement and a general older age of onset are seen among patients with type A fibrils, while patients with type B fibrils in general has no cardiomyopathy and have a younger age of onset. The underlying mechanisms behind the existence of two different fibril composition types are unknown.

Objectives
To investigate if the fibril composition type changes with time, especially if fibrils containing only full-length TTR (type B fibrils) can change over time to fibrils containing C-terminal TTR fragments (type A fibrils)

Methods
One hundred and twenty-three patients who underwent an abdominal subcutaneous adipose tissue biopsy at the Amyloidosis Centre, Umeå University Hospital or at the FAP-team, Piteå Hospital between 2004-2012, whose fibril type was determined, were identified. Of the present day survivors, 29 patients were eligible for inclusion in the study (i.e., not standing on anticoagulation medication, were able to travel for a repeated biopsy and living in northern Sweden) and were asked to volunteer for a new biopsy. Sixteen agreed and underwent a follow-up biopsy. All biopsies were analysed at the amyloidosis diagnostic unit at Uppsala University Hospital, using western blot (with an antibody directed at TTR50-127) to determine the fibril composition type.

Results
Amyloid deposits were detected in all biopsies, and all patients had the same fibril type in the follow-up biopsy as in the initial biopsy. The duration between the initial and follow-up biopsies was 6 to 13 years among type B patients and 7 to 12 years among type A patients. The majority (14) of the patients included in the study had type B fibrils whereas only 2 patients had type A fibrils.

Conclusions
This study shows that the amyloid fibril composition type does not change over time, and especially that the presence of large amounts of C-terminal TTR fragments in the amyloid deposits is not related to long-standing disease.

Keywords: transthyretin, fibril type, fibril composition
ANALYSIS OF THE DISTRIBUTION OF AMYLOID IN PATIENTS REVEALS DISCRETE PATTERNS OF ORGAN INVOLVEMENT

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Introduction

Amyloidosis is a multi-organ system disorder. The distribution of amyloid in abdominothoracic organs and peripheral nerves of light chain amyloid (AL) patients impacts treatment options and prognosis. Organ involvement, based on clinical presentation, has led to the concept of organ tropism, wherein the light chain determines organ-restricted deposition. In patients with transthyretin amyloidosis (ATTR), the primary symptoms, which are presumed to reflect amyloid distribution, are used to stratify patients for specific therapies – cardiomyopathy (tafamadis) and neuropathy (patisiran, inotersen).

We have used a novel peptide radiotracer, designated 124I-p5+14, to image 12 AL patients and 4 ATTR patients (2 cardiomyopathy and 2 neuropathy) using PET/CT (NCT03678259). Organ-specific amyloid load was quantified and a correlation analysis performed.

Objectives

The aim of this analysis was to study the organ-specific distribution of amyloid in AL and ATTR patients, which could impact factors such as the concept of organ tropism and the single organ involvement paradigm of ATTR patients.

Methods

Sixteen patients with AL (n=12) or ATTR (n=4) amyloidosis received <2 mg of 124I-p5+14 (<2 mCi) administered as a single IV bolus. PET/CT images were acquired at ~5 h post injection. Images were analyzed using Inveon Research Workplace (Siemens) and standard uptake value ratios were obtained by using the lumen of the thoracic aorta (blood pool) as the reference tissue. Images were reviewed by a nuclear medicine physician and uptake of 124I-p5+14 in each tissue assessed as positive or negative. A non-parametric correlation analysis was performed using a matrix of all tissues.

Results

In each AL patient, at least two major organs contained amyloid by PET imaging of 124I-p5+14; uptake was observed in the heart or kidneys (83%), spleen (63%) and liver (25%). A non-parametric correlation analysis that included quantitation of uptake in the heart, lung, liver, spleen, kidneys, gluteus and deltoiis muscles, and myocardium showed positive correlations between the heart with myocardium (r=0.93, p<0.001) and muscles (r=0.61, p=0.01). Additionally, radiotracer uptake in the kidney correlated with spleen (r=0.73, p=0.001) and liver (r=0.85, p<0.001). A negative correlation existed between heart uptake with that of liver and spleen.

Analysis of ATTR patients revealed cardiac uptake of 124I-p5+14, indicative of amyloid, in all four patients, regardless of whether the patient was diagnostically characterized as cardiomyopathic or neuropathic.

Conclusions

Quantitative PET imaging of amyloid load using 124I-p5+14 suggests that cardiac amyloidosis may be a specific manifestation of a generalized muscle-associated amyloid deposition. Renal amyloidosis is associated most favorably with hepatic and splenic involvement. Additionally, although based on a small population, we have shown that in ATTR patients with neuropathy, asymptomatic cardiac amyloidosis can be detected.
PM042

GENE EXPRESSION SETS AND RENAL PROFILING FROM THE RAIN (RENAI AL AMYLOID INVOLVEMENT AND NEOD00) TRIAL

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2 University of Michigan.
3 Vanderbilt University Medical Center.
4 Mayo Clinic.
5 University of Michigan.
6 Karmanos Cancer Institute.

Introduction: Renal involvement is present in 70% of patients presenting with AL amyloidosis. Despite the achievement of hematologic responses, organ improvement is not assured. NEOD001 is a humanized monoclonal antibody recognizing an epitope on the misfolded light chains, promoting clearance of amyloid fibrils. The RAIN trial (NCT03168906), a phase 2b study, evaluated NEOD001 versus placebo in patients with stable hematologic response but persistent proteinuria (> 500 mg daily). Herein, we will present an update on all available data from this study.

Objectives: The primary endpoint of the trial was achievement of a renal response. Ancillary studies included systematic histologic evaluation and transcriptional profiling of renal biopsies. Measurements of urinary epidermal growth factor to creatinine ratio (uEGF/Cr), a non-invasive biomarker that predicts patients at high risk of kidney progression, were also performed.

Materials & Methods: RAIN subjects were randomized to receive either NEOD001 (24mg/kg) or placebo every 28 days. Renal biopsies were scored by 2 expert renal pathologists employing a novel histologic tool that assessed glomerular scarring, interstitial fibrosis and the distribution of amyloid, and were successfully evaluated by transcriptional profiling in the Michigan Kidney Translational Medicine Core lab. Clinical data and histologic scores (CSIC = composite scarring injury score, AS = amyloid score) were correlated to the expression profiles of tubular and glomerular gene sets.

Results: Ten patients were enrolled and randomized on RAIN (5 NEOD001 and 5 placebo) prior to its early closure in April 2018 when Prothena halted development of NEOD001. Baseline characteristics and histologic scores are in Table 1. Of the 6 patients with available data, 3 had a renal response documented at study termination, 1 of whom had a drop of >8.5g of urinary protein. When correlating gene expression to clinical data, numerous genes of interest were identified using a false discovery rate corrected P-value of < 0.10 (Table 2). Transcriptional profiling revealed 2 distinct patient clusters (G1 and G2) within the tubular and glomerular gene expression sets. The histopathologic features were different between G1 and G2; the AS was significantly higher in G2 in both the tubular (7.0 vs. 4.25; \( p=0.03 \)) and the glomerular (6.92 vs. 4.38; \( p=0.04 \)) compartments. The differential expression for the genes of interest between G1 and G2 was also determined (Table 2).

Conclusion: The RAIN trial closed prematurely, and no conclusions can be reached about the activity of NEOD001; however, this effort has generated multiple genes of interest correlated with the distribution of amyloid. We are currently collecting long-term follow-up data. The expression profiles of “responders” and “non-responders” will be compared in an attempt to suggest predictors of response. These data will be available at the ISA meeting in Spain in March 2020.
Table 1. Baseline characteristics and renal responses

<table>
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<th>Cr (mM)</th>
<th>eGFR</th>
<th>24h UTP</th>
<th>uEgfr/Cr</th>
<th>CSIS (0-200)</th>
<th>AS (0-12)</th>
<th>NEOD001 No</th>
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eGFR = estimated glomerular filtration rates, uEGFR = urinary epidermal growth factor, Cr = creatinine, UTP = urinary total protein, EOS = end-of-study, CSIS = composite scarring injury score, AS = amyloid score, NA = not applicable
*empty spaces denote missing information, data will be available for ISA 2020

Table 2. Genes of interest and their differential expression

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<tr>
<td>NSFLC1</td>
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<td>+</td>
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<tr>
<td>PODXL</td>
<td>+</td>
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All associations significant at q value <= 0.1
EXPLORING THE AMYLOID INHIBITION POTENTIAL OF APOLIPOPROTEIN E PEPTIDE-ANALOGUES

G. Nasi1, P. Spatharas1, V. Iconomidou1

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Introduction: Many strategies are currently being assessed for preventing the formation of amyloid fibrils. Among them, peptide-based inhibition of polymerization has been described as the most attractive approach for therapeutic intervention [1]. Both “aggregation-prone” peptides of amyloid-forming proteins and randomly generated sequences from peptide libraries have been used to inhibit the aggregation of amyloid fibrils [1]. However, another promising source of peptide-based inhibitors is co-localized proteins found on amyloid deposits. In this study, we examine in vitro the ability of two amyloidogenic segments of Apolipoprotein E (ApoE) — a protein found in the amyloid deposits of several amyloidoses [2] — to minimize or entirely block the aggregation of Aβ peptide.

Materials & Methods: Lyophilized aliquots of two peptide-analogs of ApoE were incubated with Aβ in a 7:1 molar ratio. In order to monitor the kinetics of the aggregation process, a droplet of each suspension from days 0, 1, 3, 7 and month 5 of incubation was applied to glow-discharged carbon-coated copper grids and was stained with 2% (w/v) aqueous uranyl acetate. The fibril containing grids were examined with transmission electron microscopy (TEM). Additionally, Congo Red stain was utilized to verify the presence of amyloid fibrils on day 7.

Results: The experimental results show that both peptide-analogues can halt the formation of amyloid fibrils of Aβ peptide in vitro, since in both cases no amyloid fibrils are detected, either by using TEM or with Congo Red staining.

Conclusions: In this work, we investigated the ability of two amyloid-forming segments of ApoE to inhibit amyloid formation of Aβ peptide in vitro. Our preliminary results are encouraging and lead the way for more experimental work, to test the inhibition properties of other “aggregation-prone” peptides derived from proteins found co-localized on amyloid deposits.

Acknowledgement: The present work was co-funded by the European Union and Greek national funds through the Operational Program “Competitiveness, Entrepreneurship and Innovation”, under the call “RESEARCH-CREATE-INNOVATE” (project code: T1EDK-00353).

References:
INVESTIGATING THE AGGREGATION PATHWAYS OF APP AND TAU IN H. SAPIENS AND C. ELEGANS VIA NETWORK ALIGNMENT

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Introduction: Protein aggregation is an active area of research and an ever-increasing number of proteins forming amyloid fibrils in vivo is being constantly recorded [1]. Today, 37 proteins are characterized as amyloid-forming, amongst them Aβ peptide (a cleavage product of APP) and microtubule-associated Tau protein, the deposition of which is one of the main hallmarks of Alzheimer Disease [2]. Since the direct study of Aβ and Tau accumulation in humans is difficult, model organisms, such as Caenorhabditis elegans, are often used instead [3]. Protein-Protein Interactions (PPIs) are crucial for amyloid formation and a useful tool for studying them are molecular networks. The aim of this work was to compare the human network of APP and Tau with the network of C. elegans orthologs APL-1 and PTL-1, identify common pathways and help guide experimental studies on the nematode model.

Methods: Three network datasets were created for this study, each consisting of a human and a nematode network. Firstly, the human network of APP and Tau was extracted from the Amyloid Interactome [4] and the network of C. elegans orthologs was created using STRING [5]. Secondly, the APL-1 and PTL-1 network and its human ortholog network were created, and lastly, as a less biased approach without ortholog mapping, the PPI networks of APP & Tau and of APL-1 & PTL-1 were produced. Every pair of networks was visualized and compared using network alignment [6].

Results: The first and second pair of networks allowed the selection of appropriate parameters for network alignment. Afterwards, emphasis was put in the investigation of the third pair. We show that protein pairs which are commonly aligned in the second and third approach are highly conserved between the two organisms. Amongst these were proteins involved in the APP processing pathway, such as the α- and γ- secretases and in the phosphorylation of APP and Tau, a process that affects aggregation.

Conclusions: Conserved pathways were revealed between H. sapiens and C. elegans in the two networks. With many of these pathways remaining unexplored in C. elegans, this work provides the impetus for the experimental investigation of amyloid fibril formation in this model organism, that could also serve in the study of other less studied amyloidoses.

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References:
AMYLOID STRUCTURE DETERMINATION AT LIQUID INTERFACES USING VIBRATIONAL SUM-FREQUENCY GENERATION SPECTROSCOPY

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Keywords: Amyloid structure, vibrational spectroscopy, adsorption, phospholipids

Category: Amyloid fibril formation, deposition and clearance

Introduction. Characterization of amyloid secondary structures is of paramount importance for understanding amyloid formation in vivo. A vast amount of techniques is being used for this purpose, such as ssNMR, cryo-EM, FTIR, CD, and others. Most of these techniques can only determine the structure of amyloids formed in an aqueous solution. Structure determination at liquid surfaces, especially at the phospholipid/water interface, which resembles a natural cell environment, is still a challenging task. Vibrational sum-frequency generation (VSFG) spectroscopy can be used to detect a single molecular layer at any liquid surface and to determine the secondary structure simultaneously. VSFG has been successfully used to study protein aggregation at various surfaces, however, it is still a relatively new technique and has never been applied yet to study the adsorption of amyloids to lipid/water interface.

Objectives. We aim to understand and compare the adsorption behaviour of protein aggregates to air/water and lipid/water interfaces using the VSFG technique. Moreover, we focus on identifying spectral signatures, which correspond to different amyloid structures.

Methods. Hen egg-white lysozyme (HEWL) was used as a model amyloid system. HEWL aggregates formed in the bulk (heated at 62°C, pH 2.2) were monitored using FTIR spectroscopy and AFM. For the VSFG measurements, aliquots of HEWL incubation solution were injected into the water beneath the DOPG lipid monolayer.

Results and Conclusions. VSFG spectra of lysozyme and its aggregates adsorbed at air/water and lipid/water interfaces were recorded. The structure of adsorbed protein aggregates was determined from recorded VSFG spectra. We found that aggregates with a parallel and anti-parallel β-sheet structure together with smaller unordered aggregates and a denatured protein were adsorbed to both interfaces. Due to the electrostatic interaction between charged protein’s groups and lipid’s head-groups, a substantially larger amount of aggregates were adsorbed to lipid monolayer than to the water’s surface. Even after 0.5 h of heating, when aggregated protein’s form was not detectable at the water/air surface, the lipid/water surface was saturated with protein aggregates. This finding shows that there is a strong driving force for protein aggregates to be deposited at lipid monolayer even when mature fibrils are not formed yet.

Acknowledgements: Author S.S. gratefully acknowledges European Comission for funding this research through a Marie-Skłodowska Curie individual fellowship.
ORGAN-SPECIFICITY OF APOA-I AMYLOIDOGENIC VARIANTS IS DRIVEN BY TISSUE-SPECIFIC EXTRACELLULAR MILIEUS

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Apolipoprotein A-I (ApoA-I) amyloidosis is a rare autosomal dominant, degenerative disease characterized by the accumulation of fibrils constituted by the N-terminal domain of the protein in several organs, leading to their progressive dysfunction and failure. Although ApoA-I-related amyloidosis is a systemic disease, the twenty-three amyloidogenic variants so far identified show a preferential tissue specificity that seems to correlate with the location of the mutation in the protein sequence and with the different interactions with tissue-specific extracellular environments. However, the factors that lead to protein destabilization, and the processes responsible for cells/tissues damage as well as the mechanisms behind the observed organ specificity are mostly unknown. Therefore, we investigate the impact of amyloidogenic ApoA-I variants on cell physiology and the mechanisms that drive the observed tissue-specificity.

For our investigations, we used G26R and L75P variants that accumulate preferentially in liver and kidney [2, 3], and L174S and L178H that form fibrils mainly in heart and skin [4, 5]. First, the cytotoxicity of the ApoA-I variants, their ability to alter redox homeostasis, and their intracellular localization were tested on cell lines from different tissues (liver, kidney, heart, skin). Then, variant-specific interactions with extracellular matrix (ECM) components were measured by ELISA assay. Finally, variants’ aggregation propensity and fibril morphology were studied by synchrotron radiation circular dichroism and transmission electron microscopy, respectively.

We found that G26R and L75P variants showed selective cytotoxicity on renal and liver cells, whereas L174S and L178H exerted a more pronounced toxic effect on skin- and heart-derived cells. However, this was not due to altered redox homeostasis since intracellular reactive oxygen species levels were not increased, but rather seemed to be due to protein accumulation in the lysosomal compartment. Interestingly, the ApoA-I variants exhibited specific preferential binding to the ECM components, reflecting their tissue accumulation pattern in vivo. While the binding appeared to not affect protein conformations in solution, extended incubation of the ApoA-I variants in the presence of different ECM components resulted in different aggregation propensity and aggregation patterns.

In conclusion, ApoA-I amyloidogenic variants exert a cytotoxic effect in a selective, cell-specific and time-dependent manner. The variants’ affinity for the ECM components and their aggregation patterns agree with the clinically described tissue preferences. Our findings provide a deeper mechanistic understanding on how a specific mutation dictates tissue specificity and the knowledge gained will be fundamental for the development of drugs able to prevent or control this rare disease.

FC-BASED FUSION PROTEIN FOR THE TREATMENT OF AMYLOIDOSIS


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Background: Systemic amyloidosis is a group of life-threatening diseases diagnosed histologically by the presence of amyloid fibrils deposits in tissues which share typical optical properties after Congo red staining. The classification of amyloidoses rely on the biochemical nature of the amyloid protein involved in the formation of deposits (more than 30 different proteins) and on a range of clinical signs. Serum amyloid P component (SAP), a circulating protein produced by the liver, shows significant avidity for any form of amyloid deposits. It is thus systematically found in deposits where it can represent up to 15% of the total mass.

Methods: We designed chimeric proteins associating SAP with IgG1 Fc domains. While the SAP domain targets the molecule to deposits, the Fc domain marks them for recognition and elimination by the immune system. Since dimeric Fcs are required for functional recruitment of immune cells, we designed two different fully human fusion proteins, SAP-Fc, which required dimerization to be functional, and SAP-scFc (single chain Fc), which ensured a functional Fc domain in any configuration.

Results: In AA amyloidosis models, immunohistochemical, SPECT imaging and microautoradiography studies showed high accumulation of SAP-Fc molecules in spleen and liver, with specific colocalization with amyloid deposits. The therapeutic efficacy of SAP-Fc and SAP-scFc molecules was evaluated in AA-amyloidosis induced immunodeficient mice (no immunoglobulins) to avoid immunization against the human proteins. A single i.v. injection of SAP-scFc led to a significant clearance of amyloid but we did not observe any significant effect with the SAP-Fc protein probably due to an abnormal conformation of the SAP-Fc dimers. Electron microscopy studies showed the presence of fibrils into lysosomal vesicles of macrophages.

Conclusion: Such a molecule can be potentially used for the treatment of any form of amyloidosis.
EFFECTS OF SERPINA1 KNOCKDOWN IN TTR PROTEOLYSIS IN THE HEART OF TRANSGENIC MICE CARRYING HUMAN TTR V30M

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TTR proteolysis has been increasingly recognized as alternative mechanism contributing to amyloid formation. Accordingly, amyloid deposits can be composed mainly by full-length TTR or contain a mixture of both cleaved and full-length TTR, particularly in the heart. The protease responsible for TTR cleavage has not yet been identified. However, the specific fragmentation at Lys48 in the TTR polypeptide suggests that it could be a trypsin-like serine protease [1, 2]. Recent studies revealed that the serine protease inhibitor, SerpinA1, was differentially expressed in hepatocyte-like cells (HLCs) from ATTR patients comparatively to controls and a high correlation between SerpinA1 and TTR was also observed (R>0.9) [3].

In a collaborative study to investigate the relation of SerpinA1 and TTR in TTR amyloidosis we propose to unravel the mechanisms underlying TTR-related cardiac amyloidosis, particularly studying the role of SerpinA1 on TTR proteolysis in the heart.

Transgenic mice carrying human TTR V30M (HM30, n=4 per group) were subcutaneously injected with antisense oligonucleotides (ASOs) targeting SerpinA1 during 6 weeks. Then, SerpinA1 levels were estimated in plasma and heart by Western blot (WB) analysis. In plasma, TTR was quantified by ELISA, while in heart homogenates was determined by WB. Immunohistochemical analysis was used to evaluate TTR deposition in the heart. TTR fragmentation in the heart and plasma was studied by WB. Proteolytic activity was evaluated in plasma and in the heart using a fluorescence assay. Our results confirm that SerpinA1 was effectively knocked-down in both plasma and cardiac tissue of treated HM30 mice. In addition, increased human TTR levels in both plasma and heart were observed, which was accompanied by an increase in TTR deposition in the heart. Furthermore, immunoblotting analysis, using an antibody produced in our lab (AD7F6), revealed the existence of fragments (not recognized with commercial DAKO antibody) in mice cardiac tissue upon SerpinA1 knockdown. Additionally, increased proteolytic activity was also detected in mice plasmas while no proteolytic activity was found in heart.

Overall, our results indicate that downregulation of SerpinA1 increases proteolytic activity in mice plasmas promoting TTR fragmentation in vivo. In addition, these data also suggest that TTR proteolysis might occur in blood being then TTR fragments incorporated into TTR deposits in the heart.

Further studies are underway to validate these data and to contribute to a deeper knowledge of TTR cardiac amyloidogenesis.

UNDERSTANDING THE ROLE OF MATRIX METALLOPROTEINASES, TISSUE INHIBITOR OF MATRIX METALLOPROTEINASE-1 AND TISSUE RESIDENT MACROPHAGES IN RENAL AL AMYLOIDOSIS

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INTRODUCTION: Matrix metalloproteinases (MMPs) are zinc-dependent endopeptidases that regulate tissue remodeling and repair through degradation of various extracellular matrix (ECM) proteins. Gelatinases MMP-2 and MMP-9 cleave a wide range of ECM substrates; their activities are regulated by endogenous tissue inhibitors of metalloproteinases (TIMPs). In the kidneys, glomerular and interstitial expression of MMP-2 and -9, and TIMP-1 are associated with macrophages and neutrophils. Recent evidence suggests that different macrophage phenotypes express characteristically different spectra of MMPs and their inhibitors. In renal AL amyloidosis, the roles of MMPs, TIMPs, and tissue-resident macrophages remain unclear. Previous immunohistochemical (IHC) evaluations of kidney specimens have reported conflicting results. The aim of this study was to investigate the histopathology of MMP-2, MMP-9, and TIMP-1, as well as the distribution of tissue-resident macrophages in renal AL amyloidosis.

METHODS: Autopsy kidney tissues from 5 cases of renal AL amyloidosis (ALpos) and 3 cases without amyloidosis (ALneg) were included in the study. Histological sections were stained with H&E and Congo red; amyloid deposits were evaluated under bright and polarized views. IHC staining was performed for MMP-2, MMP-9, and TIMP-1 with heat induced epitope retrieval (HIER) using pH 9. Expression of each protein was assessed within the total renal parenchyma with a focus on glomeruli. Tissue-resident macrophages and inhibitory M2 macrophages were visualized with CD68+ and CD163+ markers, respectively. For each case, the total number of positively stained CD68+ and CD163+ cells was counted in 25 individual glomeruli; the median number of cells per glomerulus and the CD68+/CD163+ ratio were calculated. Clinical data were obtained from retrospective review of charts and the Amyloidosis Center IRB-approved clinical database.

RESULTS: Demographic and clinical data for the study cases are summarized in Table 1. All ALpos cases had advanced stage renal disease; two cases had end stage renal disease (ESRD) prior to death. Congo red positive staining was identified in all kidney compartments, including glomeruli, tubules, blood vessels, and interstitium in every ALpos case; amyloid fibril typing was performed in three ALpos cases and matched other testing results. Congophilic material was not found in ALneg cases. None of the ALpos cases demonstrated MMP-2 and MMP-9 staining within or adjacent to the amyloid deposits; MMP-2 staining was observed in the renal tubular epithelium of both study groups. Diffuse expression of TIMP-1 within glomerular amyloid deposits was observed in one ALpos case (Figure 1); no staining for TIMP-1 was noted for TIMP-1 was noted in ALneg cases. CD68+ and CD163+ staining were found in ALpos and ALneg glomeruli, tubules and interstitium. The number of CD68+ macrophages per glomerulus appeared decreased in ALpos compared to ALneg resulting in increased CD163+/CD68+ ratios (Table 1). Cases with ESRD demonstrated a higher CD163+/CD68+ ratio (1.16 and 1.24) compared to those with no ESRD (0.65, 1.01 and 1.06). Contrary to intra-glomerular staining, peri-glomerular macrophage staining along the Bowman capsule was found more often in ALpos compared to ALneg with staining for CD163+ more intense than CD68+. ESRD cases demonstrated marked CD163+ staining (Figure 1F), while renal stage II cases showed less intense or no staining.

Table 1: Demographics, clinical features, and histological characteristics

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>ALpos, n=5</th>
<th>ALneg, n=3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age at presentation, median (range), yrs</td>
<td>63 (57-70)</td>
<td>62 (56-73)</td>
</tr>
<tr>
<td>Gender, M/F</td>
<td>4/1</td>
<td>2/1</td>
</tr>
<tr>
<td>LC restriction</td>
<td>4x1κ</td>
<td>-</td>
</tr>
<tr>
<td>Cells per glomerulus, median (range), n</td>
<td></td>
<td></td>
</tr>
<tr>
<td>CD68+ positive</td>
<td>2.76 (1.72-4.16)</td>
<td>5.68 (4.3-6.97)</td>
</tr>
<tr>
<td>CD163+ positive</td>
<td>3.20 (1.12-4.76)</td>
<td>3.40 (2.36-3.72)</td>
</tr>
<tr>
<td>CD163+/CD68+ ratio, median (range)</td>
<td>1.06 (0.65-1.24)</td>
<td>0.65 (0.34-0.71)</td>
</tr>
</tbody>
</table>

CONCLUSION: Our findings show no increased MMP-2 and MMP-9 in the glomerular compartment of kidney tissues from autopsied cases of AL amyloidosis. Strong staining for TIMP-1 was observed within and adjacent to glomerular and perivascular amyloid deposits in one of the 5 ALpos cases. The over representation of CD163+ in peri-glomerular areas of ALpos kidney appeared to correlate to renal staging. Together, these data suggest that an imbalance in MMP/TIMP, possibly a reflection of inhibitory tissue-resident macrophage involvement, plays a role in the pathogenesis of renal AL amyloidosis.
Figure 1: Congo red histology and IHC results for AL study case. Congo red (A) standard and (B) polarized views; (C, D) no staining observed for MMP-2 or MMP-9; (E) TIMP-1 positive stain in glomerulus and blood vessel; and (F, G) CD68+ and CD163+ staining within and adjacent to amyloid infiltrated glomerulus. (400X)

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CLUSTERIN, A MOLECULAR CHAPERONE WITH AMYLOIDOGENIC PROPERTIES

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INTRODUCTION: Clusterin is a heterodimeric glycoprotein (α- and β-chain), which has been described as an extracellular molecular chaperone. In humans, clusterin is an amyloid associated protein, co-localizing in amyloid fibrillar deposits in several amyloidoses, including Alzheimer’s disease. Its implication in them though, remains a mystery to this day [1]. In an effort to elucidate clusterin’s role in amyloid formation, we aimed to experimentally examine clusterin’s amyloidogenic potential.

MATERIALS & METHODS: AMYLPRED, a consensus algorithm for the prediction of amyloid propensity [2], was used to locate “aggregation-prone” segments in mature clusterin’s sequence. Four peptide-analogues from the predicted segments were chemically synthesized and typical in vitro aggregation assays — namely, transmission electron microscopy (TEM), X-ray fiber diffraction, ATR FT-IR spectroscopy and Congo red staining — were used to monitor their amyloidogenic profiles.

RESULTS: The experimental assays showed that all four peptide-analogues exhibit characteristic amyloidogenic properties in vitro. TEM reveals amyloid-like fibrils, which bind the Congo red dye and exhibit apple green birefringence under crossed polars of a polarized light microscope. Oriented fibers of the peptide-derived fibrils produce cross-β X-ray diffraction patterns and ATR FT-IR spectra reveal the dominance of β-sheet secondary structure.

CONCLUSIONS: Based on our results, clusterin could potentially form amyloid fibrils in vitro. This is contradictory to its function as a molecular chaperone, that being the inhibition of protein aggregation or amyloid formation. Moreover, there is a number of proteins that are described as molecular chaperones and show amyloidogenic potential, but at the same time have inhibitory effects on amyloidogenesis [3]. The intrinsic amyloidogenicity of these molecular chaperones may indicate a common mechanism by which the cell prevents amyloid formation.

REFERENCES:

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SERUM PROTEOLYTIC ACTIVITY IN RENAL AL AMYLOIDOsis

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INTRODUCTION: Proteolytic remodelling of the immunoglobulin light chain (LC) amyloid precursor protein is a key pathogenic event in AL amyloidosis leading to formation of truncated N-terminal LC species that can aggregate into amyloid fibrils under physiologic conditions. An extremely heterogeneous pattern of LC fragmentation suggests contribution from a broad enzymatic repertoire in LC remodelling in AL amyloidosis.

OBJECTIVE: The objective of this study was to measure overall serum proteolytic activity (SPRA) in AL patient sera and assess the correlations with renal features and renal treatment outcomes. In addition, we examined the contribution from four major classes, specifically, metalloproteases (MMP), as well as serine, cysteine, and threonine proteases to SPrA.

METHODS: Thirty-eight untreated patients (16M/22F; median age 56.9, IQR 50.0-64.1 yrs) with biopsy proven AL amyloidosis, in the absence of multiple myeloma or B cell lymphoproliferative diseases, were selected from 311 consecutive patients evaluated between September 2009 and May 2016. At presentation, patients had evidence of a plasma cell dyscrasia as demonstrated by serum or urine immunofixation electrophoresis and free light chain (FLC) testing. Renal stage was assessed using the recently published staging system¹. All patients underwent high-dose melphalan and autologous stem cell transplantation (HDM/SCT); 16 patients received induction with bortezomib-based therapy prior to HDM/SCT. The evaluation of hematologic and renal organ responses was performed at 1.34 (IQR 1.21-2.05) yrs post-treatment using standard criteria. Clinical information and biomarker data were collected at baseline and post-treatment. Pre- and post-treatment sera were stored at -80°C until measurement; hemolyzed samples, as indicated by a direct spectrophotometric scanning procedure for quantification of hemoglobin, were excluded. SPrA was measured using the Enzchek Protease kit (Thermo Fisher) and expressed in ug/mL as trypsin activity equivalents. The contribution of each class of proteases to SPrA was evaluated using protease inhibitors: doxycycline for MMP; AEBSF for serine proteases; bortezomib for threonine proteases; E-64 for cysteine proteases; and DOX+Halt for a broad-spectrum of proteases. Forty-five sera from self-reported healthy individuals served as a control group. All data were non-parametric and expressed as median (IQR); correlation analysis was performed using Spearman’s Rho test.

RESULTS: SPrA was significantly elevated in pre-treatment (0.387; IQR 0.348-0.451) compared to post-treatment (0.360; IQR 0.336-0.418; P=0.004) AL, as well as control (0.347; IQR 0.310-0.439; P=0.030); no difference was noted between post-treatment and control (P=0.411). SPrA results did not correlate with dFLC levels at pre-treatment (P=0.560). Sera from cases with renal involvement (RI⁺, n=29) showed significantly elevated SPrA (0.410; IQR 0.353-0.471) vs. RI⁻ (n=9) cases (0.350; 0.327-0.412; P=0.039) at pre-treatment. SPrA levels were similar between the three renal stage groups and significantly correlated with 24-hr proteinuria (r=0.391; P=0.015) at pre-treatment. Patients with a hematologic complete response (hCR⁺; n=19) showed no significant change in SPrA levels at post-treatment (P=0.131), while hCR⁻ cases (n=19) demonstrated a significant decrease in SPrA (P=0.013). In contrast, cases with renal organ response (n=19) showed a significantly decreased SPrA at post-treatment (P=0.004), whereas SPrA was unchanged (P=0.322) in cases with renal progression (n=10). The most significant decrease in SPrA was noted with the MMP inhibitor doxycycline, followed by serine, threonine, and cysteine protease inhibitors (Table 1).

<table>
<thead>
<tr>
<th>Protease class inhibition</th>
<th>DOX+Halt</th>
<th>DOX</th>
<th>AEBSF</th>
<th>Bortezomib</th>
<th>E-64</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum samples tested, n</td>
<td>25</td>
<td>27</td>
<td>27</td>
<td>21</td>
<td>21</td>
</tr>
<tr>
<td>Proteolytic activity</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>decrease, median (IQR), %</td>
<td>100-100</td>
<td>100</td>
<td>100-100</td>
<td>54.6-100</td>
<td>37.8-82.3</td>
</tr>
</tbody>
</table>

CONCLUSIONS: Our data suggests that SPrA is elevated at baseline in untreated patients with AL renal amyloidosis; moreover, SPrA in these patients appears to be associated mainly with MMPs and serine proteases. Elevated SPrA may result in the creation of aggregation-prone truncated LC forms in patients with renal AL amyloidosis. A significant decrease of SPrA in cases with renal organ response following HDM/SCT therapy may reduce the supply of aggregation-prone LC fragments to renal amyloid fibril deposits, thus assisting with attainment of organ response.


This research was supported by the Wildflower Foundation, Gruss Foundation and Boston University Amyloid Research Fund.
SYNTHESIS AND EVALUATION OF A NOVEL PEPTIDE-IMMUNOGLOBULIN FUSION FOR TARGETING AND PHAGOCYTOSIS OF AMYLOID

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Introduction

Clearance of tissue amyloid remains an important, unmet need for patients with systemic amyloidosis. Despite the non-native structure of amyloid, phagocytic cells of the innate immune system do not effectively eliminate this material in patients without appropriate immunoglobulin (Ig) opsonization. This has been shown in vitro, in mouse models, and in patients with hepatic AL amyloidosis (Richards, D.B. et al. [2015] NEJM, 373, 12). Ideally, a single opsonin could target diverse amyloids.

We have reported on synthetic peptides (p5 and p5+14) that adopt an α-helical structure in the presence of polycationic ligands and bind many forms of amyloid in vitro and in vivo, as seen in animal models and in patients with AL-, ATTR- and ALECT2-associated amyloidosis.

Herein, we describe the generation of a novel peptide-Ig fusion where peptide p5 is fused to an Ig light chain (LC), which when associated with an Ig heavy chain (HC) yields a multi amyloid-reactive opsonin.

Objectives

Our aim was to create and characterize a new opsonin for targeting many amyloid types and enhancing macrophage-mediated phagocytosis.

Methods

The peptide-Ig fusion comprised an Ig LC sequence fused to peptide p5. The IgHC sequence comprised IgG1 variable, IgG1 CH1, and IgG2a CH2 and CH3 domains expressed in a second vector system. Vectors were co-transfected into HEK 293T/17 cells and the Ig product isolated using Protein A-conjugated beads. Igp5 and Ig control were radiolabeled with 125I and analyzed by gel electrophoresis. Binding of 125I-Igp5 and Ig control with synthetic amyloid fibrils and amyloid extracts was assessed using pulldown assays. 125I-Igp5 was injected IV into healthy (WT) mice and those with severe systemic AA amyloidosis (H2/IL-6 transgenic). Tissue localization was assessed by SPECT/CT imaging, biodistribution and microautoradiography. Opsonization of synthetic fibrils and amyloid extracts (20 µg) was evaluated ex vivo using pHrodo-red labeled substrates and human THP-1 cells.

Results

Purified Igp5 appeared as an intact Ig with both IgH and IgL peptides following gel electrophoresis. 125I-Igp5 bound rVλ6Wil and Aβ(1-40) amyloid-like fibrils at ~64% (21 fold higher than the control Ig, murine 11-1F4). Binding to amyloid extracts was also 10 – 30 fold higher and correlated positively with that of 125I-p5 (r = 0.9, p = 0.01). When injected into AA mice, 125I-Igp5 was detected in the liver and spleen by SPECT/CT imaging with >15% injected dose per gram (20 h post injection). In WT mice, the value was > 3-fold lower and associated with blood pool as seen in autoradiographs. Phagocytosis of rVλ6Wil fibrils by THP-1 cells was 10-fold higher than an irrelevant Ig (MOPC-31c) and twice that of the control Ig.

Conclusions

Opsonization of amyloid and clearance by cells of the innate immune system remains an important treatment goal. We have developed a novel peptide-Ig fusion that binds many forms of amyloid and is capable of enhancing phagocytosis.
EVALUATION OF ANTI-IMMUNOGLOBULIN LIGHT-CHAIN ANTIBODIES FOR ELIMINATION OF CIRCULATING INVOLVED FREE LIGHT CHAINS

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Background: In systemic light-chain (AL) amyloidosis, involved free light chains (iFLC) are toxic and form amyloid, causing organ damage. After therapy and hematologic response, low levels of iFLC may persist and cause progressive organ failure. Therefore, clearing iFLC using monoclonal antibodies may halt organ damage and provide maintenance therapy. In this study, we assessed the selective binding of AKM1 (anti-κ LC) and ALM1 (anti-λ LC) antibodies to both κ and λ FLC. Furthermore, the biological activity of AKM1 and ALM1 was examined in NOD scid γ (NSG) mice xenografted with FLC secreting multiple myeloma (MM) cells.

Methods: Hybridoma cells secreting anti-κ (AKM1) and anti-λ (ALM1) were cultured and supernatant affinity purified (Cell Essentials Inc, Boston, MA). To confirm binding of AKM1 and ALM1 to their target FLC, intracellular flow cytometry and surface plasmon resonance (SPR) were performed. The presence of immune complexes (IC) was assessed using both FLC incubated with AKM1 and ALM1 at 37°C for 2 hours and sera from treated mice. Immunoprecipitation (IP) was performed with Sepharose A/G, followed by immunoblots (IB) for both light chains and AKM1 and ALM1. NSG mice were injected intraperitoneally (IP) with JJN-3 or MM.1S luciferase reporter cells to create κ or λ FLC mouse models and were treated with AKM1 or ALM1 or control PBS. We hypothesized that the level of circulating IC would increase in treated mice because of the longer half-life of IC than FLC and deficiency of Fc receptors in NSG mice. Luciferin signal (FLUX) was obtained from xenografted mice and ELISA assays for κ or λ LC, sBCMA and murine IgG were performed (Bethyl Laboratories, Montgomery, TX and R&D Systems, Minneapolis, MN). Statistical analysis was done with Prism (GraphPad, San Diego, CA).

Results: We assessed the binding of ALM1 and AKM1 within MM cells by flow cytometry. AKM1 bound κ FLC but not λ and ALM1 did the opposite. With SPR both bound their targets but not controls (AKM1, KD 2 x10⁻⁸; ALM1, KD 5 x10⁻⁹). AKM1 formed IC with κ LC only while ALM1 did so with λ LC only. In NSG mice the T₁/₂ of AKM1 and ALM1 was 14 days. Xenografted NSG mice were sorted into 2 equal groups based on day 8 FLUX; FLC and sBCMA levels were also same in both groups. Mice were injected with AKM1 (n=15) or ALM1 (n=15) or control PBS (n=15). On day 12, FLUX and sBCMA in treated and control mice were the same but FLC levels increased 10-fold in the treated mice compared to the controls. Presence of circulating IC in treated mice was confirmed through IP/IB of sera and also by a modified ELISA designed to capture IC.

Conclusions: AKM1 and ALM1 are murine monoclonal antibodies with the potential to bind and possibly deplete free light chains in vivo from the circulation. The results of these in vitro and in vivo assays confirming high binding affinities and activity in xenograft models support the humanization and further preclinical testing of AKM1 and ALM1.
MASS SPECTROMETRY CHARACTERIZATION OF THE LIGHT CHAIN FRAGMENTATION SITES IN AL AMYLOID FIBRILS

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Introduction: The mechanisms leading to fibrillogenesis in vivo are still largely obscure; proteolytic processing by tissue or cell proteases may play an important role in modulating aggregation and fibril stability. The pathogenic role of fragmentation, however, is established only for some amyloidoses, whereas the site and timing of proteolysis in other forms, including light chain (AL) amyloidosis is still debated. Precise identification of the N- and C-termini of the deposited light chain (LC) proteoforms is instrumental to understand the enzymes and processes involved in fragmentation.

Objectives: The purpose of this study was to obtain, through a dedicated proteomic approach, a map of the N- and C-termini of the LC fragments in amyloid fibrils extracted from the heart of AL cardiomyopathy patients.

Methods: Myocardial tissue was obtained from two AL λ amyloidosis patients during, respectively, autopsy and cardiac transplantation. The tissue was repeatedly homogenized in Tris EDTA buffer [1] with protease inhibitors, followed by centrifugations, in order to remove tissue soluble proteins and to enrich amyloid fibrils. The extracted material was analyzed by polyacrylamide gel electrophoresis and immunoblotting. To characterize the “N-terminome” and “C-terminome” of the LC proteoforms in the amyloid fibrils, primary amines and free carboxyl groups were derivatized. Samples were analyzed by liquid chromatography-tandem mass spectrometry (LC-MS/MS), and N- and C-terminal labeled residues were identified by database search and manual spectra interpretation.

Results: In the two samples, both the full length and fragmented LCs from fibrils were detected by polyacrylamide gel electrophoresis. The detectable fragments have molecular weights ranging from approximately 20 kDa to approximately 10-12 kDa. Through analysis of the labeled N- and C-terminal residues, multiple cleavage sites were confidently identified and characterized by LC-MS/MS in both light chains. Fragmentation sites were documented both in the constant and in the variable domains of the LCs. However, while fragmentation points in the variable regions are rare, proteolysis sites in the constant domain are widely scattered along the sequence and partially overlapping between the two LCs. The cleavage sites are being mapped on the previously obtained [1-3] structures of native and fibrillar LCs.

Conclusions: This study adds novel important information to the characterization of the post-translational processing of LCs forming AL amyloid fibrils. Analysis of the cleaved bonds and mapping of the cleavage sites on the native and fibrillar LCs structures will allow dissecting involved enzymes and speculating on timing of cleavage with respect to fibrillogenesis.


Keywords: mass spectrometry, post-translational modifications, proteolysis
THE FREQUENCY OF LOCALISED AND SYSTEMIC AMYLOIDOSIS IN PATIENTS RECEIVING CARPAL TUNNEL RELEASE


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Introduction: Retrospective observations have established that as many as 50% of patients with cardiac amyloidosis caused by transthyretin (ATTR) have been diagnosed with carpal tunnel syndrome (CTS) 5-10 years prior to diagnosis of their cardiac condition. However, only few studies have investigated the presence of localized and systemic amyloidosis in patients undergoing carpal tunnel release (CTR).

Objectives: The aim of this study was to investigate the prevalence of amyloidosis among patients undergoing CTR by histological investigations of fatty and tenosynovial tissue obtained during surgery.

Methods: 100 consecutive patients eligible for CTR without a recognized cause of their neural compression were investigated. Biopsies of tenosynovial and fatty tissue were stained with Congo Red. Amyloid positive biopsies were subtyped by immunoelectron microscopy (IEM) and mass spectrometry (MS). Patients with amyloid positive biopsies underwent investigations for cardiac amyloidosis, including ECG-recording, echocardiography with strain imaging, cardiac magnetic resonance imaging, and whole-body scintigraphy (99m-tc-DPD). Genetic investigation was done in amyloid positive patients (ATTR, and amyloid of unspecified subtype). Furthermore, amyloid positive patients were investigated by tests including NT-proBNP, troponins, immunoglobulins, M-protein in serum and urine, and free light kappa and lambda chains in serum. Subcutaneous fat aspirates were taken from the abdominal wall, and a bone marrow aspirate and biopsy were performed in cases suspected for light-chain (AL) amyloidosis.

Results: Seventeen percent (17/100) of patients had amyloid positive biopsies. Amyloid positive patients were significantly older (73 years vs 55 years, p<0.001) and the prevalence of males was significantly higher in the amyloid positive group (47% vs 22%, p=0.038).

The subtype of amyloid was shown to be ATTR in 13 patients, one patient had localized AL amyloidosis and one patient had fibrinogen alpha amyloidosis. For two patients the amyloid subtype was undetermined due to: 1. Insufficient material for MS, 2. Not representative material for IEM, or 3. A limited panel of antibodies, hampering detection of rare amyloidosis by IEM.

None of the amyloid positive patients were identified with organ involvement. Subcutaneous fat aspiration was positive for amyloid in one ATTR patient. All ATTR patients had normal genetic investigations of the gene for ATTR.

Conclusions: A considerable number (17%) of CTR patients had amyloidosis confined to the carpal tunnel, with wild-type ATTR being the most common finding (76%). None of the subjects had signs of cardiac or other organ involvement. However, the proportion of CTR patients with localized amyloidosis who may eventually develop systemic disease is unknown. Therefore, it is necessary to perform long term follow-up of these patients before routine investigations for amyloidosis in CTR patients may be considered.

Keywords: Carpal tunnel syndrome, amyloid screening, amyloid subtyping
PM058

CHANGES IN THERMODYNAMIC STABILITY OF FIBRILS MADE WITH DIFFERENT METHODS

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Introduction: Methods suitable for the creation of amyloid in vitro may provide crucial clues to understand the biophysical basis of amyloid formation and offer important tools for drug discovery. A desirable method for amyloid formation in vitro should use biocompatible conditions to generate fibrils with structural characteristics closely resembling those of natural fibrils.

Objectives: Our aim was to compare the thermodynamic stability of V122I transthyretin (TTR) fibrils prepared in vitro at low pH or in physiological buffer containing traces of a protease with the stability of natural fibrils.

Methods: V122I TTR fibrillogenesis was carried out in vitro using the mechano-enzymatic mechanism recently described (Marcoux et al, Embo Mol Med 7:1137-49, 2015) and the widely used low pH procedure established by Colon and Kelly (Colon & Kelly, Biochemistry 31:8654-60, 1992). Both the two types of synthetic V122I TTR aggregates were characterized in comparison with natural fibrils extracted from the heart of a patient carrying the V122I TTR mutation following the classical water extraction procedure. The stability of fibrils obtained through different procedures and/or sources was analysed by incubating them at increasing concentrations of the chaotropic agent, guanidine thio-cyanate (Gdn-SCN). The disassembly of the fibrils was followed monitoring the loss of the specific fluorescence of the thioflavin T/ fibrils complex.

Results and conclusions: A remarkable difference in thermodynamic stability was observed between the two types of in vitro fibrils. Whilst the aggregates at low pH did not show any significant change in their stability compared with the precursor, the fibrils prepared by the mechano-enzymatic mechanism were much more stable than the native TTR and they appeared thermodynamically similar to those extracted from natural sources. Our data strongly suggest that the combination of proteolysis and biomechanical forces generates aggregates with thermodynamic features that more closely resemble the amyloid fibrils occurring in patients.
TAKING AIM AT AMYLOID DISEASES BY MEANS OF SERUM AMYLOID P COMPONENT-SPECIFIC NANOBODIES

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Introduction: The pentraxin protein serum amyloid P component (SAP) decorates amyloid fibrils of any origin like beads on a string. This small glycoprotein, produced in the liver, protects fibrils from phagocytosis. SAP is elevated in amyloidosis patients. We have generated so-called nanobodies which are the antigen binding part of camelid heavy chain antibodies and excel in small size, high solubility, specificity and affinity and ease of cloning. These properties have triggered numerous applications in fundamental research, diagnostics and therapy. These nanobodies can learn us more about the mode of action of SAP in amyloid diseases. Their susceptibility to post-production manipulation allows them to develop into a cheap diagnostic amyloid imaging agent. Nanobodies can access cryptic epitopes in their target, unaccessible to other antibodies.

Objectives: Our hypothesis claims that SAP nanobodies represent a novel instrument to help explain how SAP partakes in amyloid buildup by perturbing its activity, and to understand the biology of SAP in a broader context. We aim to test this hypothesis from a fundamental perspective and from a pre-clinical angle, with a focus on SAP in amyloid disorders. SAP knock-out mice have reduced amyloid deposition. SAP has also been shown to promote fibrillogenesis in vitro. Hence, SAP acts as a fibril-protecting component. SAP nanobodies could potentially have anti-amyloidogenic effects and even if not also be further developed as a diagnostic tool. We have used ⁹⁹mTc-labeled nanobodies before to image amyloid deposits in mice that recapitulate familial gelsolin amyloidosis.

Methods:
- Co-immunoprecipitation reactions: the ability to extract SAP from human/mouse serum (73% aa identity)
- Thioflavine T assay: study anti- or pro-amyloidogenic effects
- X-ray crystallography: epitope determination combined with ThT-assays to determine the mode of action of SAP
- Mass spectrometry approach to detect amyloid peptides bound to serum SAP
- Techniques to enhance nanobody delivery to the brain (Alzheimer’s)

Results: By means of co-immunoprecipitation and western blotting, 30 nanobodies were shown to be capable of pulling SAP down from human serum. Yet, none were capable of depleting SAP out of mouse serum. For this reason, nanobodies were generated against mouse SAP. This resulted in 5 distinct nanobodies capable of pulling down SAP out of mouse serum, but not out of human serum.

Conclusions: Anti-SAP nanobodies can provide a new tool to investigate SAP’s biology in regard to amyloid diseases. Furthermore, these nanobodies can exert anti-amyloidogenic effects which can lead to potential therapeutic consequences. Nanobodies can also be used as a diagnostic tool, either by detecting amyloid-bound SAP in vivo or by identifying peptides bound to serum SAP with mass spectrometry enabling early stage diagnosis of amyloid diseases such as Alzheimer’s.
CONFORMATION OF HUMAN APOLIPOPROTEIN A-I AMYLOID VARIANTS: FOLDING AND THE ROLE OF INFLAMMATION IN THE PROTEIN AGGREGATION DISEASE

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Background: The reasons that determine the pathological deposition of human apolipoprotein A-I variants inducing organ failure have been under research since the early description of natural mutations in patients. Different protein conformations may be involved in the development of clinical manifestations associated with human amyloidosis. Although a fibrillar conformation is usually the signature of damage in the tissues, it is not clear whether this species is per se the cause or the consequence of the disease. From the more than 20 amyloidogenic variants of apoA-I described1, a mutation leading to a deletion at position 107 (Lys107-0) has a unique pattern, as patients carrying this variant show amyloidosis and severe atherosclerosis.

Objectives: Here we set out to characterize protein aggregation structures, and to test the hypothesis that a pro-inflammatory microenvironment could favor protein misfolding.

Methods: Soluble, freshly folded proteins were obtained and purified. Both, Wt and Lys107-0 were oxidized under controlled concentrations of hydrogen peroxide and incubated by 30-day. Structure of soluble and incubated species was analyzed by fluorescence, circular dichroism and microscopy approaches. In order to answer whether these structures may induce cellular events associated with the chronic inflammatory scenario, we incubated apoA-I variants (either soluble or aggregated) with human neutrophils and analyzed by confocal microscopy the release of Neutrophil elastase traps (NETs).

Results: Lys107-0 was more unstable, contained less α helix secondary structure (14 % vs 63% for the Wt) and showed a more flexible spatial arrangement. In addition, it had a higher tendency to aggregate and fibrils in a high yield were obtained after oxidation of this variant (which were not present for the Wt). These fibrils bound ThT, lost α helix content and were able to induce the release of Neutrophils Extracellular Traps. This effect was lower or not significant for soluble species.

Conclusions: We conclude that a pro-inflammatory microenvironment could result in the formation of aggregation-prone species, which, in addition may induce a positive feed-back in the activation of an inflammatory response.


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Keywords: amyloidosis, protein folding, apolipoprotein A-I, inflammation
Introduction: Uncommonly, a collection of amyloid can grow large enough to be classed as an amyloidoma, a macroscopic tumor like deposition of amyloid that can cause mass effect. It may present as isolated nodule in the absence of a plasma cell dyscrasia or represent a complication of light-chain (AL) amyloidosis with systemic involvement. Amyloid deposits can appear in tongue, lymph nodes, bone, skin, larynx, bladder, eyes and gastrointestinal system. Primary or secondary spine amyloid deposition represents one of the rarest forms. Only a few cases of Amyloidosis affecting cervical spine have been reported.

Clinical summary: A case of A 77-year-old man is herewith described. Two years earlier he received a biopsy, for pain and swelling in his right shoulder. The histological examination documented amyloid deposition and subsequent examinations led to the diagnosis of kappa light chain amyloidosis associated with smouldering myeloma with shoulder infiltration but no heart or kidney involvement. Baseline staging showed FLC k 1756 mg/L, λ 10,40 mg/L, Bence-J 1250 mg/24h. FISH showed a CCND1/IGH fusion, t(11;14) in 58% of abnormal plasma cells. He was treated with first line CVD showing a refractory disease, then salvage with Lenalidomide was started and patient was able to achieve a partial response. One year after diagnosis he complained of tingling in the feet. Electromyographic examination (EMG) documented peripheral sensory motor neuropathy. For this reason, the patient underwent an outpatient visit. Neurological examination showed also hyperreflexia and spastic hypertonia. Four-limb muscle motor evoked potential (PEM) demonstrate abnormal motor system conduction. Magnetic resonance imaging (MRI) revealed a T2, contrast-enhancing mass at joint C1 – C2 with severe mass effect and initial signal alteration of the corresponding spinal cord. Fluorodeoxyglucose Positron emission tomography and CT-scan could not differentiate between amyloidoma and myeloma. The patient underwent C2 laminectomy and lesion resection with decompression, furthermore, a stabilization of the structures with screws and plates was performed. Histological examination revealed amyloid deposit in the tumor-like lesion.

Conclusion: Tumor-like localized amyloid deposits are a rare amyloidosis-related condition. This work suggests the possibility that amyloid may settle even in non-typical areas. Early diagnosis and treatment are crucial for patient prognosis.

PM062

**DISSECTING RENAL AMYLOIDOSIS WITH MALDI - IMAGING MASS SPECTROMETRY AND SHOTGUN PROTEOMICS ON PARAFFIN EMBEDDED RENAL BIOPSY TISSUE SECTION**

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**Introduction:** The amyloidoses are a group of disorders in which soluble proteins aggregate and deposit extracellularly in tissues as insoluble fibrils. The kidney is one of the most frequent sites of amyloid deposition in AL, AA. The fibrils have a characteristic appearance by electron microscopy and generate birefringence under polarized light when stained with Congo red dye. While current diagnosis relies on histopathological examination, commercially available antibodies lack specificity and sometimes failed to diagnose properly.

**Objectives:** In this study, we aimed to identify and to subtype amyloid proteins with high accuracy in paraffin-embedded tissue sections fixed in formalin-acetic acid-alcohol (FAA) fixative, using matrix-assisted laser desorption/ionization imaging mass spectrometry (MALDI-IMS). Furthermore, we established an in depth tissue proteomics at single nephron level integrating MALDI-IMS and shotgun analysis.

**Methods:** Formalin-acetic acid-alcohol (FAA) fixed paraffin-embedded tissue samples were transferred from Georges-Pombidou European Hospital and Necker Children’s Hospital. Four sample groups of AL (kappa), AL (lambda), AA amyloidosis, and non-pathological control were analyzed. Continuous slide thickness of 10 μm was made using ITO coated slide glass. Pretreatment was carried out as 1) Dewaxing, 2) Antigen retrieval, 3) On-tissue digestion with trypsin, 4) Deposition of MALDI matrix HCCA and α-cyano-4-hydroxycinnamic acid (CHCA) using TM-Sprayer TM. MALDI-IMS was done by using rapifleX MALDI Tissue typer with a spatial resolution of 50μm and 20μm. Statistical analysis of mass spectra was performed with SCiLS Lab 2018 software. Shotgun Proteomics from serial sections of MALDI - IMS were attempted using TOF Pro with nanoElute system.

**Results:** Renal FFPE biopsies from patients affected of AL amyloidosis and AA amyloidosis and patients negative for amyloid used as control were retrieved. Ethical approval for the use of renal biopsy tissue was obtained. Ten micron thick frozen sections of renal biopsy samples were cut and transferred to conductive Indium-Tin-Oxide coated glass slides. Samples are digested with trypsin. Matrix used was α-cyano-4-hydroxycinnamic acid. MALDI-imaging was done with rapifleX tissuetyper with spatial resolution of 50μm and 20μm. For the serial section of the tissues exactly prepared with the same protocol for IMS, proteins and peptides were extracted to be analyzed with timsTOF Pro with nanoElute. Obtained mass spectra as well as annotated proteins and peptides were visualized with flexImaging and SCiLS Lab 2019 Software. Renal amyloidosis and non-amyloidosis biopsies were processed for histological and MS analysis. Mass spectra corresponding to amyloid positive areas were directly linked to the histological and MS images for correlation studies. Peptides for SAA and AL were detected by MALDI IMS associated to DFS staining-positive areas. Sequence determination of amyloid peptides by LC-TIMS-MS/MS analysis provided protein distribution and identification. Visualization of deposited proteins such as Serum amyloid P component, apolipoprotein E, vitronectin with several uncharacterized proteins were identified in both AA and AL amyloidosis as an amyloid genic protein. Furthermore, we have succeeded in visualization of proteins with 20μm resolution to realize a single nephron proteomics for the studies of renal pathology.

**Conclusions:** MALDI-imaging in combination with a shotgun proteomic analysis is a powerful approach as a new method to type amyloidosis in histopathological routine material and characterize amyloid-associated proteins that may provide insights into the pathogenetic process of amyloid formation. Especially, high resolution MALDI-IMS enabled single nephron proteomics on renal biopsy samples.
Introduction: Preclinical evaluation of peptide p5+14 revealed its ability to bind the hypersulfated glycosaminoglycans and amyloid fibrils resulting in reactivity with many forms of amyloid. In 2018, we began a Phase 1 PET/CT imaging trial of iodine-124 (124I)-labeled peptide p5+14 in patients with systemic amyloidosis (clinicaltrials.gov NCT 03678259). To date, the following non-AL forms have been imaged: 4 ATTR patients (variants T60A, A81V and L58H) and one ALECT2 patient. Presently, cardiac ATTR amyloidosis is detected clinically by imaging of 99mTc-labeled bone-seeking agents such as PyP (in the US) and DPD (in Europe). For patients with ALECT2, there are no routine methods for imaging the whole-body distribution of amyloid. Thus, the 124I-p5+14 radiotracer was developed to provide quantitative detection of systemic amyloid in patients with diverse forms of the disease.

Objectives: The aim of this study was to determine the efficacy of 124I-p5+14 as a multi-amyloid imaging agent capable of visualizing amyloid deposits in patients with diverse forms of systemic amyloidosis, including ATTR and ALECT2. Methods: Patients >18 y of age with a diagnosis of amyloidosis and not requiring heparin therapy are eligible. Subjects received <2mg of 124I-p5+14 (<2 mCi) administered as a single IV infusion. PET/CT images were acquired at 5 h and 24 h post injection. Patients were imaged using a Biograph 16 PET/CT scanner using a low-dose CT. Whole body images were acquired using 5 min bed positions, followed by 10 min over the lower legs (to assess peripheral nerve amyloid) and a 10 min gated dynamic cardiac dataset. Radiotracer uptake in abdominothoracic organs was quantified using region of interest analysis and expressed as organ-to-blood ratios, which served to control for differences in the rate of renal tracer clearance.

Results: All ATTR patients retained 124I-p5+14 in the heart, with a mean heart-to-blood ratio of 2.7, as compared to 1.0 for the spleen (an organ not involved in patients with ATTR). The NT-proBNP was elevated in only one patient (1510 pg/mL), whereas the 3 other patients had normal levels. In one patient with crippling peripheral neuropathy in the legs and one with neuropathy of the left hand, binding of the radiotracer was observed in both sites. Lastly, half of the ATTR patients revealed uptake in the articular cartilage region of the humerus and femur. In the ALECT2 patient, uptake of radiotracer was observed in the kidneys, spleen, liver and (possibly) adrenal gland consistent with the established distribution of the disease.

Conclusions: Despite the small cohort of patients, we have shown pathologically consistent uptake of 124Ip5+14 in patients with ATTR and ALECT2 amyloidosis. All ATTR patients had positive cardiac uptake independent of NT-proBNP levels and diagnostic symptomology (neuropathy vs cardiac amyloidosis), indicating that this tracer may be sensitive enough to detect asymptomatic cardiac amyloidosis in ATTR patients.
ABDOMINAL FAT PAD BIOPSIES EXHIBIT A GOOD DIAGNOSTIC ACCURACY IN PATIENTS WITH SUSPECT TRANSTHYRETIN AMYLOIDOSIS.

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Background: Amyloidosis is traditionally diagnosed by demonstration of amyloid deposits in tissue samples; however, the diagnostic procedures vary between countries. In Sweden, we have a long tradition of performing abdominal fat pad biopsies using a skin punch. Our clinical experience is that this is a reliable method for diagnosing transthyretin amyloid (ATTR) amyloidosis but the diagnostic accuracy of the procedure has been questioned, especially for patients with ATTR cardiomyopathy.

Objectives: To evaluate the diagnostic accuracy of abdominal fat pad biopsies in patients with suspect amyloidosis.

Methods: Data from patients who had undergone abdominal fat pad biopsies using an 8 mm skin punch due to suspect amyloidosis at the Amyloidosis Centre, Umeå University Hospital from 2006 to 2015 were evaluated. The biopsies had been stored in a saline solution and later analysed at the department of Clinical Pathology, Uppsala University Hospital using Congo red staining to determine presence of amyloid, and immunohistochemistry or Western blot analysis to determine the type of amyloidosis (i.e. precursor protein and amyloid fibril type). Final diagnosis was based on symptoms, biopsy results, DNA sequencing and, in some cases, DPD scintigraphy. Minimum follow-up after first biopsy was 3 years.

Results: Two hundred seventy-four patients (61 % males) were identified. Mean age at examination was 60 years, and 133 (49 %) patients got a final diagnosis of amyloidosis. A majority (92 %) were diagnosed with variant ATTR (ATTRv) amyloidosis, of which 95 % had ATTRV30M amyloidosis. Due to the low number of patients with other types of amyloidosis, subsequent analyses were focused on patients with ATTRv amyloidosis. Nineteen patients were excluded due to an established amyloidosis diagnosis before 2006. Overall, our data showed a test specificity of 99 % and a sensitivity of 87 % (positive and negative predictive values of 0,99 and 0,86, respectively). Ninety-eight (93 %) of the patients had neuropathic symptoms at diagnosis, whereas 32 (31 %) had gastrointestinal symptoms and 56 (53 %) had signs of amyloid cardiomyopathy. Subgroup analyses showed that the patients with merely neuropathic symptoms (n = 48) displayed the highest test sensitivity of 90%, whereas patients with pure cardiomyopathy (n = 6) displayed the lowest sensitivity of 83 %. However, no significant differences in sensitivity were found between patients with or without cardiomyopathy or between the sexes.

Conclusions: Abdominal fat pad biopsies exhibit a good diagnostic accuracy in patients with suspect ATTRv amyloidosis when histopathology is performed at a specialized centre. Further, test sensitivity was not significantly lower in patients with cardiomyopathy. We therefore conclude that abdominal fat pad biopsies is a good diagnostic tool in ATTRv amyloidosis that enables typing of the precursor protein and of the amyloid fibril type, which is related to the phenotype and to the outcome of the disease.
PATIENT-REPORTED OUTCOMES ON FAMILIAL AMYLOID POLYNEUROPATHY (ATTR-FAP)

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Background and Objectives: Transthyretin familial amyloid polyneuropathy (ATTR-FAP) is a rare autosomal dominant inherited disease affecting multiple organ systems. ATTR-FAP patients’ experiences have rarely been documented. The aim of this study was to collect patient reported outcomes across different countries to address unmet needs and challenges.

Methods: An anonymous survey was conducted at the 2nd European meeting on ATTR amyloidosis in Berlin from September 1st to 3rd 2019. Survey questions captured information on demographics, clinical characteristics, diagnostic experience, quality of life (PROMIS-10), disability (WHODAS 2.0 12-item instrument) and therapies in ATTR-FAP.

Results: A total of 38 ATTR-FAP patients from 15 different countries participated in the survey. The mean age of patients with ATTR-FAP was 53.3 years and 79% were men. Median patient reported time from first symptom onset to diagnosis was 2 years (IQR 1-4, range from 1 to 10); 25% of patients required more than 4 years for an ATTR-FAP diagnosis. During this time, patients required a median of 3 visits to health care professionals (IQR 1-15, range from 1 to 50), yet 25% of patients required more than 15 visits. Following conversion of the PROMIS10 global physical health score into T-score values, the global physical health of ATTR-FAP patients was half a standard deviation worse than the general US population. The functioning level of ATTR-FAP patients (WHODAS 2.0 Disability score: 0 = full function and 100 = no function) ranged tremendously from 0 to 77 with substantial impact on patients’ day-to-day life, including difficulties in standing, walking, and participation in community activities. It also had negative effects on the mental health of patients. Almost half of the patients participating in the survey were currently being treated with gene silencing drugs such as Patisiran (34.4%) and Inotersen (9.4%) and the other half with TTR stabilizers such as Tafamidis (28.1%) and Diflunisal (21.9%). 18.8% of patients underwent liver transplantation.

Conclusions: This global patient survey highlighted several unmet needs and challenges from a patients’ perspective, including (i) a need for increased disease awareness, (ii) a standardized care pathway and (iii) a need for better supportive care. Moreover, it provides valuable findings to further the goal of patient-centered care in ATTR-FAP by setting individual treatment goals and monitor the treatment response independent of neurologic and cardiologic evaluations.
ANTICIPATION IN HEREDITARY TRANSTHYRETIN AMYLOIDOSIS IN MALLORCA

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Introduction: Hereditary transthyretin (ATTRv) amyloidosis is a rare multisystemic disease caused by amyloid deposition and characterized by a heterogeneous presentation and symptomatology. Anticipation is described as the decrease in age at onset with each generation.

Objectives: Our aim was to study anticipation in ATTRv amyloidosis kindreds in Majorca (Spain).

Methods: In a cohort of 262 subjects with ATTRv, we found 37 affected pairs for inclusion in our study. Disease onset (age at the first symptom) and anticipation (parent’s age at disease onset minus that of the offspring) were calculated. Chi-square test, t-test, and paired t-test were used for comparisons between groups. Association between age at onset of parents and offsprings were assessed by Pearson’s correlation coefficient.

Results: Offspring mean age at onset was 16 years (1-52) lower than that of the parents (p<0.001), regardless of the sex of the parents and the offspring (table 1). Anticipation occurred in 31 out of the 37 pairs, with no differences related to the sex of parents or offspring. There was a moderate correlation (r=0.49; p<0.001), between age at onset of the parents and that of the offsprings.

<table>
<thead>
<tr>
<th>Sex</th>
<th>Parents (n=27)</th>
<th>Offsprings (n=37)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>AO</td>
<td>55 (18)</td>
<td>39 (12)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Sex (%)</td>
<td>41.3 %</td>
<td>43.2 %</td>
<td>NS</td>
</tr>
<tr>
<td>AO (females)</td>
<td>59 (19)</td>
<td>40 (15)</td>
<td>0.006</td>
</tr>
<tr>
<td>AO (males)</td>
<td>52 (16)</td>
<td>38 (11)</td>
<td>0.003</td>
</tr>
</tbody>
</table>

Table 1: Comparison of age at onset and sex in the study cohort.

Results are expressed as means (SD) unless otherwise stated. AO: age at onset; NS: not significant

Conclusion: Anticipation was common in our cohort, and age at onset tended to decrease in successive generations. This observation has important consequences for the management of asymptomatic carriers.
CLINICAL PICTURE OF IATROGENIC TRANSTHYRETIN AMYLOIDOSIS AFTER DOMINO LIVER TRANSPLANTATION


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Introduction: Domino liver transplantation (DLT) with liver grafts from patients with hereditary ATTR (ATTRv) amyloidosis has been performed for patients with fatal hepatic disorders. According to the Familial World Transplant Registry, 1,254 domino liver transplantations (DLTs) were done from 1995 to 2017. More than 20 years have now passed since the first DLT, and the number of patients with iatrogenic ATTR amyloidosis after DLT is increasing.

Objective: The objective of this study was to elucidate the precise clinical picture of iatrogenic TTR amyloidosis.

Methods: We evaluated the presence of amyloid deposits and clinical symptoms in 30 recipients of DLTs (24 men and 6 women) who underwent liver transplantation with liver grafts explanted from patients with ATTRv amyloidosis. We analyzed symptoms and clinical parameters of 7 cases with symptomatic iatrogenic ATTR amyloidosis and compared those with 30 patients with ATTRv amyloidosis who gave a domino liver.

Results: We found amyloid deposition in 13 of 30 domino liver recipients. The mean time from DLT to amyloid first appearance in DLT recipients were 8.5 years. Our 7 symptomatic cases and the literature cases with iatrogenic ATTR amyloidosis presented with clinical features that differed from patients with ATTRv amyloidosis who gave a domino liver. Patients with iatrogenic ATTR amyloidosis showed markedly milder autonomic disturbance, which is one of the main symptoms of ATTRv amyloidosis.

Conclusions: Careful periodic monitoring is required for DLT recipients of ATTRv liver grafts because the time from DLT to disease onset has a wide range and the clinical picture of these DLT recipients is distinct from that of liver donors.
FREQUENCIES OF GENETIC MUTATIONS AND CLINICAL ASPECTS IN PATIENTS WITH ATTR: THE ITALIAN HOSPITAL OF BUENOS AIRES LABORATORY EXPERIENCE

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Background: In Argentina, there is limited data of prevalence of TTR amyloidosis, mutations or phenotype-genotype relationship.

Objectives: Describe the prevalence of transthyretin genetic mutations in Argentina and the clinical characteristics of patients with hereditary TTR amyloidosis in Italian Hospital of Buenos Aires (HIBA).

Methods: Cross sectional study of all consecutive blood samples referred to HIBA laboratory for TTR gene sequencing between 2012 and 2019. Our laboratory is a reference center for TTR sequencing in Argentina. Mutations in TTR were identified in peripheral blood by direct Sanger sequencing in all samples received but clinical characteristics were assessed only in those that were evaluated by a local physician at HIBA.

Results: In the period of interest, 576 patients were analyzed for TTR mutations. Twenty four percent (141) were positive for a TTR mutation. The frequency distribution of the mutations were: Val50Met 78% (2% were Homozygotes), Thr80Ala 11%, Ala117Ser 6%, Val142Ile 1%, Phe84Leu 1%, Ile127Val 1%, Tyr134Cys 1% and Ala56Pro 1%. Of the 141 mutated TTR patients, 20 were evaluated by a physician at HIBA. Female represented 30%, average age at diagnosis was 54 yo, 70% with family history and with a median of pedigree 4. Six different TTR variants were identified: Thr80Ala 9, Val50Met 6, Ala56Pro 2, Val142Ile 1, Phe84Leu 1 and Tyr134Cys 1. Most frequent organ involvement were: in Thr80Ala gastrointestinal 56%, cardiac 33%, neurologic 33%; in Val50Met neurologic 83%, gastrointestinal 83%, cardiac 33%, and ocular 17%; in Ala56Pro neurologic 50%, ocular 50%. The mortality rate was low of 5% (n = 1, CI 0.1% to 25%), the patient died of cardiac attack post a hepatic transplant.

Conclusion: In our knowledge, this is the first national report of the TTR mutations epidemiology in Argentina.
AMYLOIDOSIS TYPING AT AMYLOIDOSIS MEDICAL PRACTICE CENTER IN KUMAMOTO UNIVERSITY HOSPITAL

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Introduction: Recently novel therapies had been applied to several types of amyloidosis such as hereditary transthyretin (ATTR, ATTR\textsuperscript{v}) amyloidosis, ATTR\textsuperscript{wt} amyloidosis, AL amyloidosis, and AA amyloidosis. Early and accurate diagnosis is important for the effective therapies.

Objective: The purpose of this study was to analyze clinical characteristics of patients with amyloidosis diagnosed by Amyloidosis Medical Practice Center, Kumamoto University Hospital, which conducts histopathologic, proteomic, and genetic analysis for amyloidosis as an amyloidosis center in Japan.

Patients and Methods: Diagnosis results and the clinical manifestations of amyloidosis patients diagnosed during Apr, 2012 and Mar, 2019 were analyzed. Out of 3,393 patients who were consulted to our center, 1937 patients whose clinical information were obtained were included in this study.

Results: The average age of the patients was 65.9 (median 69) y.o. Male patients were 66.6%. Results of type diagnosis were follows; ATTR\textsuperscript{v}: 13.4%, ATTR\textsuperscript{wt}: 14.3%, ATTR (no request of genetic analysis): 4.6%, AL\textsubscript{\textlambda}: 19.7%, AL\textsubscript{\textkappa}: 8.8%, AA: 3.0%, A\textbeta2M: 0.8%, others (including EFEMP1 and semenogelin): 1.9%, no mutation in TTR gene (no request of histopathological analysis): 23.2%, (including duplicate consultations). Concerning ATTR\textsuperscript{v} amyloidosis, 7.4% was V30M from endemic area, 51.2% was V30M from non-endemic area, and 41.4% was non-V30M. Sixty-six cases needed laser microdissection (LMD) and liquid chromatography tandem-mass spectrometry (LC-MS/MS) analysis for diagnosis. Initial manifestations were follows: polyneuropathy: 13.2%, carpal tunnel syndrome: 5.6%, autonomic dysfunction: 2.7%, heart failure: 28.4%, cardiac hypertrophy: 2.2%, arrhythmia: 5.0%, renal impairment: 11.1%, stroke: 1.8%.

Conclusions: Type and clinical manifestations of amyloidosis were variable. For early diagnosis and appropriate intervention, more enlightenment activities and more development of diagnosis systems for amyloidosis are needed.
ELECTROPHYSIOLOGICAL FEATURES OF HEREDITARY ATTR AMYLOIDOSIS MIMICKING CHRONIC INFLAMMATORY DEMYELINATING POLYNEUROPATHY

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Introduction: Sporadic patients with hereditary ATTR amyloidosis are occasionally misdiagnosed as those with chronic inflammatory demyelinating polyneuropathy (CIDP). However, Electrophysiological features of hereditary ATTR amyloidosis mimicking chronic inflammatory demyelinating polyneuropathy (CIDP) have not been fully elucidated.

Objectives: To elucidate the electrophysiological features in patients with hereditary ATTR amyloidosis misinterpreted as CIDP.

Methods: In 102 patients with hereditary ATTR amyloidosis (85 Val30Met and 17 nonVal30Met; 37 and 65 from endemic and non-endemic areas, respectively), we retrospectively investigated motor nerve conduction studies (MNCSs) with a 2-Hz low-cut filter in the unilateral ulnar and tibial nerves and assessed whether MNCS parameters fulfilled the European Federation of Neurological Societies/Peripheral Nerve Society Electrodiagnostic (EFNS/PNS EDX) criteria for CIDP. Distal compound muscle action potential (DCMAP) duration is dependent on low-cut filter settings. However, the EFNS/PNS EDX criteria for CIDP defines the cut-off value of DCMAP duration with a 20-Hz low-cut filter setting only. Therefore, we evaluated DCMAP duration using not only the EFNS/PNS EDX criteria but also the cut-off value in a 2-Hz low-cut filter setting proposed by Mitsuma et al. (2015).

Results: Thirteen patients satisfied the definite EFNS/PNS EDX criteria for CIDP. There were no significant differences in clinical background between patients with and without the definite EFNS/PNS EDX criteria. All thirteen of these patients showed prolongation of DCMAP duration in the ulnar nerve and marked reduction of compound muscle action potential (CMAP) amplitude in the tibial nerve (0.7 ± 0.7 mV). Prolonged distal latency and abnormal temporal dispersion were observed in 5 in the tibial nerve patients. Reduced motor conduction velocity was observed in 1 patient in each nerve only. No patient exhibited conduction block in any nerve. Under the cut-off value proposed by Mitsuma et al, ten of 13 patients with the definite CIDP criteria were downgraded as those with the possible CIDP criteria.

Conclusions: Severe axonal degeneration shows electrophysiological demyelinating features without conduction block in patients with hereditary ATTR amyloidosis. To minimize misinterpreting hereditary ATTR amyloidosis as CIDP, analysis of DCMAP duration based on low-cut filter settings would be needed.

Keywords: Hereditary ATTR amyloidosis, nerve conduction study, axonal degeneration
EVALUATION OF RENAL TUBULAR FUNCTION IN HEREDITARY TRANSTHYRETIN AMYLOIDOSIS PATIENTS

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Introduction: Patients with hereditary transthyretin (ATTRv) amyloidosis usually present with neurological (sensorimotor and autonomic neuropathy) and cardiology manifestations, but may also present with renal manifestations due to glomerular or tubulointerstitial amyloid infiltration. There are rare studies evaluating tubular renal function in this population due to subclinical initial presentation of renal tubulointerstitial diseases.

Objective: To evaluate tubular renal function by urinary concentration and acidification tests in a cohort of hereditary transthyretin amyloidosis patients.

Methods: We included patients with ATTRv amyloidosis and preserved glomerular renal function with glomerular filtration rate (GFR) > 60ml/min/1.73m2 measured by CKD-EPI equation. Urinary concentration capacity was evaluated by measuring urine osmolality (uOSM) (freezing point technique) and urine specific gravity (dipstick) after a 12h water deprivation test. Proximal and distal renal tubular acidification capacities were evaluated by urinary excretion of bicarbonate (urine gas analysis) and by measuring urine pH (upH) (urine gas analysis and dipstick) hourly after a screening test using furosemide and fludrocortisone, respectively. Urinary ammonium (uNH4 + ) and titratable acids (uTA) excretion were also measured for diagnostic of Renal Tubular Acidosis (RTA). Statistical analysis was performed by Mann-Whitney test or Spearman’s Rho coefficient of rank correlation.

Results: We selected 32 outpatients (43±9 years old, 38% male, 81% white skin color, BMI 24.3±4.1 kg/m2, and 7.3±7.8 years since ATTRv diagnosis). 29 patients had V30M mutation, 7 patients had history of liver transplantation and 3 patients were in use of tafamidis. The patients were divided in 2 groups according to the presence or absence of neurologic/cardiac symptoms: symptomatic (n=20) or asymptomatic (n=12). The two groups were homogenous (except for age: 50±13 x 35±11 years old, p=0.003). In comparison between symptomatic versus asymptomatic groups, we made the following diagnostics: impaired urinary concentration (65.0 x 16.6%, p=0.008); type 2 Proximal RTA (8.3 x 0%, p=ns); type 1 Distal RTA (20 x 8.3%, p=ns); type 3 Distal RTA (8.3 x 0%, p=ns). The values of uNH4 + and uTA excretions were similar in the 2 groups. There was good correlation between uOSM x specific gravity (Spearman’s Rho=0.783; p<0.001).

Conclusions: Urinary concentrating impairment was highly frequent in symptomatic ATTRv patients. Different types of RTA also can be diagnosed in these patients. Fasting urine specific gravity and upH (dipstick) were shown to be simple tools to exclude urinary concentration or acidification disorders. Early diagnosis of these disorders would allow preventive treatment to maintain renal function in these patients.

Keywords: ATTR, renal tubular acidosis, kidney tubules.
**PM072**

**11C-PITTSBURGH COMPOUND B - PET IMAGING AS A BIOMARKER FOR ATTR-TYPE CEREBRAL AMYLOID ANGIOPATHY**

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**Background:** Hereditary ATTR (ATTRv) amyloidosis is an autosomal dominant disease caused by transthyretin (TTR) gene mutation. Liver transplantation markedly improves survival in ATTRv amyloidosis. Ironically, however, the prolonged disease duration induces de novo central nervous system (CNS) amyloidosis, ATTR-type cerebral amyloid angiopathy (CAA), as choroid plexus continues to produce variant TTR.

**Objective:** The aim of this study is to investigate the utility of 11C-Pittsburgh compound B (PiB)-PET imaging as a biomarker of to evaluate radiological ATTR amyloid deposition in the brain and CNS symptoms in posttransplant ATTRv amyloidosis patients.

**Methods:** Brain PiB-PET was performed in 19 posttransplant V30M (p.V50M) ATTRv amyloidosis patients. Seven of them underwent follow-up second scan with 3 to 5 years interval. Correlations in disease duration and CNS symptoms and PiB-PET findings were analyzed.

**Results:** Increased brain 11C-PiB retention was observed in 12 patients and most of them had long disease duration longer than 10 years. Six of them showed CNS symptoms, i.e, five patients had transient focal neurological symptoms (TFNEs) and two patients had cerebellar hemorrhage. All patients with CNS symptoms had a long disease duration (> 15 years). Higher standardized uptake value ratio (SUVR) was correlated with longer disease duration. Among the seven patients with second scan, six patients showed no significant change in brain accumulation but one patient with the longest scan interval (approximately five years) showed stronger brain retention as compared with the first scan.

**Conclusion:** CNS amyloid deposition become detectable by PiB-PET approximately 10 years after the disease onset. Subsequent CNS symptoms, including TFNEs and brain hemorrhage, develop in the next 5-10 years. 11C-PiB-PET may be a useful biomarker for the assessment of ATTR-type CAA.
HISTOPATHOLOGICAL AND BIOCHEMICAL ANALYSES OF CEREBRAL AMYLOID ANGIOPATHY IN HEREDITARY ATTR V30M AMYLOIDOSIS WITH AND WITHOUT LIVER TRANSPLANTATION

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Background: To date, more than 150 different mutations in the transthyretin (TTR) gene have been reported, most of which result in the development of hereditary ATTR amyloidosis. The V30M mutation, which is predominantly associated with a polyneuropathy phenotype, is most frequently form found in the worldwide. TTR is synthesized mainly in the liver and in the choroid plexus of the brain and retinal pigment epithelium. Liver transplantation (LT) can prevent the production of variant TTR in the liver. However, LT cannot prevent the production of TTR in the choroid and retinal pigment epithelium. Hereditary ATTR V30M amyloidosis rarely shows the central nervous system related symptoms corresponding to cerebral amyloid angiopathy (CAA) as the natural course of the disease. In addition, symptoms of CAA are often observed in ATTR V30M amyloidosis patients with long time course after LT.

Objectives: To elucidate the pathomechanism of CAA in hereditary ATTR V30M amyloidosis

Methods: To examine the frequency of CAA, we performed Congo red staining of autopsy brain tissue samples obtained from patients with ATTR V30M amyloidosis who underwent LT (n= 4) and did not undergo LT (n= 4). To investigate the biochemical features of CAA, we performed mass spectrometric analysis of samples isolated from amyloid-laden vessels of brain tissues using laser microdissection. We also performed the same analyses using heart tissues to compare the biochemical features of CAA with those of amyloid-laden vessels in the heart.

Results: Amyloid deposition around the brain vessel walls compatible with CAA was observed in 8 (97 %) of 9 brain samples. The proportions of variant TTR in the total TTR in CAA amyloid fibrils were 100 % in patients who did not undergo LT and 96 % in patients who underwent LT. In contrast, the proportions of variant TTR in the total TTR in amyloid fibrils of the heart vessels were 53 % in patients who did not undergo LT and 4.3 % in patients who underwent LT. Conclusions: Amyloid formation in the brain vessels strongly depends on variant TTR synthesized in the choroid plexus. Except for the therapy for systemic organs amyloid deposition, disease modifying therapy targeted at the choroid plexus is needed to prevent CAA in hereditary ATTR V30M amyloidosis.
**PM074**

**THE ROLE OF PATIENT INPUT IN DEVELOPMENT OF THE ATTR AMYLOIDOSIS PATIENT SYMPTOM SURVEY**

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**Introduction:** Amyloid transthyretin (ATTR) amyloidosis is a rare, progressive, and often fatal disease characterized by deposits of misfolded protein aggregates. Clinical characteristics and symptoms of ATTR amyloidosis vary greatly across patients, and primarily depend on which organs are affected by the disease. Patients may also experience change over time in the type and severity of symptoms. Standardized tracking of symptoms is imperative to understanding disease progression and response to treatment.

**Objective:** This study incorporated 2 separate patient focus groups to support the development of a new patient-reported outcome measure, the ATTR Patient Symptom Survey (ATTR-PSS), which can be used by patients with either wild-type or hereditary ATTR amyloidosis.

**Methods:** Information to inform the development of the ATTR-PSS was gathered through literature review, discussions with 2 US-based patient advocacy group leaders, and input from a patient focus group. This focus group was held during a day-long in-person patient advisory board meeting with 10 patient participants; it included a 2-hour concept elicitation session during which patients discussed ATTR amyloidosis symptoms and impacts. Feedback was used to create an initial draft of the ATTR-PSS, which was presented to a second patient focus group that included 7 participants. This 90-minute focus group included a concept elicitation session to yield spontaneous reports of symptoms and impacts, followed by a worksheet exercise during which patients checked all symptoms they had ever experienced due to their ATTR amyloidosis, and ranked which of those symptoms were most bothersome. Patients also participated in a cognitive debriefing of the ATTR-PSS. Transcripts were coded and analyzed, and the ATTR-PSS was revised based on analysis of the patient data.

**Results:** Feedback provided by patients across both focus groups resulted in the expansion of the survey’s symptom list and informed the best way to word questions in the ATTR-PSS. Cognitive debriefing confirmed the ATTR-PSS was relevant, comprehensive, and understandable; patient feedback provided at this stage resulted in the addition of 11 new symptoms and improvements to the clarity of instructions, item text, and response choices.

**Conclusions:** Patient focus groups provided important input to inform the development of the ATTR-PSS beyond what was identified through literature review. The addition of a second focus group likely reduced the number of changes that would result from an ongoing content validation study with clinicians and patients.

**Keywords:** Patient-Reported Outcomes; Symptoms; Survey
PM075

GENOTYPE AND PHENOTYPE OF HATTR IN SOUTHWESTERN PENNSYLVANIA AND TRI-STATE AREA (PA/OH/WV)

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Introduction/background:
Previous studies describe hereditary transthyretin amyloidosis (hATTR) in the US as a disorder of predominantly older men. Most common TTR mutations associated with hATTR in the US in the THAOS registry were Val122Iso (45.3%), Thr60Ala (20.4%) and Val30Met (6%). However, regional differences in the distribution of TTR mutations are not known. Cardiac-predominant phenotype is dominant in most world regions, except in Portugal where there is high prevalence of early-onset Val30Met which has predominant neuropathic manifestations.

Objectives:
To describe genotype and phenotype of hATTR patients in southwestern Pennsylvania and tri-state area (PA/OH/WV).

Methods:
Review of hATTR patients and carriers followed in two amyloid clinics at University of Pittsburgh Medical Center and Allegheny General Hospital in Pittsburgh, PA, USA.

Results:
There were 38 subjects with hATTR, including 28 manifesting patients and 10 asymptomatic carriers. Manifesting patients were 54% men with mean age of 68 years (range 30-81), and asymptomatic carriers were 10% men with mean age of 57 years (range 36-85). Most of the patients with symptomatic hATTR (93%) reported late onset of symptoms, after age of 50. Most common mutations were Val122Iso (n=14, 36%), Thr60Ala (n=10, 26%), Val30Met (n=8, 21%), Phe64Leu and Arg54Ser (n=2 each, 5.3%), Ala36Pro and Phe33Leu (n=1 each, 2.6%). Clinical phenotype included 23 patients with predominant amyloid cardiomyopathy and 7 patients with predominant neuropathy. There were 2 patients who received organ transplants (liver – 1, liver + heart – 1; 7% of manifesting patients)

Conclusions:
Our study demonstrates regional differences of genotype in southwestern Pennsylvania when compared to rest of United States. The most common mutation was Val122Ile, similar to previous reports for US population. Additionally, there was a greater prevalence of Val30Met mutation compared to rest of US, in the absence of significant endemic populations with high prevalence of Val30Met (e.g. Portuguese, Swedish, Japanese). Most patients presented with cardiac-dominant phenotype similarly as in other regions. Additional studies are needed to define regional variances of hATTR in different areas in US.
PERIPHERAL NEUROPATHY OUTCOME AFTER 2-YEARS OF TREATMENT WITH TAFAMIDIS IN
PAUCI-SYMPTOMATIC NON-VAL30MET HATTR AMYLOIDOSIS PATIENTS.

A. Gonzalez-Duarte

Introduction / Background: HATTR amyloidosis results in rapid progressing polyneuropathy. Tafamidis has demonstrated neurologic function improvement, but is still unclear its role in patients with minimal symptoms at the time of treatment onset.

Objective: To describe peripheral neuropathy progression in patients with minimal neurological findings after two years of treatment with tafamidis.

Design and Methods: Thirty patients (56% female, mean age 34.4 ± 10.8 years) with rare TTR mutations (73% Ser50Arg, 13% Ser52Pro, and 13% Gly47Arg) were included in an open-label clinical trial of 20 mg of tafamidis per day. Examination scores, quantitativative sensory testing and nerve conduction studies were performed. Punch skin biopsies were taken from 2 sites on the leg with intra-epidermal nerve density (IENFD) and Congo red staining for quantitation of amyloid deposition.

Results: Baseline Polyneuropathy Disability (PND) score was 0 in 21 patients, stage 1 in 8, and stage 2 in one; after two years was unchanged in 53%, improved in 10%, and worsened in 36%. Worsening was seen from PND 0 to I in 7(23%), from I to II in 1(3%), from I to IIIA in 2(6%), and from II to IIIB in 1(3%). At entry, neuropathy impairment scores (NIS) ranged from 0 to 38 points. Two years later, 7(23%) of patients had an improvement in NIS score (-4.1, -1 to -8points), and 11(36%) patients worsened (+7.6, 2 to 38 points). Small fiber function, assessed with UENS, QST CT, and WT in feet, improved in 30%, 23%, and 26% of patients and worsened in 40%, 26%, and 30%, respectively. Large fiber function assessed with QST VT, NCV sural, and peroneal amplitude improved in 43%, 26%, and 23% and worsened in 33%, 46%, and 30%, respectively. Change in biopsy IENFD and amyloid deposition index will be reported.

Conclusions: Tafamidis improved NIS scores in 7(23%), remained stable in 18(60%), and worsened in 11(36%) patients. Both small and large nerve fiber worsened equally, through clinical evaluations (NIS and UENS) and laboratory studies; however, mean NIS worsening was less than expected when compared to the natural history. Eight patients with PND of 0 converted to stage I, proving that disease progression begins early despite discrete or null neurological signs.
PM077

WHAT SHOULD AN ATTR AMYLOIDOsis PATIENT-REPORTED OUTCOME SURVEY MEASURE?
RESULTS FROM A LITERATURE REVIEW AND INTERVIEWS WITH KEY OPINION LEADERS

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Background: Patient-reported outcome (PRO) measures are a key aspect of patient-focused drug development and clinical care. In a complex disease like ATTR amyloidosis (ATTR), fully understanding the patient experience can be challenging without a disease-specific PRO. Such an instrument—created using input from the literature, experts, and patients to ensure the inclusion of relevant, comprehensive, and understandable concepts—may improve the measurement of disease and treatment impacts and the comparison of results across clinical studies. This study presents findings from a literature review and interviews with key opinion leaders (KOLs) conducted as part of a larger effort to develop an ATTR-specific PRO measure.

Objectives: This qualitative study used a literature review and interviews with KOLs to identify important concepts for inclusion in an ATTR disease-specific PRO meant for use in clinical practice and clinical trials.

Methods: Signs, symptoms, and impacts of ATTR were extracted from 26 peer-reviewed articles and used in the development of a semi-structured KOL interview guide. Clinicians and patient advocates with expertise in ATTR were identified and invited to participate in 60-minute telephone interviews. During the interviews KOLs reviewed a list of ATTR symptoms; provided input on wording, organization, and content; and answered questions about potential impacts of living with ATTR. Interview transcripts were analyzed to identify themes related to signs, symptoms, and impacts of the disease; how to identify and measure changes in disease activity; and best practices in collecting patient feedback.

Results: Sixteen KOLs were interviewed: 3 patient advocates and 13 clinicians specializing in hematology/oncology (n=5), cardiology (n=4), neurology (n=3), and gastroenterology (n=1). KOLs reported that patients experience a wide variety of symptoms and that symptom severity is best measured by asking patients about physical functional impacts. KOLs also described the need to measure the other major impacts that patients experience: declines in social, emotional, and financial well-being; and the inability to perform activities of daily living. KOLs advised that an ATTR-specific PRO should be available in paper and electronic form, take no more than 10 minutes to complete, and be available for patients to complete at home. They had conflicting opinions on format options for the survey, frequency of administration, and ideal recall period.

Conclusions: A literature review and interviews with KOLs revealed that an ATTR-specific PRO should measure symptoms and functional impairment as it pertains to physical, social, emotional, and financial well-being as well as activities of daily living. These findings will inform the next phase of the PRO development in which the research team will conduct a focus group and interviews with patients with ATTR amyloidosis to ensure the patient perspective is accurately represented in the PRO.

Keywords: ATTR amyloidosis, patient-reported outcome, qualitative research
PATIENTS WITH P.V142I/V122I HEREDITARY TRANSTHYRETIN AMYLOIDOSIS: INSIGHTS FROM A GENETIC TESTING PROGRAM

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Background: Cardiomyopathy and heart failure are frequent manifestations of hereditary transthyretin mediated (hATTR) amyloidosis, a progressive and fatal disease that results from the deposition of misfolded transthyretin (TTR) protein in major organs and systems, leading to multisystem dysfunction. Other common symptoms include polyneuropathy and carpal tunnel syndrome. The hATTR Compass Program offers confidential genetic testing and counseling to patients suspected of having, or with a family history of, hATTR amyloidosis.

Objectives: Report real-world data from the hATTR Compass Program for patients suspected of having hATTR amyloidosis.

Methods: This study analyzed patients with TTR mutations identified by the hATTR Compass Program. DNA samples were analyzed for TTR mutations associated with hATTR amyloidosis using a single-gene test, a 92-gene panel or an 81-gene panel targeting patients with a mixed phenotype or predominantly polyneuropathy, respectively.

Results: Of 165 patients identified with TTR mutations, 130 had the p.V142I/V122I ATTR mutation; 44.6% were female (n=58/130), with an average patient age of 67.5 years (range: 28–87 years), and, of patients with confirmed ethnicity, the majority were African American (n=108/113; 95.6%). Overall, 35 patients (26.9%) had a known family history, while 82 (63.1%) and 13 (10.0%) patients had no family history or did not know, respectively. Of patients who had symptoms reported, 62 (52.5%) had symptoms of both cardiomyopathy (CM) and polyneuropathy (PN), 40 (33.9%) had CM alone, and 12 (10.2%) had PN alone. Note that symptoms reported may be underrepresented due to limitations of data collection and program participation.

Conclusion: Many patients with the p.V142I/V122I mutation were identified by genetic testing through the hATTR Compass Program. In this study, most patients with p.V142I/V122I were African American and commonly presented with a mixed phenotype, including both cardiomyopathy and polyneuropathy. It is critical to recognize symptoms of hATTR amyloidosis and refer patients for genetic testing to facilitate diagnosis of this debilitating, fatal disease.

Key words: genetics, V122I, clinical
PATIENTS WITH HEREDITARY TRANSTHYRETIN AMYLOIDOSIS: INSIGHTS FROM A GENETIC TESTING PROGRAM

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Background: Hereditary transthyretin (hATTR) amyloidosis is a progressive and fatal disease that results from the deposition of misfolded transthyretin (TTR) protein in major organs and systems, leading to multisystem dysfunction, including peripheral neuropathy, cardiomyopathy, and autonomic dysfunction. The hATTR Compass Program offers anonymous, confidential genetic testing and counseling to patients suspected of having, or with a family history of, hATTR amyloidosis in the United States, Canada, and Puerto Rico.

Objective: To report real-world data from the hATTR Compass Program for patients suspected of having hATTR amyloidosis.

Methods: This study analyzed data from 165 patients with TTR mutations sequentially identified by the hATTR Compass Program. DNA samples were scanned for TTR mutations associated with hATTR amyloidosis using a single-gene test, a 92-gene panel or an 81-gene panel targeting patients with a mixed phenotype or predominantly polyneuropathy, respectively.

Results: The most commonly identified mutations were p.V142I/V122I (n=130), p.V50M/V30M (n=10), and p.T80A/T60A (n=12). The average patient age was 64.8 years (range, 25–87 years) and 53.9% were male (n=89/165). Within the patients testing positive for a TTR mutation, 62 (37.6%) had a known family history, while 88 (53.3%) and 15 (9.1%) patients had no family history or did not know, respectively. The TTR mutation-positive patients were 66.7% (110/165) African American, 16.4% (27/165) white, 6.1% (10/165) other ethnicities, and 10.9% (18/165) unknown. The majority of patients who tested positive for TTR mutations were referred by a cardiologist (110/165; 66.7%), while neurologists referred 8/165 (4.8%) patients. Patients were reported to have a clinical history of sensory, motor, and autonomic dysfunction, along with gastrointestinal dysfunction, heart disease, bilateral carpal tunnel syndrome, and lumbar spinal stenosis. Note that symptoms reported may be underrepresented because of limitations of data collection and program participation.

Conclusion: Diagnosis of hATTR amyloidosis is challenging, but recognition of its symptoms and subsequent genetic testing through the hATTR Compass Program can facilitate diagnosis of this debilitating, fatal disease.

Key words: genetics, clinical, demographics

Category: Hereditary amyloidosis (ATTR and others)
MOLECULAR MECHANISMS OF ATTR AMYLOIDOSIS

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The development of newer and efficient forms of treatment in the ATTR amyloidoses encompasses several steps: i) in one hand to counteract TTR aggregation either by stabilization of the native conformation, modification of fibril intermediates, disruption of amyloid, or by lowering the serum precursor; ii) counteract maladaptative pathways, such as inflammation, ER stress, ECM dysregulation; iii) regenerate the organ. We are in great demand to know in depth the comparative biology associated with organ injury and regeneration upon various pathological stimuli. Such is the case of the peripheral nerve in FAP when is exposed to TTR aggregates and or fibrils. In this regard, comparative molecular studies involving nerve injury and regeneration in V30M animals show us impairment of regeneration that guide us to maximize intervention measures in amyloid neuropathies.
PM081

A SEVERE AND AGGRESSIVE AMYLOIDOTIC POLYNEUROPATHY IN A MALAY MALAYSIAN FAMILY WITH A RARE TRANSTHYRETIN MUTATION (GLU54LYS)

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Introduction: Familial amyloid polyneuropathy (FAP) is a rare autosomal dominant peripheral neuropathy in Malaysians, the majority of FAP patients were ethnic Chinese1. We report the first case of FAP in an ethnic Malay Malaysian family with a rare mutation (Glu54Lys) with severe and aggressive phenotype.

Objective: To describe the clinical, electrophysiological, cardiac imaging and genetic mutation findings of Malay Malaysian family with FAP.

Case report: The proband was a 35-year-old Malay gentleman with non-consanguineous parents and no known family history of peripheral nerve or cardiac disease. He initially presented at the age of 31 years with progressive dysphagia, but no odynophagia causing significant loss of weight. He had no hyperthyroidism. At age 33, he developed distal lower limbs weakness and difficulty in walking. There was chronic diarrhea and bowel incontinence.

On examination revealed he was cachectic, pale with multiple purpura over all the limbs. He had postural hypotension of more than 30 mm Hg. There was hypotonia, wasting of the intrinsic muscles of the hands and feet, and weakness of distal muscles of the lower limbs. There was patchy sensory loss and impaired proprioception in all limbs.

Nerve conduction study showed length-dependent axonal sensorimotor polyneuropathy. Cerebrospinal fluid protein was high (1.25 mg/dL). Serum and urine electrophoresis were normal. Thyroid hormone, connective tissue screening, tumour markers, infectious disease and pulmonary tuberculosis workup were negative. Esophagastroduodenoscopy was normal. Rectum, skin and abdominal fat pad biopsies were negative for amyloid. Echocardiogram showed concentric left ventricular hypertrophy with mild tricuspid regurgitation. Cardiac MRI revealed normal left ventricular wall thickness, ejection fraction: 55% with abnormal myocardial nulling in the late gadolinium images. Technetium-99m scintigraphy showed moderate intense uptake in the myocardium.

Next generation sequencing revealed Glu54Lys mutation in transthyretin (TTR) gene in the proband and his young brother who presented with dizziness and syncope at age 31. Three other siblings were asymptomatic and did not have the mutation and patient’s three children (aged 12, 8 and 6) had not tested yet.

Discussion: Glu54Lys mutation has been reported in Japanese, Costa Rican and Turkish families2-4. Our family ethnic Malay, with no history of ancestors from other countries. This mutation causes rapid progression, with severe motor and autonomic neuropathy, early vitreous opacity with severe heart failure, with all reported patients dying under the age of 40 years2-4. Our patient had severe sensorimotor and autonomic neuropathy, and cardiac amyloidosis. He died at age 35, 4 years after presentation after developing intracranial bleeding.

Conclusion: We report of an ethnic-Malay Malaysian FAP family with Glu54Lys mutation, with severe, progressive and fatal phenotype.

Keywords: Familial amyloid polyneuropathy, Transthyretin, Glu54Lys

Reference:


Visualisation of Central Nervous Amyloid Deposits by 18F-Flutemetamol Positron Emission Tomography in Long-Term Surviving Transthyretin Val30Met Patients.


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Background. Hereditary transthyretin amyloid (ATTRv) amyloidosis caused by the Val30Met (p.V50M) mutation is characterised primarily by peripheral neuropathy and a shorter life expectancy than the general population. Liver transplantation has prolonged the patients’ survival, however, CNS complications attributed to amyloid angiopathy caused by amyloid formation from variant transthyretin (TTRv) synthesised within the brain have been reported.

Objective. The aim of the study was to ascertain CNS amyloid deposition by 18F-flutemetamol PET/CT examination in long-term ATTRv amyloidosis survivors and do a clinical evaluation of neurological signs and symptoms.

Patients and Methods. Twenty ATTR Val30Met patients with symptoms from the CNS and a median disease duration of 16 years (8-25 years) together with five Alzheimer (AD) patients, who served as positive controls were included in the study. The 18 ATTRv amyloidosis patients were clinically evaluated with Montreal cognitive assessment (MoCA) test and the neurological impairment lower limb (NIS-LL) scores. Amyloid CNS deposits were ascertained by 18F-flutemetamol PET/CT examination utilising relative z scores with pons as reference.

Results. Expectedly, all AD patients had a clearly increased global composite z score above 2.0 compared with about half (55%) of the ATTRv patients (Table 1). There was an increased cerebellar Z score in 12 ATTRv patients compared to only one in the AD group. Four ATTRv patients with increased cerebellar z score had a global composite z score within the normal range. No correlation between duration after 8 years and amyloid CNS deposition was noted. There was no correlation between MoCA score and PET/CT uptake. Notably, 9 of 20 amyloidosis patients had atrial fibrillation including 4 patients with radiologically diagnosed ischemic stroke.

Conclusions. Amyloid deposition within the brain after long-standing ATTRv amyloidosis is common and is often noted in the cerebellum. However, not all patients display amyloid CNS deposition and a correlation between amyloid deposition within the brain and disease duration or CNS symptoms could not be established in this study. Thus, additional causes for CNS complications should always be considered.

Table 1. 18F-flutemetamol retention in the brain of ATTR Val30Met patients and Alzheimer patients

<table>
<thead>
<tr>
<th></th>
<th>ATTR Val30Met</th>
<th>Alzheimer</th>
</tr>
</thead>
<tbody>
<tr>
<td>Composite global Z-score</td>
<td>2.2 (-0.3–5.0)</td>
<td>6.1 (5.0–9.0); P = 0.001</td>
</tr>
<tr>
<td>Correlation Z-score and disease duration from diagnosis</td>
<td>( \tau_s = -0.34 ) (NS)</td>
<td>NA</td>
</tr>
<tr>
<td>Z-score cerebellum</td>
<td>2.5 (-1.2 – 5.5)</td>
<td>0.4 (-0.2 – 1.1); P = 0.025</td>
</tr>
<tr>
<td>Correlation Z-score cerebellum and disease duration from diagnosis</td>
<td>( \tau_s = 0.11 ) (NS)</td>
<td>NA</td>
</tr>
</tbody>
</table>

Key words. Hereditary transthyretin amyloid amyloidosis, central nervous system
THE PATIENT JOURNEY PRIOR TO DIAGNOSIS OF HEREDITARY TRANSTHYRETIN AMYLOIDOSIS

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Introduction/Background: Despite emerging treatments for hereditary transthyretin (ATTRv) amyloidosis, the disease is often misdiagnosed, with reported diagnostic delays of up to several years. Knowledge of the patient journey leading up to diagnosis may help to promote earlier intervention.

Objectives: To examine patient clinical characteristics and healthcare utilization prior to ATTRv amyloidosis diagnosis.

Methods: Patients ≥18 years and newly diagnosed with ATTRv amyloidosis identified in IBM® MarketScan® Commercial and Medicare Supplemental data using a claims-based algorithm as follows: diagnosis required ≥1 medical claim with relevant amyloidosis diagnosis code (ICD-10-CM: E85.0-.4, E85.89, E85.9; excludes light chain and wild type) during identification (ID) period (1/1/16-12/31/17), and ≥1 occurrence of qualifying criteria during 2011-2017: ≥15 days diflunisal use without >30-day gap, liver transplant, or claim with specific codes E85.1 or E85.2. Index date was defined as the date of first claim with an amyloidosis diagnosis code in ID period. Patients had continuous enrollment ≥5 years pre-index date (look-back period). Occurrence of selected comorbidities (potential symptoms) and healthcare utilization (testing, procedures, and visits) measured during the look-back; demographics, physician specialty, and Charlson comorbidity index (CCI) measured 1 year pre-index. Patients with an ICD-9/10 amyloidosis code during look-back were excluded. An ATTRv-free cohort was created with patients without ATTRv matched 3:1 to ATTRv patients on age, gender, and region to provide reference values; same index and enrollment requirement as match.

Results: For the 141 qualifying ATTRv patients and 423 matched controls, mean (standard deviation) age was 62.5 (14.2) years and 53.9% were female. Mean CCI for ATTRv cohort was 2.7 (3.0) vs. 1.1 (1.9) among controls. Primary care providers were the most common specialty pre-index for both cohorts (46.1% and 54.4%, respectively). Selected comorbidities, testing, and visits were common among ATTRv patients during the look-back with higher rates vs. controls: neuropathy (26.2% vs. 5.9%), nausea/vomiting (27.0% vs. 13.0%), congestive heart failure (23.4% vs. 5.9%), ventricular hypertrophy (19.9% vs. 5.7%), diarrhea (17.7% vs. 11.1%), cognitive decline (17.0% vs. 9.2%), vitreous opacity (8.5% vs. 4.3%); blood/urine testing (34.8% vs. 9.5%) and biopsy/genetic testing (34.8% vs. 20.8%); ED visits (60.3% vs. 47.0%) and hospitalization (47.5% vs. 24.3%). In general, first occurrence of comorbidities and diagnostic testing was low during the look-back until a steep rise in the year before index diagnosis. First hospitalization and ED visit were fairly constant over the look-back period.

Conclusions: Patients with ATTRv amyloidosis experience myriad of comorbidities, testing, and hospitalization prior to diagnosis. Occurrence of potential markers of illness is most common in the year before diagnosis.
QUANTIFICATION OF SMALL FIBER NEUROPATHY AT THE TRUNK IN PATIENTS WITH HEREDITARY TRANSTHYRETIN AMYLOIDOSIS

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Background
Hereditary transthyretin (ATTRv) amyloidosis is an autosomal-dominant disorder caused by mutations in TTR gene, characterized by systemic accumulation of amyloid fibrils in peripheral nerves and various organs. Patients with ATTRv amyloidosis typically present with small fiber neuropathy (SFN), starting with a loss of sensation in the distal regions of the feet. Although similar sensory deficits at the anterior trunk are also observed at the early stage of this disease, truncal neuropathy has not been well investigated and quantified in detail. Skin biopsy quantifying intra-epidermal nerve fiber density (IENFD). It is a useful diagnostic method for evaluating SFN.

Objective
To elucidate truncal neuropathy in ATTRv amyloidosis.

Patients and Methods
We included 24 patients with ATTRv amyloidosis, 2 asymptomatic mutation carriers, and 3 healthy volunteers who visited Kumamoto University Hospital from April 2017 to March 2019. We investigated IENFD in the skin punch biopsy specimens from the anterior trunk, lateral trunk, and distal leg. Furthermore, clinical findings and the heat-pain detection thresholds were subsequently measured through Computer Aided Sensory Evaluator IV and nerve conduction studies (NCS).

Results
Significant decrease in the IENFD values (numbers/mm) was observed in patients with ATTRv amyloidosis (anterior trunk: 5.4 ± 3.6/mm; lateral trunk: 9.1 ± 3.3/mm; distal leg: 2.3 ± 2.3/mm). IENFD at the anterior trunk was lower than that at the lateral trunk in those patients. The value at the anterior trunk was significantly correlated with disease duration and peripheral neuropathy parameters, including sensory impairment evaluated by Kumamoto clinical score, heat-pain detection thresholds, and various parameters of NCS. There were considerably high rates of TTR amyloid deposits in skin punch biopsy specimens at the anterior trunk.

Conclusions
ATTRv amyloidosis causes length-dependent SFN at the trunk as the same way as the lower limb. Skin biopsy may be a useful biomarker to evaluate SFN and TTR amyloid deposits at the trunk in patients with ATTRv amyloidosis.

Keywords: small fiber neuropathy, intra-epidermal nerve fiber density, transthyretin
EFFECT OF ETHNIC BACKGROUND ON PHENOTYPE AT PRESENTATION IN V122IRELATED HEREDITARY CARDIAC AMYLOIDOSIS

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Background: V122I-related hereditary Transthyretin Amyloidosis (hATTR) is commonly associated with a cardiac phenotype. While mostly reported in patients of African descendant, a cluster of Caucasian V122I carriers has been identified in central Italy and sporadic cases have been reported worldwide.

Objectives: The aim of this study was to evaluate whether ethnic background is associated with differences in clinical and cardiac phenotype at presentation.

Methods: Clinical files of three amyloid referral centres were reviewed and V122I patients identified. Only patients with definite cardiac involvement (as defined by mean of endomyocardial biopsy or by a combination of echocardiography and bone tracer scintigraphy) were included. Baseline demographic and clinical data at diagnosis were collected, together with echocardiographic and electrocardiographic (ECG) parameters.

Results: 89 patients with V122I hATTR cardiac amyloidosis were identified: 70 black and 19 caucasian. Most patients were male in both groups (63% vs 79% respectively; p:0.188), and black patients were younger at presentation (72±8 vs 75±5; p:0.026). Heart failure was the common clinical presentation in both groups, with overall 61% of patients presenting with NYHA class ≥ 3 and no differences in levels of NTproBNP (median: 3124 pg/mL vs 2744 pg/mL, p: 0.878). Both groups presented with a similar degree of wall thickening (interventricular septum [IVS]: 17±4mm vs 17±3mm, p:0.823; posterior wall [PW]: 17±4mm vs 16±2mm, p:0.408; for black vs caucasian, respectively), but black patients presented with a smaller cavity size (left ventricle diastolic diameter [LVEDD]: 44±7mm vs 47±5mm, p:0.021) as a result of a more extensive concentric remodelling (relative wall thickness [RWT]: 0.79±0.23 vs 0.67±0.13, p:0.033). There were no differences in measures of systolic function (ejection fraction [EF]:36±16% vs 42±15%, p:0.150; myocardial contraction fraction [MCF]: 14±7 vs 13±7, p:0.515; for black vs caucasian respectively). Atrial fibrillation was prevalent in both groups (black: 27%, caucasian: 37%; p: 0.385), as was low voltage on ECG (black: 48%, caucasian: 37%; p:0.373). A higher prevalence of pseudo-infarct pattern was noted among black patients (51% vs 22%, p: 0.031). Carpal tunnel syndrome, a marker of tenosynovial amyloid infiltration, was almost two-fold more prevalent in black patients (64% vs 31%; p:0.018).

Conclusion: Irrespective of their ethnic background, patients with V122I present with a relatively uniform and highly symptomatic phenotype of infiltrative cardiomyopathy. The younger age of black patients at presentation, together with a higher degree of left ventricular remodelling and a higher prevalence of extracardiac markers of amyloid infiltration, may suggest a more aggressive disease course for black patients, resulting in earlier symptom onset. Further studies are warranted to address the role of ethnic background in influencing the phenotypic expression of TTR gene mutations.
THE EFFECTS OF RACE IN VAL122ILE AND WILD-TYPE PATIENTS IN THE TRANSTHYRETIN AMYLOIDOSIS OUTCOMES SURVEY


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8 Cleveland Clinic Foundation, Cleveland Ohio, USA.
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14 Pfizer Inc, Collegeville, PA, USA.

Introduction: Transthyretin (TTR) amyloidosis (ATTR amyloidosis) is caused by a destabilizing mutation in TTR (ATTRv) or age-related TTR stability changes in the wild-type (ATTRwt) form of the disease. The Transthyretin Amyloidosis Outcomes Survey (THAOS) is an ongoing, global, longitudinal, observational survey of patients with ATTR amyloidosis, including both inherited and wild-type disease, and asymptomatic patients with TTR mutations. ATTR cardiomyopathy (ATTR-CM) is widely associated with the Val122Ile mutation and more prevalent in those of African Descent than other races. The exact prevalence of ATTRwt is unknown, but this is the most common cause of ATTR-CM. Whether race impacts disease presentation in ATTR-CM is unknown.

Objective: To examine differences in clinical presentation as a function of self-reported race amongst Val122Ile vs ATTRwt patients in THAOS.

Methods: Descriptive analysis of demographic and clinical characteristics at enrollment as a function of the most common self-reported race with all other races of all Val122Ile and ATTRwt patients from THAOS (data cut-off: April 1, 2019). African-American and other African descent categories were combined for analysis presuming common African ancestry. Val122Ile were stratified as African vs Non-African, with the latter including Caucasian patients. Wild-type were stratified as Caucasian vs Non-Caucasian with the latter including African descent.

Results: There were 263 Val122Ile (197 male) and 874 ATTRwt (827 male) patients at data cut-off. Self-reported race was predominantly African descent (201, 76.4%) for Val122Ile vs Caucasian (752, 86.0%) for ATTRwt. Median (10th – 90th percentile) age at disease onset (65.0 [45.7-76.1] vs 63.1 [43.3-77.9] years) and age at diagnosis (70.8 [57.8-80.1] vs 70.5 [51.1-82.1] years) was similar for African vs non-African descent Val122Ile patients, respectively. Median (10th – 90th percentile) age at disease onset (69.6 [54.1-80.7] vs 70.2 [54.7-82.4] years) and age at diagnosis (75.5 [66.2-83.9] vs 76.5 [67.3-83.8] years) was also similar for Caucasian vs non-Caucasian ATTRwt patients, respectively. African vs non-African descent Val122Ile patients had: a higher incidence of abnormal ECG (88.4% vs 83.3%) and heart failure (79.1% vs 55.3%); similar median interventricular septal thickness (17.6 mm in both groups); and lower left ventricular (LV) ejection fraction (41% vs 46%). Caucasian vs non-Caucasian ATTRwt patients had: a similar incidence of abnormal ECG (97.0% vs 100.0%); higher incidence of heart failure (84.6% vs 75.0%); thinner median interventricular septum (17.0 mm vs 18.0 mm) and similar LV ejection fraction (50% each).

Conclusions: Self-reported race did not impact age at disease onset or diagnosis in Val122Ile or ATTRwt patients. By some measures, African descent Val122Ile patients and Caucasian ATTRwt patients appeared to have slightly more advanced cardiac disease than other races although overall phenotypes were similar.

Keywords: ATTR; variant ATTR; Val122Ile
EFFECT OF GENDER ON THE RISK OF CARDIAC INVOLVEMENT IN PATIENTS FROM THE TRANSTHYRETIN AMYLOIDOSIS OUTCOMES SURVEY

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7 Department of Neurology, Alexandrovskia University Hospital, Sofia Medical University, Sofia, Bulgaria.
8 Amyloidosis Network, Department of Cardiology, all at CHU Henri Mondor, INSERM U955 and Clinical Investigation Center 006, and DHU A-TVB all at Creteil, Creteil, France.
9 Pfizer Inc, New York, NY, USA.
10 Pfizer Inc, Collegeville, PA, USA.

Introduction: The Transthyretin Amyloidosis Outcomes Survey (THAOS) is an ongoing, global, longitudinal, observational survey of patients with inherited (ATTRv) and wild-type (ATTRwt) transthyretin amyloidosis (ATTR amyloidosis) as well as asymptomatic carriers with TTR mutations. There are currently few studies and limited information available on gender differences in myocardial involvement in ATTR amyloidosis.

Objective: To assess the impact of gender on cardiac phenotype and clinical manifestations in ATTRv patients with ATTR cardiomyopathy (ATTR-CM) enrolled in THAOS, exploring if female gender confers a protective effect on myocardial involvement in ATTR amyloidosis.

Methods: Data from THAOS (data cut-off: April 1, 2019) were analyzed to determine any gender-based differences in ATTRv patients diagnosed with ATTR-CM in terms of genotype, phenotype, and presence of cardiac and neurologic symptoms. Univariate and multivariate (including the parameters of gender, age at onset and genotype) regression analyses assessed the impact of male gender on the presence of cardiac phenotype and measures of myocardial involvement in ATTRv patients diagnosed with ATTR-CM.

Results: According to main genotype categories, male prevalence progressively increased among ATTRv patients diagnosed with ATTR-CM from early onset Val30Met (51.1%; 95% CI 47.9, 54.2) to non-Val30Met non-cardiac (59.2%; 54.7, 63.6%) to late onset Val30Met (63.8%; 59.0, 68.5%) to non-Val30Met cardiac (72.4%; 67.1, 77.2%). In contrast, this pattern was not observed in patients not diagnosed with ATTR-CM. Multivariate analysis demonstrated that male gender was a significant, independent risk factor for development of ATTR-CM (estimate [95% CI], 2.0947 [1.5996, 2.5898]; P<0.001) and increased LV septum thickness/height (estimate [95% CI], 0.5198 (0.2421, 0.7975); P<0.001). However, males and females with ATTR-CM (n=596) had a similar clinical/echocardiographic phenotype based on NYHA class, E wave deceleration time and E wave/A wave ratio, which were similar in each gender.

Conclusions: In ATTRv patients diagnosed with ATTR-CM participating in THAOS, gender has a significant impact on clinical phenotype. In particular, myocardial involvement was more frequent in ATTRv males, suggesting both genetic and non-genetic causes. These data suggest that currently unidentified biological characteristics either prevent, delay, or down-regulate myocardial amyloid infiltration in females, or facilitate it in males.

Keywords: ATTR; variant ATTR; gender
A DESCRIPTIVE ANALYSIS OF ATTR AMYLOIDOSIS IN SPAIN FROM THE TRANSTHYRETIN AMYLOIDOSIS OUTCOMES SURVEY

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Introduction: In Spain, there are two large endemic foci of transthyretin amyloidosis (ATTR) amyloidosis and additional cases occur across the country; however, there is no centralized patient registry. The Transthyretin Amyloidosis Outcomes Survey (THAOS) is an ongoing, global, longitudinal, observational registry of patients with ATTR amyloidosis, including both inherited (ATTRv) and wild-type (ATTRwt) disease, and asymptomatic patients with TTR mutations. The largest database of patients with ATTR amyloidosis in Spain comes from THAOS.

Objective: To gain a deeper understanding of the clinical profile of patients with ATTR amyloidosis in Spain enrolled in THAOS.

Methods: A descriptive analysis of the demographic and clinical characteristics of symptomatic patients enrolled in all six Spanish sites participating in THAOS was conducted (data cut-off: April 1, 2019).

Results: At data cut-off, 333 patients were enrolled (58.9% male). Most patients had a Val30Met mutation (combined total with or without Gly6Ser polymorphism; 70.6%), followed by ATTRwt (13.2%). Of the 213 symptomatic patients (61.0% male), the genotype distribution was Val30Met (63.8%), ATTRwt (19.7%) and other (16.4%); median (10th-90th percentile) age at symptom onset was 50.5 (25.6-70.8) years. Most symptomatic Val30Met patients had a neurologic phenotype (69.1%), with others cardiac (8.8%) or mixed cardiac and neurologic (22.1%). Notably, most symptomatic ATTRwt patients had a mixed phenotype (37.7%), followed by cardiac (37.5%) and neurologic (3.6%). In symptomatic patients, signs and symptoms of cardiac disorder were present in 100% with a cardiac, 11.6% with a neurologic, and 81.8% of a mixed phenotype, respectively. Autonomic neuropathy was present in 70.8% of cardiac, 75.9% of neurologic and 76.6% of mixed phenotype symptomatic patients, respectively. Sensory neuropathy was present in 54.2% of cardiac, 78.6% of neurologic and 61.0% of mixed phenotype symptomatic patients, respectively. The mean ([SD]) derived Neuropathy Impairment Score of the Lower Limbs score was 6.0 (11.5) in those with a cardiac, 11.0 (20.4) in those with a neurologic and 8.2 (13.6) in those with a mixed phenotype. In symptomatic patients, the mean (SD) left ventricle (LV) septum thickness was 16.8 (3.1) in those with a cardiac, 13.1 (4.2) in those with a neurologic and 15.5 (4.9) in those with a mixed phenotype, respectively; mean LV ejection fraction was 58% (9) in cardiac, 61% (7) in neurologic, and 57% (11) in the mixed phenotype, respectively.

Conclusions: In Spain, 31% of symptomatic Val30Met patients experience cardiac symptoms either alone or with neurological symptoms, while the majority of ATTRwt patients experience neurologic symptoms together with cardiac symptoms. These results highlight the phenotypic heterogeneity associated with ATTR amyloidosis in Spain and the importance of comprehensive neurologic and cardiac evaluations in all patients with ATTR amyloidosis.

Keywords: ATTR; variant ATTR; wild type ATTR
CHARACTERISTICS OF PATIENTS WITH AUTONOMIC DYSFUNCTION IN THE TRANSTHYRETIN AMYLOIDOSIS OUTCOMES SURVEY


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Introduction:
Transthyretin amyloidosis (ATTR amyloidosis) is a phenotypically heterogeneous disease. Autonomic dysfunction often presents at onset or early-stage disease, and significantly impacts disease burden. The Transthyretin Amyloidosis Outcome Survey (THAOS) is an ongoing, global, longitudinal survey of patients with ATTR amyloidosis, including inherited (ATTRv) and wild-type (ATTRwt) disease, and asymptomatic patients with TTR mutations.

Objective:
To characterize autonomic dysfunction in ATTR amyloidosis patients in THAOS.

Materials & Methods:
Descriptive analysis of demographic and clinical characteristics at enrollment of patients in THAOS with vs without autonomic dysfunction regardless of treatment received (data cut-off: April 1, 2019). Autonomic dysfunction was defined as the presence of orthostatic hypotension, diarrhea/constipation, dry eye, dyshydrosis, early satiety, erectile dysfunction, fecal/urinary incontinence, nausea, urinary retention or vomiting. Except where noted, results are shown as mean [SD].

Results:
Autonomic dysfunction was present in 1811/3943 (45.9%) symptomatic patients, and was more common in ATTRv (1676/2978, 56.3%) than ATTRwt (135/815, 16.6%). Median age at enrollment was similar for ATTRv with vs without autonomic dysfunction (49.4 vs 50.2 yrs), but dissimilar for ATTR symptom onset (40.5 vs 45.9 yrs). A similar pattern was seen in ATTRwt with vs without autonomic dysfunction: median age at enrollment 75.9 vs 76.6 and 66.6 vs 70.5 at symptom onset. The most common autonomic dysfunction symptoms in ATTRv were erectile dysfunction (66.3%), early satiety (34.8%) and diarrhea/constipation (30.7%). In ATTRwt they were erectile dysfunction (44.9%), orthostatic hypotension (35.1%), and early satiety (17.0%). ATTRv with vs without autonomic dysfunction had poorer quality of life (QoL) (Norfolk TQoL score, 38.0 [31.8] vs 14.5 [20.6]), whereas in ATTRwt QoL was similar with and without autonomic dysfunction (19.5 [17.9] vs 22.3 [19.8]). Years from ATTR amyloidosis symptom onset to first symptom of autonomic dysfunction were shorter in ATTRv (3.4 [6.0]) than ATTRwt (7.1 [9.6]). Time from autonomic dysfunction onset to diagnosis of ATTR amyloidosis was shorter in ATTRv (2.7 [3.6]) than ATTRwt (5.4 [6.1]). Autonomic dysfunction was more common in Val30Met (1230/1965 [62.6%]) than in non-Val30Met/non-cardiac (312/637 [49.0%]) or cardiac mutation (Vall122Ile, Leu 111Met, Thr60Ala or Ile68Leu) (134/376 [35.6%]) patients.

Conclusions:
Autonomic dysfunction is more common in ATTRv than ATTRwt patients in THAOS, with the most common symptoms being erectile dysfunction, early satiety and diarrhea/constipation. Poorer QoL was observed in ATTRv patients with autonomic dysfunction vs ATTRwt patients with autonomic dysfunction and vs ATTRv and ATTRwt patients without autonomic dysfunction. A greater understanding of characteristics of autonomic dysfunction will aid physicians diagnosing and treating patients with ATTR amyloidosis.
UNCOVERING CLINICAL SIGNS OF AMYLOIDOSIS IN UNDIAGNOSED TTR CARRIERS: FROM BIOBANKS TO BEDSIDE

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Background: Heritable TTR amyloidosis (hATTR) is caused by pathogenic variants in the transthyretin (TTR) gene, the most common of which in the United States (V142I) confers a 60% risk of heart failure. Systemic amyloid fibril deposition causes the “red flag” conditions carpal tunnel syndrome (CTS), spinal stenosis (SS) and polyneuropathy (FAP), preceding the diagnosis of hATTR by up to 7 years. Little is known, however, about the natural history of cardiac amyloidosis (CA). While echocardiography with global longitudinal strain (GLS) is highly sensitive and specific for fully manifest CA, it is unknown whether GLS can detect subclinical myocardial amyloid deposition. Since new therapies for hATTR have emerged, early recognition of disease in individuals harboring pathogenic TTR variants (TTR+) is paramount.

Objective: We retrospectively assessed systemic and cardiac features of TTR+ individuals from a large, ethnically diverse and unselected population-based Biobank in New York City to identify early features that might precede the diagnosis of hATTR.

Methods: Among 30,223 Biobank participants with available sequencing data linked to the electronic health record (EHR), we identified TTR+ subjects. Of those, we focused on 44 adults (V142I, N=43; V64L, N=1; ages 23-90, median 63; 75% female) with available echocardiograms and without known diagnoses of amyloidosis or cardiomyopathy by querying: 1) EHR for systemic diagnoses (CTS, SS and FAP) and 2) electrocardiographic (EKG) and echocardiographic data. Speckle Tracking Echocardiography (STE, Philips aCMQ) was performed by post-processing stored echocardiographic images to calculate GLS and segmental strain.

Results: Of 44 TTR+ subjects without a diagnosis of hATTR, 15 (31.8%) had ≥1 associated systemic diagnosis and 9 (20.5%) had low voltage on EKG. Left ventricular (LV) hypertrophy was present in 16 (36.4%) and maximal LV wall thickness was similar in females vs. males (0.7-1.4cm vs. 1.1-1.6cm, p=0.09). Overall, the majority of TTR+ subjects (52%, 55%, 57% and 83% in those aged <60, 60-69, 70-79 and 80+ respectively) had sub-normal GLS. Interestingly, GLS was lower in males vs. females (mean 15% vs. 18.6%, p=0.01). Moreover, average apical strain was higher than basal in 37 (90.2%) subjects, consistent with the known phenomenon of apical sparing in CA.

Conclusions: We investigated systemic and cardiac phenotypes in TTR+ adults across all ages in an unselected population-based biobank. “Red flag” hATTR diagnoses or LV hypertrophy are present in roughly 1/3 of cases. Derangements in myocardial deformation are prevalent, even in the 5-6th decades, when CA is rarely considered clinically relevant. Furthermore, the “cherry on top” pattern of apical strain sparing is present in >90% of TTR+ subjects. Additional studies are needed to further characterize subclinical features of hATTR in patients harboring pathogenic TTR variants and to determine who may benefit from early treatment with new disease-modifying therapies.
RARE TRANSTHYRETIN GENE MUTATIONS: PHE33LEU, GLU89LYS AND ALA81VAL IN PATIENTS WITH CARDIAC TRANSTHYRETIN AMYLOIDOSIS—SINGLE CENTRE EXPERIENCE

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2 Department of Medical Biology, Institute of Cardiology, Warsaw, Poland.
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4 Department of Neurology, Medical University of Warsaw, Poland.
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6 Unit for Screening Studies in Inherited Cardiovascular Diseases, The Cardinal Stefan Wyszynski Institute of Cardiology, Warsaw, Poland.

Background: More than 140 different mutations of transthyretin (TTR) gene is related to hereditary form of transthyretin amyloidosis (hATTR). The prevalence of mutations varies according to ethnicity and geographic region.

Objectives: We describe patients with cardiac hATTR diagnosed in 2017-2019 in a single cardiology centre.

Methods: Consecutive patients with suspected cardiac amyloidosis were evaluated, including NT-proBNP, high-sensitive cardiac troponin T (hs-cTnT), free light chain testing, standard 12-lead electrocardiography (ECG) and transthoracic echocardiography. ATTR was confirmed histologically or non-invasively using 99mTc-DPD scintigraphy. TTR gene sequencing was performed.

Results: Of the 58 patients, 6 (10.3%) unrelated male patients had hATTR (Table 1). All patients had very rare TTR mutations: Phe33Leu, Glu89Lys and Ala81Val mutations. The most prevalent was Phe33Leu mutation identified in 4 patients. The age of onset ranged from 48 to 67 years. On ECG, most patients had pseudoinfarct pattern and/or low QRS voltage. Echocardiography showed concentrically increased thickness of left ventricular (LV) wall and reduced LV ejection fraction (LVEF). The maximal wall thickness (MWT) varied from moderate (16 mm) to massively increased (30 mm). Elevated levels of hs-cTnT and NT-proBNP were present in all patients, even with initially mild heart failure (HF) symptoms. Four patients had polyneuropathy and four had carpal tunnel syndrome. On follow-up, we observed progressive HF in all cases. First patient with Phe33Leu mutation died of HF, second died suddenly, third deteriorated to NYHA IV and underwent successful combined heart and liver transplantation. The patient with Ala81Val mutation died of stroke.

Conclusions: We report unexpectedly rare TTR mutations in patients from a single cardiology centre. Phe33Leu mutation was previously described only in 7 unrelated families, including 2 with origin from our country. The high incidence of Phe33Leu mutation reported in our study (4 unrelated families) and previous literature data imply that this TTR mutation might be endemic in our population.

Table 1. Patients characteristics and outcome.

<table>
<thead>
<tr>
<th>Patient</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
</tr>
</thead>
<tbody>
<tr>
<td>TTR mutation</td>
<td>Phe33Leu</td>
<td>Phe33Leu</td>
<td>Phe33Leu</td>
<td>Phe33Leu</td>
<td>Glu89Lys</td>
<td>Ala81Val</td>
</tr>
<tr>
<td>Age of onset</td>
<td>64</td>
<td>57</td>
<td>48</td>
<td>58</td>
<td>57</td>
<td>67</td>
</tr>
<tr>
<td>Age of diagnosis</td>
<td>67</td>
<td>57</td>
<td>48</td>
<td>58</td>
<td>57</td>
<td>70</td>
</tr>
<tr>
<td>NYHA</td>
<td>III</td>
<td>I</td>
<td>I</td>
<td>II</td>
<td>III</td>
<td>III</td>
</tr>
<tr>
<td>ECG</td>
<td>ventricular pacing, AF</td>
<td>SR, nsVT</td>
<td>SR</td>
<td>SR</td>
<td>AF</td>
<td>AF</td>
</tr>
<tr>
<td>low QRS voltage</td>
<td>n.a.</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Pseudoinfarct pattern</td>
<td>n.a.</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>NT-proBNP, pg/ml</td>
<td>10954</td>
<td>1199</td>
<td>925</td>
<td>1049</td>
<td>2340</td>
<td>6070</td>
</tr>
<tr>
<td>hs-cTnT, ng/l</td>
<td>103</td>
<td>98</td>
<td>28</td>
<td>51</td>
<td>50</td>
<td>78</td>
</tr>
<tr>
<td>MWT, mm</td>
<td>30</td>
<td>20</td>
<td>16</td>
<td>18</td>
<td>23</td>
<td>27</td>
</tr>
<tr>
<td>LVEF, %</td>
<td>30</td>
<td>40</td>
<td>40</td>
<td>55</td>
<td>45</td>
<td>35</td>
</tr>
<tr>
<td>Restrictive LV filling pattern</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Pericardial effusion</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Polyneuropathy</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Carpal tunnel syndrome</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Outcome</td>
<td>Death of HF at age 67</td>
<td>SCD at age 59</td>
<td>Heart and liver transplant</td>
<td>Alive</td>
<td>Awaiting heart and liver transplant</td>
<td>Death of stroke at age 71</td>
</tr>
</tbody>
</table>

AF—atrial fibrillation; SR—sinus rhythm; nsVT—nonsustained ventricular tachycardia; n.a.—not applicable
PM092

PEAK DELAY PHENOMENON OF GASTROCOLIC REFLEX AND SERUM GASTRIN LEVELS: USEFUL PRE-SYMPTOMATIC MARKERS OF GASTROINTESTINAL SYMPTOMS IN HEREDITARY TRANSTHYRETIN AMYLOIDOSIS

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Background: Gastrointestinal (GI) symptoms are common in hereditary transthyretin (ATTRv) amyloidosis. We previously reported that our cutaneous electrointestinogram (EIG) recording method could detect the peak delay phenomenon of gastrocolic reflex in these patients. However, we have not proved the usefulness of the phenomenon as a pre-symptomatic marker of gastrointestinal symptoms from the limitation of the number of examined cases.

Objective: To assess the usefulness of peak delay phenomenon of gastrocolic reflex as a pre-symptomatic marker of GI symptoms in ATTRv amyloidosis.

Patients and Methods: Eight Japanese ATTRm patients and 27 healthy controls (19 women and 8 men, mean age 22 ± 1.89 years) were available for this study. We evaluated the patterns of gastrocolic reflex after meal (high-caloric (600 kcal) oral liquid nutritional supplement) start using a portable EIG recorder (MP150 + EGG100C system, BIOPAC Co., U.K.) in all subjects. Moreover, we also measured the serum gastrin levels after meal start in all subjects because gastrin was an important mediator of gastrocolic reflex.

Results: Not only 5 patients with GI symptoms but also 3 patients without GI symptoms showed remarkable peak delay of gastrocolic reflex (35-60 minutes after meal start) compared with 27 healthy controls (15-20 minutes after meal start). Moreover, the serum gastrin levels of these 8 patients are significantly higher (>620 pg/ml) than those of 27 healthy controls (<200 pg/ml).

Conclusion: Peak delay phenomenon of gastrocolic reflex and elevation of serum gastrin levels that seems to be compensatory may be useful pre-symptomatic markers of GI symptoms in ATTRv amyloidosis.
NEUROLOGICAL MANIFESTATIONS IN PATIENTS WITH HEREDITARY TRANSTHYRETIN AMYLOIDOSIS: A MAJOR MULTIDISCIPLINARY CENTER EXPERIENCE

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Background: Hereditary Transthyretin Amyloidosis (hATTR) is a clinically heterogeneous disease with various neurological, cardiac and other manifestations. Improved characterization of neurological involvement is critical for earlier diagnosis and prognostication in an era of commercially available therapies.

Objectives: The goals of this study are to characterize the prevalence of neurological manifestations of hATTR stratified by major mutation types and characterize delays in diagnosis.

Methods: A retrospective chart review was performed of all patients with hATTR evaluated at a tertiary Amyloidosis Center between January 1, 2016 and December 31, 2018. One-hundred-eighteen independent variables were coded when available including (1) demographics information, (2) symptoms and signs of neurological disorders including carpal tunnel syndrome (CTS), large fiber polyneuropathy (PN) and small fiber polyneuropathy (SFN), dysautonomia, myopathy, cervical and lumbar radiculopathy along with year of onset, (3) results of diagnostic testing including neurophysiological studies and biopsy results, (4) initiation of therapies and (5) recommendations regarding neurophysiological studies at the initial visit.

Results: Among 92 patients with hATTR amyloidosis evaluated, the most common mutations were V122I (36), T60A (20) and V30M (12), followed by L58H (5), F64L (3), Y114C (3), S77Y (2), F46L, Y144C, E89Q, E42G, T69H, A97S, I107V, H88R, V30R, I107M, K35N and I73V. Seventy-two percent had symptoms of CTS with an average 6.7-year-delay between symptom onset and definitive diagnosis of amyloidosis. Thirty-one patients had carpal tunnel release, five had tenosynovial staining with two revealing congophilia. There was a trend towards greater prevalence of CTS in patients with V30M (83%) than T60A (75%) and V122I (64%) without meeting statistical significance (p = 0.11). Only 66% and 36% of patients had symptoms of PN and SFN, respectively, while 75% and 65% had findings of PN and SFN on examination, respectively. Prevalence of PN was greater in patients with V30M (92%) than T60A (80%) and V122I (53%) upon multivariate analysis with age and diabetes as confounders (p = 0.048). Forty-seven percent had dysautonomia, 18% lumbar radiculopathy and 11% cervical radiculopathy. Electrophysiological studies were recommended at the initial examination in the majority of patients with symptoms of hand numbness (95%) and toe numbness (86%).

Conclusions: This study identified (1) prevalence of CTS and PN as well as spinal radiculopathy; (2) different profiles of neurological manifestations in the most common hATTR mutations; (3) new reporting of the delay between onset of CTS and diagnosis of amyloidosis. Early electrophysiological studies in appropriate cases prior to initial visit at an amyloidosis center would expedite disease characterization and treatment initiation.

Keywords: hereditary transthyretin amyloidosis, carpal tunnel syndrome, peripheral neuropathy
**PM094**

**CLINICAL PROGRESSION OF LIVER TRANSPLANTED PATIENTS WITH V30M TRANSTHYRETIN-RELATED HEREDITARY AMYLOIDOSIS (ATTRV30M).**

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**Introduction:** Precursor of amyloid deposits in ATTRV30M disease, mutant TTR, is mainly produced in the liver. Consequently, liver transplantation (LT), although being an invasive technique, became the first therapeutic approach for these patients. LT increased survival of ATTRV30M patients and changed the course of this disease unraveling symptoms and affection of different organs as TTR is not only produced in the liver but also in choroid plexus and retinal pigment epithelium.

**Objectives:** To analyze the evolution and complications of the patients in follow-up in a tertiary hospital in Mallorca with ATTRV30M that underwent LT between 2001 and 2019.

**Methods:** We performed a retrospective, descriptive study of patients with ATTRV30M and LT in the last 20 years. We collected information regarding transplant complications, morbidity and data about the progression of the disease. The PND status was registered before the transplant and today (for the patients still alive). Results are expressed in means and standard deviation (continuous) or in percentages (categorical).

**Results:** 67 ATTRV30M patients underwent a LT in the last two decades in our Hospital, 35 men (53 %) and 32 women (47 %). Mean (SD) age at transplant was 42 (13) years. Mean (SD) time between diagnosis and liver transplant was 15 (10) months. In the first year after LT, the most frequent complications were rejection (2 patients), hepatobiliary infections (3 patients), non-hepatobiliary infections (2 patients) and complications of the anastomoses (2 patients). After the first year, the major complication was non-hepatobiliary infections (6 patients), followed by rejection (5 patients) and hepatobiliary infections and brain stroke (3 patients each).

The progression of the disease after LT is shown in the table. As can be seen, ocular manifestations are the last ones in appear, while the polyneuropathic and gastrointestinal manifestations are the first ones. The PND status hasn’t change before the transplant and to date on 53 % of the patients; worsened on 27 % and improved on 20 % of the patients.

**Conclusion:** Liver transplant on ATTRV30M patients is associated with a high morbidity and, in many patients, with progression of the disease. However, neurological symptoms remain stable in the majority of the patients.

**Table.** Progression of the disease after the transplant

<table>
<thead>
<tr>
<th>N (%)</th>
<th>Most frequent manifestations</th>
<th>Mean time progression (years)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Polineuropathy</strong></td>
<td>41 (60 %)</td>
<td>Paresthesia (78 %)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Lost of thermalgesic sensitivity (34%)</td>
</tr>
<tr>
<td><strong>Gastrointestinal</strong></td>
<td>36 (53 %)</td>
<td>Diarrhea (78 %)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Constipation (6 %)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Both (8 %)</td>
</tr>
<tr>
<td><strong>Disautonomy</strong></td>
<td>17 (25 %)</td>
<td>Erectile dysfunction (47 %)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Orthostatic hypotension (41 %)</td>
</tr>
<tr>
<td><strong>Ocular</strong></td>
<td>31 (46 %)</td>
<td>Vitreous opacity (65 %)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Scalloped iris (29 %)</td>
</tr>
<tr>
<td><strong>Renal</strong></td>
<td>19 (28 %)</td>
<td>UTI (63 %)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>CKD (37 %)</td>
</tr>
<tr>
<td><strong>NCS</strong></td>
<td>14 (21 %)</td>
<td>Ischemic Stroke (36 %)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Stroke (21 %)</td>
</tr>
<tr>
<td><strong>cardiovascular</strong></td>
<td>8 (12 %)</td>
<td>Atroventricular block (7 %)</td>
</tr>
</tbody>
</table>
COGNITIVE FUNCTION MEASURED BY THE MOCA SCALE IN PATIENTS WITH ATTRV30M TREATED WITH LIVER TRANSPLANTATION OR WITH TAFA MIDS: AN OBSERVATIONAL STUDY

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1 Hospital Universitario Son LLàtzer.

Introduction: 30M Hereditary transthyretin amyloidosis (ATTRV30M) is a rare, multisystem disease caused by amyloid deposition, characterized by heterogeneous presentation and symptomatology. TTR is mainly produced in the liver, however it is also produced in choroid plexus and retinal pigment epithelium. The involvement of CNS in patients with ATTRV30M was described more than 20 years ago, but the cognitive dysfunction in patients treated or untreated has been less studied.

Objective: To assess the cognitive function in patients with hereditary transthyretin amyloidosis with a previous liver transplant, or in treatment with tafamidis.

Methods: We conducted a retrospective observational study of patients with ATTRV30M that underwent a liver transplantation (LT) between 1997 and 2011, and those in treatment with tafamidis. We collected demographic data, treatment duration and years since the transplant. We performed on October 2019 the Montreal Cognitive Assessment test (MoCA), and the Lawton and Brody test. As established, we considered the presence of a mild cognitive impairment when the MoCA test was 25 or lower. Results are expressed in mean and standard deviations (SD) for continuous variables and count and percentages for categorical variables.

Results: 36 patients were included in the study, 17 that underwent a LT and 19 in current treatment with tafamidis. The main characteristic of the patients in both groups can be seen in the table. The percentage of women and the level of education were similar in both groups. However, patients in the tafamidis group were older than those in the transplant group, and we believe this is the reason why the MoCA test yielded slightly lower results in the tafamidis group compared with the transplant group. Overall, cognitive impairment was not frequent in our sample considering the age of the participants, and all of them were independent in their daily activities (only 3 participants had a score of 7 out of 8 in the Lawton and Brody test).

Conclusions: Cognitive dysfunction may be a late manifestation of the disease. We have observed a similar cognitive dysfunction in our patients than those reported in the literature. The percentage of patients with mild cognitive impairment was higher in the tafamidis group due to the higher age on that group. Overall, patients of this cohort are independent in their daily activities, with independence of the treatment received. Further studies with more specific and complete neuropsychological studies are necessary to ascertain the role of transplant/tafamidis in cognitive impairment of these patients.
TRANSTHYRETIN-RELATED VARIANT AMYLOIDOSIS IN AN CAUCASIAN MALE WITH TTR VAL142ILE (PREV. VAL122ILE) MUTATION.

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Background: Transthyretin-related variant amyloidosis (ATTRv) is an autosomal dominant inherited disease caused by mutations in the transthyretin gene (TTR). TTR amyloidosis is an under-diagnosed cause of heart failure in the elderly. TTR amyloidosis can result from a mutant TTR protein with Val142Ile, frequently described in Afro American population. There are few reports of ATTRv in non-Afro Americans.

Aim: We describe the clinical characteristics of a caucasian male affected by ATTRv with Val142Ile mutation.

Methods/Patient: We present a 71 year-old male war veteran, with post-traumatic anxiety syndrome and a history of 8 years of carpal tunnel syndrome, hypoacusia, omalgia and back pain. The patient is caucasian, and he has Italian ancestry. The patient had effort dyspnea with NYHA class II. In a preoperative evaluation for lumbar stenosis, infiltrative cardiomyopathy was diagnosed by an echocardiogram with an ejection fraction of 41% and reduced global longitudinal strain with typical apical sparing pattern. At physical examination, vital signs were unremarkable, neurological exam was normal and mild ankle edema on the cardiovascular assessment. Pyrophosphate scintigraphy was positive. NT-ProBNP was 1184 pg/ml and ultrasensitive troponin T was 60 pg/ml. TTR cardiomyopathy was diagnosed. TTR was sequenced and evidenced a Val142Ile mutation. He was medicated with diflunisal, doxycycline, ursodeoxycholic acid and low dose of furosemide. After one year of follow up, he developed deglutory symptoms, constipation, paresthesia, axonal neuropathy and hyperreactive bladder. There are several differences in this case with the typical clinical picture of this mutation, the age at onset, the racial origin, the slow progression of cardiopathy and the peripheral and autonomic neuropathy.

Conclusions: Mutated Val142Ile TTR amyloidosis can be presented in elderly and caucasian patients. In a patient with suspicion of senile TTR cardiac amyloidosis, a broad range of non-cardiac symptoms and disorders might require a molecular test to rule out a mutated TTR amyloidosis. There are few reports of this mutation in patients of caucasian ancestry and the clinical picture in this ethnic group is different from Afro American ancestry.

Key words: ATTRv, Val142Ile, Amyloidosis cardiomyopathy
**FLUOCINOLONE ACETONIDE (FAc, 0.2mg/DAY) IMPLANT IN THE TREATMENT OF AMYLOID RETINAL ANGIOPATHY MACULAR EDEMA IN ATTR V30M PATIENTS – 1-YEAR FOLLOW-UP**

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**Introduction/Background:** Hereditary transthyretin V30M amyloidosis (ATTRV30M) is an autosomal dominant disorder caused by extracellular deposition of variants of transthyretin (TTR) in several tissues. Liver transplantation is one of the treatments for ATTRV30M amyloidosis once it removes mutant TTR and interrupts the progression of the disease. However, the intraocular production of TTRV30M remains unchanged, even after liver transplantation, contributing to the progression of amyloid deposition-associated ocular manifestations such as glaucoma and retinal angiopathy.

**Objectives:** The goal of the present study was to determine the effectiveness and safety of fluocinolone acetonide (0.2 µg/day FAc) implant in amyloid retinal angiopathy macular edema in ATTR V30M patients.

**Methods:** Clinical data review of eyes from liver transplanted ATTR V30M patients treated with an FAc implant. The efficacy and safety of the FAc implant over a mean follow-up period of 8.5±4.1 months were evaluated. Demographic data was collected concerning ATTR V30M diagnosis, retinal angiopathy development and surgical assessments such as Ahmed valve implantation and vitrectomy pre implant, as well as recording of best corrected visual acuity (BCVA), central retinal thickness (CRT) and intraocular pressure (IOP). The standard assessment included follow-up measurements of these parameters in order to estimate their variation over time. The percentage of eyes with functional gains greater than 5, 10 and 15 ETDRS letters was estimated as well as the percentage of eyes achieving a CRT <300 µm.

**Results:** A total of 15 eyes/15 patients were included. Disease was diagnosed 20 years ago with liver transplant being performed 3.7±2.3 years after diagnosis. Thirteen eyes out of 15 had an Ahmed valve implanted prior to the development of retinal angiopathy (mean time between valve implantation and retinal angiopathy onset was 22.3±10.8 months). Vitrectomy was performed in all eyes, but 3, prior to FAc implant and the mean time between vitrectomy and FAc implant injection was 30.9±21.6 months.

The mean baseline BCVA was 44.7±14.2 ETDRS letters and had an average increase of +22.5 ETDRS letters in the last observation (p<0.01). All patients improved BCVA compared to baseline. 93.3%, 93.3% and 100% of eyes gained ≥15, 10 and 5 letters, respectively, compared to baseline. The mean CRT decreased 281.0 µm in the last observation (p=0.363). Twelve eyes (80%) achieved a reduction in CRT ≤300 µm. IOP remained stable through all the follow-up (from 12.1±2.3 mmHg in baseline to 11.5±2.9 mmHg at last observation, p<0.01)

**Conclusions:** FAc implant was very beneficial in the treatment of amyloid retinal angiopathy macular edema in ATTR V30M patients with no additional safety concerns. These satisfactory results allowed FAc implants to be used, at Centro Hospitalar do Porto, as a first-line treatment in retinal amyloidotic angiopathy macular edema with previously vitrectomized eyes and submitted to glaucoma surgery.
THE AGE OF THE FINNISH GELSO LINAMYLOIDOSIS MUTATION


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Background: Finnish gelsolin amyloidosis (AGel amyloidosis) is an autosomal dominantly inherited systemic disorder with ophthalmologic, neurologic and dermatologic symptoms. The gelsolin (GSN) variant c.640G>A, p.D187N has been only found in Finnish patients thus far. Genealogic studies have suggested that the mutation has descended from a common ancestor instead of separate mutational events. Recently we demonstrated, using SNP array-based haplotype analysis, that all 68 patients from 62 nuclear families shared a common haplotype encompassing at least 361 kb. This GSN haplotype was the longest shared haplotype block in a genome-wide analysis among the AGel amyloidosis patients, but not in controls. These results provided strong evidence that although there is a known mutational hotspot at GSN c.640G, all studied 62 families are genetically linked to a common ancestor (1).

Objectives: Here, we wanted to estimate the age of the GSN c.640G>A mutation based on the previous study (1).

Methods: We used two approaches. First, we genotyped four microsatellite markers around the mutation and analyzed the decay of linkage disequilibrium (LD) in 7 parent-offspring pairs with the mutation, 20 unrelated AGel amyloidosis patients and 60 controls. Second, we analyzed our previous high-density SNP data in 68 patients from 62 families and estimated the age of the variant based on the lengths of haplotypes shared between individuals.

Results: Preliminary analysis of the SNP data indicates that the age of the mutation is from 15 to 21 generations old (375-525 years). The results on the mutation age using the two approaches will be presented.

Conclusions: The estimated age of Finnish gelsolin amyloidosis mutation is presented in the symposium for the first time.

References:

Key words: Finnish gelsolin amyloidosis, mutational age, genealogy
BASELINE DISEASE CHARACTERISTICS OF PATIENTS WITH ATTR VAL122ILE AMYLOIDOSIS FROM THE BRAZILIAN NATIONAL AMYLOIDOSIS REFERRAL CENTER (CEPARM).

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Introduction: Transthyretin amyloidosis (ATTR) is characterized by the deposit of mutant or wild-type transthyretin that forms amyloid fibrils, which are extracellularly deposited within tissues and organs. Clinical manifestations of familial amyloid disease vary according to the mutation, age at onset and geographical location.

Objective: This study aimed to describe baseline disease characteristics of ATTR Brazilian patients with Val122Ile mutation enrolled in our center.

Methods: Patients with ATTR Val122Ile amyloidosis seen in the Brazilian National Amyloidosis Referral Center from April 2012 to September 2019 were included. Demographics (age at symptom onset, gender, time from onset of symptoms to diagnosis, family history), and clinical characteristics (presence of amyloid deposit, frequency of misdiagnosis, presenting symptomatology, ECG and echocardiogram) were extracted. Descriptive data is presented as frequencies and percentages for categorical variables and median and ranges for continuous variables.

Results: Twenty two patients with Val122Ile mutation were found (13 symptomatic and 9 asymptomatic). The asymptomatic patients were excluded from this analysis. Median age at symptom onset was 64 years (60.5 to 81.7), and 10 (77%) were male. Misdiagnosis was observed in 11 (84.6%) patients. The median time from symptoms onset to diagnosis was 1.8 years (0.3 -6). Only 1 (7.7%) patient reported a positive family history of ATTR. At presentation, 100% of patients had cardiac disease and autonomic neuropathy (69.2% had gastrointestinal manifestations), 61.2% had sensory neuropathy and only 23% had motor neuropathy. ECG was abnormal in 84.6% of the patients and 69.2% had Intraventricular septum hypertrophy (≥12 mm).

Conclusions: ATTR Val122Ile amyloidosis in Brazil is a late-onset disorder with cardiac manifestations and autonomic neuropathy. The majority of patients have gastrointestinal symptoms and sensory nerves involvement. Positive family history of ATTR is rarely encountered and most patients are diagnosed more than one year after symptom onset.

Keywords (proposed): ATTR, VA1122Ile
AMBULATORY GAIT ANALYSIS FOR SUPPORTING CLINICAL ASSESSMENT OF HEREDITARY TRANSTHYRETIN AMYLOIDOSIS

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Introduction/Background: Hereditary amyloidosis associated with transthyretin V30M (hATTR V30M) is a progressive length-dependent polyneuropathy involving walking impairment [1], in which movement is analyzed mostly by observation [2]. Recently, Vita et al. have described the 6-Minute Walk Test as a reliable tool to monitor hATTR V30M patients [3]. However, to the best of our knowledge, there is no study comparing of patients with different disease profiles based on their gait characteristics. Until a few years ago, motion capture was mostly carried out using expensive and intrusive multi-camera marked-based systems [4]. Nowadays, vision-based systems relying on low-cost, portable, markerless RGB-D cameras are being developed for inexpensive and minimally intrusive motion analysis in clinical practice [5].

Objective: The aim of this work is to explore the possibility of distinguishing between hATTR V30M patients with different clinical characteristics, based on quantified gait information obtained using an RGB-D camera-based system.

Methods: An experiment was carried out at Centro Hospitalar Universitário do Porto (Porto, Portugal) with 20 hATTR V30M patients: 5 asymptomatic carriers (AC); 5 patients taking a 20 mg daily dose of tafamidis for 1 year (1T), with normal nerve conduction studies (NCS) in baseline and at the 1-year evaluation; and two groups of 5 patients taking tafamidis for 3 or 4 years, with normal (4TN) and abnormal (4TA) NCS in baseline and at the last evaluation. The exclusion criteria were the presence of comorbidities, feet or leg injuries. The study was authorized by the local Ethics Committee and complies with the Declaration of Helsinki. All subjects signed an informed consent form.

The experimental protocol consisted of a 2-minute walking task, during which 3-D body data were acquired using our NeuroKinect RGB-D camera-based system [6, 7]. For each automatically identified gait cycle [8, 9], we computed the 20 gait parameters defined in [7] and performed the Kruskal-Wallis test [10] for each parameter. Since the p-value was always lower than or equal to 0.05, we carried out the Conover-Iman test for multiple pairwise comparison [11].

Results: Table 1 presents the obtained results, which show that AC and 4TA are the groups that differ the most, as expected, since all parameters except one may be used for distinction (p-value ≤ 0.05). Moreover, 3 parameters may be used to distinguish between all groups: stance duration, step length, and foot swing velocity. All other parameters (except TBCM sway y-component) may also be used together for group distinction.

Conclusions: Several gait parameters can potentially be used to distinguish between asymptomatic and symptomatic hATTR V30M carriers at different disease stages, which may allow an early access and/or optimization of treatment, and a consequent improvement of the patients’ quality of life. We are currently acquiring data from more patients, which will be used in the future to confirm this study’s results and develop disease management tools.

Keywords: Hereditary amyloidosis; Gait; Clinical Assessment
Table 1. Results of the Conover-Iman Test (p-value) for each gait parameter, when comparing the different combinations of the following groups: asymptomatic carriers (AC), symptomatic patients with 1 year tafamidis (1T), with 3 or 4 years on tafamidis with normal nerve conduction studies (4TN) and, with 3 or 4 years on tafamidis with abnormal nerve conduction studies (4TA). N.S. stands for “Non-significant” (p-value > 0.05).

<table>
<thead>
<tr>
<th>Gait Parameter</th>
<th>AC - 1T</th>
<th>AC - 4TN</th>
<th>AC - 4TA</th>
<th>1T - 4TN</th>
<th>1T - 4TA</th>
<th>4TN - 4TA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stride duration (s)</td>
<td>≤ 0.001</td>
<td>≤ 0.001</td>
<td>≤ 0.001</td>
<td>≤ 0.001</td>
<td>N.S.</td>
<td>N.S.</td>
</tr>
<tr>
<td>Step duration (s)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Stance duration (s)</td>
<td>N.S.</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Single support duration (s)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Double support duration (s)</td>
<td>N.S.</td>
<td>0.007</td>
<td>≤ 0.001</td>
<td>0.023</td>
<td>≤ 0.001</td>
<td></td>
</tr>
<tr>
<td>Stride length (cm)</td>
<td>≤ 0.001</td>
<td>0.002</td>
<td>≤ 0.001</td>
<td>0.046</td>
<td>≤ 0.001</td>
<td></td>
</tr>
<tr>
<td>Step length (cm)</td>
<td>≤ 0.001</td>
<td>0.007</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Step width (cm)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TBCM(^a) sway x-component (mm(^2))</td>
<td>0.024</td>
<td>N.S.</td>
<td>0.011</td>
<td>N.S.</td>
<td>N.S.</td>
<td>N.S.</td>
</tr>
<tr>
<td>TBCM(^a) sway y-component (mm(^2))</td>
<td>N.S.</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gait speed (m/s)</td>
<td>≤ 0.001</td>
<td>≤ 0.001</td>
<td>≤ 0.001</td>
<td>≤ 0.001</td>
<td>0.024</td>
<td>N.S.</td>
</tr>
<tr>
<td>Foot swing velocity (m/s)</td>
<td>N.S.</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Arm swing velocity (m/s)</td>
<td>N.S.</td>
<td>0.012</td>
<td>0.005</td>
<td>≤ 0.001</td>
<td>N.S.</td>
<td></td>
</tr>
<tr>
<td>Spine shoulder angle (deg.)</td>
<td>N.S.</td>
<td></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Maximum elbow angle (deg.)</td>
<td>≤ 0.001</td>
<td>0.012</td>
<td>≤ 0.001</td>
<td>0.004</td>
<td>≤ 0.001</td>
<td>N.S.</td>
</tr>
<tr>
<td>Minimum elbow angle (deg.)</td>
<td>N.S.</td>
<td></td>
<td>N.S.</td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Maximum knee angle (deg.)</td>
<td>≤ 0.001</td>
<td>≤ 0.001</td>
<td>≤ 0.001</td>
<td></td>
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</tr>
<tr>
<td>Minimum knee angle (deg.)</td>
<td>N.S.</td>
<td>0.01</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Hip angle range (deg.)</td>
<td>≤ 0.001</td>
<td>0.006</td>
<td></td>
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</tbody>
</table>

\(^a\)TBCM stands for Total Body Center of Mass

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11. Dinno, A., Conover-Iman Test of Multiple Comparisons Using Rank Sums, in Package 'conover.test' version 1.1.5. 2017, CRAN.
PM101

MANAGEMENT OF THROMBOCYTOPENIA IN PATIENTS WITH HEREDITARY TRANSTHYRETIN AMYLOIDOSIS TREATED WITH INOTERSEN


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Introduction: Hereditary transthyretin amyloidosis (hATTR) is a rare, progressive, and fatal disease that causes debilitating autonomic and sensorimotor neuropathy, often with overlapping cardiomyopathy. Inotersen is an antisense oligonucleotide that specifically binds and degrades TTR messenger RNA, reducing production of the amyloid-forming transthyretin protein. In NEURO-TTR (NCT01737398), a randomized, placebo-controlled phase 3 study of patients with hATTR polyneuropathy, grade 4 thrombocytopenia (platelet count <25 × 10^3/µL) by Common Terminology Criteria for Adverse Events occurred in 3 (3%) patients receiving inotersen prompting implementation of enhanced (weekly) platelet monitoring.

Objective: To analyze and describe the effect on platelets in patients treated with inotersen for the polyneuropathy of hATTR in the open-label extension (OLE; NCT02175004) of NEURO-TTR.

Methods: In NEURO-TTR, patients were randomly assigned 2:1 to receive inotersen or placebo once weekly via subcutaneous injection. Patients who completed NEURO-TTR were eligible to enter the ongoing OLE study. Weekly platelet monitoring began during NEURO-TTR (May 2016) and is continuing throughout the OLE. Platelet count decreases are confirmed by repeat testing within 7 days, if available. Data are presented for all patients who received at least 1 dose of inotersen in NEURO-TTR or the OLE.

Results: Of 139 patients who completed NEURO-TTR, 135 (97.1%) enrolled in the OLE, with 85 patients continuing to receive inotersen and 50 patients switching from placebo to inotersen. As of May 31, 2018, the longest cumulative inotersen exposure was 5.2 years, with a mean (standard deviation) cumulative exposure of 2.3 (1.4) years.

Confirmed platelet counts reached <100 × 10^3/µL at some time during the OLE in 48/135 (35.6%) patients. Maximum confirmed platelet count decrease reached grade 1b (≥75 to <100 × 10^3/µL) in 33/135 (24.4%) patients, grade 2 (≥50 to <75 × 10^3/µL) in 13/135 (9.6%) patients, and grade 3 (≥25 to <50 × 10^3/µL) in 2/135 (1.5%) patients during the OLE study. Both (2/2) patients with platelet count <50 × 10^3/µL were treated with corticosteroids. No patients experienced confirmed grade 4 thrombocytopenia.

Inotersen treatment was paused (≥1 missed dose) during the OLE in 26/135 (19.3%) patients due to prespecified holding criteria (platelet count <75 × 10^3/µL) for median 70.5 days (range, 16–169 days). A majority of patients (19/26; 73.1%) resumed inotersen treatment after dose pause.

Conclusions: No case of grade 4 thrombocytopenia has been observed with inotersen treatment for the polyneuropathy of hATTR in the analysis period covering approximately 2 years following implementation of weekly platelet monitoring in May 2016. Platelet count decreases, if they occur with inotersen treatment, can be identified with routine hematologic testing and should be managed with the recommended dose pause and/or corticosteroid treatment, as indicated in the relevant regulatory guidelines.
Introduction: Hereditary transthyretin amyloidosis (hATTR) is a rare, progressive, and fatal disease that causes debilitating autonomic and sensorimotor neuropathy. Efficacy and safety of inotersen, an antisense oligonucleotide inhibitor of TTR protein production, were demonstrated in patients with hATTR polyneuropathy in the phase 3 NEURO-TTR (NCT01737398) study and its open-label extension (OLE; NCT02175004). Following the occurrence of a few serious adverse events each of thrombocytopenia and glomerulonephritis, enhanced safety monitoring was implemented with the goal of preventing occurrence of these events.

Objective: To review efficacy and assess enhanced monitoring for thrombocytopenia and glomerulonephritis in patients treated with inotersen in clinical and real-world datasets.

Methods: Patients who completed NEURO-TTR were eligible to enter an OLE. Patients with hATTR have also received inotersen through a United States expanded access program (EAP) and a French Compassionate Use Program (ATU). An ongoing, investigator-sponsored trial (IST) includes patients with the cardiomyopathy of hATTR or wild-type ATTR. Data from these 5 studies plus ~3 patient-years of post-marketing exposure were evaluated from 7/6/2018 to 1/5/2019.

Results: In total, 267 unique patients have been treated with inotersen in NEURO-TTR (N=112), OLE (N=135), EAP (N=67), ATU (N=2), and IST (N=36) as of 1/5/2019. Interim analyses of the OLE show slowing of neurologic disease progression and stabilization of health-related quality of life in inotersen-treated patients. Of patients who began receiving inotersen after introduction of weekly platelet monitoring, 23.6% had confirmed platelet counts <100 × 10^9/L during the OLE. 5.2% of patients in the OLE experienced a ≥50% decrease in estimated glomerular filtration rate; all cases were transient or had alternative explanations. No cases of grade 4 thrombocytopenia and no severe acute glomerulonephritis have been reported since implementation of enhanced monitoring of platelet levels and kidney function in ongoing clinical trials and noninterventional studies.

Conclusions: Patients showed sustained slowing of disease progression from extended dosing of inotersen. These data show that with enhanced safety monitoring, events of grade 4 thrombocytopenia and acute glomerulonephritis have been successfully mitigated across a number of studies and treatment programs.

Key words: inotersen, safety, monitoring
PM103

PATIENTS WITH MIXED PHENOTYPE HEREDITARY TRANSTHYRETIN AMYLOIDOSIS: INSIGHTS FROM A GENETIC TESTING PROGRAM

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Background: Hereditary transthyretin (hATTR) amyloidosis is a progressive and fatal disease that results from the deposition of misfolded transthyretin (TTR) protein in major organs and systems, leading to multisystem dysfunction, including peripheral neuropathy, cardiomyopathy, and autonomic dysfunction. Patients with hATTR often experience symptoms of both cardiomyopathy (CM) and polyneuropathy (PN); this is referred to as a mixed phenotype. The hATTR Compass Program offers anonymous, confidential genetic testing and counseling to patients in the United States, Canada, and Puerto Rico suspected of having hATTR amyloidosis with polyneuropathy or with a family history of hATTR amyloidosis.

Objective: To analyze real-world data from the hATTR Compass Program in patients suspected of having hATTR amyloidosis to determine postreferral prevalence and characteristics of patients with a mixed phenotype.

Methods: This study analyzed data from 165 patients with TTR mutations sequentially identified by the hATTR Compass Program. DNA samples were scanned for TTR mutations associated with hATTR amyloidosis using a single-gene test, a cardiomyopathy gene panel, or a polyneuropathy gene panel.

Results: Seventy (42.4%) patients with hATTR had both PN and CM symptoms. Patients with a mixed phenotype had an average age of 70 years (range, 29–87 years), and the majority were male (57.1%). Most patients with a mixed phenotype were African American (n=54; 77.1%). Ten patients (14.3%) had a known family history of hATTR, 49 (70.0%) had no known family history, and 11 (15.7%) did not know. Most patients with a mixed phenotype had the p.V142I/V122I mutation (n=62; 88.6%), while 4 (5.7%) had p.T80A/T60A, 2 (2.9%) had p.S97F/S77F, 1 (1.4%) had p.V50M/V30M, and 1 (1.4%) had p.H108R/H88R. For this mixed phenotype group, cardiologists diagnosed 57 (81%), physician assistants and nurse practitioners working in cardiovascular clinics diagnosed 6 (9%), and various other specialties diagnosed 7 (10%) patients. Patients with a mixed phenotype presented with a variety of symptoms including sensory, motor, and autonomic dysfunction, alongside gastrointestinal dysfunction, heart disease, and bilateral carpal tunnel syndrome. Note that symptoms reported may be underrepresented because of limitations of data collection and program participation.

Conclusion: Diagnosis of hATTR amyloidosis is challenging, but recognition of its symptoms and subsequent genetic testing through the hATTR Compass Program can facilitate diagnosis of this debilitating, fatal disease. This study demonstrates that the mixed phenotype is fairly common in patients with hATTR and, regardless of mutation, patients should be assessed for both PN and CM symptoms.

Key words: genetics, mixed phenotype, demographics
Introduction: Hereditary ATTR amyloidosis (hATTR) is a progressive and fatal disease caused by the deposition of misfolded transthyretin-derived amyloid fibrils in nerve, heart, and other tissues. Early diagnosis is paramount to optimal disease management; however, the multisystemic involvement, phenotypic variability, and nonspecific symptoms can hinder hATTR recognition and delay diagnosis.

Objective: To describe neurological and other clinical features that should raise suspicion of hATTR polyneuropathy.

Methods: Descriptive summary of three neurologists’ clinical experience of diagnosing patients with hATTR polyneuropathy.

Results: Because hATTR is a rare neuropathy that may resemble many other forms of axonal neuropathy in its early stages, a high index of suspicion is needed, and widespread testing should be performed. Numerous factors in a patient’s medical history and clinical presentation should increase clinical suspicion for hATTR polyneuropathy. Important clues include heterogeneous presentations of peripheral neuropathy with varying degrees of pain, numbness, weakness and autonomic features; bilateral carpal tunnel syndrome (CTS); unexplained weight loss (recent or past); and/or a personal or family history of congestive heart failure. Patients may have substantial autonomic involvement with orthostatic hypotension, sexual dysfunction, and/or gastrointestinal disturbances. Lumbar spinal stenosis can also be present. Often patients look ill with cachexia, malaise, and lethargy. The symptoms and changes in the neurological examination are often rapidly progressive. Variable combinations of neuropathic pain, sensory loss, muscle weakness, weight loss and autonomic symptoms should make a physician suspect hATTR polyneuropathy. hATTR polyneuropathy is sometimes mistaken for chronic inflammatory demyelinating polyneuropathy because both can exhibit rapid progression with weakness and elevated CSF protein; thus, progressive neuropathy not responsive to immunotherapy is another indicator of hATTR. Once clinical suspicion is raised, genetic testing and biopsy of fat pad, skin, salivary gland, nerve, or other tissue may confirm diagnosis.

Conclusions: Early recognition of the clinical indicators of hATTR polyneuropathy (combination of sensory with pain, motor, and/or autonomic neuropathies, rapidly progressive neuropathy unresponsive to immunotherapy, appearing ill, weight loss, and a history of bilateral CTS) may improve earlier diagnosis of this progressive and fatal disease.

Key words: Hereditary ATTR amyloidosis; diagnosis; clinical
PM105

IMPACT OF INOTERSEN ON NEUROPATHY SYMPTOM AND CHANGE (NSC) SCORES INCLUDING AUTONOMIC SCORES FOR HEREDITARY TRANSTHYRETIN AMYLOIDOSIS POLYNEUROPATHY


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9 Indiana University.
10 New York University.
11 Otonomy, Inc.
12 Ionis Pharmaceuticals Inc.
13 Aurora Bio.
14 Akcea Therapeutics.

Background: Hereditary transthyretin amyloidosis (hATTR) is a rare, progressive, and fatal disease that manifests as buildup of transthyretin (TTR) protein in major organ systems, resulting in organ failure. Inotersen, a transthyretin-directed antisense oligonucleotide indicated for the treatment of hATTR polyneuropathy, demonstrated therapeutic benefit on sensorimotor peripheral neuropathy [modified Neuropathy Impairment Score +7 (mNIS+7)] and quality of life [Norfolk Quality of Life–Diabetic Neuropathy questionnaire (Norfolk QOL-DN)] in a global, randomized, double-blind, placebo-controlled phase 3 study (NEURO-TTR, NCT01737398). In the present analysis, we evaluated the impact of inotersen on Neuropathy Symptoms and Change (NSC) scores, an exploratory outcome in the NEURO-TTR study. The NSC score is a standardized questionnaire measuring motor, sensory, and autonomic symptoms in generalized polyneuropathy that is useful for the conduct of clinical practice, epidemiology surveys, and therapeutic trials. The NSC is used to quantitate and assess the distribution and severity of muscle weakness, sensory symptoms, and autonomic symptoms.

Objective: To report the impact of inotersen on NSC scores in patients with hATTR in the NEURO-TTR trial

Methods: Adults with stage 1 or 2 hATTR amyloidosis were randomized (2:1) to receive 300 mg weekly subcutaneous inotersen or placebo for 15 months. The NSC score (total, subdomains, and individual items) was assessed with the primary end points at baseline, 8 and 15 months.

Results: In total, 165 patients were analyzed. Inotersen-treated patients experienced significant therapeutic benefit versus those receiving placebo in NSC total score from baseline to 8 months (least-square mean [LSM] difference, −3.28; 95% CI, −5.70 to −0.86; \( P = 0.008 \)) and 15 months (LSM difference, −6.33; 95% CI, −9.12 to −3.55; \( P < 0.001 \)). Inotersen-treated patients also experienced therapeutic benefit at 15 months in subdomains of muscle weakness (−3.07; \( P < 0.001 \)), sensory (−1.90; \( P = 0.005 \)), pain (−1.59; \( P < 0.001 \)), and autonomic symptoms (−1.36; \( P = 0.008 \)) when compared with baseline status. Notably, autonomic symptoms also showed therapeutic benefit at 8 months (−1.03; \( P = 0.022 \)). Additionally, ten individual items showed statistical significance in favor of inotersen vs placebo at 15 months.

Conclusion: Inotersen conferred therapeutic benefit on neuropathic symptoms in patients with hATTR amyloidosis consistent with previously reported benefits in mNIS+7 and Norfolk QOL-DN. The improvement of autonomic symptom scores is notable as minimal changes in autonomic testing outcomes were observed with the mNIS+7.

Keywords: Amyloidosis; axonal biology; clinical trials; human genetics.
LOW SENSITIVITY OF BONE SCINTIGRAPHY IN DETECTING PHE64LEU MUTATION RELATED TRANSTHYRETIN CARDIAC AMYLOIDOSIS


Objectives Aim of our study is to assess diagnostic accuracy of bone scintigraphy in a large multicenter cohort of patients with cardiac amyloidotic involvement and Phe64Leu transthyretin (TTR) mutation.

Background Diagnostic accuracy of bone scintigraphy for transthyretin related cardiac amyloidosis (TTR-CA) is considered extremely high, enabling this technique as the non invasive diagnostic standard for TTR CA. Nevertheless this approach has not been systematically validated across the entire spectrum of TTR mutations.

Methods We retrospectively analyzed 55 patients with Phe64Leu TTR mutation, evaluated between 1993 and 2018 at seven specialized Italian tertiary centres. Cardiac involvement was defined in presence of an end-diastolic interventricular septum thickness≥12 mm, without other possible causes of left ventricular hypertrophy (i.e. arterial hypertension or valvulopathies). A technetium-99m-diphosphonate (99mTc-DPD) or technetium-99m-hydroxyl-methylene-diphosphonate (99mTc-HMDP) bone scintigraphy was reviewed and visual scoring was evaluated according to Perugini’s method.

Results Among 26 patients with definite cardiac involvement, 19 underwent 99mTc-DPD or 99mTc-HMDP bone scintigraphy. Of them, 17 (89.5%) patients had low or absent myocardial bone tracer uptake, while only 2 (10.5%) showed high grade myocardial uptake. The sensitivity and the accuracy of bone scintigraphy in detecting TTR-CA were respectively 10.5% and 37%. Characteristics of patients with high grade and low or absent bone tracer heart retention were reassumed in the table. Patients with cardiac involvement and low or absent bone tracer uptake were similar to those with high grade myocardial uptake, in terms of age, gender, electrocardiographic and echocardiographic findings.

Conclusions Sensitivity of bone scintigraphy in detecting TTR CA is extremely low in patients with Phe64Leu TTR mutation suggesting the need to assess diagnostic accuracy of bone scintigraphy to identify cardiac involvement across a wider spectrum of TTR mutations.
**PM107**

**DIVERSITY OF TRANSTHYRETIN PATHOGENIC VARIANTS AMONG SPANISH PATIENTS WITH TRANSTHYRETIN-RELATED HEREDITARY AMYLOIDOSIS**

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**Introduction:** Transthyretin-related hereditary amyloidosis (ATTR) is a dominantly-inherited form of systemic amyloidosis consequence of pathogenic variants in the transthyretin (TTR) gene. These variants modify the normal structure of transthyretin, facilitate its deposition in several tissues and cause variable degrees of neurological and cardiological manifestations. The p.Val30Met TTR variant represents the most frequent and widely studied pathogenic variant among the 130 already described.

**Objective:** To describe the diversity of TTR variants in a large Spanish cohort of families with ATTR.

**Methods:** The enrolled families were received in our unit in the period 1998-2019. Genomic DNA was isolated from the peripheral blood of affected individuals, and all coding exons of the TTR gene were analyzed using the Sanger method of DNA sequencing. Only families carrying potential TTR pathogenic variants were included in the present study.

**Results:** A total of 217 individuals carrying heterozygous TTR variants from 116 unrelated families of Spanish ancestry were enrolled. TTR genotyping revealed 15 different variants, which were absent or nearly absent in databases of genomic diversity (gnomAD) and most of them were classified as pathogenic variants in the ClinVar database (Table 1). The most frequent detected variants were p.Val30Met (64.98%), p.Val122Ile (11.98%) and p.Glu89Lys (9.22%). A systematic review of pedigrees revealed incomplete genetic evaluations in first-degree relatives of patients in nearly all enrolled families.

**Conclusions:** Herein we report the genetic data of the largest Spanish cohort of patients with ATTR described to date, with p.Val30Met TTR variant accounting for only 2/3 of them. Despite these large figures, the real incidence of ATTR in these families should be larger than here described as they have been incompletely studied.

**Key Words:** transthyretin, genetics, systemic amyloidosis

**Table 1: TTR gene variants detected in enrolled families.**

<table>
<thead>
<tr>
<th>Nucleotide Substitution (RefSeq: NM_000371.3)</th>
<th>Amino Acid Substitution</th>
<th>gnomAD (Allele Frequency)</th>
<th>ClinVar Classification</th>
<th>Patients n (%)</th>
<th>Families n (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>c.136G&gt;A</td>
<td>p.Ala25Thr</td>
<td>0</td>
<td>Pathogenic</td>
<td>2 (0.92)</td>
<td>1  (0.86)</td>
</tr>
<tr>
<td>c.148G&gt;A</td>
<td>p.Val30Met</td>
<td>0.00010</td>
<td>Pathogenic</td>
<td>141 (64.98)</td>
<td>81 (69.83)</td>
</tr>
<tr>
<td>c.199G&gt;C</td>
<td>p.Gly47Arg</td>
<td>0</td>
<td>Pathogenic</td>
<td>2 (0.92)</td>
<td>1  (0.86)</td>
</tr>
<tr>
<td>c.206C&gt;T</td>
<td>p.Thr49Ile</td>
<td>0</td>
<td>Not reported</td>
<td>3 (1.38)</td>
<td>2  (1.72)</td>
</tr>
<tr>
<td>c.220G&gt;C</td>
<td>p.Glu54Gln</td>
<td>0</td>
<td>VUS</td>
<td>1 (0.46)</td>
<td>1  (0.86)</td>
</tr>
<tr>
<td>c.221A&gt;G</td>
<td>p.Glu54Gly</td>
<td>0</td>
<td>Not reported</td>
<td>1 (0.46)</td>
<td>1  (0.86)</td>
</tr>
<tr>
<td>c.238A&gt;G</td>
<td>p.Thr60Ala</td>
<td>0.000004</td>
<td>Pathogenic</td>
<td>3 (1.38)</td>
<td>1  (0.86)</td>
</tr>
<tr>
<td>c.242A&gt;G</td>
<td>p.Glu61Gly</td>
<td>0</td>
<td>VUS</td>
<td>4 (1.84)</td>
<td>2  (1.72)</td>
</tr>
<tr>
<td>c.272T&gt;C</td>
<td>p.Val71Ala</td>
<td>0</td>
<td>Pathogenic</td>
<td>1 (0.46)</td>
<td>1  (0.86)</td>
</tr>
<tr>
<td>c.290G&gt;A</td>
<td>p.Ser77Tyr</td>
<td>0</td>
<td>Pathogenic</td>
<td>5 (2.3)</td>
<td>1  (0.86)</td>
</tr>
<tr>
<td>c.325G&gt;A</td>
<td>p.Glu89Lys</td>
<td>0</td>
<td>Not reported</td>
<td>20 (9.22)</td>
<td>7  (6.03)</td>
</tr>
<tr>
<td>c.325G&gt;C</td>
<td>p.Glu89Gln</td>
<td>0</td>
<td>Pathogenic</td>
<td>1 (0.46)</td>
<td>1  (0.86)</td>
</tr>
<tr>
<td>c.381T&gt;G</td>
<td>p.Ile107Met</td>
<td>0</td>
<td>Not reported</td>
<td>3 (1.38)</td>
<td>1  (0.86)</td>
</tr>
<tr>
<td>c.424G&gt;A</td>
<td>p.Val122Ile</td>
<td>0.001538</td>
<td>Pathogenic</td>
<td>26 (11.98)</td>
<td>14 (12.07)</td>
</tr>
<tr>
<td>c.424_426del</td>
<td>p.Val122del</td>
<td>0</td>
<td>Pathogenic</td>
<td>1 (0.46)</td>
<td>1  (0.86)</td>
</tr>
</tbody>
</table>

VUS: Variant of uncertain significance
**PM108**

**hATTR ITALIAN REGISTRY: CLINICAL AND EPIDEMIOLOGICAL DATA**


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**Introduction:** Hereditary transthyretin amyloidosis (ATTRv) is a life-threatening multisystemic disease with sensory-motor peripheral neuropathy, cardiomyopathy and dysautonomia. hATTR is considered to be endemic in Portugal and Sweden, with foci in Japan, Brasil, Maiorca, and Cyprus. Its global prevalence is traditionally estimated as 5000 to 10,000, but a recently published analysis reported that it may be as high as 38,000 persons.

**Objectives:** A national registry was planned to better characterize the epidemiology and the clinical phenotype of hATTR in Italy. Another aim of the registry was to monitor the clinical course of the disease and response to treatments.

**Methods:** Fifteen Italian referral centers for amyloidosis spread all over the country have contributed to the registry. Patients gave a written informed consent.

**Results:** Four-hundred-forty-seven living subjects were enrolled in the registry. Among these, 187 are asymptomatic carriers, while 260 are affected patients. Clinical characteristics are summarized in table 1. Thirty-four different mutations were recorded. The seven most represented genetic variants were significantly different in terms of age at onset, clinical features and geographical distribution. Met30 is the third most frequent variant, including carriers, found in 23% of symptomatic patients. According to region of birth, hATTR is more frequent in South Italy, with a high range of regional prevalence (from 2.5/million in Piedmont to 9-10/million in Calabria and Sicily), and a global prevalence of 4.33/million. Diagnostic delay is 3.4 years in probands, but is relatively short in their relatives (1.2 years) The most frequent symptoms at the diagnosis are sensory disturbances in lower limbs, followed by cardiological symptoms (dyspnea and lower limb edema). However often patients present at onset with a complex clinical picture including neuropathic, cardiological and dysautonomic symptoms in various combination.

**Conclusions:** Data reported in the Italian registry highlight that hATTRv has a high genetic heterogeneity and a great regional difference. The high number of different mutations with several different phenotypes is the cause of diagnostic difficulty among general practitioners and medical specialists, leading to the still long diagnostic delay. Increased medical awareness can allow easier recognition of the disease and early access to treatment. Moreover, knowledge of the regional prevalence in the national territory can lead to better planning appropriate health service policies. This study was funded by Italian Telethon Foundation (GUP15010)
A NEW COMPOUND TRANSTHYRETIN MUTANT, T60I AND V122I ASSOCIATED WITH CARDIAC AND AUTONOMIC NEUROPATHY PHENOTYPE

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Introduction: Transthyretin amyloidosis (ATTRm) is the most common form of familial amyloidosis. More than 100 amyloidogenic TTR mutations have been identified throughout the coding regions of the gene1. The amyloid-associated mutant, V122I, occurs in 4% of the African-American population and features a clinical phenotype of heart failure. While heterozygosity for TTR-V122I predominates, several compound heterozygous cases have been reported2. Here, we present a new compound mutation comprised of V122I along with the previously unreported T60I mutation manifesting as cardiac ATTRm amyloidosis.

Methods: A diagnosis of ATTR amyloidosis was established by Congo red staining of a tissue biopsy followed by immunohistochemical staining. Isoelectric focusing (IEF) was used to screen serum for a TTR variant protein. Bidirectional nucleotide sequencing of exons 1-4 in the TTR gene using DNA extracted from blood leukocytes detected gene mutations. Possible impact of T60I on TTR function was assessed using the prediction tool, PolyPhen2 (http://genetics.bwh.harvard.edu/pph2/).

Results: A 63-year-old African-American female presented with symptoms of cardiac involvement, autonomic neuropathy and sensory peripheral neuropathy. At visit, the patient reported significant dyspnea on exertion; orthopnea; early satiety; dizziness; numbness in her toes; and fatigue. The patient’s mother died of lung cancer and cause of death in the father was unknown. Among nine siblings, two brothers died of end-stage renal disease at ages 72 and 30, a sister succumbed to non-Hodgkin lymphoma, and cause of death for two other brothers was unknown. In living siblings, two sisters had hypertension, and two brothers had coronary artery and chronic kidney diseases, respectively. Nineteen months prior to evaluation, the patient underwent the first of two thoracenteses for pleural effusion. Nine months later, she noted dyspnea on exertion and increased fatigue. Atrial fibrillation and congestive heart failure with diastolic dysfunction were diagnosed. Two months later amyloidosis was identified by Congo red staining of rectal biopsy. At evaluation, the blood pressure measured 93/63 mm Hg without orthostatic changes; an ECG demonstrated sinus rhythm and low voltage in the limb leads; echo was notable for increased interventricular wall thickness of 15mm and left ventricular ejection fraction of 55%; and cardiac MRI documented diffuse delayed enhancement indicative of cardiac amyloidosis and severe mitral regurgitation. BNP and Troponin I measured 193 pg/mL and 0.085 ng/mL, respectively consistent with cardiac amyloidosis. Congo red staining of a fat pad aspirate was strongly positive (3+) for amyloid deposits. Immunohistochemical staining of the rectal biopsy identified TTR as the amyloid fibril protein; the stains for kappa and lambda light chain immunoglobulins and AA protein were negative. Serum screening by IEF showed the presence of two variant forms of TTR and absence of wild-type protein. DNA sequencing of the TTR gene revealed heterozygosity for both T60I (p.T80I; c.239C>T) in exon 3 and V122I (p.V142I; c.424G>A) in exon 4. PolyPhen2 predicted that the T60I mutation would cause functional disruption with the score of 0.998, a value that closely matched the predicted result of 1.0 obtained for V122I mutation. The patient died of unknown causes three years after the initial evaluation.

Conclusions: This is the first report of the new compound mutation T60I and V122I in the TTR gene with a phenotype of ATTRm featuring amyloid heart and nervous system involvements.

REFERENCES:

This research was supported by the Boston University Amyloid Research Fund, the National Institutes of Health RO1AG031804 (LHC), and the Young Family Amyloid Research Fund.
LIVER TRANSPLANTATION AND ORAL DRUG TREATMENT IN THE YEAR 2020. DATA FROM THE FAP WORLD TRANSPLANT REGISTER

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Background: Three decades ago, liver transplantation (LTx) was introduced as the only treatment with capacity to halt the progress of disease in transthyretin amyloidosis (ATTR). For the commonest mutation, Val30Met, the effect of transplantation is well known – stabilizing most patients. Some mutations require combined LTx/heart transplantation (HTx). However, not all patients are helped by transplantation. Large variations in survival, not only between different mutations but also between mutations with similar phenotypes, has been noted and it has become evident that each mutation needs individual consideration. Some mutations have similar long-term survival as the Val30Met, while LTx is not to be recommended for other mutations. Novel pharmaco-therapeutical approaches have also emerged over the last years providing a more attractive and less risky therapeutic procedure than transplantation.

Methods: Registry data concerning outcome after LTx for ATTR amyloidosis was evaluated. The material consisted of data from more than 2250 patients from 82 collaborating liver transplant centers. Survival was analyzed by the Kaplan-Meier method and Log-Rank test.

Results: In total, 66 separate mutations were treated by LTx alone or by LTX/HTx. Overall, 20-year survival after LTx was 55%. The introduction of pharmacotherapy in Europe has led to a significant drop in transplantation. Of the 224 reported transplantations between 2012 and 2018, one third of the patients had received pharmacotherapy before transplantation. There was no difference in disease duration before transplantation between the patients receiving pharmacotherapy and those not treated with drug therapy. Heart related deaths are the most common death cause at all timepoints. Deaths within the 1st year after transplantation are related to the operation procedure. Deaths after more than 10 years are more related to the amyloidosis (Table).

<table>
<thead>
<tr>
<th>Causes of death</th>
<th>Death &lt; 1 yr (n=263)</th>
<th>Death &gt; 10 yrs (n=100)</th>
<th>Total (n=695)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heart</td>
<td>21%</td>
<td>17%</td>
<td>20%</td>
</tr>
<tr>
<td>Sepsis</td>
<td>29%</td>
<td>9%</td>
<td>19%</td>
</tr>
<tr>
<td>PNF</td>
<td>5%</td>
<td>-</td>
<td>2%</td>
</tr>
<tr>
<td>MOF</td>
<td>10%</td>
<td>1%</td>
<td>7%</td>
</tr>
<tr>
<td>Intra-op</td>
<td>5%</td>
<td>-</td>
<td>2%</td>
</tr>
<tr>
<td>HAT</td>
<td>4%</td>
<td>-</td>
<td>2%</td>
</tr>
<tr>
<td>Pulm</td>
<td>6%</td>
<td>5%</td>
<td>5%</td>
</tr>
<tr>
<td>Cerebrov</td>
<td>3%</td>
<td>12%</td>
<td>19%</td>
</tr>
<tr>
<td>Rec of FAP</td>
<td>-</td>
<td>10%</td>
<td>6%</td>
</tr>
<tr>
<td>Not available</td>
<td>8%</td>
<td>30%</td>
<td>18%</td>
</tr>
</tbody>
</table>

Conclusions: Long-term survival after LTx for many TTR variants is excellent. The increased use of pharmacotherapy warrants careful surveillance regarding its outcome compared to LTx, because of the potential risk of less favorable surgical outcome if transplantation is merely delayed.
MAGNETIZATION TRANSFER RATIO QUANTIFIES POLYNEUROPATHY IN HEREDITARY TRANSTHYRETIN AMYLOIDOSIS – INTRODUCTION OF A NEW IMAGING BIOMARKER

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Introduction: A rapidly progressive, distal-symmetric, sensorimotor polyneuropathy (PNP) is one of the main manifestations in hereditary transthyretin (ATTRv) amyloidosis. Novel cost-intensive drug therapies were recently approved in the United States and Europe with promising initial results showing a decrease or cessation of disease progression when therapy was started early. Sufficient biomarkers for the detection of early or subclinical lesions as well as for therapy monitoring are still missing. We quantified peripheral nerve lesions by analyzing the magnetization transfer ratio (MTR) of the sciatic nerve in symptomatic and asymptomatic ATTRv-PNP, and tested its potential as a novel biomarker that indicates macromolecular changes.

Methods: We prospectively included 25 patients with symptomatic ATTRv-PNP (18 male, 7 female, mean age 58.8 years, range 33-76), 30 asymptomatic carriers of the mutant transthyretin gene (mutTTR) (13 male, 17 female, mean age 43.3 years, range 22-62), and 20 age-/sex-matched healthy controls (11 male, 9 female, mean age 44.3 years, range 22-73) in this study. All participants underwent magnetization transfer contrast (MTC) imaging in a 3 Tesla MR scanner, and two axial 3D gradient-echo sequences with and without an off-resonance saturation rapid frequency pulse were conducted at the right distal thigh. Sciatic nerve regions of interest were manually drawn on ten consecutive axial slices in the images without off-resonance saturation, and then transferred to the corresponding slices that were generated by the sequence with the off-resonance saturation pulse. Subsequently, we evaluated the MTR and cross-sectional area (CSA) of the sciatic nerve. All symptomatic ATTRv-PNP patients and asymptomatic mutTTR-carriers received detailed neurologic and electrophysiologic examinations including assessment of the Neuropathy Impairment Score of the Lower Limbs (NIS-LL).

Results: Sciatic nerve MTRs and CSAs reliably differentiated between symptomatic ATTRv-PNP, asymptomatic mutTTR-carriers, and controls. MTR was lower in symptomatic ATTRv-PNP (26.4±0.7%; p<0.0001) and in asymptomatic mutTTR-carriers (32.6±0.8%; p=0.0005) versus controls (39.4±2.1%), and was also lower in symptomatic ATTRv-PNP versus asymptomatic mutTTR-carriers (p=0.0009). The MTR correlated negatively with the NIS-LL, and positively with compound muscle and sensory nerve action potentials. Sciatic nerve CSA was higher in symptomatic ATTRv-PNP (34.3±1.7 mm³) versus asymptomatic mutTTR-carriers (26.0±1.1 mm³; p=0.0005), and versus controls (20.4±1.2 mm³; p<0.0001). CSA was also higher in asymptomatic mutTTR-carriers versus controls.

Conclusions: Magnetization transfer ratio is a novel imaging biomarker that i) can quantify macromolecular changes in ATTRv amyloidosis, ii) differentiates between symptomatic ATTRv-PNP patients, asymptomatic mutTTR-carriers, and non-amyloidotic controls, and iii) correlates with electrophysiologic results and clinical scores.
EARLY BIOMARKER IN HEREDITARY TRANSTHYRETIN AMYLOIDOSIS

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Background: Hereditary transthyretin (ATTRv) amyloidosis is an autosomal-dominant disease in which genetic mutations of transthyretin (TTR) cause amyloid fibrils deposits in various organs such as peripheral nerves and the heart. Disease-modifying therapies such as liver transplantation, tetramer stabilizers, and gene silencing drugs are more effective in the early stages of this disease. To diagnose this disease earlier, we need biomarkers detecting early pathological changes of the disease. Circulating growth and differentiation factor-15 (GDF-15) and soluble suppression of tumorigenicity-2 protein (sST2) were reportedly implicated in cardiovascular stress, fibrosis, inflammation, and remodeling.

Objective: To investigate whether plasma GDF-15 and sST2 levels are associated with early pathological changes of ATTRv amyloidosis.

Materials & Methods: We employed 32 patients with ATTRv amyloidosis, 8 asymptomatic TTR mutation carriers, and 8 healthy volunteers. We evaluated plasma GDF-15 and sST2 levels in those subjects and compared these levels to serum BNP, high-sensitivity troponin T (hsTnT), creatinine (Cr), eGFR, and CRP levels. In addition, we investigated whether plasma GDF-15 and sST2 levels are correlated with echocardiographic findings such as ventricular septal wall thickness (IVSTd), left atrial diameter (LAD), left ventricular ejection fraction (EF), E/ e’.

Results: Plasma GDF-15 levels were 6481.5 ± 763.1 pg/ ml, 2662.8 ± 162.5 pg/ ml, and 1196.5 ± 373.2 pg/ ml in ATTRv amyloidosis patients, asymptomatic mutation carriers, and healthy volunteers, respectively. We did not find significant differences of plasma sST2 levels among those three groups. In addition, plasma GDF-15 levels were significantly correlated with BNP, Cr, eGFR, hsTnT, CRP, IVSTd, and E/ e’ in patients with ATTRv amyloidosis.

Conclusion: GDF-15 may be a useful for the early diagnosis of ATTRv amyloidosis.
TRANSVERSE MYELITIS IN PATIENTS WITH TTRVAL30MET MUTATION: CLINICAL SPECTRUM OF THE DISEASE?

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Background: Transthyretin Familial Amyloid Polyneuropathy (TTR-FAP) is an hereditary amyloidosis caused by extracellular deposition of transthyretin, leading to systemic manifestations including in central nervous system. However, little is known about medullar manifestations. We describe 3 patients carrier of TTRVal30Met mutation, two of them siblings, with acute transverse myelitis.

Case report:  
Case 1: Male of 65 years-old (yo), with familial history of TTR-FAP (mother and 3 siblings), presented with asymmetric transverse myelitis at 47 yo. The CSF analysis was normal; medullar T2-weighted MRI revealed a hypersignal in D1-D2 level. Partial recovery from motor abnormalities was observed without acute treatment. By the age of 58 yo, he presented dysautonomic symptoms followed by sensory symptoms in lower limbs. Neurophysiological studies confirmed an axonal sensory polyneuropathy (PNP) and salivary gland biopsy was positive for amyloid. He started tafamidis with 60 yo, and is stable since then (stage 1 and PND score 1), without other signs of central nervous system involvement.

Case 2: Female of 52 yo, with large familial history of TTR-FAP, and personal history of hypothyroidism since 18 yo, presented with acute myelitis at 45 yo. T2-weighted MRI showed a medullar hypersignal in D2-D3 level. CSF analysis revealed a mild pleocytosis (10 mononuclear cells). Acute treatment with intravenous methylprednisolone was done without improvement. By the age of 49 yo she began sensory and disautonomic signs and amyloidotic ophthalmopathy was diagnosed. Neurophysiological studies showed a bilateral tunnel carpal syndrome, and salivary gland biopsy was positive for amyloid. She initiate tafamidis with 51 yo, and is stable since then.

Case 3: Female of 57 yo (patient 2 sister) began with symptoms of sensory polyneuropathy and dysautonomia by 38 yo. TTR-FAP diagnosis was made and she was submitted to liver transplant at 41yo. At the age of 44 yo she was diagnosed with serious heart conduction disturbance, and was submitted to pacemaker implantation. In terms of polyneuropathy she was stable since the transplant until the age of 57 yo. By the age of 57 she presented with a transverse myelitis. CSF study was normal. T2-weighted MRI showed a hypersignal in D8-9 level. Treatment with intravenous methylprednisolone was performed with improvement.

Conclusion: We describe 3 case reports of acute transverse myelitis of undetermined cause in patients known to have TTRVal30Met mutation. In two cases the event was previous to polyneuropathy. The presence of this phenotype in siblings (case 2 and 3) may point to an etiology related to TTR-FAP and that genetic factors may influence phenotypic heterogeneity.

Keywords: Transthyretin Familial Amyloid Polyneuropathy; Transverse myelitis
EXTERNAL QUALITY ASSESSMENT (EQA) SCHEME FOR GENETIC TESTING IN ATTR AMYLOIDOSIS

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Introduction: The TTR gene is highly polymorphic with more than 140 variants identified, of which 130 are amyloidogenic. With the exception of a valine residue deletion at position 142 and the 6 nucleotides duplication in exon 3 of the TTR gene all variants result from nucleotide substitutions. Recent advances in the treatment for ATTR amyloidosis, which has revolutionised patient care and prognosis, will result to a higher demand for genetic testing.

Objective and Methodology: Diagnostic genetic testing requires quality control procedures to ensure consistency and accuracy of results. To date there is no standardised criteria for genetic analysis and reporting in hereditary ATTR amyloidosis, which can influence genetic outcome and compromise patient care. We are proposing a new external quality assessment (EQA) scheme for genetic testing in ATTR amyloidosis across European laboratories. This will be done in collaboration with the European Molecular Genetics Quality Network (EMQN) and will help in establishing best practice guidelines for molecular diagnosis ensuring appropriate patient care.

Participating in EQA scheme will help in obtaining information on molecular techniques used, quality and performance of the laboratories undertaking genetic testing and help implementing the use of nomenclature in accordance with current recommendations (HGNC and HGVS nomenclature guidelines). This is particularly useful since laboratory practices are changing rapidly due to the implementation of next-generation sequencing (NGS) technologies and an increased demand for DNA screening in the era of treatment-oriented genetic testing.

Discussion: With advances in treatment for ATTR amyloidosis, in particular targeted gene silencing therapies we are on the threshold of a new era in the treatment of TTR amyloidosis. Previously TTR gene screening was used in the diagnostic work up – to avoid misdiagnosis of AL amyloidosis, which can be treated with chemotherapy, to provide information for family members and to allow highly selected patients to be considered for liver transplantation. Genetic testing provides early and accurate diagnosis and is a logical and feasible way to corroborate clinical diagnosis.
LONG-TERM IMPACT OF INOTERSEN ON NEUROPATHY QUALITY OF LIFE FOR TRANSTHYRETIN AMYLOIDOSIS & POLYNEUROPATHY: NEURO-TTR OPEN-LABEL EXTENSION AT 2 YEARS

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Background: A randomized, controlled phase 3 trial (NEURO-TTR) of patients with hereditary transthyretin amyloidosis with polyneuropathy (hATTR-PN) evaluated the efficacy of inotersen on neuropathic-specific quality of life (QOL), as measured by the Norfolk QOL-Diabetic Neuropathy (DN) survey. At 66 weeks of the trial’s randomized phase, patients receiving inotersen experienced statistically significant improvements from baseline, relative to placebo, in total score and several domains of the Norfolk QOL-DN. In the NEURO-TTR open-label extension (OLE) phase, all patients received inotersen. The Norfolk QOL-DN was assessed with data available for two years of OLE.

Objective: To examine changes in Norfolk QOL-DN domain scores for patients with hATTR-PN treated with inotersen for two years in the NEURO-TTR OLE phase.

Methods: The NEURO-TTR OLE phase enrolled 135 (of 139) randomized-phase completers who received inotersen (inotersen-inotersen; n=85) or placebo (placebo-inotersen; n=50) during the randomized phase. In the OLE phase, all patients received 300 mg inotersen once-weekly. The Norfolk QOL-DN, which captures neuropathic-specific QOL on five domains – activities of daily living (ADL; score range: 0 to 20), autonomic neuropathy (0 to 12), large fiber neuropathy/physical functioning (-4 to 56), small fiber neuropathy (0 to 16), and symptoms (0 to 32), with higher scores indicating worse QOL. The Norfolk QOL-DN was administered at OLE baseline and weeks 26, 78, and 104. Descriptive analyses examined observed mean Norfolk QOL-DN domain scores during the OLE phase for inotersen-inotersen and placebo-inotersen subgroups.

Results: For both subgroups, Norfolk QOL-DN domain scores from OLE baseline to week 104 were stable, with relatively small mean changes for inotersen-inotersen and placebo-inotersen subgroups for ADL (mean changes = 2.0, 2.3 points, respectively), autonomic neuropathy (0.3, 0.1), large fiber neuropathy (2.1, 1.5), small fiber neuropathy (0.2, 1.2), and symptoms domains (0.4, -0.5). Further, subgroup differences were similar at OLE baseline and week 104: the mean difference for the inotersen-inotersen and placebo-inotersen subgroups was 2.2 at OLE baseline vs. 2.4 at week 104 for ADL, 0.7 vs. 0.5 for autonomic neuropathy, 7.2 vs. 6.6 for large fiber neuropathy, -0.3 vs. 0.8 for small fiber neuropathy, and 2.5 vs. 1.6 for symptoms, indicating that the gaps between inotersen and placebo arms observed at the end of the randomized phase were sustained even after the placebo-inotersen subgroup received inotersen for two years.

Conclusions: Treatment with inotersen stabilized neuropathic-specific QOL for patients with hATTR-PN over two years, regardless of previous inotersen treatment status. The gaps in QOL between those receiving inotersen versus placebo during the randomized phase did not close over the OLE phase, indicating the importance of early treatment for maintaining QOL in these patients.
INOTERSEN DELAYS IMPACT OF POLYNEUROPATHY SYMPTOMS AND IMPAIRED DAILY ACTIVITIES FOR 3 YEARS IN PATIENTS WITH HEREDITARY TRANSTHYRETIN AMYLOIDOSIS

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Background: A randomized, controlled phase 3 trial (NEURO-TTR) of patients with hereditary transthyretin amyloidosis with polyneuropathy (hATTR-PN) evaluated efficacy of inotersen on neuropathic-specific quality of life, as measured by the Norfolk QOL-Diabetic Neuropathy (DN) survey. After the 66 week randomized phase, completers could enroll in the NEURO-TTR open-label extension (OLE) phase for continued inotersen treatment. The Norfolk QOL-DN was assessed with data available at Week 104 of the OLE phase, or 170 weeks (3.3 years) after the randomized-phase baseline.

Objective: To examine changes in polyneuropathy symptoms and impairment of daily activities and functioning, as captured by responses to selected items on the Norfolk QOL-DN, for patients with hATTR-PN treated with inotersen for over 3 years during the NEURO-TTR randomized and OLE phases.

Methods: Eighty-five patients with hATTR-PN who received inotersen during the 66-week NEURO-TTR randomized phase were enrolled in the OLE phase, with Norfolk QOL-DN data available at 104 weeks. Patients received 300 mg inotersen once weekly in each phase. Items analyzed were within the following Norfolk QOL-DN domains: activities of daily living (ADL; e.g., ‘difficulty dressing’, ‘difficulty using eating utensils’), autonomic neuropathy (e.g., ‘diarrhea’, ‘fainting/dizziness’), large fiber neuropathy/physical functioning (PF; e.g., ‘pain kept you awake at night’, ‘symptoms prevented usual activities’, ‘difficulty walking’), and small fiber neuropathy (e.g., ‘unable to feel feet when walking’, ‘unable to tell hot from cold water [hands/feet]’). Response choices for all items captured ‘severe’, ‘moderate’, ‘mild’, ‘very mild’, or ‘no’ problem; ‘severe’ or ‘moderate’ were classified as substantial impairment. For each of 18 items, descriptive analysis compared changes in the prevalence of substantial impairment between randomized-phase baseline and Week 104 of the OLE phase (170 weeks total). Positive percentage point changes represented increased prevalence.

Results: Changes in the prevalence of substantial impairments (in percentage points) between randomized-phase baseline and Week 104 of the OLE phase were quite small. The change in prevalence ranged from -1.9% to 11.5% for ADL items, -4.5% to 0.1% for autonomic neuropathy items, -3.3% to 10.1% for large fiber/PF neuropathy items, and -7.1% to 3.7% for small fiber neuropathy items. The prevalence in substantial impairments increased by more than 10 percentage points for only two items: ‘symptoms prevented usual activities’ (10.1%) and ‘difficulty getting on/off the toilet’ (11.5%).

Conclusions: Very few patients with hATTR-PN receiving continuous treatment with inotersen developed substantial impairments in any of the 18 assessed daily activities or function items over the course of 3 years. Long-term treatment with inotersen is thus associated with preservation of functioning and ability to carry out activities of daily living.
**PM118**

**PREDICTORS OF COGNITIVE DYSFUNCTION IN HEREDITARY TRANSTHYRETIN AMYLOIDOSIS WITH LIVER TRANSPLANT**

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**Introduction/Background:** Characteristics and predictors of central nervous system involvement of ATTRV30M amyloidosis are still largely unknown. Cognitive dysfunction has been shown to be part of the broad spectrum of clinical manifestations in older untreated hereditary TTR amyloidosis patients with peripheral polyneuropathy.

**Objectives:** To systematically explore cognitive dysfunction in patients whose disease course was modified by liver transplant (LT).

**Methods:** A series of 270 carriers of the TTRVal30Met mutation treated with LT (55% men, mean age at assessment=46.1±7.9, years of education=8.2±3.9, age at disease onset=31.1±7.0, disease duration at assessment=14.9±5.2 years and at transplant=3.6±2.0, and stage of disease=1.2±0.5) underwent a neuropsychological assessment, which included the Dementia Rating Scale-2 (DRS-2), Auditory Verbal Learning Test, Semantic Fluency, Phonemic Fluency, and Trail Making Test. Cognitive deficits were identified at the individual level. The 5th percentile of regression-based norms adjusted for demographic characteristics (sex, age, and/or education) was applied as a cut-off of deficit. Presence of cognitive dysfunction was determined by deficit in DRS-2 and/or multiple cognitive domains. Logistic regressions were applied to explore predictors of cognitive dysfunction. For multiple logistic regressions, a backward selection method was applied, with a threshold for variable removal of p>0.100.

**Results:** Cognitive dysfunction was found in 21 patients (7.8%). These patients had older age at assessment (odds=1.084, p=0.002) and at disease onset (odds=1.091, p=0.001) and, lower education (odds=0.755, p=0.007), and higher stage of disease (odds=2.826, p=0.002). No association was found with sex (p=0.823) or disease duration at assessment (p=0.933) or at transplant (p=0.663). Age at disease onset (adjusted odds=1.059, p=0.040), education (adjusted odds=0.819, p=0.049), and stage of disease (adjusted odds=2.353, p=0.022) remained statistically associated with cognitive dysfunction when analyzed as covariates. Though, age at assessment did not (p=0.100). Nine patients had ≥50 years at disease onset and 81 patients were ≥50 years at assessment. The frequency of cognitive dysfunction was higher in patients with ≥50 years at disease onset (44.4% vs. 6.5%, odds=11.482, p=0.001). The association between cognitive dysfunction and age at disease onset ≥50 years remained statistically significant (adjusted odds=5.070; p=0.039) when education (adjusted odds=0.804; p=0.033) and stage of disease (adjusted odds=2.218; p=0.036) were taken into account.

**Conclusions:** This cross-sectional study in LT patients confirms that cognitive dysfunction in ATTRV30M amyloidosis is closely related to age at disease onset and neuropathy stage, but not with disease duration. These results support the findings of the natural history of the disease.

**Keywords:** Familial amyloidotic polyneuropathy, Liver Transplant, Cognition

**Category:** Hereditary amyloidosis (ATTR and others)
In-vivo reflectance confocal microscopy of Meissner corpuscle for early detection of hereditary transthyretin amyloid polyneuropathy

Background: Early detection of neuropathy in patients with TTR mutations is essential for starting treatment in a timely manner. However, nerve conduction studies lack sensitivity for early detection of distal peripheral neuropathy. Confocal microscopy of Meissner Corpuscle (MC) is a noninvasive and painless method to evaluate the density of MC which could help detect very early neuropathy in patients with TTR mutations and affect treatment. 

Objective: To investigate the role of MC using RCM in patients with hereditary transthyretin amyloidosis (hATTR) polyneuropathy and asymptomatic TTR carriers.

Design/method: Sixty patients (40 healthy controls, 10 TTR amyloid neuropathy patients and 10 carriers of TTR mutations with clinical evidence of disease) will undergo in vivo RCM at the fingertip (Digit V) and medial longitudinal arch. Images analyzed for MC found in the dermal papillae and assessed for density at each site. Patients with TTR amyloid neuropathy and carriers also undergo Nerve Conduction Studies (NCT), Quantitative Sensory Testing (QST), Neuropathy Impairment Score (NIS), and Norfolk QOL.

Results: Results will be available in spring 2021 and for presentation at the meeting.

Conclusion: Confocal microscopy of MC could serve as a noninvasive and painless method to detect the very early hATTR polyneuropathy and follow patients over the progression of their disease.
A CASE REPORT OF HEREDITARY TRANSTHYRETIN AMYLOIDOSIS IN THE NOVEL VARIANT TRANSTHYRETIN V121A(P.V141A)

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Background: In hereditary transthyretin amyloidosis, amyloid cardiomyopathy and carpal tunnel syndrome were the primary and initial clinical manifestation of cardiac mutations. We describe a case of ATTRV121A amyloidosis, it is a novel mutation showed cardiac amyloidosis. In whom the diagnosis was settled by histopathological examination and genetic analysis of TTR DNA and mass spectrometry. The aim of this study was to assess the amyloidogenecity of this mutation.

Patients and Methods: A 71-year-old male with a non-family history of amyloidosis. At his age 50s, he was noticed hypertension and hyperuricemia, hyperlipidemia. He started a feeling of chest tightness at his age 70. The patient gradually noticed palpitation, and was referred to a general hospital. In the admission, the development of cardiomegaly was seen in X-ray, and presenting with orthopnea and dyspnea on exertion followed by swelling of the extremities. The patient had clinical signs and symptoms of heart failure (New York Heart Association grade3). The echocardiogram showed wall thickness at IVs and low voltage on electrocardiogram. He also showed the elevation of serum creatinine and his renal function deteriorate fast. During the course, the endomyocardial biopsy was performed and amyloid cardiomyopathy was suspected. There were no overt signs of peripheral neuropathy. The DNA analysis of TTR was performed, and heterozygous V121A (p.V141A) mutation was identified. It is considered as a novel mutation and it need further examination to pathogenicity of mutation, so we checked the amino acid substitution effects by using Polyphen2, the laser microdissection and tandem-mass spectrometry for cardiac amyloid deposition was performed. And the stability of tetramer dissociation is under investigation in present lab.

Results: The amino acid substitution effect showed damaging, and the extract amyloid from specimens showed the mixture of mutant TTR and wild-type TTR peptides and identified it as pathogenic mutation. Preliminary biophysical experiments suggest that this mutation is kinetically compromised

Conclusions: Heterozygous V121A is pathogenic mutation, showing the amyloid cardiomyopathy.

Disclosure statement: No potential conflict of interest was reported by the authors

Keywords: cardiac amyloidosis, hereditary ATTR amyloidosis, transthyretin
CARDIAC BIOMARKERS IN PATIENTS WITH HEREDITARY TRANSTHYRETIN AMYLOIDOSIS

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Background: Transthyretin (TTR) amyloid (ATTR) amyloidosis is either hereditary and caused by a TTR mutation (ATTRv amyloidosis) or required and related to aging (ATTRw amyloidosis). ATTRv amyloidosis is a systemic disease with amyloid depositions in various organs, notably the heart and nervous system. Heart involvement in ATTR amyloidosis is characterized by left ventricular hypertrophy and heart failure with preserved ejection fraction and/or cardiac arrhythmia. Soluble suppression of tumorigenicity 2 (sST2), galectin 3, Growth differentiation factor-15 (GDF-15) are promising biomarkers for heart failure and have been reported to have prognostic value in other cardiac diseases but have not yet been studied in ATTR amyloidosis patients.

Objectives: To examine if Soluble suppression of tumorigenicity 2 (sST2), galectin 3, Growth differentiation factor-15 (GDF-15), NTpro-BNP and Troponin-T are elevated in patients with hereditary ATTR amyloidosis.

Methods: Ninety patients with hereditary ATTR amyloidosis (73 ATTRV30M and 17 other mutations) and 27 healthy controls were included in the study. The serum levels of sST2, galectin-3, GDF-15, NTpro-BNP and Troponin-T were measured by enzyme-linked immunosorbent assay (ELISA).

Results: The patient group’s mean age was 64.7 years (range 34-86 years) and 56 % were males. The serum sST2 was significantly elevated in patients with ATTR amyloidosis compared to controls (p < 0.008). There was no statistical significance regarding Galectin-3, GDF-15, Pro-BNP or Troponin-T in ATTR patients compared to controls. sST2 was significantly higher in males compared to females (p<0.004) whereas galectin-3 concentration showed a tendency to be higher in males compared to females (p<0.06). There were significantly higher sST2 and Pro-BNP concentrations in patients with non-V30M mutations compared to controls (p<0.02 and 0.01 respectively). Pro-BNP was significantly elevated in patients with non-V30M mutations compared to patients with ATTRV30M. Pro-BNP correlated to interventricular septum thickness (r= 0.288 and p= 0.006). There was no correlation between the concentration of sST2, galectin-3, GDF-15, pro-BNP and amyloid fibril type.

Conclusions: Novel biomarkers such as ST2, GDF-15, galectin-3 may be useful for evaluation of patients with cardiac ATTRv amyloidosis. sST2 was the most sensitive marker of ATTRv amyloidosis in this study and may reflect ongoing myocardial damage.

Keywords: Biomarker, Soluble suppression of tumorigenicity 2, Galectin-3
CLINICAL CHARACTERISTICS OF ATTR-TYPE LEPTOMENINGEAL AMYLOIDOSIS/CEREBRAL AMYLOID ANGIOPATHY

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Background: Hereditary ATTR (ATTRv) amyloidosis is an autosomal dominant genetic disorder caused by transthyretin (TTR) gene mutation. The main clinical symptoms of ATTRv amyloidosis are polyneuropathy, autonomic neuropathy, and cardiomyopathy. Disease modifying therapies including liver transplantation, TTR tetramer stabilizers, and gene therapies ameliorate these clinical symptoms, however, efficacy of these therapies for ATTR-type CNS amyloidosis is limited. Therefore, understanding of ATTR-type CNS amyloidosis (i.e., leptomeningeal amyloidosis/cerebral amyloid angiopathy (LMA/CAA)), is becoming increasingly important.

Objectives: To elucidate the clinical characteristics of LMA/CAA in patients with ATTRv amyloidosis.

Methods: In this study, LMA/CAA symptoms are defined as clinically detectable objective and subjective CNS symptoms observed in the patients with ATTRv amyloidosis. CNS symptoms which can be clearly ascribable to common atherosclerotic/hypertensive cerebrovascular events or common epilepsy were excluded. We retrospectively analyzed medical records of ATTRv amyloidosis patients who developed LMA/CAA symptoms between 1996 and 2018 and their clinical features were investigated.

Results: Twelve patients (6 men, 6 women) were found to develop LMA/CAA. Seven were long-surviving post-liver transplant V30M (p.V50M) ATTRv patients. In these post-transplant patients, duration of illness from onset of ATTR amyloidosis to LMA/CAA onset were 17.1 ± 2.7 (mean ± SD) years. Another group consisted of five non-transplanted younger patients with non-V30M genotype (D18G (p.D38G), 2; G47R (p.G67R), 1; Y69H (p.Y89H), 1; Y114C (p.Y134C), 1), having short disease duration. The patients with D18G or Y69H genotype developed LMA/CAA as the initial symptom of ATTR amyloidosis. Durations of illness from onset of ATTR amyloidosis to LMA/CAA onset in the patients with Y114C and G47R were 5 and 6 years, respectively. Characteristic clinical manifestations of LMA/CAA were transient focal neurological episodes (TFNEs) (observed in 10 patients, 83%) and intracranial hemorrhage (observed in 4 patients, 33%). TFNEs included hemisensory disturbance, hemiparesis, motor aphasia, dysarthria, scintillating scotoma, abnormal behavior, consciousness disturbance, seizure, dizziness and black out. Four of the 10 patients with TFNEs were treated with antiepileptic drugs and three of them showed decrease of TFNEs frequency.

Conclusion: LMA/CAA is one of the most important complication in ATTRv amyloidosis patients. LMA/CAA occurs in long-surviving post-transplant V30M patients, as well as patients with several rare specific TTR gene mutations. TFNEs are the most common symptom in LMA/CAA and antiepileptic drugs might reduce frequency of TFNEs.
THE CLINICAL COURSE OF TRANSTHYRETIN FAMILIAL AMYLOID POLYNEUROPATHY WITH p.Ala117Ser MUTATION

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Background: Transthyretin Familial Amyloid Polyneuropathy (FAP) is an autosomal dominant inheritable disease resulted from a mutation in the TTR gene that encodes protein transthyretin (TTR). Typically, TTR-FAP patients develop progressive peripheral polyneuropathy, autonomic neuropathy, and restrictive cardiomyopathy secondary to the amyloid deposits in the tissue because of the mutated TTR. The epidemiology of FAP shows great variation across countries with p.Val50Met being the most common mutation worldwide in endemic regions. Hong Kong and mainland China are not FAP endemic regions and the disease is very rare. Recently, it is found that a missense mutation in TTR gene p.Ala117Ser is particularly prevalent among FAP in Southern China. We aim to study the phenotype of FAP in Hong Kong Chinese carrying this mutation.

Objectives: To study the phenotype of Hong Kong Chinese FAP with p.Ala117Ser mutation.


Results: Six symptomatic and 5 presymptomatic mutation carriers, being identified through cascade screening, were included. The median age of presymptomatic carriers was 42. For symptomatic patients, the median age of symptom onset was 56 (range 33-62). Two remained in FAP stage I and 15 years after symptom onset, 1 in FAP stage II 3 years after symptom onset and 3 in stage III after disease progression for 4, 17, and 19 years respectively. The first presenting symptom was limb numbness in 4 and gastrointestinal (GI) discomfort in 2. Three patients had carpal tunnel syndrome, two bilateral and one unilateral, as initial presentation of neuropathy, and all of them underwent decompression surgery. All 6 patients developed full-blown peripheral neuropathy and different degrees of GI upset in the course of their disease. Five had heart failure in the later years with 2 in NYHA class II, 2 in class III and 1 in class IV. The average lapse time from neuropathy to heart failure was 8.4 (range 1-14) years. One old lady with cardiomyopathy, in FAP stage I and NYHA class II had sudden cardiac death at age 70 without prior episode of syncope or palpitation, which raised concern on whether cardiac amyloidosis could be more susceptible to malignant arrhythmia at a relatively preserved cardiac function.

Conclusions: FAP patients with p.Ala117Ser tend to present initially with neuropathic form with carpal tunnel syndrome a common feature. Most patients are accompanied with different GI upsets and they develop cardiomyopathy after an average of 8.4 years. There is a wide variation on age of symptom onset, making the disease course and prognosis less predictable. Pre-symptomatic mutation carriers would require regular physical examination and investigations as early as mid-20s in order to capture the disease emergence.

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¹Subjects from the same family are grouped in the same cell; ²Not applicable.

Keywords: FAP, Chinese, Phenotype
PM124

CLINICAL AND RADIOLOGICAL CHARACTERIZATION OF CENTRAL NERVOUS INVOLVEMENT IN HEREDITARY TRANSTHYRETIN-MEDIATED AMYLOIDOSIS

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Background: Hereditary transthyretin-mediated amyloidosis (hATTR) results from mutations in the TTR gene, provoking TTR amyloid deposits. Central nervous system (CNS) involvement, particularly focal neurological events (FNE), including migraine with aura, seizures and stroke, have been seldom described and remain poorly understood.

Objectives: To describe the clinical CNS manifestations of hATTR and characterize brain imaging abnormalities.

Methods: Amongst a cohort of 351 symptomatic hATTR patients followed at Bicetre University Hospital, we investigated for patients with CNS symptoms and/or brain imaging (computed tomography, CT scan or magnetic resonance imaging, MRI). All brain images were blindly analyzed by a trained neuroradiologist.

Results: We identified 69 patients with CNS symptoms (20%); 61 had brain MRI, and 8 brain CT. Mean age was 57 years. Mean disease duration was 9.4 ± 6.7 years. CNS symptoms were: headache (38%), cognitive impairment (25%), depression (17%), ischemic stroke (16%), epilepsy (12%), migraine with aura (12%), transient ischemic attack (6%), hemorrhagic stroke (3%), subarachnoid hemorrhage (3%), and transient CNS symptoms of undetermined origin (14%). When combined together, FNE represented 52% of CNS events. Vascular complications occurred in 28% of cases with a higher incidence of ischemic events. When considering brain MRI, a high rate of microbleeds (27%) and hemosiderosis (12%) was observed. Meningeal contrast enhancement was found in 19% of cases. Enhancement was more frequent in patients with FNE, regardless of disease duration: 40% in FNE patients vs 0% in patients without FNE.

Conclusion: This study demonstrates that CNS involvement is clinically relevant in hATTR, most especially when considering the incidence of vascular complications (28% of cases). Microbleeds, hemosiderosis, and leptomeningeal enhancement were frequently observed and were associated with FNE. Clinicians should be aware of CNS involvement in hATTR in order to improve clinical management.
PM125

ANALYSIS OF THE RESPONSE TO TAFAMIDIS IN HEREDITARY TRANSTHYRETIN AMYLOIDOSIS WITH NEUROPATHY IN A NON-ENDEMIC AREA CENTER

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1 Hospital Clínico San Carlos.
2 Hospital Universitario Puerta de Hierro.
3 Hospital Infanta Sofía.

Introduction: Hereditary TTR amyloidosis (AhTTR) is a progressive disease that can be fatal in the absence of treatment. Till recently the only pharmacological treatment approved in Europe was tafamidis. Response to tafamidis appears to be best in the early-onset Val30Met patients. In this subset of patients an algorithm to predict response has been recently proposed. Having a NIS above 10 at the beginning of the treatment and being male appear to be predictors for non response.

Objective: To analyze the characteristics of the patients in treatment with tafamidis in our center (mostly late-onset patients) and the characteristics of responders and non-responders.

Methods: We selected all the patients followed up in a center that have received tafamidis for at least 6 months. In our country tafamidis is only approved for FAP I neuropathy patients. We consider non responders (NR) all patients in which their neurologist has considered that have no longer benefit by tafamidis. This included patients with progression of more than 6 points in two consecutive evaluations but also other criteria as EMG progression. We analyzed retrospectively age, sex, NIS, Norfolk, Compass and RODS at the beginning of the treatment. Statistical analysis was performed with non parametric test (U Mann Whitney and Chi square) with Prism software.

Results:
- 28 patients were selected. 11 (39% NR vs 61% responders R);
- 11 patients (39%) have a NonVal30Met mutation (4 Glu89Lys, 4 Ser97Tyr, 2 Val122Ile, 1 Glu89Gln), (3/11 NR, Glu89Gln and Ser77Tyr) vs (8/17 R, Glu89Lys, Ser77Tyr and Val122Ile) ns, p=0.29;
- 3 patients (10%) were early onset all of them responders (ns, p=0.15);
- 12 (43%) were females and 16 males (57%). NR 4 females (36%) and 7 males (64%) and R 8 females (47%) and 9 males (53%), ns (p=0.5);
- Mean age at the beginning was 62.4 (38-85), 66.2 NR vs 60 R (ns, p=0.29);
- Mean of treatment duration 34 months (6-86), NR 37.6 vs R 35.2 (ns p=0.69);
- Mean NIS at the beginning 22.74 (1-64), NR 34.72 vs R 16.00, p=0.01. 75% of R have a NIS<26.5;
- Norfolk at the beginning 39.17 (1-108), NR 50.4 vs R 33.2 (ns, p=0.11);
- First Compass-31 41.75 (26-88), NR 43.14 vs R 41.75 (ns, p=0.15);
- First RODS 39.1 (17-46), NR 36.8 vs R 40.1, (ns p=0.11);
- 1/11 NR was previously followed up as a carrier vs 9/17 R (p=0.04).

Conclusions: In our population, Tafamidis was effective in 61% of patients. The two factors clearly related with response were lower NIS and having been followed-up as a carrier. Although our sample is small and statistical power limited, we propose for late onset patients with NIS>30 to consider another treatment than Tafamidis as first-line choice or alternatively if Tafamidis is started carefully and tighten follow up should be implemented.

Keywords: Hereditary TTR Amyloidosis, Tafamidis, Treatment
SAFETY AND EFFICACY OF TAFAMIDIS IN PATIENTS WITH LATE-ONSET ATTRv AMYLOIDOSIS WITH POLYNEUROPATHY: SURVIVAL ANALYSIS AND LONG-TERM CLINICAL OUTCOMES

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2 Medical Department V, Heidelberg University Hospital, Heidelberg, Germany.
3 Institute of Human Genetics, Heidelberg University Hospital, Heidelberg, Germany.
4 Department of Neuroradiology, Heidelberg University Hospital, Heidelberg, Germany.

Background: Treatment with tafamidis has become the standard of care in ATTRv amyloidosis with polyneuropathy (ATTRv-PNP). However, little is known on the efficacy of tafamidis in late-onset ATTRv-PNP which is associated with a rapid disease course and worse prognosis.

Objectives: To identify predictors for survival and response to treatment with tafamidis in pts with mostly late-onset ATTRv-PNP.

Methods: We conducted a retrospective study of 122 consecutive patients with ATTRv-PNP treated at our center since 2000. Data on genotype, clinical presentation and disease progression were collected from medical records. We contacted family physicians of pts with no recent follow-up since January 2018 for information on long-term treatment outcome. Pts treated with tafamidis received regular neurological follow-up examinations at 6-12 month intervals, and disease progression under tafamidis was identified by a clinical expert, based on profound clinical examination and neurophysiology results, and resulted in termination or change of treatment. Primary endpoints were the identification of predictors for mortality and disease progression under tafamidis.

Results: Medical records of 348 patients with ATTRv amyloidosis were reviewed. 122 pts with ATTRv-PNP were identified (Table 1). Mean observation period (first to last contact) was 54±39 months. 38 (31.1%) pts died within the observation period following a mean period of 44±26 months. Cox regression analysis demonstrated an association between age >65 years at first contact (HR 2.33 [95% CI 1.21-4.52]; p=0.012) and cardiac involvement (HR 4.01 [95% CI 1.23-13.08]; p=0.021) with death.

We identified 65 pts who received treatment with tafamidis. It was well tolerated by all patients. Mean age at beginning of treatment was 61±11 years with only 8 pts being younger than 50 years. Mean follow-up period was 40±27 months. Four pts had a progression free follow-up period of over 6 years. Mean time until disease progression was 25±15 months in 33 (50.8%) pts. Progression rates within the first 2 years were 26.2% for all pts treated with tafamidis and 18.2% for pts with a baseline Peripheral Neuropathy Disability (PND) score of I. Time-to-progression was found to be associated with baseline modified Rankin Scale (mRS) (HR 1.94 [CI 1.18-3.19]; p=0.009) but was not associated with age (p=0.54).

Conclusion: Our study is unique in that it focuses on long-term clinical results in 122 pts with mostly late-onset ATTRv-PNP. Cardiac involvement and age represented the main risk factors for death. We found that early treatment outcomes on tafamidis depended on baseline neurological status and were not associated with age. Response rates to treatment were similar when compared to the results of other trials once baseline PND scores are taken into account. In summary, treatment with tafamidis is safe and efficacious in patients with early-stage and late-onset ATTRv-PNP.
Table 1: Characteristics of 122 patients with ATTRv-PNP according to the most common mutations

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<tbody>
<tr>
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<td>9</td>
<td>9</td>
<td>8</td>
<td>122</td>
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<tr>
<td>Age at first presentation, mean (SD)</td>
<td>57.88 (14.07)</td>
<td>58.33 (4.44)</td>
<td>66.11 (8.42)</td>
<td>67.38 (5.21)</td>
<td>58.52 (12.27)</td>
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<td>Liver Tx, n (%)</td>
<td>10 (19.6)</td>
<td>1 (11.1)</td>
<td>0 (0)</td>
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<td>20 (16.4)</td>
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<tr>
<td>Cardiac involvement, n (%)</td>
<td>34 (66.7)</td>
<td>5 (55.6)</td>
<td>7 (77.8)</td>
<td>8 (100)</td>
<td>94 (77)</td>
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<td>First line Treatment regimen</td>
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<tr>
<td>Tafamidis, n (%)</td>
<td>28 (54.9)</td>
<td>7 (77.8)</td>
<td>6 (66.7)</td>
<td>6 (75)</td>
<td>65 (53.3)</td>
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<tr>
<td>Diflusinal, n (%)</td>
<td>4 (7.8)</td>
<td>1 (11.1)</td>
<td>1 (11.1)</td>
<td>0 (0)</td>
<td>15 (12.3)</td>
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<td>Patisiran, n (%)</td>
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<td>0 (0)</td>
<td>0 (0)</td>
<td>0 (0)</td>
<td>1 (0.8)</td>
</tr>
<tr>
<td>No specific, n (%) treatment</td>
<td>19 (37.3)</td>
<td>1 (11.1)</td>
<td>2 (22.2)</td>
<td>2 (25)</td>
<td>41 (33.6)</td>
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<tr>
<td>Mortality</td>
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<td>Death, n (%)</td>
<td>16 (31.4)</td>
<td>1 (11.1)</td>
<td>3 (33.3)</td>
<td>4 (50)</td>
<td>38 (31.1)</td>
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<tr>
<td>Time until death within observation period (months), mean (SD)</td>
<td>52.94 (28.94)</td>
<td>107</td>
<td>41.67 (28.45)</td>
<td>41.50 (12.01)</td>
<td>44.18 (26.09)</td>
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<td>Clinical outcome</td>
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<td>Follow-up period (months), mean (SD)</td>
<td>58.47 (37.40)</td>
<td>57.56 (47.32)</td>
<td>38.89 (23.72)</td>
<td>44.87 (17.26)</td>
<td>54.66 (39.18)</td>
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<td>FAP stage, median (IQR)</td>
<td>1 (1-1.5)</td>
<td>1 (1-2)</td>
<td>1.5 (1-2.25)</td>
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*Abbreviations:* FAP, familial amyloidotic polyneuropathy; IQR, interquartile range; SD, standard deviation; Tx, transplantation

*Keywords:* Hereditary Transthyretin (Attrv) Amyloidosis, Polyneuropathy, Tafamidis
THE CHANGING FACE OF ATTR AMYLOIDOSIS: A 12 YEAR PERSPECTIVE FROM THE TRANSTHYRETIN AMYLOIDOSIS OUTCOMES SURVEY (THAOS)


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8 Columbia University College of Physicians and Surgeons, New York, NY, 9 Division of Hematology, Mayo Clinic, Rochester, MN, USA.
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10 Hospital Son Llatzer, Palma de Mallorca, Spain.
11 INCMNSZ, Mexico.
12 Institute for Neurological Research, FLENI, Buenos Aires, Argentina.
13 Penn Presbyterian Medical Center, University of Pennsylvania Health System, Philadelphia, Pennsylvania, USA.
14 Hospital Universitario Puerta de Hierro Majadahonda, CIBERCF, Madrid, Spain; and University Francisco de Vitoria (UFV), Pozuelo de Alarcon, Madrid, Spain.
15 Hospital Universitari de Bellvitge, Barcelona, Spain.
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17 Department of Transplant Medicine, University Hospital Muenster, Muenster, Germany.
18 Department of Neurology, Alexandrovska University Hospital, Sofia Medical University, Sofia, Bulgaria.
19 Nemo Sud Clinical Centre for Neuromuscular Disorders, Messina University Hospital, Messina, Italy.
20 Pfizer, Sydney, Australia.
21 Pfizer Inc, New York, NY, USA. 22 Pfizer, Collegeville, PA, USA.

Introduction: The Transthyretin Amyloidosis Outcomes Survey (THAOS) is an ongoing, global, longitudinal, observational survey of patients with transthyretin amyloidosis (ATTR amyloidosis). THAOS was established in 2007 and currently includes over 5,000 patients with inherited and wild-type ATTR as well as asymptomatic carriers with TTR mutations.

Objective: To examine the changing face of ATTR amyloidosis over the past 12 years by comparing patient demographics and genotype-phenotype relationships across the same group of THAOS study sites from the initial registry period to the present day.

Methods: Descriptive analysis of ATTR amyloidosis in THAOS at enrollment (data cut-off: April 1, 2019) comparing patient profiles at study sites between 2007 and 2009 versus profiles at those same study sites between 2010 and 2019. Site selection was restricted to those in THAOS from the initial registry period to present day to control for sampling bias due to growth in the number of sites over the analysis period.

Results: The THAOS population from 2007-2009 comprised 176 patients from 7 countries. Patients were primarily symptomatic for ATTR amyloidosis (70.2%) and 52.2% were male. Median age at enrollment was 49.1 years and 43.9 years at symptom onset. Genotype distribution was: 65.9% Val30Met, 11.4% ATTRwt, 15.9% cardiac mutations (Val122Ile, Leu 111Met, Thr60Ala or Ile68Leu), and 6.8% non-Val30Met/non-cardiac mutations. Phenotype distribution was: 21.2% cardiac, 46.0% neurologic, and 32.9% mixed. The majority of patients (58.9%) had early onset (aged <50 years) disease. Median NIS-LL score was 4.0, Karnofsky Index was 80.0%, and Norfolk QoL score was 28.0. The THAOS population from 2010-2019 comprised 2176 patients: 77.8% symptomatic and 60.8% male. Compared with 2007-2009, median age at enrollment was lower (41.9 years) as was the median age of disease onset (38.6 years). Genotype distribution differed notably with more Val30Met patients (85.6%), and fewer ATTRwt (6.2%), cardiac mutations (3.6%), and non-val30Met/non-cardiac mutations (4.6%). Phenotype distribution also differed: 11.0% cardiac, 66.3% neurologic, and 22.8% mixed. The proportion of patients with early onset disease was greater (65.9%). Median NIS-LL score was lower (2.0), Karnofsky Index was greater (90.0%), and Norfolk QoL score was lower (9.0).

Conclusions: This 12 year perspective of the changing face of ATTR amyloidosis in the THAOS dataset has revealed several key findings. Improved disease awareness is reflected in the growth of THAOS over time and earlier diagnosis is evident in the lower age and disease severity at enrollment in the 2010-2019 population vs the 2007-2009 population. As the 2010-2019 population only included those sites that were enrolling in 2009 and were still active, changes seen in genotype over time may reflect site selection bias. THAOS continues to be a valuable resource for elucidating the natural history of ATTR amyloidosis and its changing face over time.
PM128

ESCOR-TTR: SUPPORT PROGRAM FOR ALL PATIENTS TREATED FOR HEREDITARY TRANSTHYRETIN AMYLOIDOSIS (HATTR)


1 CHU Bicetre.
2 CHU Marseille.
3 CHU Bichat.
4 CHU Toulouse.
5 CHU Nantes.
6 CHU Strasbourg.
7 CHU Lyon.
8 CHU Lille.
9 CHU Fort de France.
10 CHU Montpellier.
11 CHU St Etienne.
12 French Association for Amyloidosis.

Introduction: Hereditary transthyretin amyloidosis (ATTRv) is a rare, debilitating, rapidly evolving and potentially life-threatening disease. It is linked to pathogenic transthyretin gene mutations responsible for amyloid fibril deposits in various organs such as the somatic and autonomic peripheral nerves, heart, gastrointestinal tract and eyes.

Objectives: The “ESCOR-TTR” Patient Support Program aims to provide personalized support for treated patients, and to help them prepare for consultations. This program suggests also to the HCPs to follow some standardized clinical parameters defined by experts to monitor patient’s evolution.

Methods: On signing the consent form, the patient enrolled by the doctor is contacted by a dedicated nurse trained in this condition. Four more telephone interviews are scheduled for M3, M6, M9 and M12. In addition to scheduled calls, patients may call a toll-free number to contact their dedicated nurse. At baseline and every 6 months before the consultation, patients are asked to complete 4 self-questionnaires: RODS (assessment of neurological disability), NORFOLK (quality of life assessment), COMPASS 31 (assessment of dysautonomia), Kansas City (impact of cardiac failure on the patient’s life). Nurses also report spontaneous calls. Therefore, at the beginning of the consultation, the doctor is provided with information reported over the last 6 months. Doctors are able to document the physical examination data in a “standardized consultation data” file. Information from referent neurologist, cardiologists and ophthalmologist for each patient will be updated every six to 12 months and shared among the specialists.

Results: From March to mid December 2019, 10 expert centers enrolled 40 patients in ESCOR-TTR. Fourteen were called during Month 3 and 6 during Month 6. Initial results show that 90% of the 4 self-questionnaires were easily completed by the patients. 6 alerts predefined by the scientific committee were sent to the doctor.

Conclusions: These initial results show the strong involvement of patients in the follow-up of their condition through the 4 self-questionnaires completed and confirm the importance of support Albby a dedicated nurse. This program can harmonize at national level both clinical practice and the management of the course of hereditary ATTR amyloidosis. ESCOR-TTR is proving to be an additional aid for doctors in terms of patient follow-up. Updated results will be presented at the time of ISA meeting.
HEREDITARY TRANSTHYRETIN AMYLOIDOSIS: EXPERIENCE FROM THE AMYLOIDOSIS CENTER OF EXPERTISE OF THE NETHERLANDS

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1 Amyloidosis Center of Expertise, University Medical Center Groningen, The Netherlands.

Introduction: Pathogenic variants in the gene encoding transthyretin (TTR) result in hereditary ATTR (ATTRv) amyloidosis, a clinically heterogeneous multisystem disorder. The Amyloidosis Center of Expertise of the University Medical Center Groningen (UMCG) in the Netherlands, was established in the 1960s as a national referral center for diagnosis and treatment of patients with all types of amyloidosis.

Objectives: To provide an overview of the different TTR variants, number of families, disease characteristics, and genotype-phenotype correlations of patients and presymptomatic carriers who presented to the UMCG Amyloidosis Center of Expertise for genetic and/or clinical evaluation.

Methods: This study retrospectively analyzed patients and presymptomatic carriers with a pathogenic TTR variant who presented to the UMCG Amyloidosis Center of Expertise for genetic and/or clinical evaluation from 1985 to 31 August 2019.

Results: We identified 231 carriers of a pathogenic TTR variant. Of them 71 (31%) are symptomatic ATTRv patients who are currently being treated at the UMCG. 47 carriers are asymptomatic and undergo periodical screening. A total of 96 patients (42%) are deceased, including 27 obligate carriers identified through pedigree analysis. A total of 17 carriers were either awaiting their first visit or were evaluated elsewhere. The most common pathogenic variant was TTRV30M (p.Val50Met) in 112 patients (48%). The largest single family carried the pathogenic TTRY114C p.(Tyr134Cys) variant (34 carriers). The six most common variants were TTRV30M, TTRY114C, TTRV122I p.(Val142Ile), TTRV71A p.(Val91Ala), TTRG47E p.(Gly67Glu), and TTRE89K p.(Glu109Lys). Median age of onset for all ATTRv patients was 51.8 years. Genotype-phenotype correlation studies are ongoing; of the 72 symptomatic TTRV30M patients (deceased and alive), 90% showed polyneuropathy, 68% autonomic neuropathy and a diagnosis of cardiomyopathy was made in 65%. Of eleven TTRV122I carriers, six (54%) had polyneuropathy.

Conclusions: Our data illustrate the phenotypical heterogeneity of ATTRv amyloidosis in a large national cohort. Variant-and organ-specific analyses of disease penetrance and prognosis are ongoing. Since ATTRv amyloidosis is a rare disorder, the size of our cohort demonstrates the importance of a Center of Expertise for a large referral area. This enables us to perform this study and to continuously improve the management of ATTRv amyloidosis patients and their presymptomatic family members.

Keywords: ATTR amyloidosis, Phenotypes, Pathogenic variants

Abstract category: Hereditary amyloidosis (ATTR and others)
RENAL INVOLVEMENT IN EREDITIONARY TRANSTHYRETIN AMYLOIDOSIS (HATTR): MYTH OR REALITY?

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4 Neurology Dept, Policlinico Gemelli, Rome, Italy.
5 Neurology Dept Italian Auxological Institute of Verbania, Italy.
6 Laboratory of Genetics Careggi University Hospital Florence, Italy.
7 Internal Medicine and and Tuscan Amyloid Center, Careggi University Hospital, Florence, Italy.
8 Interventional Structural Cardiology Division, Careggi University Hospital, Florence, Italy.
9 Cell therapy and transfusion medicine, BMT Unit, Careggi University Hospital, Florence, Italy.
10 Nephrology Dept. S. Orsola University Hospital, Bologna, Italy.
11 Nephrology Dept. Careggi University Hospital, Florence, Italy.
12 Fondazione IRCCS Policlinico S. Matteo, Pavia.

Background: TTR amyloidosis (ATTR) is the most common form of hereditary amyloidosis. Up to now we know more than one hundred mutations most of them showing a prevalent neurological, cardiological or mixed phenotype, respectively Familial Amyloid Polineuropathy (FAP), Familial Amyloid Cardiomiopathy (FAC) or both (1). Renal phenotype occurs much less frequently and is caused by at least 15 variants of whom V30M is the more common (2). Abroad, particularly in Portugal, renal involvement is reported in 1 out of 3 cases, whereas in mutations observed in Italy it would seem less common. Renal injury outbreaks with microalbuminuria which develops in overt proteinuria and later renal failure which in one third of patients progresses to ESRD in about 10 years (3). The aim of our study is to investigate frequency, characteristics and distribution of renal phenotype of hereditary ATTR (hATTR) in our country.

Materials and Methods: We performed a retrospective investigation among some of the main Italian centers caring for ATTR by sending a specific questionnaire concerning renal involvement in patients with amyloidosis due to TTR mutations. Questions present in the survey concerned the number of pts with TTR mutations, the types of mutations observed and characteristics of renal injury (24h urinary protein excretion, renal function, date of onset of disease etc.). Particularly it was asked whether there were comorbidities and if a renal biopsy was executed. In the absence of a renal biopsy, all patients who showed a renal injury not attributable to any known disease, were classified as doubtful/possible cases of renal hATTR.

Results: We recruited a total of 474 patients with at least 34 different TTR mutations. 35 out of 474 (8%) showed a renal injury (variable degree of urinary protein excretion and/or renal insufficiency) not attributable in 19 of them to any known disease. These patients were considered as possible renal involvement secondary to TTR mutation. In four cases renal biopsy showed the presence of TTR amyloidosis. Three of them had a V30M mutation with onset of renal injury at 41,43 and 46 years of age respectively. The Fourth patient showed a T78P variant with onset at the age of 69 years (Tab.1)

Discussion: Our data show that renal involvement in hATTR is apparently rare in our country. However our data may be the results of an underdiagnosis either due to the inadequate and not diffuse employment of common markers for renal injury including renal biopsy or to the diffuse presence of underlying comorbidities. A further explanation may be a possible different expression in our country of TTR gene variants, particularly the V30M one. Only a prospective study including most centers involved in hATTR which will check systematically the markers of renal injury like eGFR, urinalysis, microalbuminurina and 24h urinary protein excretion and possibly renal biopsy in doubtful cases, will be able to outline better the frequency and characteristics of hATTR in our country.

<table>
<thead>
<tr>
<th>Centers Involved</th>
<th>Patients with hATTR</th>
<th>Ratio Val30/Met all variants</th>
<th>Patients with renal injury</th>
<th>Patients with “possible” renal hATTR (renal injury not related to any other disease)</th>
<th>Patients with biopsy proven renal hATTR</th>
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<tr>
<td>Italian Auxological Institute</td>
<td>39</td>
<td>3 / 34</td>
<td>6</td>
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<td>Verbania Piemonte</td>
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<td>Policlinico S. Matteo Pavia</td>
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<td>51</td>
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<tr>
<td>Policlinico S. Orsola Bologna</td>
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<td>1 / 14</td>
<td>11</td>
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<tr>
<td>Policlinico di Careggi Firenze</td>
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<td>2 / 12</td>
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<tr>
<td>Fatebenefratelli Hospital &amp; Policlinico Gemelli Roma</td>
<td>64</td>
<td>8 / 42</td>
<td>5</td>
<td>3</td>
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</tr>
<tr>
<td>Total</td>
<td>474</td>
<td>20 / 139</td>
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</tbody>
</table>
PATISIRAN IN CLINICAL PRACTICE – EXPERIENCE FROM THE UK NATIONAL AMYLOIDOSIS CENTRE

J. D. Gillmore ¹

¹ National Amyloidosis Centre, Division of Medicine, UCL.

Introduction: Three weekly intravenous administration of patisiran, a TTR-specific RNA inhibitor, suppresses plasma TTR concentration by approximately 85% for the duration of therapy. Patisiran has been shown in a phase 3 clinical trial (APOLLO) to benefit patients with hereditary transthyretin (ATTR) amyloidosis and is licensed for treatment of hereditary ATTR amyloidosis with Stage 1 or 2 polyneuropathy.

Objectives: To determine the safety, tolerability and efficacy of patisiran for the treatment of hereditary ATTR amyloidosis in a clinical practice setting.

Methods: Patisiran was administered to 28 patients with hereditary ATTR amyloidosis via the UK National Amyloidosis Centre. All had stage 1 or 2 polyneuropathy and 26/28 had cardiac amyloid at the time of commencement of patisiran. Eighteen patients received concomitant diflunisal as a ‘TTR stabilising’ drug. Patients underwent serial monitoring of their neurological (PND stage, NIS score) and cardiac amyloidosis (cardiac magnetic resonance imaging (CMR), cardiac biomarkers and Tc-DPD scintigraphy) as well as functional assessment by 6 minute walk test. Cardiac outcomes were compared with 13 matched patients with hereditary ATTR amyloidosis followed for a similar time period who did not receive TTR-lowering therapy.

Results: Patisiran was generally well tolerated and there were few adverse events. Median (range) time per patient from commencement of patisiran to censor was 12.3 (5.0-25.9) months. One patient discontinued the drug due to persistent post-infusion neuropathic and generalised body pain; the remainder continued to receive the drug at censor, mostly via infusion at home. Median (range) plasma TTR suppression from baseline was 86% (33-93%) with 82% showing suppression of >80%. Neuropathy was stable or better both by subjective and objective measures in 28/28 patients. The was a median reduction in extracellular volume (ECV) by CMR of 0.02 in patients receiving patisiran accompanied by a reduction in cardiac uptake by Tc-DPD scintigraphy compared to a median increase in ECV of 0.04 among control patients (p=0.001). The CMR findings were corroborated by a fall in NT-proBNP concentration and stable 6MWT distance among those receiving patisiran whilst both measures declined in control patients.

Conclusions: Patisiran is well tolerated, can be successfully administered by home infusion, and benefits both the nerves and hearts of patients with hereditary ATTR amyloidosis who have a mixed cardiac and neuropathic phenotype.

Keywords: ATTR, amyloidosis, patisiran, RNAi
LONG-TERM IMPACT OF INOTERSEN ON GENERIC QUALITY OF LIFE FOR TRANSTHYRETIN AMYLOIDOsis & POLYNEUROPATHY: NEURO-TTR OPEN-LABEL EXTENSION AT 2 YEARS

A. Yarlas¹, A. Lovley¹, M. Pollock², M. Vera Llonch², K. McCausland¹, L. Obici³

¹ Optum.
² Akcea Therapeutics.
³ Amyloidosis Research and Treatment Center, Fondazione IRCCS Policlinico San Matteo, University of Pavia.

Background: A randomized, controlled phase 3 trial (NEURO-TTR) of patients with hereditary transthyretin amyloidosis with polyneuropathy (hATTR-PN) evaluated the efficacy of inotersen on generic health-related quality of life (HRQOL), as measured by the SF-36v2® Health Survey (SF-36v2). At 66 weeks in the randomized phase, inotersen showed statistically significant improvements from baseline, relative to placebo, in several SF-36v2 domains. In the NEURO-TTR open-label extension (OLE) phase, all patients received inotersen. The SF-36v2 was assessed with data available for 2 years of OLE.

Objective: To examine changes in SF-36v2 domains for patients with hATTR-PN treated with inotersen for 2 years in the NEURO-TTR OLE phase.

Methods: The NEURO-TTR OLE phase enrolled 135 (of 139) randomized-phase completers who received inotersen (inotersen-inotersen [I-I]; n=85) or placebo (placebo-inotersen [P-I]; n=50) during the randomized phase. In the OLE phase, all patients received 300 mg inotersen once-weekly. The SF-36v2 captures 8 domains of generic HRQOL – physical functioning (PF), role-physical (RP), bodily pain (BP), general health (GH), vitality (VT), social functioning (SF), role-emotional (RE), and mental health (MH) – all represented as norm-based T scores (mean=50, standard deviation=10) with higher scores indicating better HRQOL. The SF-36v2 was administered at OLE baseline and weeks 26, 78, and 104. Descriptive analyses examined observed mean SF-36v2 domain scores during the OLE phase for I-I and P-I subgroups.

Results: Most SF-36v2 domain scores were stable between OLE baseline and week 104, with relatively small mean changes for I-I and P-I subgroups for PF (mean changes = -1.6, -0.8 points, respectively), RP (-1.0, -0.7), BP (1.6, 3.3), RE (-1.0, -0.7), and MH (-1.2, -0.6). Further, mean differences between I-I and P-I subgroups were similar at OLE baseline and week 104 for these domains –5.8 at OLE baseline vs. 5.0 at week 104 for PF, 4.9 vs. 4.6 for RP, 4.8 vs. 3.0 for BP, 4.2 vs. 3.8 for RE, and 2.6 vs. 2.0 for MH – indicating that the gaps between inotersen and placebo arms observed at the end of the randomized phase were sustained even after the P-I subgroup received inotersen for 2 years. Different patterns for subgroups were observed for GH, VT, and SF domains, with small changes for I-I (-1.9, 1.3, and 0.1 points, respectively) but substantial declines for P-I (-5.9, -2.6, -3.8), with these decreases mostly occurring after the first year of treatment.

Conclusions: Treatment with inotersen stabilized many aspects of HRQOL, including physical functioning, role limitations due to physical and emotional health, pain, and mental health for patients with hATTR-PN over two years. For all domains, the gaps between treatment arms observed at the end of the randomized phase did not diminish following two years of subsequent inotersen treatment, indicating the importance of early treatment for maintaining HRQOL.

Keywords (up to 3): hATTR amyloidosis, quality of life, inotersen
Category: Hereditary amyloidosis (ATTR and others)
INOTERSEN DELAYS IMPAIRMENTS IN PHYSICAL, ROLE, AND SOCIAL FUNCTIONING FOR 3 YEARS IN PATIENTS WITH HEREDITARY TRANSTHYRETIN AMYLOIDOSIS

K. McCausland¹, A. Yarlas¹, A. Lovley¹, M. Pollock², M. Vera Llonch¹, L. Obici³

¹ Optum.
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³ Amyloidosis Research and Treatment Center, Fondazione IRCCS Policlinico San Matteo, University of Pavia.

Background: A randomized, controlled phase 3 trial (NEURO-TTR) of patients with hereditary transthyretin amyloidosis with polyneuropathy (hATTR-PN) evaluated efficacy of inotersen on health-related quality of life, as measured by the SF-36v2® Health Survey (SF-36v2). After the 66 week randomized phase, completers could enroll in the NEURO-TTR open-label extension (OLE) phase for continued inotersen treatment. The SF-36v2 was assessed with data available at Week 104 of the OLE phase, or 170 weeks (3.3 years) after the randomized-phase baseline.

Objective: To examine changes in impairment of physical functioning, role limitations, and social functioning, as captured by responses to selected items on the SF-36v2, for patients with hATTR-PN treated with inotersen for over 3 years during the NEURO-TTR randomized and OLE phases.

Methods: Eighty-five patients with hATTR-PN who received inotersen during the 66-week NEURO-TTR randomized phase were enrolled in the OLE phase, with SF-36v2 data available after 104 weeks of additional treatment. Patients received 300 mg inotersen once weekly in each phase. Items analyzed were within the following SF-36v2 domains: physical functioning (PF; e.g., ‘lifting/carrying groceries’, ‘walking several hundred metres’), role-physical (RP; e.g., ‘accomplished less at work due to physical problems’), and social functioning (SF; ‘physical or emotional health interfered with social activities’). For each domain, response choices were: PF items - ‘limited a lot’, ‘limited a little’, and ‘not limited at all’ (‘limited a lot’ was classified as substantial impairment); RP items - ‘all’, ‘most’, ‘some’, ‘a little’ and ‘none of the time’ (‘all’ or ‘most’ were classified as substantial impairment); SF items - ‘extremely’, ‘quite a bit’, ‘moderately’, ‘slightly’, and ‘not at all’ (‘extremely’ or ‘quite a bit’ were classified as substantial impairment). For each of 15 items, a descriptive analysis compared changes in the prevalence of substantial impairment between randomized-phase baseline and Week 104 of the OLE phase (170 weeks total). Positive percentage point changes represented increased prevalence.

Results: Changes in the prevalence of substantial impairments (in percentage points) between randomized-phase baseline and Week 104 of the OLE phase were quite small. The change in prevalence ranged from -5.3% to 13.1% for PF items and -2.7% to 4.7% for RP items, while the change for the single SF item was -4.4%. The prevalence in substantial impairments increased by more than 10 percentage points for only one item: the PF item ‘bending/kneeling/stooping’ (13.1%).

Conclusions: Very few patients with hATTR-PN receiving continuous treatment with inotersen developed substantial impairments in any of the 15 assessed physical, role, or social function items over the course of 3 years. Long-term treatment with inotersen is thus associated with preservation of physical, role, and social functioning that impact patients’ everyday lives.

Keywords (up to 3): hATTR amyloidosis, quality of life, inotersen
Category: Hereditary amyloidosis (ATTR and others)
PT001

PRELIMINARY DATA ON THE SAFETY AND EFFICACY OF A NOVEL PET RADIOTRACER, 124I-P5+14, FOR IMAGING PATIENTS WITH AL AMYLOIDOSIS

J. S. Wall 1, E. B Martin 1, A. Stuckey 1, T. Richey 1, A. D. Williams 1, D.C. Wooliver 1, S. Macy 1, J. S. Foster 1, B. Whittle 2, R. Geldrich 2, M. Powell 2, Y. Fu 2, D. Powell 2, D. Osborne 2, R. E. Heidel 3, B. Marine 1, M. Besozzi 2, R. Ramchandren 1, R. Lands 1, S. Kennel 1

1 Department of Medicine, University of Tennessee Medical Center.
2 Department of Radiology, University of Tennessee Medical Center.
3 Department of Surgery, University of Tennessee Medical Center.

Introduction: Deposition of immunoglobulin light chain-associated amyloid (AL) in abdominothoracic organs, notably the heart, liver, spleen and kidneys leads to organ dysfunction and significant morbidity. At present, determining the distribution of systemic AL can only be achieved through organ biopsy as well as inferences based on anatomic imaging or noting shifts in serum and urine biomarkers. Currently, there are no approved radiotracers in the US for the non-invasive, quantitative estimation of tissue amyloid load in all major organs. To address this, we have developed a synthetic, polybasic peptide radiotracer, designated 124Ip5+14, suitable for PET/CT imaging. This peptide binds many forms of amyloid through multivalent electrostatic interactions with the amyloid associated glycosaminoglycans and fibrils. Based on positive preclinical data, peptide p5+14 was labeled with iodine-124 for a Phase 1 PET/CT imaging trial (clinicaltrials.gov NCT 03678259).

Objectives: The goal of the Phase 1 clinical trial is to assess the safety and efficacy of 124I-p5+14 as an amyloid imaging agent in patients with systemic amyloidosis.

Methods: Patients >18 years of age with biopsy proven amyloidosis and not requiring heparin therapy are eligible. Subjects received <2 mg of 124I-p5+14 (<2 mCi) administered as a single IV bolus. PET/CT images for the initial cohort (n = 3) were acquired from 25 min to 48 h post injection. The second cohort of patients (n = 6) were imaged at ~5 h and 24 h post injection. Image data were acquired using a Biograph 16 PET/CT scanner with a low dose CT and 5 min acquisitions per PET bed position. The primary endpoints include recording adverse events and dosimetry, with a secondary endpoint of organ-specific sensitivity.

Results: To date, 9 AL patients have been evaluated. No serious adverse events of any grade were noted. Organ-specific and whole-body effective dosimetry were calculated, and the gender-averaged mean whole-body effective dose was estimated to be 0.235 mSv/MBq. Blood pool clearance of 124I-p5+14 was analyzed using a two phase exponential equation which yielded fast serum half-life values of 12 - 22 min, and corresponding elimination half-life values of 668 - 747 min. PET images indicated patient-specific retention of 124I-p5+14 in the heart, kidneys, liver, spleen, pancreas, bone marrow, adrenals, and lung. Cardiac uptake of the radiotracer was observed in 78% of patients, with a mean myocardium:blood pool ratio of 2.2 ± 0.5 for positive images (negative ratio is estimated to be ~ 1.3). Additionally, hepatic, splenic and renal uptake of 124Ip5+14 was observed in 20%, 40%, and 60% of AL patients, respectively, with an overall organspecific sensitivity of 0.88, based on clinical presentation.

Conclusions: PET/CT image data indicate that 124I-p5+14 can provide quantitative detection of systemic AL amyloidosis in multiple organ systems and will be clinically important for detecting and monitoring amyloid load.
PRESENCE OF t (11;14) IN AL AMYLOIDOSIS AS A MARKER OF RESPONSE WHEN TREATED WITH BORTEZOMIB BASED REGIMEN

B. Dumas¹, H. Yameen¹, S. Sarosiek¹, J. M. Sloan¹, V. Sanchorawala¹

¹Amyloidosis Center, Boston University School of Medicine and Boston Medical Center.

Introduction/background: The proteasome inhibitor bortezomib has become a backbone for first line treatment of AL amyloidosis for those not eligible for autologous stem cell transplantation (SCT). While promising overall hematologic response rates have been demonstrated, the presence of t(11;14), seen in up to 40-60% of patients with AL amyloidosis, may be associated with poorer response in those treated with bortezomib based regimens.

Objectives: Here we compare hematologic response to bortezomib based treatment regimens in a cohort of 88 patients with newly diagnosed AL amyloidosis, based on t(11;14) status. Specifically, we evaluate overall survival (OS), hematologic event free survival (hEFS), and organ response.

Methods: A cohort of 88 patients, who had a new diagnosis of AL amyloidosis and were treated with a bortezomib based regimen as first line therapy, were evaluated at the Amyloidosis Center between 2013 and 2017. Patients were followed through August 2019. Hematologic response to treatment, overall survival (OS), and hematologic event free survival (hEFS) were compared based on t(11;14) status at the time of diagnosis. Presence of t(11;14) was determined by FISH on plasma cell rich fresh bone marrow specimens. A hematologic event was defined as hematologic relapse, hematologic progression, start of second line therapy, or death. Kaplan Meier analysis was used for OS and hEFS analysis. Cardiac, renal, and hematologic response were classified using consensus criteria with two proportion hypothesis testing for statistical significance.

Results: Of the 88 patients with AL amyloidosis who received treatment with bortezomib based regimens, 44 had presence of t(11;14) and 44 were negative for the translocation. The majority of the patients (75%) received CyBorD/VCD, followed by bortezomib/dex in 23% and only 1 patient received VRD as the first line. Hematologic complete and very good partial responses (CR+VGPR) were achieved in 41% of patients with t(11;14) when treated with bortezomib based therapy and 75% of patients without t(11;14) (p=0.012). The median OS was 48 months in patients with t(11;14) and was not reached for patients without t(11;14). Five-year OS was 72% in patients without t(11;14). The median hEFS was 17 months for patients with t(11;14) compared to 34 months for the patients without t(11;14) when treated with bortezomib based therapy. Cardiac response was achieved in 11% of those with t(11;14) and 24% of those without t(11;14) after first line therapy (p=0.234). Renal response was achieved in 28% of those with t(11;14) vs 40% without t(11;14) (p=0.303).

Conclusion: Patients with AL amyloidosis and the presence of t(11;14) have inferior outcomes with respect to hematologic response, OS and hEFS when treated with bortezomib based regimens as first line therapy. Alternative treatments should be considered when possible in this population.

Table 1: Patient Characteristics

<table>
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<th>Characteristic</th>
<th>Presence of t(11;14)</th>
<th>Absence of t(11;14)</th>
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</thead>
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<tr>
<td></td>
<td>N=44</td>
<td>N=44</td>
</tr>
<tr>
<td>Male (%)</td>
<td>28 (64)</td>
<td>26 (59)</td>
</tr>
<tr>
<td>Median age, years</td>
<td>63.5 (42-80)</td>
<td>64 (41-87)</td>
</tr>
<tr>
<td>Median plasmacytosis</td>
<td>12.5 (1-50)</td>
<td>12.5 (0.31-30)</td>
</tr>
<tr>
<td>Median dFLC (range)</td>
<td>196.3 (3.1-9942)</td>
<td>112 (4.8-1091)</td>
</tr>
<tr>
<td>Cardiac involvement (%)</td>
<td>28 (64)</td>
<td>26 (59)</td>
</tr>
<tr>
<td>BU 2018 cardiac stage (%)</td>
<td>*N=22</td>
<td>*N=15</td>
</tr>
<tr>
<td>I</td>
<td>2 (9)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>II</td>
<td>10 (45)</td>
<td>9 (60)</td>
</tr>
<tr>
<td>III</td>
<td>3 (14)</td>
<td>2 (13)</td>
</tr>
<tr>
<td>IIIb</td>
<td>7 (32)</td>
<td>4 (27)</td>
</tr>
<tr>
<td>Renal involvement (%)</td>
<td>37 (84)</td>
<td>32 (73)</td>
</tr>
<tr>
<td>Renal stage (%)</td>
<td>*N= 32</td>
<td>*N= 26</td>
</tr>
<tr>
<td>I</td>
<td>7 (22)</td>
<td>6 (23)</td>
</tr>
<tr>
<td>II</td>
<td>16 (50)</td>
<td>15 (57)</td>
</tr>
<tr>
<td>III</td>
<td>9 (28)</td>
<td>5 (77)</td>
</tr>
</tbody>
</table>

*Not all patients with cardiac/renal involvement were able to be classified due to missing baseline data.
Figure 1: Kaplan Meier survival curves for OS and hEFS

<table>
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<tr>
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<th>Value</th>
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<td>Male (n = 182) (74%)</td>
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<tr>
<td>Median age, years (range) (n = 342)</td>
<td>74 (70-90)</td>
</tr>
<tr>
<td>Organ involvement (n = 335)</td>
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<tr>
<td>Renal</td>
<td>229 (68.3%)</td>
</tr>
<tr>
<td>Cardiac</td>
<td>167 (49.8%)</td>
</tr>
<tr>
<td>Neurologic</td>
<td>98 (29.2%)</td>
</tr>
<tr>
<td>Gastrointestinal</td>
<td>63 (19.4%)</td>
</tr>
<tr>
<td>BI cardiac staging (n = 215)</td>
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</tr>
<tr>
<td>I</td>
<td>45 (20.9%)</td>
</tr>
<tr>
<td>II</td>
<td>92 (42.8%)</td>
</tr>
<tr>
<td>IIb</td>
<td>38 (18.8%)</td>
</tr>
<tr>
<td>Median IVS4, mm (range) (n = 318)</td>
<td>11 (7 - 66)</td>
</tr>
<tr>
<td>Median LVEF, % (range) (n = 318)</td>
<td>62 (8 - 77)</td>
</tr>
<tr>
<td>Median B-type native renal peptide IgM (range), (n = 267)</td>
<td>185 (7 - 7684)</td>
</tr>
<tr>
<td>Median troponin I, ng/ml, (range), (n = 206)</td>
<td>0.01 (0.06 - 4.4)</td>
</tr>
<tr>
<td>Renal staging (n = 239)</td>
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<tr>
<td>I</td>
<td>98 (41%)</td>
</tr>
<tr>
<td>II</td>
<td>103 (43.1%)</td>
</tr>
<tr>
<td>IIb</td>
<td>38 (15.9%)</td>
</tr>
<tr>
<td>Median creatinine, mg/dl. (range)</td>
<td>0.7 (0.4 - 9.7)</td>
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<td>Median eGFR, ml/min/1.73m² (range)</td>
<td>88 (5 - 121)</td>
</tr>
<tr>
<td>Median 24 hour urine total protein, mg/24 hr (range)</td>
<td>3918 (4 - 20,515)</td>
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<tr>
<td>Treatment administered, n (%)</td>
<td>223 (65%)</td>
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<td>First line therapy, (n = 223)</td>
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<tr>
<td>Melphalan-Dexamethasone</td>
<td>70 (35.4%)</td>
</tr>
<tr>
<td>Proteasome inhibitor based regimens</td>
<td>90 (40.4%)</td>
</tr>
<tr>
<td>HDI/SCI</td>
<td>20 (11.7%)</td>
</tr>
<tr>
<td>Immuno modulatory drugs based regimens</td>
<td>17 (7.5%)</td>
</tr>
<tr>
<td>Other</td>
<td>11 (4.9%)</td>
</tr>
<tr>
<td>Second line therapy, (n = 81)</td>
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<tr>
<td>Melphalan-Dexamethasone</td>
<td>6 (7.4%)</td>
</tr>
<tr>
<td>Proteasome inhibitor based regimens</td>
<td>28 (34.6%)</td>
</tr>
<tr>
<td>HDI/SCI</td>
<td>6 (7.4%)</td>
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<tr>
<td>Immuno modulatory drugs based regimens</td>
<td>31 (38.3%)</td>
</tr>
<tr>
<td>Daratumab</td>
<td>4 (4.9%)</td>
</tr>
<tr>
<td>Other</td>
<td>6 (7.4%)</td>
</tr>
</tbody>
</table>
BUILDING A REGIONAL MULTIDISCIPLINARY CENTER FOR THE CARE OF PATIENTS WITH AMYLOIDOSIS.

J. Nativi-Nicolau1, J. Cowley1, J. Waldron1, K. Caballero1, K. Barrel1, J. Abraham1, L. Gessel1, C. Reddy1, M. Revelo1, K. Swade1, A. Rooks1, J. Hong1, L. Dranow1, A. Siu1, I. Sierra1, E. Montalvan1, J. Conte1, C. Rodriguez-Correa1, T. Kovacsovic1.

1 University of Utah, Salt Lake City, Utah.

Introduction: Amyloidosis is a systemic disease which requires complex diagnostic tools from different specialties, in order to provide an accurate and expedite diagnosis. In 2014 we organized a multidisciplinary group comprised of specialists from hematology, cardiology, nephrology, neurology, pulmonology, gastroenterology and pathology for the care of patients with amyloidosis.

Objectives: Our goal is to describe the impact of a multidisciplinary practice in the diagnosis of patients with amyloidosis. Methods: Retrospective analysis of patients treated with amyloidosis at the University of Utah and Huntsman Cancer Institute in Salt Lake City, Utah from 1997 to 2018. Patient volumes, characteristics, diagnostics tools and referral patterns were analyzed.

Results: A total of 391 patients with amyloidosis were seen from 1997 to 2018. Of them, 51% had light chain, 24% transthyretin and 26% other types of amyloidosis. After the introduction of the multidisciplinary practice and diagnostic algorithms in 2014, patients with light chain amyloidosis increased from an average of 5 to 22 patients per year (x4 fold), patients with transthyretin amyloidosis increased from an average of 1 to 16 patients per year (x16 fold) and other types increased from 4 to 6 patients per year – Table.

Patients with light chain amyloidosis, were predominately males (62%), who were diagnosed using a combination of bone marrow (100%), heart (34%) and fat pad biopsies (30%). The majority of patients with transthyretin amyloidosis were males (89%), only 11% had hereditary amyloidosis, and the diagnosis was confirmed using heart biopsy (58%) and/or nuclear scintigraphy with the absence of a monoclonal protein (49%). Genetic sequencing was performed for the majority (76%) of patients with transthyretin amyloidosis; testing rates increased from 50% (16/32 patients from 20014-2016) to 98% (39/40 patients from 2016-2018) when a designated genetic counselor joined the group in 2016. Referrals originated predominately from cardiology (55%) and hematology (25%), and approximately 49% were from states other than Utah.

Table. Types of amyloidosis diagnosed at the Utah Amyloidosis Program per Year

<table>
<thead>
<tr>
<th>Year</th>
<th>Sum of Total</th>
<th>Sum of AL</th>
<th>Sum of TTR</th>
<th>Sum of Other</th>
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<tbody>
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<td>1997</td>
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Conclusion: The introduction of a regional multidisciplinary center for the care of patients with amyloidosis increased the number of all types of amyloidosis; though the growth was considerably higher in patients with transthyretin amyloidosis. The diagnosis of amyloidosis required several multidisciplinary tools, diagnostic algorithms and collaboration among different specialists.

Key words: amyloidosis, light chains, transthyretin
PT005

AL AMYLOIDOSIS: SERUM AND URINE IMMUNOCHEMISTRY, COMPARISON WITH MULTIPLE MYELOMA AND MONOCLONAL GAMMOPATHY OF UNDETERMINED SIGNIFICANCE.

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Background and aims: AL Amyloidosis (AL AM) is a serious and rare disease whose diagnosis requires tissue biopsy although it may be suspected in the presence of some serum/urine immunochimistry findings. Our objective is to describe the immunochemical patterns (IC) found in serum / urine of AL AM patients at diagnosis and compare them with those found in myeloma (MM) and monoclonal gammapathies of undetermined significance (MGUS) to improve the early diagnosis of AL AM.

Methods: Patients: 89 AL AM, 100 MM and 100 MGUS (unselected, consecutively diagnosed, prospectively registered). Parameters analyzed (retrospective collection, computerized laboratory): immunofixation (IF) in serum / urine, serum free light chains (FLC, Freelite®).

Results:

Table 1. Results of IC data at diagnosis (AL AM, MM, MGUS).

<table>
<thead>
<tr>
<th>DIAGNOSIS</th>
<th>SERUM IF</th>
<th>URINE IF</th>
<th>ABNORMAL FLC</th>
</tr>
</thead>
<tbody>
<tr>
<td>AL AM</td>
<td>53.00%</td>
<td>65.00%</td>
<td>93.00%</td>
</tr>
<tr>
<td>MM</td>
<td>82%</td>
<td>89%</td>
<td>61%</td>
</tr>
<tr>
<td>MGUS</td>
<td>96%</td>
<td>61%</td>
<td>62%</td>
</tr>
</tbody>
</table>

Table 2. Results of serum immunofixation.

Table 3. Results urine immunofixation.

Table 4. Summary of IC results (serum/urine) in AL/AM.

Over 90% of AL AM show a positive IF (serum and / or urine) but 6.5% just an increase in one of the FLC. Only 3.5% of the cases show negative IF in serum and urine study and normal FLC. Clonality is lambda in 80% of cases. Among MM, 98% of cases show a positive IF (in serum and / or urine) while only 1% present with just an increase in any of the FLC (hyposecretory MM) or with complete absence of IC data (1%, non-secretory MM). Clonality is kappa in 65% of MM and 59% of MGUS.
Conclusions: The diagnosis of AL AM requires a high degree of suspicion and expertise because there is no single laboratory technique which is diagnostic, except a tissue biopsy. Comparative analysis between the patterns found in AL AM, MM and MGUS may help data interpretation and diagnosis. Most MM and MGUS show a positive IF in serum, with a kappa clone being the most prevalent. Only rarely do MM show negative IF in serum and urine. Immunochemistry results in AL AM are entirely different: high percentage of cases with negative IF in serum and / or urine, but almost always an increase in one of the FLC and a predominance of lambda clonality. The high percentage of patients with biclonal components is striking. If AL AM is suspected, IF in serum and / or urine and FLC quantification are mandatory. A diagnosis of AL AM is unlikely (but not impossible) if both serum and urine IF are negative and FLC are normal (3.5% in ours and up to 5% in other series). If a diagnosis of AL AM is suspected, tissue biopsy of an involved organ should be performed even if IC data are negative.

Keywords: Immunochemistry, differential diagnosis. Category: Diagnosis and prognosis of AL amyloidosis.
THE IMPORTANCE OF ABDOMINAL FAT ASPIRATION IN ADDITION TO BONE MARROW BIOPSY IN THE DIAGNOSIS OF AL AMYLOIDOSIS

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1 National Amyloidosis Centre, Royal Free Hospital, London.

Introduction: Systemic AL amyloidosis has variable clinical features and is challenging to diagnose. Histological confirmation of amyloid subtype by immunohistochemistry or mass spectrometry remains the gold standard. Biopsy of a clinically affected organ is invasive, costly and requires technical expertise to perform, which can lead to further diagnostic delays. Screening biopsies represent a low–risk approach to histological diagnosis and differentiation between ATTR (transthyretin) and AL amyloidosis without visceral organ biopsy.

Objectives: To report combined sensitivity and intrapatient concordance of abdominal fat aspirates (AFA), bone marrow (BM) and gastrointestinal (GI) biopsies in addition to the impact of organ involvement and amyloid load.

Methods: All confirmed cases of AL and ATTR amyloidosis whereby both an AFA and either a BM or GI biopsy had been performed (2006-2019) were identified from the database at the UK National Amyloidosis Centre. Baseline characteristics including organ involvement and amyloid load by 123I-SAP scintigraphy were recorded.

Results: Four hundred and seventy-one patients were identified. Median age was 75 (41-95) years; 338 (71.6%) were male. In AL amyloidosis, a lambda clone was present in 231 (72.0%) patients whilst 90 (28.0%) patients had a kappa clone. 98.1% patients underwent 123I-SAP scintigraphy. Organ involvement of the heart, liver and kidney was 68.6%, 15.8% and 34.2% respectively. In AL amyloidosis, diagnostic sensitivity of AFA, BM and GI biopsy were 73.5%, 62.4% and 76.7% whilst in ATTR, sensitivities were 27.3%, 41.4% and 44.6% respectively. The sensitivity of AFA in patients with wild-type ATTR was significantly lower than in those with AL amyloidosis (p<0.0001) or hereditary ATTR amyloidosis (p=0.0001). In ATTR amyloidosis, presence of amyloid was detected in 35.4% BMs and 33.3% of GI biopsies when AFA was negative for amyloid. When amyloid was present in AFA, amyloid was also present in 66.6% BMs and 68.2% GI biopsies. In AL amyloidosis, when AFA was positive, 68.4% of BM and 78.7% of GI biopsies were also positive. Amyloid was detected in AFA in 60% of patients with AL amyloidosis who had a negative BM for amyloid. Conversely, amyloid was detected in 43.7% of BM biopsies from patients with AL amyloidosis when AFA was negative.

In AL amyloidosis, patients with a visceral organ amenable to biopsy (cardiac, hepatic, renal), diagnostic sensitivities for AFA, BM and GI biopsies were 85.3%, 96.2% and 86.9% respectively. In combination, AFA and BM biopsies led to diagnostic sensitivities of 89.5% in this patient group (83.5% in all AL amyloid patients inclusive of those without a visceral organ amenable to biopsy e.g. soft tissue involvement). All screening biopsies were significantly more likely to be positive in patients with a moderate/large amyloid load (AFA: p=0.001, BM: p=0.014, GI: p = 0.037).

Conclusion: AFA is a simple, low risk procedure that can be performed at the bedside in addition to the BM trephine, which is part of standard diagnostic practice in patients with AL amyloidosis, to maximize avoidance of visceral organ biopsy. Screening biopsy review in a specialist center leads to a need for further biopsy in just 16.5% patients reducing to 10.5% in those with cardiac, hepatic or renal involvement, likely reflecting disease burden. The sensitivity is limited in the diagnosis of ATTR amyloidosis and yet, in AL amyloidosis, only a relatively small number of patients truly require biopsy of a clinically affected organ.
PT007

CLINICAL CHARACTERISTICS, TREATMENT REGIMENS AND SURVIVAL IN ELDERLY PATIENTS WITH AL AMYLOIDOSIS IN A TERTIARY REFERRAL CENTER

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1 Boston University Medical Center.

Background: AL amyloidosis is the most common form of systemic amyloidosis, characterized by an associated plasma cell dyscrasia leading to extracellular fibril deposition causing organ dysfunction. In these patients there is a fine balance between treatment toxicities and tolerability due to frailty and presence of multiorgan involvement.

Methods: A retrospective analysis of all patients 70 years or older who were evaluated at a tertiary referral center from 2000 until 2018 was performed to determine demographics, clinical characteristics, treatments and outcome measures. Kaplan Meier method was used to perform an overall survival (OS) of all patients from the time of diagnosis. Further analysis of OS was performed based on whether treatment was received before or after 2010 (when proteasome inhibitors were incorporated in the treatment algorithm for AL amyloidosis), whether administered treatment regimens consisted of proteasome inhibitors (PI), and whether or not treatment with high dose melphalan and stem cell transplantation (HDM/SCT) was received.

Results: A total of 342 patients with AL amyloidosis who were older than 70 years of age at the time of diagnosis had their initial evaluation at a tertiary referral center from 2000 to 2018. There were 215 (63%) men. The median age was 74 years (range, 70 – 90), and 55 (16%) were older than 80 years of age. The majority of patients were Caucasians (90%). The median number of organs involved by AL amyloidosis was 2 (range, 1- 7).

The most common organ involvement was renal in 229 patients (68.3%), followed by cardiac in 167 patients (48.8%), and neurologic in 98 (29.2%). The majority of patients had renal stage II disease and BU cardiac stage II disease.

Of the 342 patients, 223 (65%) received systemic treatment, the remainder (35%) received only supportive treatment. Of the 342 patients, 32 (9.4%) received treatment with HDM/SCT.

The median overall survival was 3.4 years (95% CI 2.9-4.1) with a median follow-up of 2.6 years (range, 0.02-15.2). The median OS of patients treated with PI based therapy was 6.0 years vs 3.7 years for those not treated with PI based regimens (p=0.01) (Figure 1). The median OS of patients was 6.8 years for patients treated with HDM/SCT and 4.0 years for those not receiving HDM/SCT (p=0.08). Moreover, the median OS of patients diagnosed prior to 2010 was 3.4 years which is similar to 3.9 years for those diagnosed after 2010 (p=0.07).

Conclusions: In summary, the clinical presentation of elderly patients with systemic AL amyloidosis is similar to that of younger patients in general. Overall survival of patients receiving a proteasome inhibitor-based therapy is better than those not receiving proteasome inhibitor-based therapy. In addition, HDM/SCT was offered to highly selected patients (9.4%) older than 70 years, however, the overall survival was no different than those who did not receive SCT.
DOES TIME TO DIAGNOSIS AFFECT OUTCOMES IN AL AMYLOIDOSIS?

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Introduction: Symptomology of AL amyloidosis can be vague, with a broad range of manifestations and potential etiologies. Despite advances in care and treatment over the past two decades, 6-month mortality has largely remained unchanged. A recent study utilizing patient-reported experiences found that diagnosis within 6 months of symptom onset occurred in only 37% of patients, whereas another 37% had a significant delay in diagnosis of more than 12 months.

Objective: We sought to determine whether time from symptom onset (patient-reported or appearance of first laboratory abnormality associated with AL amyloidosis) to diagnosis was associated with survival.

Methods: The Boston University Amyloidosis Patient Database is a comprehensive record of clinical and laboratory data on patients who present for evaluation at the Center. The database was queried for patients with AL amyloidosis who presented for an initial evaluation from 2010 to 2015. Patients with other forms of amyloidosis and/or coincident multiple myeloma or associated B cell lymphoproliferative disorders were excluded. Medical records were reviewed to confirm eligibility and acquire dates of symptom onset. Data cut-off date was Oct 1, 2019.

Results: A total of 324 patients with AL amyloidosis were included in this analysis. Median age was 63 years (range, 21 – 90), 59% were male (n = 196), and 83.9% were Caucasian (n = 271). The most common involvements were cardiac (n = 89, 27.5%) and renal (n = 170, 52.5%), with 85 patients (26.2%) having both cardiac and renal organs affected. A total of 222 patients (68.5%) had ≥ 2 organ systems involved. The median time to diagnosis from initial symptom onset was 7.1 months (range, 0 – 61). At the time of data cut-off, 195 (60.2%) patients were alive; of those, the majority were diagnosed less than 6 months from initial symptom onset (n = 102, 52.3%). In comparison, time to diagnosis from symptom onset in the non-survivor cohort was greater than 6 months in 63.6% (n = 82) of patients, a statistically significant result (p = 0.0005). Survival analysis by Kaplan Meier Log-Rank testing and Cox Proportional Hazards regression of time from diagnosis to death or censor stratified by time from patient-reported symptom onset to diagnosis (<6, 6-12, and >12 months) was also significant (p = 0.001). (Table 1)

Conclusions: AL amyloidosis is a complex disease, and non-specific symptoms may reduce the likelihood of a prompt diagnosis. Moreover, previous reports have described the importance of early diagnosis on prognosis for these patients. Our data supports this concept, revealing that the time to diagnosis from initial symptoms may be associated with overall survival. While a limitation of our study may be selection bias, further investigation to determine additional variables (age, cardiac/renal biomarker status, first line therapy) affecting this finding should confirm our results.

Table 1: Summary of Result

<table>
<thead>
<tr>
<th>Time to Diagnosis From Initial Symptoms</th>
<th>Total (n=324)</th>
<th>Survivors (n=195)</th>
<th>Non-Survivors (n=129)</th>
<th>P value</th>
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<tbody>
<tr>
<td>Median (range)</td>
<td>7.1 (0 – 61)</td>
<td>5.7 (0 – 52.8)</td>
<td>9.4 (0.5 – 61)</td>
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<td>&lt; 6 months (%)</td>
<td>149 (46)</td>
<td>102 (52)</td>
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<td>6-12 months (%)</td>
<td>81 (25)</td>
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<td>&gt;12 months (%)</td>
<td>94 (29)</td>
<td>42 (22)</td>
<td>52 (40)</td>
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Survival Analysis

<table>
<thead>
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<th>Time to Diagnosis From Initial Symptoms</th>
<th>Kaplan Meier Log-Rank</th>
<th>Cox Proportional Hazards</th>
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<tr>
<td></td>
<td>P value</td>
<td>HR (95% CI)</td>
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<tr>
<td>&lt; 6 months</td>
<td>0.001</td>
<td>1.43 (1.16 – 1.74)</td>
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<tr>
<td>6-12 months</td>
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<td>&gt;12 months</td>
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¹Wilcoxon Rank-Sum Test
SURVIVAL OUTCOME OF AL AMYLOIDOSIS A SINGLE-CENTER REGISTRY DATA ANALYSIS BETWEEN 1995 AND 2018

S. E. Yoon¹, S. J. Kim¹, K. Kim¹, J. H. Min¹, D. Kim¹, J. Choi¹, M. Park¹, E. Kim¹, J. Y. Choi¹, J. Kim¹, J. E. Lee¹

¹Samsung Medical center.

Introduction: Amyloid light-chain (AL) amyloidosis is a rare disease diagnosed by biopsy and immunofluorescence studies to identify the extracellular deposition of misfolded immunoglobulin light chains. The present incidence of amyloidosis in Korea has increased approximately 25 times compared with the 1990s. Thus, we analyzed the characteristics of AL amyloidosis during the last 20 years.

Methods: We have conducted a registry study for amyloidosis patients from 1995 to 2018 in Samsung Medical Center. At the time of analysis, a total of 302 patients registered. We excluded AA amyloidosis and hereditary amyloidosis. We also excluded patients without enough data or short follow up due to transfer to other medical centers.

Results: Ninety-six patients enrolled in 1995-2011, and 206 patients registered in 2012-2018. Among the 302 patients, the median age was 62 (36-83), 192 (63.6%) of those were aged younger than 65 years, and 110 (36.5%) of those were aged older than 65 years. The ratio of men and women was similar (male vs. female, 51.7%vs. 48.3%). Cardiac involvement found in 207 patients (68.8%), renal involvement in 171 patients (57.4%), and both involvement in 94 patients (31.1%). Besides that, 40 (13.3%) patients had hepatic involvement, 145 (54.4%) with autonomic neuropathy, and 51 (17.0%) with gastrointestinal amyloidosis. There were 122 (40.4%) patients with more than three organ involvements. Cytogenetic abnormality of AL amyloidosis was identified in 190 patients, for example Amp 1q (16.8%), t (11;14) (13.6%), and del 13 (10.5%). There were 160 patients (53.0%) with light chain only AL amyloidosis, 84 patients (27.8%) with IgG heavy chain, and 51 patients (16.9%) with IgA heavy chain. The light chain had more lambda (n=227, 75.1%) than kappa type (n=74, 24.5%). In the plasma cell disorder characteristics associated with AL amyloidosis, patients with increased bone marrow plasma cells were 42.1% (n=123), and among them, 27.4% (n=80) had symptomatic multiple myeloma (MM) with CRAB.

The median follows up duration in our study was 19 months (range 0-154), and the median overall survival was 41 months (95% CI 28.6-53.4). In survival comparison every 4 years, the median survival in 2009-2016 was 47 months (range 28.7-65.3), longer than in 2002-2008 (24 months, range 2.5-45.5) and 1995-2001 (5 months, range 0-28.8, p-value < 0.00).

Of the 193 patients that could be evaluated by Mayo 2004 criteria, there were 30 patients (15.5%) in stage I, 32 patients in stage II (16.6%), and 131 patients in stage III (67.9%). In addition, stage identification using Mayo 2012 was possible in 190 patients (stage I: n=26, 13.7%, stage II: n=23, 12.1%, stage III: n=50, 26.3%). In the comparison of survival according to stage based on Mayo 2004 and 2012, as previously reported in several studies, patients with stage III showed inferior outcome than stage I and II (P-value < 0.00).

Conclusion: Despite the shortcoming as a single-center data, we compared the patient characteristics of amyloidosis over time. In recent years, the number of patients has increased due to the elaboration of diagnostic methods, and the survival rate has confirmed by the wide application of anticancer drugs used for multiple myeloma.
PT010

CLINICAL FEATURES AND OUTCOME OF PATIENTS WITH AL AMYLOIDOSIS INVOLVING RARE EXTRANODAL SITES OTHER THAN HEART AND KIDNEY

S. E. Yoon1, S. J. Kim1, K. Kim1, J. H. Min1, D. Kim1, J. Choi1, M. Park1, E. Kim1, J. Y. Choi1, J. Kim1

1 Samsung Medical Center.

Introduction: Light-chain (AL) amyloidosis is the most common of systemic amyloidosis, accounting for 71% of patients with cardiac involvement, followed by 58% of patients with renal amyloidosis. Amyloid deposition in organs other than the kidney and heart rarely reported. In most patients, several organs may relate to amyloid, and there are few reports of clinical data on patients with amyloid that develop alone in organs such as GI tract, nervous, and liver, without the involvement of kidney and heart. Thus, based on the medical record, we report on the clinical outcomes of patients with non-renal and non-cardiac involved amyloidosis.

Methods: We have conducted a registry study for amyloidosis patients from 1995 to 2018 in Samsung Medical Center. Twenty-one patients with lung, liver, gastrointestinal, nervous system, and soft tissue included in this study. We excluded renal amyloidosis and cardiac amyloidosis confirmed by biopsy and imaging workup. Also, we excluded AA amyloidosis and hereditary amyloidosis.

Results: The median age of 21 patients was 60 years (range 45-75), with 47.6% of men similar to 52% of women. Their presentation patterns were varied: fatigue, poor oral intake, eyelid edema, jaundice, diarrhea, hematemesis, hematochezia, dry cough, dyspnea, and tingling sensation. Except for renal and cardiac involvement, the most commonly influenced organ in our study were soft tissue (n=11, 52.4%) followed by gastrointestinal (GI) tract involvement (n=8, 38.1%) and upper/lower respiratory system (n=7, 33.3%). Light chain kappa type presented in 11 patients (52.4%) and lambda type was shown in 10 patients (47.6%) with no difference. Eleven (52.4%) patients had shown more than 10% plasma cell monoclonality with bone marrow aspiration. The t(11:14) cytogenetic abnormality, found most frequently in amyloidosis, was found in 9 (42.9%) patients in our study. After diagnosis, 17 patients had received treatment to remove toxic amyloid clones, and four patients were followed up without treatment. Among 17 patients administrated with chemotherapy, five patients received thalidomide-based chemotherapy, five patients treated with bortezomib based chemotherapy, two patients treated with Cytoxan based chemotherapy, two patients with melphalan based chemotherapy and one patient with lenalidomide based chemotherapy. A total of 8 patients underwent Autologous hematopoietic stem cell transplantation (ASCT), all except one patient received induction chemotherapy before ASCT. The median follow-up duration was 14.6 months (range 0.9-102.0). The survival did not reach the median, and there was no difference in survival with or without treatment. (P-value = 0.88).

Conclusion: We analyzed the characteristics of patients without amyloid involvement of the kidney and heart. In the present study, the number of amyloid patients without renal and cardiac involvement was 6.9%. The initial symptoms of these patients showed various patterns, and the survival outcome in the therapeutic group showed no difference compared to the untreated group. Due to the few numbers of patients with non-renal and non-cardiac involved amyloidosis, further research would be needed to explain the characteristics and predict the outcome, based on the collaboration of the multi-center.
TREATMENT PATTERNS AND OUTCOMES OF PATIENTS WITH SYSTEMIC AL AMYLOIDOSIS BY ORGAN INVOLVEMENT: A RETROSPECTIVE ANALYSIS OF AN INSTITUTIONAL REGISTRY FROM ARGENTINA

M. L. Posadas-Martínez 1, M. A. Aguirre 1, P. Sorroche 1, D. Pérez de Arenaza 1, F. Varela1, G. Gustavo 1, D. Kohan 1, E. Brule 1, A. Basquiera 1, G. Machnicki1, M. Fernandez 1, E. Nucifora 1

1 Hospital Italiano de Buenos Aires.

Background: AL amyloidosis diagnosis is often delayed, and prognosis is poor due to advanced, multi-organ (particularly cardiac) involvement at diagnosis.

Aim: Describe treatments and overall survival according to organ compromise in treated AL amyloidosis patients.

Methods: Retrospective cohort (2010-2019) of adult patients with AL amyloidosis from Italian Hospital of Buenos Aires - Institutional Registry of Amyloidosis (IRA - ClinicalTrials.gov#: NCT01347047). Baseline characteristics, organ involvement, laboratory and treatment data were collected. Organ compromise was defined as clinical and/or biomarkers and/or imaging findings. All patients were followed until death or lost to follow-up and hematologic response was evaluated. Survival rates are expressed as the % surviving at 1, 5, and 10 years by Kaplan Meier method. Cox regression was used to evaluate mortality according to organ involvement at diagnosis.

Results: 91 patients with AL amyloidosis were included, median age was 63 years (53-70) and 43% (29) were women with a median Charlson score of 2 (DS 1) and ECOG status of 2 (DS 1). The median of organ involvement was 2 (1-3). Heart and kidney were affected in 71% and 72% of patients, respectively; followed by gastrointestinal and neurological involvement. Median alkaline phosphatase was 90IU (64-140), NT-Pro BNP 2899 pg/dl (874-10106), serum creatinine 1.1mg/dL (0.7-2.1), kappa/lambda ratio 0.5 (0.1-2), serum free light chains kappa 28(10-73), lambda 73(29-466), ESR 41 (21-52), kappa/lambda urine ratio 0.68 (0.5-1.5), β2 serum microglobulin 0.4 (0.3-1.9)mg/l. Sixty-nine patients received treatment out of which 6 had a heart transplant upfront and ASCT was performed on 13 (19%). The most frequent first line regimens were: CyBorD 76% (51) and Thalidomide-Dexamethasone/Cyclophosphamide 11% (7). The mean of first line cycles was 5 (SD 5). Second line was received by 26 patients with a mean of 4 cycles (SD4) - Lenalidomide/Dexamethasone for 42% (11) and Daratumumab for 19% (5). Three patients received a third line treatment with a mean of 4 cycles (SD 3). The median follow-up for treated patients was 66 months (IC 24-128). Overall hematologic best response rate (intention-to-treat) was: complete remission 46% (32), very good partial remission 7% (5), partial remission 5% (3) and no response 26% (18). Response was not evaluable in 11 patients (2 deaths, 1 discontinued treatment by patient’s decision, 2 with insufficient follow-up time and 6 referred to other institutions). Overall mortality was 38% (26, CI 27-50). Survival was 94% (84-98) at 1 year, 75% (63-84) at 3 and 61% (46-74) at 10 years. Number of organs compromised and higher levels of proBNP were associated with higher risk of mortality (HR1.5 and HR1, respectively). Kidney, gastrointestinal and neurological involvements were not associated with mortality.

Conclusion: Number of organs involved, and cardiac compromise were associated with poorer prognosis on this cohort of patients.
PATIENT-REPORTED OUTCOMES (PROs) AT DIAGNOSIS CAN PREDICT ONE-YEAR SURVIVAL IN LIGHT CHAIN (AL) AMYLOIDOSIS

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1 Medical College of Wisconsin.
2 Marquette University.
3 Mayo Clinic.

Introduction: PROs measure patient symptoms, functioning, other aspects of quality of life, and are predictive of mortality in multiple conditions.

Objectives: 1. Understand PRO domain changes in the first year (1-y) after diagnosis, 2. Explore whether baseline PROs predict 1-y mortality in AL amyloidosis.

Methods: 59 patients with newly diagnosed systemic AL were enrolled at 2 sites and followed at 3 months (3-m), 6 months (6-m) and 1-y. Patients were staged at enrollment with the 2012 Mayo system. PROs were measured using PROMIS Global Health and PROMIS-29. Scores are described as mean T-scores with standard deviations (SD) and standardized to a mean of 50 (SD 10) where 50 represents the mean of the general U.S. population. Higher score indicates more of the concept being measured (eg. fatigue score >50 implies worse fatigue, physical function >50 means better physical function). Analysis was adjusted for missing PRO data by a latent mixed model. A score change of 3 points was considered clinically meaningful. Age, gender, stage, number of organs and type of organ involvement were analyzed as predictors for 1-y survival.

Results: Baseline features are shown in Table 1. Mortality at 1-y was 21%. Scores worsened at 3-m with physical function, fatigue, social roles, anxiety, global physical and mental health scores showing significant change. These improved by 1-y- significantly for physical domains including global physical health, fatigue and physical function (Table 2). Table 3 shows the significant baseline PRO predictors of 1-y mortality after adjusting for stage.

Conclusions: PRO trajectories show initial worsening after AL amyloidosis diagnosis followed by improvement by 1-y, particularly for physical domains. Multiple PRO domains can predict 1-y mortality even after adjusting for stage.

Table 1.

<table>
<thead>
<tr>
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<th>N=59</th>
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<tbody>
<tr>
<td>Age, median (range)</td>
<td>68 (48-83)</td>
</tr>
<tr>
<td>Male (%)</td>
<td>34 (58)</td>
</tr>
<tr>
<td>AL Lambda (%)</td>
<td>46 (78)</td>
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<tr>
<td>2012 stage (%)</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>9 (14)</td>
</tr>
<tr>
<td>2</td>
<td>18 (28)</td>
</tr>
<tr>
<td>3</td>
<td>20 (36)</td>
</tr>
<tr>
<td>4</td>
<td>10 (16)</td>
</tr>
<tr>
<td>Missing</td>
<td>2 (6)</td>
</tr>
<tr>
<td>Cardiac (%)</td>
<td>39 (66)</td>
</tr>
<tr>
<td>Renal (%)</td>
<td>34 (58)</td>
</tr>
<tr>
<td>Number of organs (%)</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>21 (36)</td>
</tr>
<tr>
<td>2</td>
<td>23 (39)</td>
</tr>
<tr>
<td>≥3</td>
<td>15 (25)</td>
</tr>
</tbody>
</table>
Table 2. Change in PRO scores over time

<table>
<thead>
<tr>
<th></th>
<th>Baseline mean (SD), N=59</th>
<th>Baseline - 3m change mean (p-value), N=43</th>
<th>3m - 1y change mean (p-value), N=31</th>
</tr>
</thead>
<tbody>
<tr>
<td>Global Physical Health</td>
<td>42.5 (1.6)</td>
<td>-3.3 (0.03)</td>
<td>3.9 (0.004)</td>
</tr>
<tr>
<td>Global Mental Health</td>
<td>48.5 (1.2)</td>
<td>-3.3 (0.006)</td>
<td>2.1 (0.1)</td>
</tr>
<tr>
<td>Anxiety</td>
<td>55.5 (1.1)</td>
<td>-2.1 (0.03)</td>
<td>-1.6 (0.3)</td>
</tr>
<tr>
<td>Depression</td>
<td>53.4 (1.2)</td>
<td>-1.3 (0.2)</td>
<td>-0.6 (0.6)</td>
</tr>
<tr>
<td>Sleep Disturbance</td>
<td>51.8 (1.3)</td>
<td>0.4 (0.8)</td>
<td>-0.5 (0.8)</td>
</tr>
<tr>
<td>Social Roles</td>
<td>47.1 (1.4)</td>
<td>-2.7 (0.05)</td>
<td>1.4 (0.3)</td>
</tr>
<tr>
<td>Fatigue</td>
<td>55.6 (1.6)</td>
<td>3.4 (0.02)</td>
<td>-4.2 (0.009)</td>
</tr>
<tr>
<td>Physical Function</td>
<td>39.8 (1.4)</td>
<td>-4.1 (0.003)</td>
<td>3.3 (0.03)</td>
</tr>
<tr>
<td>Pain Interference</td>
<td>51.2 (1.4)</td>
<td>0.4 (0.7)</td>
<td>-1.5 (0.2)</td>
</tr>
</tbody>
</table>

Table 3. Stage-adjusted survival based on PRO domain score (Hazard Ratio corresponds to every 5 unit increase in PRO score)

<table>
<thead>
<tr>
<th></th>
<th>Hazard Ratio (95% confidence interval)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Global Physical Health</td>
<td>0.68 (0.48 - 0.95)</td>
<td>0.03</td>
</tr>
<tr>
<td>Global Mental Health</td>
<td>0.65 (0.42 - 0.99)</td>
<td>0.05</td>
</tr>
<tr>
<td>Anxiety</td>
<td>1.36 (0.92 - 2.02)</td>
<td>0.1</td>
</tr>
<tr>
<td>Depression</td>
<td>1.46 (1.01 - 2.12)</td>
<td>0.05</td>
</tr>
<tr>
<td>Social Roles</td>
<td>0.70 (0.19 - 0.97)</td>
<td>0.03</td>
</tr>
<tr>
<td>Fatigue</td>
<td>1.32 (0.98 - 1.76)</td>
<td>0.07</td>
</tr>
<tr>
<td>Physical Function</td>
<td>0.73 (0.51 - 1.05)</td>
<td>0.09</td>
</tr>
</tbody>
</table>
HYPOVITAMINOSIS D IS PREVALENT IN PATIENTS WITH RENAL AL AMYLOIDOSIS AND ASSOCIATED WITH NON-T(11;14)

E. Muchtra1, M. Drake1, A. Dispenzieri1, M. Lacy1, F. Buadi1, D. Dingli1, S. Hayman1, P. Kapoor1, N. Leung1, M. Siddiqui1, A. Fonder1, M. Hobbs1, Y. L. Hwa1, W. Gonsalves1, R. Warsame1, T. Kourelis1, S. Russell1, J. Lust1, R. Go1, R. Kyle1, S. V. Rajkumar1, S. Kumar1, M. Gertz1

1 Mayo Clinic.

Introduction: Vitamin D deficiency is common, with 25-50% of the population having levels below the optimal range (<20 ng/mL). Aside from its role in maintaining serum calcium and skeletal homeostasis, vitamin D has been shown to play an important role in regulation of differentiation, proliferation, apoptosis, metastatic potential and angiogenesis in a variety of malignancies. Low serum 25(OH)D levels have been associated with a worse prognosis in several malignancies, including colorectal and breast cancer, chronic lymphocytic leukemia and non-Hodgkin’s lymphoma.

Methods: Stored serum samples from 90 patients with newly diagnosed AL amyloidosis were obtained for vitamin D studies, which included 25-hydroxyvitamin D [25(OH)D], 1,25-dihydroxyvitamin D [1,25(OH)2D] and vitamin D binding protein (DBP). All vitamin D measurements were made by liquid chromatography-tandem mass spectrometry. Total 25(OH)D and 1,25(OH)2D was assessed as the additive sum of the 25(OH)D2/25(OH)D3 and 1,25(OH)2D2/1,25(OH)2D3 components, respectively. Clinical data was extracted from the electronic medical records.

Results: All patients were seen between June 2004 and September 2013. Median age was 63 [IQR 57-70]. Heart, kidney, nerve, liver and gastrointestinal amyloidosis was seen in 80%, 60%, 19%, 17% and 13% of patients, respectively. The median dFLC and bone marrow plasma cells were 28 (IQR 12-84) mg/dL and 10% (IQR 5-20%), respectively. The median serum 25(OH)D level was 20 ng/mL (IQR 10-29; normal >20 ng/mL), while the median serum level of 1,25(OH)2D was 23 pg/mL (IQR 14-37; normal 16-64 pg/mL for men; 16-78 pg/mL for women). 25(OH)D levels <20 ng/mL were strongly associated with renal involvement (84% renal involvement vs 42% of renal involvement for levels ≥20 ng/dL, P<0.001; Table). Among all patients, heavy proteinuria (>5 gr/24-h) was strongly associated with low 25(OH)D, (P<0.001, Table). A weaker association was seen between 25(OH)D levels and estimated glomerular filtration rate (eGFR) (P=0.06). Seventy-eight percent of patients with 1,25(OH)2D levels below 20 pg/mL had renal involvement versus 48% in those with levels above 20 pg/mL (P=0.01). While heavy proteinuria was associated with 1,25(OH)2D (P=0.008), the association was stronger for eGFR, as no patient with an eGFR <30 ml/min/1.73 m2 had a 1,25(OH)2D level above 20 pg/mL (P<0.001). An association between vitamin D levels and low serum albumin was also seen, mainly in relation to 25(OH)D (Table). The median level of serum DBP was 58 μg/mL (IQR 36 – 120 μg/mL), significantly lower than the normal range (168-367 μg/mL). Low levels of 25(OH)D or 1,25(OH)2D were not associated with lower DBP levels. Surprisingly, patients with renal involvement had higher levels of DBP versus those without renal involvement (median 70.9 vs 48 μg/mL; P=0.03), although median levels were below normal in both groups.

FISH data was available in 47 of patients (52%). Patients with 25(OH)D levels <20 ng/dL were less likely to harbor t(11;14) versus patients with 25(OH)D levels above this threshold (26% vs 64%, respectively; P=0.0095). A similar, but less significant association was seen between 1,25(OH)2D levels at a 20 pg/mL threshold and t(11;14) (35% vs 59%, P=0.09).

Conclusions: Hypovitaminosis D is common among AL amyloidosis patients with renal involvement. In the general cohort, heavy proteinuria, reduced eGFR and low serum albumin were all associated with low circulating vitamin D levels. Although the majority of AL patients had low levels of DBP, urinary loss of DBP does not explain the low levels of vitamin D. Hypovitaminosis D is associated with non-t(11;14) disease, a finding which warrants further exploration.
<table>
<thead>
<tr>
<th></th>
<th>25(OH)D &lt;20 ng/mL</th>
<th>25(OH)D ≥20 ng/mL</th>
<th>P-value</th>
<th>1,25(OH)2D &lt;20 pg/mL</th>
<th>1,25(OH)2D ≥20 pg/mL</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Renal involvement</td>
<td>84%</td>
<td>42%</td>
<td>&lt;0.001</td>
<td>73%</td>
<td>48%</td>
<td>0.01</td>
</tr>
<tr>
<td>Heart involvement</td>
<td>74%</td>
<td>85%</td>
<td>0.2</td>
<td>86%</td>
<td>74%</td>
<td>0.13</td>
</tr>
<tr>
<td>Liver involvement</td>
<td>18%</td>
<td>13%</td>
<td>0.7</td>
<td>18%</td>
<td>15%</td>
<td>0.7</td>
</tr>
<tr>
<td>Nerve involvement</td>
<td>11%</td>
<td>25%</td>
<td>0.07</td>
<td>16%</td>
<td>22%</td>
<td>0.47</td>
</tr>
<tr>
<td>GI involvement</td>
<td>18%</td>
<td>10%</td>
<td>0.22</td>
<td>14%</td>
<td>13%</td>
<td>0.93</td>
</tr>
<tr>
<td>Proteinuria &gt;5 gr/24-h</td>
<td>59%</td>
<td>0</td>
<td>&lt;0.001</td>
<td>37%</td>
<td>13%</td>
<td>0.008</td>
</tr>
<tr>
<td>eGFR &lt;30 ml/min/1.73 m²</td>
<td>27%</td>
<td>12%</td>
<td>0.06</td>
<td>37%</td>
<td>0</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Serum albumin &lt;2.5 g/dL.</td>
<td>73%</td>
<td>10%</td>
<td>&lt;0.001</td>
<td>47%</td>
<td>27%</td>
<td>0.05</td>
</tr>
<tr>
<td>DBP &lt;100 μg/mL</td>
<td>71%</td>
<td>67%</td>
<td>0.7</td>
<td>68%</td>
<td>70%</td>
<td>0.88</td>
</tr>
<tr>
<td>t(11;14)</td>
<td>26%</td>
<td>64%</td>
<td>0.009</td>
<td>35%</td>
<td>59%</td>
<td>0.09</td>
</tr>
<tr>
<td>Chromosome 13</td>
<td>32%</td>
<td>35%</td>
<td>0.83</td>
<td>21%</td>
<td>42%</td>
<td>0.12</td>
</tr>
<tr>
<td>abnormalities</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Trisomies</td>
<td>37%</td>
<td>23%</td>
<td>0.31</td>
<td>40%</td>
<td>20%</td>
<td>0.14</td>
</tr>
</tbody>
</table>

**Abbreviations:** DBP, Vitamin D binding protein; eGFR, estimated glomerular filtration rate; GI, Gastrointestinal
EPIDEMIOLOGICAL AND SURVIVAL TRENDS IN AMYLOIDOSIS: 1987-2019

S. Ravichandran\(^1\), O. Cohen\(^1\), S. Mahmood\(^1\), S. Sachchithanantham\(^1\), C. Whelan\(^1\), M. Fontana\(^1\), A. Naharro Martinez\(^1\), J. Gillmore\(^1\), H. Lachmann\(^1\), P. Hawkins\(^1\), A. Wechalekar\(^1\)

\(^1\) Centre for Amyloidosis and Acute Phase Proteins, United Kingdom.

**Introduction:** Amyloidosis is a progressive and usually fatal disorder of protein folding. Epidemiological studies have estimated the incidence of amyloidosis at 3-9 cases per million; but, wild type ATTR amyloidosis may be substantially more common. A database maintained at the Centre for Amyloidosis and Acute Phase Proteins (NAC, UK) has captured data on all referrals from 1987 onwards. We analysed the available data to discern the trends in the epidemiology/outcomes over this period.

**Materials and Methods:** Details of all patients with a diagnosis of amyloidosis along with their amyloid type and follow up data was obtained from the NAC database. Survival data was censored on the date of last visit/patient contact. Results were analysed using SPSS 25.

**Results:** 11014 patients were diagnosed with amyloidosis in the period 1987-2019 (Table 1). The median age at diagnosis was 66 years (range 5 months to 97 years). There is a progressive increase in the number of new patients referred to the centre increasing from 158 in 1987-90 to 3317 in 2016-19. AL Amyloidosis remains the commonest form and accounts for 54% referrals. Survival in AL has gradually improved with a particularly noticeable improvement from 2005 onwards – median survival 17 months before 2005 vs. 51 months in 2011-15.

There is a marked increase in patients referred with wild type ATTR amyloidosis in the past decade and it now accounts for 25% of all cases. It has overtaken AA amyloidosis to become the second commonest cause of acquired amyloidosis. AA amyloidosis has seen a significant decline from 13% of total cases in 1987-2010 to 3% in 2016-19. There has been no significant change in survival for wt ATTR or AA over the years. Aβ2M is now rare with no cases referred after 2015.

**Conclusions:** Amyloidosis is being increasingly recognised. AL Amyloidosis is the commonest form and survival has improved over the past three decades. Wild type ATTR is now the second commonest amyloidosis. Diagnosis of AA amyloidosis is declining.

**Table 1: Baseline Characteristics**

<table>
<thead>
<tr>
<th>Amyloid Type</th>
<th>N (% total cases)</th>
<th>Median Age in Years (Range)</th>
<th>Male:Female ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>ALL types</td>
<td>11014</td>
<td>66 (0.5-97)</td>
<td>1.63:1</td>
</tr>
<tr>
<td>AL</td>
<td>6016 (54%)</td>
<td>66 (19-96)</td>
<td>1.39:1</td>
</tr>
<tr>
<td>AA</td>
<td>889 (8%)</td>
<td>54 (9-88)</td>
<td>1:1</td>
</tr>
<tr>
<td>AL Localised</td>
<td>1234 (11%)</td>
<td>62 (0.5-97)</td>
<td>1:1</td>
</tr>
<tr>
<td>ATTR-wild type</td>
<td>1412 (12%)</td>
<td>78 (50-94)</td>
<td>14:1</td>
</tr>
<tr>
<td>ATTR-hereditary</td>
<td>841 (7%)</td>
<td>69 (20-89)</td>
<td></td>
</tr>
<tr>
<td>AApo1</td>
<td>58 (0.5%)</td>
<td>48 (19-77)</td>
<td>1.4:1</td>
</tr>
<tr>
<td>AFib</td>
<td>152 (1.3%)</td>
<td>59 (38-80)</td>
<td>1.2:1</td>
</tr>
<tr>
<td>ALect2</td>
<td>40 (0.4%)</td>
<td>62 (37-75)</td>
<td>1.2:1</td>
</tr>
<tr>
<td>Aβ2M</td>
<td>94 (0.9%)</td>
<td>54 (30-78)</td>
<td>1.5:1</td>
</tr>
<tr>
<td>ALys</td>
<td>27 (0.2%)</td>
<td>41.5 (20-72)</td>
<td>0.5:1</td>
</tr>
</tbody>
</table>
PROGNOSTIC SIGNIFICANCE OF PULMONARY FUNCTION TESTS IN PATIENTS WITH SYSTEMIC AL AMYLOIDOSIS: UNDERRECOGNIZED LUNG DYSFUNCTION IS ASSOCIATED WITH WORSE OUTCOMES

E. Kastritis 1, D. Fotiou 1, G. Trakada 1, F. Theodorakakou 1, M. Magdalini Migkou 1, M. Gavriatopoulou 1, N. Kanellias 1, E. Eleutherakis-Papaikakou 1, I. Dialoupi 1, I. Ntanasis-Stathopoulos 1, M. Irini Tselegkidi 1, A. Papanikolaou 1, E. Terpos 1, M.A. Dimopoulos 1

1 Department of Clinical Therapeutics, National and Kapodistrian University of Athens.

Background: Lung involvement in patients with systemic AL amyloidosis is not very common and consensus criteria require either direct biopsy verification or typical radiographic changes. Dyspnea and other symptoms are commonly attributed to cardiac involvement and thus, lung dysfunction may go unrecognized. No prospective comprehensive evaluation of lung function has been performed in AL amyloidosis patients.

Aim: to prospectively assess lung function by performing comprehensive pulmonary function tests (PFTs) in consecutive patients with systemic AL amyloidosis.

Methods: The study included consecutive patients with systemic AL amyloidosis treated in the Department of Clinical Therapeutics (Athens, Greece). Patients with localized lung amyloidosis were excluded. PFTs were performed according standard ERS/ATS guidelines. We performed spirometry, lung volumes measurement, single-breath determination of carbon monoxide uptake in the lung corrected for hemoglobin (carbon monoxide diffusion capacity, DLCO) and maximal expiratory (Pe) and inspiratory (Pi) pressures; age, height and weight were used for the calculation of reference values. Smoking habits were recorded.

Results: the study included 120 patients. Median age was 65 years (range 44-84), 60% were males, median baseline dFLC was 162 mg/L; kidney involvement was present in 71% (median eGFR: 66 ml/min/1.73 m2), heart was involved in 77% (10%, 55%, 22% & 13% were Mayo stage 1,2, 3A and 3B respectively). By consensus criteria lung involvement was present in 4 (3%) patients; 47.5% of the patients were current or ex-smokers. Primary treatment was bortezomib-based in 88%. According to PFTs, breathing pattern was normal in 49%, restrictive in 37%, obstructive in 11% and mixed in 3%. A restrictive pattern was marginally associated with heart involvement (p=0.056). In univariate analysis, restrictive pattern was associated with worse survival than obstructive and normal (p=0.015); 1-year mortality was 42% for restrictive vs 13% for obstructive and 5% for normal breathing patterns.

Among individual indices, a DLCO <60% was associated with poor survival (7 months vs not reached, HR:3.3, p=0.001). Pe%, (maximum expiratory pressure, a simple to do and helpful indicator of muscle weakness and lung compliance), was associated with shorter survival when was <70% of the predicted (HR:5, p<0.001). Other indices were also associated with poor outcome in univariate analysis, however, a Pe%<70% was the most important prognostic factor in multivariate analysis (HR: 5.7, p<0.001) along with stage 3B (HR:5, p=0.001)

Conclusions: A restrictive breathing pattern is common in patients with systemic AL, while lung function indices are associated with prognosis independently of cardiac dysfunction. Pe% is a very strong prognostic factor, independent of Mayo stage (even stage 3B). Our results also point to the presence of unrecognized pulmonary involvement, despite the absence of typical imaging findings.
Introduction: Light chain (AL) amyloidosis is a rare plasma cell dyscrasia characterized by organ deposits of amyloid derived from misfolded immunoglobulin light chains. Diagnostic delays >1 year from initial AL signs and symptoms (AL-S/Sx) have been reported in surveys and single-center studies but not on the population-level. Patients with delayed diagnoses are more likely to have severe morbidity and early mortality.

Objectives: To detect patients earlier, understanding the patterns of AL-S/Sx and healthcare utilization preceding AL diagnosis is necessary.

Methods: Using the Optum De-Identified Clinformatics® Datamart healthcare claims data, we identified US commercially-insured adults with ≥1 inpatient or ≥2 outpatient codes (ICD-9-CM 277.30 or ICD-10-CM E85.81, E85.89, E85.9) and ≥1 multiple myeloma treatment within 730 days (01Jan01-31Dec18). We flagged 27 AL-S/Sx using prior diagnosis codes and grouped them by system affected (cardiac, nervous, renal, gastrointestinal, other). For each AL-S/Sx, prevalence and median days to AL diagnosis was estimated and compared with that of 1:3 matched population controls. Symptom sequences were explored with data mining. Healthcare use was summarized.

Results: Among 1173 AL patients, the median time from first AL-S/Sx to AL diagnosis was 2.7 years. Monoclonal gammopathy (MG) was detected before diagnosis in 33%. Except for impotence, neuralgia, and purpura, the AL population had a much higher AL-S/Sx prevalence than the general population (Table 1). The most common AL-S/Sx were edema (59%) and dyspnea (56%). Cardiac AL-S/Sx were observed in 86% of patients, including arrhythmia (51%), heart failure (37%), pleural effusion (29%), cardiomegaly (28%), and cardiomyopathy (22%). The median days to AL diagnosis ranged from 473 from first arrhythmia to 71 from first cardiomyopathy. Sixty percent of patients had renal AL-S/Sx, with proteinuria (38%) and nephrotic syndrome (18%) diagnoses appearing shortly before AL diagnosis (median 63 and 20 days before, respectively). Neurologic AL-S/Sx, such as peripheral nerve disease (44%), generally appeared earlier than cardiac or renal AL-S/Sx (median 1.5-2 years before AL). More than 1000 AL-S/Sx sequences were identified. In the most likely AL-S/Sx progression, 40% with peripheral nerve disease developed edema. Healthcare utilization was high between first AL-S/Sx and diagnosis, with 21% of patients visiting the ER 3+ times, 15% hospitalized 3+ times, and 92% visiting 5+ provider types. The most visited specialties were cardiology (67%) and hematology/oncology (43%).

Conclusion: AL patients have a lengthy prodrome, with 50% experiencing a first AL-S/Sx ≥2.7 years before AL diagnosis. AL patients had a higher disease burden than the general population, especially cardiac and renal disease. AL suspicion was low before diagnosis; only 33% had prior MG detected. The findings suggest that novel approaches to early diagnosis could significantly improve outcomes in AL.
<table>
<thead>
<tr>
<th>Symptoms</th>
<th>Symptom Group</th>
<th>n</th>
<th>%</th>
<th>%</th>
<th>SMD</th>
<th>Median Time</th>
<th>IQR</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cardiomyopathy (non-infectious)</td>
<td>Cardiac</td>
<td>262</td>
<td>22.3</td>
<td>4.0</td>
<td>0.56</td>
<td>-70.5</td>
<td>532.5</td>
</tr>
<tr>
<td>Cardiomegaly</td>
<td>Cardiac</td>
<td>331</td>
<td>28.2</td>
<td>8.4</td>
<td>0.53</td>
<td>-224.0</td>
<td>632.0</td>
</tr>
<tr>
<td>Pleural effusion</td>
<td>Cardiac</td>
<td>343</td>
<td>29.2</td>
<td>4.7</td>
<td>0.69</td>
<td>-95.0</td>
<td>352.0</td>
</tr>
<tr>
<td>Heart failure</td>
<td>Cardiac</td>
<td>428</td>
<td>36.5</td>
<td>11.0</td>
<td>0.63</td>
<td>-187.0</td>
<td>559.5</td>
</tr>
<tr>
<td>Arrhythmia</td>
<td>Cardiac</td>
<td>601</td>
<td>51.2</td>
<td>25.4</td>
<td>0.55</td>
<td>-473.0</td>
<td>1143.0</td>
</tr>
<tr>
<td>Difficulty breathing</td>
<td>Cardiac</td>
<td>654</td>
<td>55.8</td>
<td>22.1</td>
<td>0.73</td>
<td>-353.0</td>
<td>876.0</td>
</tr>
<tr>
<td>Edema or swelling</td>
<td>Cardiac</td>
<td>696</td>
<td>59.3</td>
<td>22.5</td>
<td>0.81</td>
<td>-375.0</td>
<td>929.3</td>
</tr>
<tr>
<td>Dysphagia</td>
<td>GL</td>
<td>216</td>
<td>18.4</td>
<td>8.0</td>
<td>0.31</td>
<td>-330.5</td>
<td>852.3</td>
</tr>
<tr>
<td>Diarrhea</td>
<td>GL</td>
<td>281</td>
<td>24.0</td>
<td>11.8</td>
<td>0.32</td>
<td>-431.0</td>
<td>1064.0</td>
</tr>
<tr>
<td>Constipation</td>
<td>GL</td>
<td>283</td>
<td>24.1</td>
<td>11.0</td>
<td>0.35</td>
<td>-433.0</td>
<td>1036.0</td>
</tr>
<tr>
<td>Nausea and vomiting</td>
<td>GI</td>
<td>343</td>
<td>29.2</td>
<td>4.7</td>
<td>0.69</td>
<td>-95.0</td>
<td>352.0</td>
</tr>
<tr>
<td>Hepatomegaly</td>
<td>Liver</td>
<td>59</td>
<td>5.0</td>
<td>1.1</td>
<td>0.23</td>
<td>-79.0</td>
<td>280.5</td>
</tr>
<tr>
<td>Paralysis/paresis/plegia</td>
<td>Nervous</td>
<td>75</td>
<td>6.4</td>
<td>2.8</td>
<td>0.17</td>
<td>-125.0</td>
<td>573.0</td>
</tr>
<tr>
<td>Impotence/Erectile Dysfunction</td>
<td>Nervous</td>
<td>116</td>
<td>9.9</td>
<td>7.6</td>
<td>0.08</td>
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<td>Neuralgia/fibromyalgia/pain disorder</td>
<td>Nervous</td>
<td>162</td>
<td>13.8</td>
<td>10.7</td>
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<td>-673.5</td>
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<td>Paresthesia or anesthesia</td>
<td>Nervous</td>
<td>231</td>
<td>19.7</td>
<td>9.4</td>
<td>0.29</td>
<td>-654.0</td>
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<td>Dizziness or syncope</td>
<td>Nervous</td>
<td>450</td>
<td>38.4</td>
<td>21.9</td>
<td>0.36</td>
<td>-537.0</td>
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<td>Peripheral Nerve Disease</td>
<td>Nervous</td>
<td>516</td>
<td>44.0</td>
<td>27.0</td>
<td>0.36</td>
<td>-729.0</td>
<td>1340.8</td>
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<td>Enlarged tongue</td>
<td>Other</td>
<td>8</td>
<td>0.7</td>
<td>0.0</td>
<td>1.80</td>
<td>-51.5</td>
<td>247.3</td>
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<td>Purpura</td>
<td>Other</td>
<td>24</td>
<td>2.0</td>
<td>1.2</td>
<td>0.07</td>
<td>-466.0</td>
<td>1032.0</td>
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<tr>
<td>Malaise or fatigue</td>
<td>Other</td>
<td>622</td>
<td>53.0</td>
<td>29.7</td>
<td>0.49</td>
<td>-725.0</td>
<td>1491.8</td>
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<tr>
<td>Abdominal Pain</td>
<td>Other</td>
<td>623</td>
<td>53.1</td>
<td>30.3</td>
<td>0.47</td>
<td>-536.0</td>
<td>1286.0</td>
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<tr>
<td>Polyclonal gammopathy</td>
<td>Protein marker</td>
<td>15</td>
<td>1.3</td>
<td>0.1</td>
<td>0.15</td>
<td>-281.0</td>
<td>859.0</td>
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<td>Monoclonal gammopathy</td>
<td>Protein marker</td>
<td>386</td>
<td>32.9</td>
<td>0.5</td>
<td>0.96</td>
<td>-78.5</td>
<td>441.3</td>
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<td>Nephrotic syndrome</td>
<td>Renal/ureter</td>
<td>206</td>
<td>17.6</td>
<td>0.3</td>
<td>0.63</td>
<td>-20.0</td>
<td>55.0</td>
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<tr>
<td>Proteinuria</td>
<td>Renal/ureter</td>
<td>447</td>
<td>38.1</td>
<td>2.8</td>
<td>0.97</td>
<td>-63.0</td>
<td>282.5</td>
</tr>
<tr>
<td>Chronic kidney disease or renal failure</td>
<td>Renal/ureter</td>
<td>559</td>
<td>47.7</td>
<td>13.6</td>
<td>0.80</td>
<td>-197.0</td>
<td>631.0</td>
</tr>
</tbody>
</table>

**Table 1.** Prevalence of AL amyloidosis signs and symptoms (Al-S/Sx) prior to AL diagnosis in comparison with 1:3 matched population controls and median days from the first Al-S/Sx diagnosis to AL diagnosis

SMD=standardized mean difference in the proportions; IQR=interquartile range

**Keywords:** prodrome, early diagnosis, AL amyloidosis

**Category:** Diagnosis and prognosis of AL amyloidosis
THE IMPACT OF DELAYED DIAGNOSIS OF LIGHT CHAIN AMYLOIDOSIS ON THE CARDIAC MAYO RISK SCORE AND SURVIVAL

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Introduction: Light-chain (AL) amyloidosis is a rare disease characterized by extracellular deposition of misfolded proteins in various organs. The diagnosis is challenging due to the rapid progression of organ manifestations and mostly the unawareness of physicians. The prognosis of patients with AL amyloidosis is mostly dependent on the cardiac manifestations expressed as the Mayo stage.

Objectives: The aim of this study was to analyze the impact of time from first cardiac symptoms to diagnosis of AL amyloidosis on Mayo stage and prognosis of the disease in patients with cardiac involvement.

Methods: We gathered retrospective data from 74 patients who were histologically diagnosed with light-chain amyloidosis between 2010 and 2019 and presented in the special consultation for amyloidosis in university hospital of Essen, Germany. For 64 patients detailed medical history was available with a special focus on the timeline of symptoms, 10 patients were excluded because of insufficient records. The cardiac involvement was assessed using the modified Mayo risk score from 2004 and survival data. In our analysis we included clinical data assessing the hematological response to induction therapy, cardiac response and survival.

Results: The study included 64 patients with light-chain amyloidosis with cardiac involvement. The median age was 66 years with a greater number of males 44 (68.8 %) than females 20 (31.3%). The underlying disease was monoclonal gammopathy MGUS in 31 (49.2%), smoldering myeloma in 22 (34.9%) and multiple myeloma in 10 (15.9%) patients. The amyloidogenic light-chain was in 50 cases lambda (78.1%). We show that the longer the time from first cardiac symptoms to diagnosis the worse the Mayo stage at beginning of the therapy. Further, we show that patients with worse Mayo stage have poorer hematological response rates to anti-plasma cell therapy. This translates to diminished cardiac response and to significantly reduced survival. These data indicate that delayed diagnosis directly affects patient’s survival.

Conclusion: Clinical experience proposes that delay in the diagnosis of AL amyloidosis with cardiac involvement worsens prognosis. However, according to our knowledge, this has not been so far studied systematically. Although with a small cohort, this is the first study that shows that time from first cardiac symptoms to diagnosis of the disease measurably affects outcome and survival of patients significantly.

Keywords: AL amyloidosis, survival, prognosis, Mayo-Stage, cardiac response, hematological response.
PT018

COAGULATION ABNORMALITIES IN LIGHT CHAIN AMYLOIDOSIS

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Background: Coagulopathy is a well-recognized manifestation of light chain (AL) amyloidosis, but is not well-studied. Strategies to correct coagulation abnormalities are mainly supportive, with modest and transient effects. There are few reports on improvement of factor (F)X deficiency with high dose chemotherapy followed by stem cell transplant (SCT).

Objectives: We conducted this study to assess the prevalence of coagulation abnormalities in patients with AL amyloidosis, their association with disease-related characteristics, and the effect of treatment.

Methods: This is a retrospective study using a preexisting database at Mayo Clinic, including patients diagnosed with AL amyloidosis from 2006 to 2016. Baseline disease characteristics were compared between patients with normal and abnormal coagulation tests using Fisher’s exact test. The relationship between an abnormal parameter and survival outcomes was assessed using univariate cox proportional hazards regression. Multivariate analysis was performed including coagulation parameters and disease characteristics significantly associated with PFS and OS. We also evaluated the association between improvements in coagulation parameters and response to first line treatment. P values < 0.05 were considered statistically significant.

Results: 411 patients were included. The median age was 63.2 (32-89) years. 267 (65%) were males. Abnormalities at diagnosis included prolonged clotting times and clotting factor deficiencies (Table 1). FX deficiency was associated with higher Mayo stage, >1 organ involvement, liver involvement, and > 10% bone marrow plasma cells. On univariate analysis, the hazard risk for disease progression or death was higher in patients with abnormal values for INR, aPTT, Factors V, VII and X, compared to those with normal values (table 1). INR was an independent predictor for disease progression or death after adjusting for Mayo Stage and > 1 organ involvement (PFS HR: 1.52 CI:1.08, 2.14 and OS HR: 1.53 CI:1.14, 2.05). 159 patients had repeat testing after treatment, with a median time of 3.4 (1.2-11.6) months from diagnosis. Twenty of 30 patients with prolonged INR had improvement, including 15 with normalization. Ten of 15 with abnormal FX had improvement, including 4 with normalization. Six of 8 had improvement in TT, including 3 with normalization. All 3 patients with abnormal aPTT had normalization. There was no association between hematologic or organ responses and improvement for any coagulation test.

Conclusion: Coagulation abnormalities occur in a significant proportion of patients with AL amyloidosis. FX deficiency is associated with advanced disease, multiorgan and liver involvement. INR is an independent predictor of disease progression and mortality. Treatment of AL amyloidosis is associated with improvement in coagulation abnormalities in a subset of patients. However, there is no clear relationship between treatment response and improvement in individual parameters.
Tables:

<table>
<thead>
<tr>
<th>Coagulation parameter</th>
<th>Normal reference range</th>
<th>Number tested</th>
<th>Abnormal N (%)</th>
<th>Median (range) of abnormal</th>
<th>PFS HR (95% CI)</th>
<th>P value</th>
<th>OS HR (95% CI)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>INR</td>
<td>0.9-1.1</td>
<td>396</td>
<td>98 (24.7)</td>
<td>1.2 (1.2-2.8)</td>
<td>1.62 (1.24-2.11)</td>
<td>0.0004</td>
<td>1.82 (1.37-2.41)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>aPTT (s)</td>
<td>28-38</td>
<td>238</td>
<td>18 (7.6)</td>
<td>42 (39-58)</td>
<td>1.72 (1.04-2.85)</td>
<td>0.0338</td>
<td>1.17 (0.63-2.17)</td>
<td>0.6176</td>
</tr>
<tr>
<td>Fibrinogen (mg/dL)</td>
<td>200-375</td>
<td>203</td>
<td>1 (0.5)</td>
<td>852.5 (255-19280)</td>
<td>1.43 (0.91-2.24)</td>
<td>0.1189</td>
<td>1.64 (0.97-2.78)</td>
<td>0.0657</td>
</tr>
<tr>
<td>D-Dimer (mg/dL)</td>
<td>0-250</td>
<td>204</td>
<td>172 (84.3)</td>
<td>592 (2-69)</td>
<td>2.48 (1.54-4.00)</td>
<td>0.0002</td>
<td>2.64 (1.62-4.29)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>SFM</td>
<td>negative</td>
<td>173</td>
<td>28 (16.2)</td>
<td>65 (29-73)</td>
<td>1.04 (0.63-1.75)</td>
<td>0.8714</td>
<td>1.21 (0.72-2.04)</td>
<td>0.4794</td>
</tr>
<tr>
<td>FII (% activity)</td>
<td>75-145</td>
<td>105</td>
<td>29 (27.6)</td>
<td>59 (26-68)</td>
<td>2.73 (1.51-4.97)</td>
<td>0.0010</td>
<td>1.96 (1.06-3.66)</td>
<td>0.0332</td>
</tr>
<tr>
<td>FV (% activity)</td>
<td>70-165</td>
<td>106</td>
<td>60 (57.1)</td>
<td>46.5 (17-63)</td>
<td>2.48 (1.54-4.00)</td>
<td>0.0002</td>
<td>2.64 (1.62-4.29)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>FVII (% activity)</td>
<td>65-180</td>
<td>105</td>
<td>60 (57.1)</td>
<td>58 (2-69)</td>
<td>1.33 (1.05-1.68)</td>
<td>0.0171</td>
<td>1.77 (1.37-2.28)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>FX (% activity)</td>
<td>70-150</td>
<td>411</td>
<td>177 (43.1)</td>
<td>58 (2-69)</td>
<td>1.49 (1.07-2.07)</td>
<td>0.0192</td>
<td>1.54 (1.08-2.20)</td>
<td>0.0166</td>
</tr>
<tr>
<td>FX &lt; 50% activity</td>
<td>411</td>
<td>51 (12.4)</td>
<td>39 (2-69)</td>
<td>1.49 (1.07-2.07)</td>
<td>0.0192</td>
<td>1.54 (1.08-2.20)</td>
<td>0.0166</td>
<td></td>
</tr>
</tbody>
</table>

Table 1: Prevalence of abnormalities in coagulation times and coagulation factors at diagnosis of AL amyloidosis, and Relationship between abnormal coagulation parameters and PFS and OS. Abbreviations: INR: international normalized ratio, aPTT: activated partial thromboplastin time, SFM: soluble fibrin monomers, F II: Factor II, F V: Factor V, F VII: Factor VII, F X: Factor X, PFS: progression-free survival, OS: overall survival, HR: hazard ratio, CI: confidence interval

Keywords: amyloidosis, coagulation, factor X deficiency
THE AMYLOIDOSIS FORUM: A PUBLIC-PRIVATE PARTNERSHIP TO ADVANCE DRUG DEVELOPMENT IN AL-AMYLOIDOSIS

I. Lousada 1, V. Sanchorawala 2, K. Hsu 1, P. Dunnmon 3, M. Maurer 4

1 Amyloidosis Research Consortium
2 Amyloidosis Center, Boston University School of Medicine
3 United States Food and Drug Administration
4 Clinical Cardiovascular Research Laboratory for the Elderly, Columbia University Medical Center

Introduction: AL amyloidosis is a rare, multi-systemic disease epitomizing a tale of two diseases: the underlying plasma cell dyscrasia and the resulting organ dysfunction caused by light chain deposition. There are currently no approved treatments directed at organ dysfunction; a high unmet medical need persists despite advances in disease management. A public-private partnership (PPP) was recently formed between the Amyloidosis Research Consortium and the US Food and Drug Administration (FDA) to bridge scientific gaps in drug development for the treatment of AL amyloidosis.

Objective: The PPP seeks to leverage expertise and resources of all stakeholders (academia, industry, patients, and regulatory agencies) for the conduct of mutually beneficial scientific activities in the precompetitive domain to support bringing new, safe and efficacious therapies to patients with AL amyloidosis.

Methods: In the first of a series of meetings at the FDA, the inaugural Amyloidosis Forum focused on achieving a broad understanding of AL amyloidosis. The forum consisted of a series of presentations and panel discussions. The panel was comprised of 11 physicians from medical institutions in the US and Europe, 2 health outcomes professionals, and representatives from 6 divisions of the FDA and 3 pharmaceutical companies. Patients provided important perspectives on the path to diagnosis, challenges of rigorous treatment, and the burden of disease.

Results: The panel reviewed the epidemiology, pathobiology, and clinical features of AL amyloidosis. Hematological characteristics, staging systems, and response criteria were examined with clear consensus that a “deep response” to plasma cell clone directed treatments was critical to overall survival. Emphasis was placed on the heterogeneous clinical phenotypes of AL amyloidosis, including impacts on the cardiovascular, renal, neurological, and gastrointestinal systems which all substantially impact morbidity, but render challenges to clinical trial endpoint selection. FDA representatives discussed regulatory perspectives in the context of rare diseases and clinical outcomes assessments. The importance of natural history data and development of ranked analyses methodology and multi-domain responder indices were considered. The panel also highlighted the importance of health-related quality of life (HRQOL) instruments, the potential of imaging technology, and survival prediction models.

Conclusions: The Amyloidosis Forum identified challenges and opportunities in the development of new treatments to decrease mortality, improve function, and/or improve HRQOL in patients with AL amyloidosis. There is a clear need for novel trial designs and clinical endpoints to address the burden of disease and therapeutic effects, the development of which necessitates prospective natural history data in an evolving therapeutic landscape. Future forums will delve into these issues and seek to include participation from additional stakeholders.
NEUROFILAMENT LIGHT CHAIN, A PROMISING BIOMARKER FOR POLYNEUROPATHY IN SYSTEMIC AL AND ATTR AMYLOIDOSIS

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Background: Detection of polyneuropathy (PNP) in an early stage of AL and ATTRv amyloidosis is crucial for prognosis and choice of treatment. There is a clear clinical need for an easily applicable biomarker for both early detection and disease severity of PNP in systemic amyloidosis. The neurofilament light chain (NfL), a major cytoskeletal protein of neurons, is released into the blood after axonal damage. Serum NfL (sNfL) has been shown to be a promising biomarker for PNP in several diseases affecting the peripheral nervous system [1,2]. Recently, NfL appeared to be also increased in serum of ATTRv amyloidosis patients with PNP [3].

Objective: To study NfL in serum of AL amyloidosis patients with PNP and to confirm the observation that sNfL is increased in ATTRv amyloidosis patients with PNP.

Methods: In this pilot study NfL levels were assessed in serum of patients with well-defined AL and ATTRv amyloidosis and healthy controls. Patients with AL amyloidosis were divided into two groups: one group with evident symptoms of PNP (AL+PNP+) and a second group with patients diagnosed with clinically manifest amyloidosis but without symptoms and signs of PNP (AL+PNP-). All samples were collected before any treatment was started. Patients with ATTRv amyloidosis and symptoms of PNP confirmed by electromyography (EMG) were also included (ATTR+PNP+). Asymptomatic individuals with a proven pathogenic mutation in the TTR gene, negative abdominal fat biopsy and normal EMG results were included as asymptomatic carriers (TTRv-carriers). Healthy controls were age- and sex-matched for both the AL+PNP- group (HCAL) and the TTRv-carriers (HCATTR). The Single-molecule array (Simoa) platform was used to assess NfL concentrations.

Results: sNfL levels were increased both in AL+PNP+ patients (median 149, IQR 64.2-329 pg/ml, P<0.001) and in AL+PNP- patients (median 22.7, IQR 18.6-37.6 pg/ml, P<0.005) compared to the HCAL group (median 13.6, IQR 9.80-17.3 pg/ml). sNfL levels were higher in AL+PNP+ patients compared to AL+PNP- patients (P<0.005). sNfL levels were also increased in ATTR+PNP+ patients (median 66.4, IQR 18.2-138 pg/ml), compared to both the HCATTR group (median 8.80, IQR 6.50-11.4 pg/ml, P<0.0001) and the TTRv-carriers (median 6.90, IQR 4.80-11.8 pg/ml, P<0.0001). sNfL levels did not differ between the TTRv-carriers and the HCATTR group (P=0.340). In a group comprising all healthy controls (median 9.9 interquartile range (IQR) 7.9-15.0 pg/ml) sNfL levels correlated with age (r= 0.57, P=0.007).

Conclusion: NfL is increased in serum of both AL and ATTRv amyloidosis patients with PNP. Our pilot study indicates NfL to be a promising biomarker for detection of PNP in systemic amyloidosis. Larger groups and longitudinal studies are needed to confirm these findings and further investigate the value of NfL as a biomarker for early detection, for disease severity, for follow-up and for response to treatment in systemic amyloidosis.

Keywords: neurofilament light chain, systemic amyloidosis, polyneuropathy

References:
AL AMYLOIDOSIS: RANDOM OR INVOLVED ORGAN BIOPSIES?

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3CNIC, Madrid.
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Background: Light chain (AL) amyloidosis is a rare deposition disease with can affect many organs and with a variable but usually bad, prognosis. Therapy requires a quick and correct diagnosis. Accurate identification of amyloid deposition and of the amyloid subtype in tissue biopsies is mandatory. Random biopsies of easily accessible tissues such as subcutaneous fat, gingivae or rectum are usually recommended but sensitivity of this approach is low.

Aims: To present our experience with tissue biopsies performed in 100 consecutive patients diagnosed of AL amyloidosis in our center.

Methods: We have prospectively registered all tissue biopsies performed during the study period (2005-2019) in 100 consecutive patients diagnosed of AL amyloidosis at the same center. A bone marrow (BM) biopsy was performed per protocol in 94/100 cases. Decisions on biopsies were taken considering organ involvement and accessibility: skin, lymph nodes, lung or tongue biopsies were performed when lesions were seen on clinical or X-ray examinations, cardiac biopsies in the presence of increased NT-proBNP (N-terminal natriuretic peptide) levels and typical echocardiographic findings, kidney biopsies in patients with nephrotic syndrome and sural nerve biopsies in cases with polyneuropathy. Biopsies were stained with Congo Red and read under polarized light with a Texas filter. Subtyping of the amyloid was done using anti-kappa, anti-lambda, anti-TTR and anti-A antisera. Samples with equivocal results were sent for mass-spectrometry. If any biopsy was positive for AL amyloid, no further biopsies were performed unless necessary for therapeutic decisions.

Results: A total of 257 biopsies were performed during the study period:

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Bone marrow</th>
<th>Heart</th>
<th>Kidney</th>
<th>Subcutaneous fat</th>
<th>Salivary glands/stomach</th>
<th>Tongue</th>
<th>Lung</th>
<th>Sural nerve</th>
<th>Liver</th>
<th>Sking</th>
<th>Lymph node</th>
<th>Tonsil</th>
</tr>
</thead>
<tbody>
<tr>
<td>Biopsy/ Amyloid+</td>
<td>94/38 (40%)</td>
<td>54/53 (98%)</td>
<td>16/16 (100%)</td>
<td>35/10 (29%)</td>
<td>19/6 (31%)</td>
<td>9/8 (89%)</td>
<td>4/4</td>
<td>3/3</td>
<td>2/2</td>
<td>2/2</td>
<td>1/1</td>
<td>1/1</td>
</tr>
</tbody>
</table>

In this table we present the distribution of biopsies performed in our center and their profitability, since they have presented with amyloid deposits. The objective of these results is to demonstrate that the highest profitability in our center is derived from biopsies of the affected organ. In our experience, amyloid deposit was more frequently found in damaged organs than in bone marrow, subcutaneous fat or salivary glands.

Conclusion: Prognosis in AL amyloidosis is slowly improving with the use of new anti-myeloma drugs and may improve further with new monoclonal antibodies. Therapy requires an early and accurate diagnosis. We do not perform random biopsies of tissues such as fat or gingivae due to low sensitivity. In our hands biopsies of tissues or organs with clinical, analytical or radiological involvement shows higher sensitivity. A bone marrow biopsy is required for diagnosis of the neoplastic disease underlying AL amyloidosis and may show amyloid in up to 50% of the cases. Cardiac biopsy is also highly sensitive and in centers with a high degree of expertise such as ours, has no complications. Our data allow us to recommend a different approach to AL amyloidosis of what is usually published. Biopsies of clinically involved organs yields almost 100% sensitivity. Random biopsies of gingivae subcutaneous fat or rectum should be discouraged.

Keywords: organ biopsies, profitability diagnosis.
COMPARISON OF 24-HOUR URINARY PROTEIN AND SPOT PROTEIN-TO-CREATININE RATIO IN PATIENTS WITH AL AMYLOIDOSIS UNDERGOING RENAL STAGING

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¹Boston University Amyloidosis Center.

Introduction: Accurate measurement of proteinuria in newly diagnosed patients with AL amyloidosis remains a critical part of their initial evaluation to assess renal involvement and determine renal staging and prognosis. Furthermore, in established patients accurate measurement of proteinuria is required to determine renal response to anti-plasma cell therapy. Currently in clinical practice proteinuria is measured from a 24 hour urine collection, however this is time consuming and cumbersome for patients and can lead to incomplete or incorrect collections.

Objectives: This study aims to investigate the correlation between 24 hour urine protein measurement and spot urine protein-to-creatinine ratio (UPCR) from a single-voided urine sample. We also examined if replacing the 24 hour urine collection with UPCR from a single-voided urine sample would affect renal staging at diagnosis.

Methods: We retrospectively analyzed the data of 95 patients with AL amyloidosis seen at the Boston University Amyloidosis Center between July 2018 and July 2019. All patients had a UPCR measured from a single voided urine sample immediately prior to starting the 24 hour urine collection. Renal involvement in AL amyloidosis was defined as over 500 mg of proteinuria in a 24 hour period. Renal staging was defined as: stage I: estimated glomerular filtration rate (eGFR) = or > 50 ml/min/1.73 m² and < 5 g/24 h proteinuria stage II: eGFR < 50 ml/min/1.75 m² or > 5 g/24 h proteinuria stage III: eGFR < 50 ml/min/1.75 m² and > 5 g/24 h proteinuria.

Results: In our cohort based on the 24 hour urine collection 31% of the patients had stage I renal disease, 49% had stage II disease and 16% had stage III disease. Median characteristics were: age 66 years (range 43-89), creatinine 1.3 mg/dl (range 0.5-10), eGFR 50 ml/min/1.73m² (range 4.7-125) and proteinuria 3.8 g (range 0.5-22.8 g/day). The correlation between UPCR and 24 hour urine protein excretion was moderate (r=0.67). Utilizing UPCR alone versus 24 hour urine protein collection would have resulted in misclassification of 20% of our cohort (13% of stage I patients, 30% of stage II patients and 6% of stage III patients).

Conclusions: Spot urine protein-to creatinine-ratio from a single-voided urine sample demonstrates a moderate correlation with 24 hour urine collection for proteinuria; this is feasible option for patients whom have difficulty with the 24 hour urine collection and renal staging has been established in prior with a 24 hour urine collection. However in new patients with AL amyloidosis undergoing renal staging UPCR is less reliable.
OUTCOME OF AUTOLOGOUS SCT IN PATIENTS WITH ALamyloidosis in Slovenia – single center experience

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Introduction: AL amyloidosis (ALA) is often diagnosed late and organ involvement is too severe to enable treatment with autologous stem cell transplantation (ASCT). The later we consider as one of the most effective treatment modalities and in our clinical practice we treat patients up to 70 years of age.

Materials and Methods: Twenty-four patients with ALA underwent ASCT at UMC Ljubljana, Slovenia. One patient was treated in KBC Zagreb, Croatia. The ALA diagnosis was immunohistochemically confirmed in all patients. Coexisting multiple myeloma was excluded. Heart involvement was assessed by Mayo 2004 cardiac staging system. Kidney involvement was assessed by proteinuria. Other organ involvement was screened with patient’s clinical status and laboratory values in the context of ALA organ involvement screening procedure. Treatment response was assessed with IMWG criteria.

Results: In the period from January 2009 and November 2019 we performed 24 ASCT. The number of transplants were equally distributed through years. 17 pts. (71 %) were males and 7 (29 %) females. Mean age during the procedure was 55 years (within range 42 – 73y). Two patients were older than 65 years. Diagnosis was confirmed with 14 kidney biopsies, 7 biopsies of subcutaneous fat, two liver biopsies and one biopsy of rectal mucosa. Other types of amyloidosis were excluded. At the time of diagnosis hearth involvement was present in 11 patients (NT pro-BNP within range 554-6870 ng/L), 4 of them had positive troponin. Proteinuria was detected in 14 patients. Regarding patient’s clinically manifested organ involvement at diagnosis; 17 pts. (70 %) had kidney, 9 pts. (37.5%) heart, 10 pts. (42 %) autonomous nervous system, 6 pts. (25 %) peripheral polyneuropathy, 6 pts. (25 %) carpal tunnel syndrome, 5 pts. (21 %) liver, 2 pts. (8%) bladder and 2 pts. (8%) spleen involvement. Five pts. had ALA with single organ involved, mostly kidney. As induction therapy 8 pts. received solely corticosteroids, 8 pts. VCD regimen (bortezomib/cyclophosphamide/dexamethason), 2 pts. Vel-Dex regimen, 2 pts. received other therapies and 5 pts. none induction therapy. Conditioning consisted of MEL200 in 21 pts. (87%). Three pts. (13 %) received MEL140. Nine pts. (38 %) continued with maintenance therapy after ASCT, mostly bortezomib based. Due to a small size of plasma clone involved, response was difficult to assess. Regarding IMWG criteria 13 pts. (54 %) reached CR and 7 pts. (29 %) reached PR. Three pts. (13 %) had disease progression. Overall six ALA patients (25 %) undergone ASCT in our center died; one patient died during the ACST procedure, one died 6 months after ASCT, other four pts. died 2-5 y after ACST. Only half of them had high NT-proBNP values at the diagnosis (range values 36 – 6870 ng/L).

Discussion Treatment with ASCT in our center proved safe and transplant mortality rate low (4 %). 1-yr OS was 90,5 %. In the EBMRT retrospective analysis of 259 centers 1-yr OS was 91% (CI 87-96), which is comparable to our results. Based on our results treatment of ALA with ASCT proved feasible and may prolongs life in patients. The number of the patient being treated with ASCT within single transplant center is crucial for establishing low single center mortality rate.

Keywords: AL amyloidosis, autologous stem cell transplantation, treatment outcome
CLINICAL FEATURES AND OUTCOME OF 292 PATIENTS WITH LOCALIZED LIGHT CHAIN AMYLOIDOSIS: A HETEROGENEOUS DISEASE

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Background: Localized amyloid light chain (AL) amyloidosis (AL L amyloidosis) is a rare disease with a favorable prognosis (Kourelis, et al. 2017) in which amyloid fibrils are produced locally by a B cell clone at deposition site. Its natural history has been studied in one large series of patients (Mahmood, et al. 2015). However, AL L amyloidosis is a very heterogeneous disease and differences between subgroups are not yet well reported.

Objective: Evaluate clinical features and outcome of patients with AL L amyloidosis stratified by subgroups.

Methods: Retrospective data were collected from 292 well characterized patients with AL L amyloidosis evaluated at our centre between 2000 and 2019. Diagnosis was confirmed by immunohistochemical typing of amyloid precursor protein (App) on tissue biopsy and systemic AL amyloidosis was ruled out in all cases. Local progression (LocP) was defined as recurrence/worsening of symptoms and/or recurrence/size increasing of amyloidoma.

Results: Patient characteristics at first diagnosis are reported in Table 1. Interestingly, respiratory tract was the most common involved site: 62 (21%) subjects had nodular pulmonary, 51 (18%) laryngeal, 30 (10%) tracheobronchial and 12 (4%) nasopharyngeal involvement. Among patients with a monoclonal component (MC) in the serum or urine, 36 (12%) showed the same light chain (LC) isotype of the App. Sjögren syndrome was present in 19 (7%) and autoimmune thyroiditis in 18 (6%) patients. Symptoms related to amyloidosis were reported by 193 (66%) patients. Amyloid mass symptoms (i.e. obstruction) were more frequent in laryngeal (92% vs. 51%; P<0.001), bleeding in urinary tract (95% vs. 13%; P<0.001) and recurrent infections in tracheobronchial involvement (30% vs. 10%; P=0.007). Nodular pulmonary and cutaneous amyloidosis were most frequently asymptomatic (84% vs. 28%; P<0.001). AL L amyloidosis was an incidental finding in 71 (24%) cases, mainly in nodular pulmonary involvement (77% vs. 11%; P<0.001). A treatment for AL L amyloidosis was performed in 156 (53%) patients and was surgery in 128 (82%) and radiotherapy in 11 (7%) subjects. LocP occurred in 59 of 156 (38%) treated patients and 36 (23%) were further treated. Overall, the median time to LocP in treated patients was 51 months. No differences in time to LocP were seen in patients with a MC (34 vs. 56 months; P=0.846), even in those with the same LC isotype of App (44 vs. 56 months; P=0.937). After a median follow-up of 26 months, 12 (4%) patients died, all with respiratory tract involvement. No progressions to systemic AL amyloidosis were observed.

Conclusion: AL L amyloidosis is a heterogeneous disease and organ involvement is associated with particular clinical features. Interestingly, a MC and an autoimmune disorder were particularly frequent. The history of the disease is characterized by the risk of a LocP. Prognosis is generally good and the presence of a MC in serum does not seem to interfere with the local course of disease.

Keywords: localized AL amyloidosis, clinical features, outcome

Category: Diagnosis and prognosis of AL amyloidosis
Table 1. Characteristics of 292 patients with localized AL amyloidosis

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>N (%) – median (range)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex, male</td>
<td>144 (49)</td>
</tr>
<tr>
<td>Age, years</td>
<td>65 (18-94)</td>
</tr>
<tr>
<td>Localization</td>
<td></td>
</tr>
<tr>
<td>Respiratory / Urinary tract / Cutaneous and soft tissues / GI</td>
<td>155 (53) / 39 (13) / 31 (11) / 34 (12)</td>
</tr>
<tr>
<td>Eye / Lymphatic tissue / CNS / other*</td>
<td>12 (4) / 8 (2) / 9 (3) / 4 (1)</td>
</tr>
<tr>
<td>NT-proBNP, ng/L</td>
<td>83 (18-9004)</td>
</tr>
<tr>
<td>Proteinuria g/24h</td>
<td>0.1 (0.4-4.2)</td>
</tr>
<tr>
<td>eGFR, mL/min x 1.73 m²</td>
<td>89 (141-16)</td>
</tr>
<tr>
<td>Alkaline phosphatase concentration/u.r.l. ratio</td>
<td>0.6 (0.3-1.8)</td>
</tr>
<tr>
<td>Monoclonal protein**</td>
<td>60 (21)</td>
</tr>
<tr>
<td>IgG / IgA / IgM / LC</td>
<td>44 (15) / 3 (1) / 15 (5) / 6 (2)</td>
</tr>
<tr>
<td>Abnormal FLCR</td>
<td>61 (21)</td>
</tr>
<tr>
<td>MC or abnormal FLCR</td>
<td>97 (33)</td>
</tr>
<tr>
<td>Amyloid typing: LC isotype***</td>
<td></td>
</tr>
<tr>
<td>Kappa / lambda</td>
<td>73 (25) / 210 (72)</td>
</tr>
<tr>
<td>iFLC, mg/L</td>
<td>15.5 (3.2-1110)</td>
</tr>
<tr>
<td>Fat aspirate, positive</td>
<td>1/181 (&lt;1)†</td>
</tr>
<tr>
<td>Autoimmune disorders</td>
<td>63 (22)</td>
</tr>
</tbody>
</table>

*2 bone involvement (cervical and thoracic vertebra), 1 parotis, 1 perineural amyloidoma,  
°32 patients had a biclonal gammopathy and 1 a triclonal gammopathy  
†1 patient with both localized AL (GI) and AIns (fat pad)  
§In 9 patients, typing was consistent with AL but it was not possible to clearly identify the amyloidogenic LC.

CNS, central nervous system; eGFR, estimated glomerular filtration rate; FLC, free light chain; FLCR, free light chain ratio; GI, gastrointestinal, iFLC, involved free light chain; LC, light chain.
**Quantitative Immunoprecipitation of Free Light Chain Mass Spectrometry Facilitates Monoclonal Protein Assessment and Adds Clinical Value in Systemic AL Amyloidosis**

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**Introduction:** Diagnosis and response assessment in AL amyloidosis are primarily based on the presence of monoclonal immunoglobulins and free light chains (FLC) as determined by turbidimetric and electrophoretic approaches, which present inherent limitations and lack sensitivity particularly in AL where the levels are typically low. Novel mass spectrometry methods provide sensitive, accurate identification of M-proteins and may prove instrumental in the timely management of patients with low-level amyloidogenic light chain production.

**Objectives:** To assess the performance of quantitative immunoprecipitation FLC mass spectrometry (QIP-FLC-MS) at diagnosis and during monitoring of AL amyloidosis patients treated with bortezomib-based regimens.

**Methods:** We included 76 serial patients with systemic AL amyloidosis diagnosed and treated at the UK National Amyloidosis Centre. All patients had detailed baseline assessments of organ function. Baseline, +6- and +12-month serum samples were retrospectively analysed by QIP-FLC-MS. Briefly, magnetic microparticles were covalently coated with polyclonal sheep antibodies monospecific for free $\kappa$ and free $\lambda$ light chains. Microparticles were incubated with patient sera, washed and FLC eluted in monomeric form. Light chain mass spectra were acquired on a MALDI-TOF-MS system. Results were compared to serum FLC and serum and urine proteins (SPE, sIFE and uIFE).

**Results:** 52(68%) and 49(65%) patients presented with cardiac and renal involvement, respectively; 30(40%) patients presented with both. In 73 patients with complete data, baseline Freelite, SPE, sIFE and uIFE measurements identified a monoclonal protein in 66(90%), 33(45%), 55(75%) and 39(53%) patients, respectively. A panel consisting of Freelite + sIFE identified the M-component in 72(99%) patients, whereas Freelite + sIFE + uIFE detected 100% cases. QIP-FLC-MS alone identified monoclonal FLC in 100% serum samples and was 100% concordant with Freelite for typing the monoclonal FLC (14 $\kappa$, 53 $\lambda$). In 8 patients with normal FLC ratios QIP-FLC-MS identified a $\kappa/\lambda$ monoclonal FLC. Furthermore, QIP-FLC-MS identified an additional M-protein in 10 patients that was not detected by the other techniques. In addition, 14(18%) patients showed a glycosylation pattern of monoclonal FLCs at baseline by mass spectrometry. During the 1-year follow-up period, a total 49 CR were assigned at 6, 12 months, or both. QIP-FLC-MS identified serum residual disease in 40(82%) of these samples.

**Conclusion:** QIP-FLC-MS offers improved sensitivity for the detection of M-proteins at diagnosis and during monitoring and renders information on secondary clones and glycosylation status. Persistence of QIP-FLC-MS positive M-component in patients otherwise in CR may allow targeted therapy. Overall, QIP-FLC-MS demonstrates potential to be exploited as a single serum test for serial assessment of patients with AL amyloidosis.
LIGHT CHAIN GLYCOSYLATION INFLUENCES ORGAN TROPISM IN SYSTEMIC AL AMYLOIDOSIS

S. Ravichandran 1, F. A Sharpley 1, H. V Giles 2, R. Manwani 1, S. Mahmood 1, S. Sachchithanantham 1, H. Lachmann 1, J. Gillmore 1, C. Whelan 1, P. Hawkins 1, A. Wechalekar 1

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Introduction: Organ involvement is the major determinant of prognosis in AL amyloidosis, yet factors concerning amyloid tissue tropism remain poorly understood. Glycosylation can modulate protein function and impact prognosis in haematological malignancies. Light chain (LC) glycosylation is over-represented in AL patients, however monoclonal immunoglobulins are not routinely investigated for glycosylation for lack of rapid evaluation methods. Quantitative immunoprecipitation free light chain mass spectrometry (QIP-FLC-MS) provides sensitive assessment of M-proteins and can identify LC glycosylation. We hypothesised that LC glycosylation influences amyloid tissue deposition in systemic AL amyloidosis.

Objectives: To investigate patterns of monoclonal LC glycosylation by QIP-FLC-MS and their impact on organ involvement in AL amyloidosis.

Methods: We included 76 serial patients with systemic AL amyloidosis diagnosed and treated at the UK National Amyloidosis Centre. All patients had detailed baseline assessments of organ function. Baseline serum samples were retrospectively analysed by QIP-FLC-MS. Briefly, magnetic microparticles were covalently coated with polyclonal sheep antibodies monospecific for free κ and free λ LC. Microparticles were incubated with patient sera, washed and FLC eluted in monomeric form. LC mass spectra were acquired on a MALDI-TOF-MS system.

Results: QIP-FLC-MS identified monoclonal LC in 100% patients at baseline. In 14(19%) cases, the monoclonal LC showed a glycosylation pattern by mass spectrometry. Glycosylation was more prevalent in κ than λ patients (50% v 13%, respectively). Median involved FLC levels were not significantly different between patients with glycosylated and non-glycosylated LC (p=0.191). 52(68%) and 49(65%) patients presented with cardiac and renal involvement, respectively. 30(40%) patients presented with both. Other organs included spleen (n=35), liver (n=17), soft tissue (n=9), gastrointestinal tract (n=5), peripheral nervous system (n=6), bone (n=4) and lungs (n=1). Median number of organs involved was 2(1-5).

The frequency of cardiac involvement was not significantly different in patients with glycosylated v non-glycosylated LC (71% v 68%, respectively; p=0.789). By contrast, renal (43% vs 69%; p=0.061) and spleen (14% vs 53%; p=0.008) involvement was lower for patients with glycosylated forms; whereas nerve (29% v 3%; p=0.001), gastrointestinal (29% v 2%; p=0.032) and soft tissue (29% v 8%; p=0.032) involvement strongly associated with the occurrence of glycosylated LC.

Conclusion: LC glycosylation associates with differential organ tropism, potentially through distinct glycan usage. QIP-FLC-MS demonstrates potential for routine assessment of the glycosylation status of monoclonal LC in patients with AL amyloidosis.
PREVALENCE OF ADRENAL INSUFFICIENCY IN AL AMYLOIDOSIS: A PROSPECTIVE SINGLE CENTER STUDY

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3 Department of Biostatistics, Boston University School of Public Health.

Background: Primary systemic (AL) amyloidosis results from a plasma cell dyscrasia producing misfolded Ig-light chains which deposit in multiple tissues. Involvement of the adrenal glands with amyloidosis can lead to adrenal insufficiency (AI) and fatal Addisonian crisis. To assess the prevalence of AI in patients with AL amyloidosis, we prospectively evaluated adrenal function in patients referred to an amyloidosis center for treatment of their disease.

Methods: A total of 135 patients with AL amyloidosis not receiving active treatment of their disease enrolled in the study. Subjects underwent orthostatic blood pressure measurements followed by cosyntropin stimulation testing. Blood samples were collected before and 60 min after intravenous administration of 250 ug cosyntropin. AI was defined as a serum total cortisol level < 18 ug/dL after cosyntropin stimulation. Measurements of cortisol binding globulin (CBG) and serum free cortisol before and after cosyntropin were examined in a subgroup of subjects.

Results: Nineteen of 135 patients (14%) had low serum total cortisol levels by cosyntropin stimulation testing, however only 7 of these patients had orthostatic hypotension (37%). Twelve of 19 patients (63%) with low total cortisol response were normotensive on postural testing, indicating discordance between cortisol levels and clinical features typical of AI. As serum cortisol is highly protein bound, we examined serum protein levels in all participants. Seventeen of 19 patients (89%) with low total cortisol response were hypoalbuminemic (< 3 g/dL), and 14 patients (74%) had nephrotic range proteinuria (> 3.5 g/24 hr).

To determine the impact of hypoproteinemia on adrenal testing, we examined cortisol binding globulin (CBG) and serum free cortisol levels in the final 39 patients enrolled. Eight patients in this group (21%) had abnormal total cortisol levels post-cosyntropin. Low CBG levels significantly associated with subnormal post-cosyntropin total cortisol levels, hypoalbuminemia and nephrotic range proteinuria. Two of the final 39 patients (5%) exhibited low baseline total cortisol (< 4 ug/dL) and serum free cortisol levels, indicating true AI.

Conclusions: Prospective evaluation of patients with AL amyloidosis identified 19/135 cases (14%) with subnormal cosyntropin testing, however, hypoproteinemia confounded accurate interpretation of standard adrenal testing. Only 2 patients had confirmed AI by baseline and free cortisol levels. CBG and free cortisol levels help differentiate true AI from hypoproteinemic state.
**AL AMYLOIDOSIS: THE PROGNOSTIC IMPACT OF CYTOGENETIC ABNORMALITIES ON ORGAN INVOLVEMENT AND SURVIVAL**

M. Ozga¹, Q. Zhao¹, D. Benson¹, Jr., MD, PhD¹, P. Elder¹, N. Williams¹, N. Bumma¹, A. Rosko¹, M. Chaudhry¹, A. Khan¹, S. Devarakonda¹, R. Kahwash¹, A. Vallakati¹, C. Campbell¹, S. Parikh¹, S. Alamaan¹, J. Prosek¹, J. Bittengle¹, K. Pfund¹, S. LoRusso¹, M. Freimer¹, E. Redder¹, CLT. Lana¹, Y. Efebera¹, N. Sharma¹.

¹ The Ohio State University, Wexner Medical Center.

**Introduction:** Systemic light chain amyloidosis (AL) is a clonal plasma-cell neoplasm that carries a poor prognosis. Efforts are being made at developing better prognostic tools as AL is frequently diagnosed at an advanced stage. Cytogenetic analysis and its prognostic relevance have been well studied in multiple myeloma, but remain relatively unknown in AL literature.

**Objectives:** The present study aims to a) evaluate the most prevalent FISH cytogenetic abnormalities in AL patients as independent prognostic factors, b) assess the impact of cytogenetics on the survival of high-risk cardiac AL patients, and c) determine the effect of the novel monoclonal antibody, daratumumab, on the hematologic response in AL patients.

**Methods:** A retrospective chart review was performed on 140 consecutive AL patients treated, 20 of which received daratumumab. Patients were divided into subgroups based on FISH data obtained within 90 days of diagnosis. Hyperdiploidy was defined as trisomies of at least 2 chromosomal loci. Primary endpoints were progression free survival (PFS) and overall survival (OS), estimated via the Kaplan Meier method. The log-rank test and Cox proportional hazard models were used to test the equality of survival functions and further evaluate the differences between groups.

**Results:** The median age at diagnosis was 62 years (range: 33-88) and 55% were male. Chromosomal abnormalities were detected in 86 (61%) patients. Translocation t(11;14) was the most prevalent aberration (40%) followed by hyperdiploidy (35%), with hyperdiploidy associated with worsening PFS (p=0.019) and OS (p=0.032), confirmed in multivariable analysis (Figure 1A). There was also a significant relationship between several FISH abnormalities and increasing plasma cell burden (PC) (≥10%), including gain (+) 5p/5q (p=0.025), del13q (0.009), +11q (p<0.001), and hyperdiploidy (p=0.001). In addition, in patients with cardiac involvement, hyperdiploidy was associated with worsening PFS (p=0.0497) and OS (p=0.006) (Figure 1B). Moreover, the overall presence of t(11;14) did not have any prognostic impact on OS (p=0.76) or PFS (p=0.41), but upon further stratification, there was a marginal difference in PFS (p=0.09) among four cytogenetic groups (Figure 1C). t(11;14) alone also showed a worse PFS when compared to those patients with normal FISH (p=0.021). Finally, we evaluated the response to daratumumab and observed an association between +1q and a trend toward better hematologic response. 100% of patients (5/5) with +1q achieved a hematologic partial response or better versus only 60% of patients without +1q.

**Conclusion:** Our findings reveal the effect of hyperdiploidy on survival, PC tumor burden, and its importance within the cardiac AL population. The results with daratumumab in our patient subset with +1q are intriguing, but identification of the mechanism by which this mutation is abrogated merits further exploration as its use only continues to grow.

**Figure 1**
DEVELOPMENT OF A NEW HYBRID STAGING SYSTEM FOR AL AMYLOIDOSIS: INCORPORATING BNP, TROPOIN, AND DFLC IN PATIENTS WITH AL AMYLOIDOSIS

M. Ozga1, Q. Zhao1, D. Benson1, Jr., MD, PhD1, P. Elder1, N. Williams1, N. Bumma1, A. Rosko1, M. Chaudhry1, A. Khan1, S. Devarakonda1, R. Kahwash1, A. Vallakati1, C. Campbell1, S. Parikh1, S. Alamaani1, J. Prosek1, J. Bittengle1, K. Pfund1, S. LoRusso1, M. Freimer1, E. Redder1, CLT. Lana1, Y. Efebera1, N. Sharma1.

1 The Ohio State University, Wexner Medical Center.

Introduction: Immunoglobulin light chain amyloidosis (AL amyloidosis) is caused by misfolded light chains that form soluble toxic aggregates that deposit in tissues and organs, leading to organ dysfunction. AL amyloidosis most commonly affects the kidney and heart, leading often to nephrotic syndrome and cardiomyopathy, respectively. Studies have emphasized the importance of cardiac involvement to the survival of AL amyloidosis patients, leading to biomarker staging systems that reflect circulating markers of cardiac, renal, and clonal disease. NT-proBNP, cardiac markers (TnT or TnI), and the concentration of circulating amyloidogenic free light chains (FLCs) are used. However, NT-proBNP is often not readily available at many centers.

Objectives: To develop a new staging system using brain natriuretic peptide (BNP), troponin, and difference between involved and uninvolved light chains (dFLC) that will pose as a hybrid between the Boston Medical Center (BMC) staging system and the Mayo 2004/2012 systems. This will be predictive of overall survival (OS) and utilize dFLC in AL amyloidosis in the absence of NT-proBNP.

Methods: A retrospective chart review was performed on 140 consecutive AL patients treated. Four stages were developed for OSU staging based on BNP ≥ 81 pg/mL, TnI ≥ 0.10 ng/mL or TnT ≥ 0.025 ng/ml, and dFLC ≥ 18 mg/dL. Patients were classified as having stage I, II, III, or IV disease based on whether they had zero, 1, 2, or 3 of the above risk factors. Patients were also staged according to BMC, Mayo 2004, Mayo 2012 (Table 1). OSU staging system was compared with BMC and Mayo 2012 staging systems. Concordance analysis between OSU and Mayo 2012 staging systems was determined using linear weighted κ statistics and percent agreement. Primary endpoint was OS, estimated using the Kaplan Meier method.

Results: The median age at diagnosis was 62 years (range: 33-88) and 55% were male. Median number of organs involved was 2, with 49% and 65% having cardiac and kidney involvement, respectively. Our OSU staging system showed that patients with stage I disease had significantly better survival outcomes compared to stages II-IV. Furthermore, the differences between stage I vs II (p=0.012) and stage II vs III (p=0.002) were statistically significant by the OSU staging system (Figure 1A). There was also a significant difference between Mayo 2012 II and III (p=0.021) (Figure 1B). However, there was no difference between OSU stage III and IV, and Mayo 2012 stage III and IV. This correlated well with our concordance analysis which showed strong overlap between OSU and Mayo 2012 (κ = 0.7, agreement = 77.14%).

Conclusion: In summary, we have created a staging system which incorporates BNP, TnT/TnI, and dFLC, thereby creating a new, hybrid AL staging system that will have utility across different centers where NT-pro BNP is not measured.
Table 1.

<table>
<thead>
<tr>
<th>Staging System</th>
<th>NT-proBNP (ng/L)</th>
<th>Trop T or I (ug/L)</th>
<th>BNP (pg/ml)</th>
<th>dFLC (mg/dL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mayo 2004</td>
<td>≥332</td>
<td>≥0.035 (trop T) or ≥0.10 (trop I)</td>
<td>x</td>
<td>x</td>
</tr>
<tr>
<td>Mayo 2012</td>
<td>≥1800</td>
<td>≥0.025 (trop T) or ≥0.10 (trop I)</td>
<td>x</td>
<td>≥18</td>
</tr>
<tr>
<td>BMC</td>
<td>x</td>
<td>≥0.10 (trop I)</td>
<td>≥81</td>
<td>x</td>
</tr>
<tr>
<td>OSU</td>
<td>x</td>
<td>≥0.025 (trop T) or ≥0.10 (trop I)</td>
<td>≥81</td>
<td>≥18</td>
</tr>
</tbody>
</table>
SHOULD THE REPORTING OF BONE MARROW POSITIVITY FOR AMYLOID BE REVISED? A CRITICAL ASSESSMENT OF BONE MARROW BIOPSIES

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Introduction: Amyloidoses are rare but heterogeneous disorders where diagnosis is contingent upon the detection of deposits by Congo-red stain while amyloid protein typing determines the treatment options. While the criteria for the definition of organ involvement in kidney, heart, liver, nerve, gastrointestinal, lung and soft tissue have been established, the criteria for bone marrow (BM) involvement have not been conclusively addressed.

Objectives: We sought to address the reporting of BM involvement by amyloid in relation to the spatial distribution of deposits and to explore whether the location of deposits may have clinical relevance.

Methods: We examined 66 BM biopsies positive for amyloid with regard to the location and type of amyloid, the % and clonality of plasma cells, other organ involvement and relevant clinical information.

Results: In this study, out of 809 consecutive BM biopsies where Congo red stain was performed, deposits of amyloid were detected in 85 cases (10.5%). In 66/85 cases, we were able to review the spatial distribution of deposits. In 21 cases, amyloid deposits involved BM stroma while in 45 cases were non-stromal. All cases of stromal involvement were typed as AL-amyloidosis (or presumed AL) while non-stromal involvement was associated with at least 3 types of amyloidoses: AL, ATTR and AA. The initial diagnosis of amyloidosis was made in a BM specimen in 21/66 (31.8%) cases.

Plasma cells ranged from 1-80%; mean 13.4%, median 8%. In 44/66 [66.6%] specimens plasma cells were <10%, eight of which were post-therapy bone BM specimens. Twenty two of the 66 cases showed ≥10% plasma cells, among which 17 cases met the WHO criteria for plasma cell myeloma. Plasma cells were polyclonal in 8/66 (12.1%). In this study, 4 cases of AL amyloidosis were associated with near normal counts (<5% plasma cells) and polyclonal plasma cells prior to receiving treatment. The 2 cases with AA amyloidosis had 6% and 11% polyclonal plasma cells and the 2 cases with ATTR amyloidosis had 1% and 5% polyclonal plasma cells, respectively.

In 54 of 66 (81%) cases there were amyloid deposits in at least one other organ, mostly in kidneys (n=18) and abdominal fat (n=16). Other organs (n=31) included heart, gastrointestinal tract and liver, salivary gland, thyroid, lung, bone, prostate, urinary bladder, spleen, lymph node and skin. Ten patients were shown to have more than one organ involvement.

Conclusions: This study demonstrates that there is significant heterogeneity in the spatial distribution of amyloid in BM biopsy specimens with medullary, extramedullary, purely vascular or combined involvement. While stromal deposits were exclusively associated with AL, non-stromal and purely vascular deposits were seen in at least 3 types of systemic amyloidoses (AL, AA and ATTR). We discuss the reporting of BM biopsy tissue positivity for amyloid deposits and postulate that reporting the location of amyloid deposits may have clinical relevance.
ANALYSIS OF THE DIAGNOSTIC PROCESS IN PATIENTS WITH LIGHT CHAIN AMYLOIDOSIS

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Introduction: Light-chain (AL) amyloidosis is a rare disease, characterized by deposition of misfolded proteins in different organs leading to organ failure, often associated with a poor outcome. Clinical experience and prior studies from Lousada et al. (Lousada et al. 2015, Adv Ther Journal) let us suggest that due to the rarity of the disease and insidious, often unspecific initial symptoms, the diagnosis of AL amyloidosis tends to be missed or delayed.

Objectives: The purpose of this study was to analyze the diagnostic circumstances of this disorder in Germany.

Methods: We collected retrospective data from 90 patients with AL amyloidosis which were admitted between 2010 and 2019 to the special consultation for amyloidosis, Germany. A group of 49 patients filled out an 18-question survey covering the clinical history with special focus on the onset of symptoms and physician visits. Further, we gathered data from 41 patients where accurate clinical documentation was available.

Results: The median age of the cohort was 66 years with more males 63% than females 37%. The most common underlying disease was monoclonal gammopathy in 50% of the cases, smoldering myeloma in 33%, multiple myeloma in 14% and Waldenström disease in 1%. The most affected organs were the heart (82%) and the kidneys (82%). The diagnosis of AL amyloidosis was established within the first 6 months after onset the initial symptoms in 35% of the cases, within the first year in 65%, in 35% of the cases time from first symptoms to diagnosis lasted more than one year. The median number of visited physicians from begin of symptoms until correct diagnosis was 4 and the median number of different involved specialties was 3. The diagnosis was confirmed histologically in 82% of the cases by nephrologists, hematologists or cardiologists. Although that the most visited physicians after the general practitioner were the cardiologists, the diagnosis was mostly histologically confirmed by nephrologists in 44% of the cases. However, our data further show that cardiologists more often suspect amyloidosis as a possible cause as indication for biopsy, whereas the nephrologists performed the biopsies rapidly but rarely referred the amyloidosis as a possible reason for the proteinuria/kidney failure.

Conclusions: Our findings widely confirm the data published by Lousada et al. in the US-population. However, the correct diagnosis in our cohort was mostly performed by nephrologists in nearly half of the cases and only 17% of the patients were primary diagnosed by hematologists. Further, no patients obtained the correct diagnosis during the first physician visit. The delayed diagnosis of AL amyloidosis seems to be regardless of geography a persistent medical issue, which still remains one of the main challenges in AL amyloidosis. Sensitizing the physicians and raising awareness of patients may be helpful to solve this problem.

Keywords: AL amyloidosis, diagnosis, physicians, organ involvement, awareness
A NATIONAL, POPULATION-BASED REGISTER FOR SYSTEMIC AL AMYLOIDOSIS IN THE NETHERLANDS

M. Brink

Introduction/Background: Systemic AL amyloidosis is a rare disease with poor morbidity and survival, especially in patients with symptomatic heart failure. The cause of the disease is an underlying plasma cell dyscrasia in the bone marrow. Although treatment for systemic AL amyloidosis has greatly improved over the last decades, early referral and diagnosis are crucial to improve outcome even more.

At present, it is unknown what the annual incidence of systemic AL amyloidosis is in the Netherlands, what the most common symptoms are, and whether outcome improves due to advances in AL amyloidosis management over time. Therefore, all patients with systemic AL amyloidosis are, from 2017 onwards, nationally recorded in the systemic AL amyloidosis register, embedded in the Netherlands Cancer Registry (NCR).

Objectives: The objective of the systemic AL amyloidosis register is to gain insight in incidence, primary treatment and outcome in the Netherlands at population-based level.

Methods: Nationwide since 1989, the population-based NCR has a coverage of at least 95% of all malignancies in the Netherlands. All patients with systemic AL amyloidosis are reported in the population-based NCR via the Nationwide Network of Histopathology and Cytopathology and the National Registry of Hospital Discharges (i.e. inpatient and outpatient discharges). Topography and morphology are coded according to the International Classification of Diseases for Oncology (ICD-O). Items regarding diagnosis and first-line treatment are based on the itemset of the Amyloidosis Expert Centre in Pavia, Italy and are routinely recorded by trained registrars of the NCR through retrospective medical records review. Diagnostic items are type of underlying plasma cell dyscrasia, cardiac biomarkers such as ejection fraction, B-type natriuretic peptide (BNP) or N-terminal proBNP (NT-proBNP), troponins T or I, and organ involvement. Moreover, type and initiation of first-line treatment is recorded, including kidney dialysis, and, in addition, hematological and overall organ response. Last known vital status (i.e. alive, death, or emigration) is obtained through annual linkage with the Nationwide Population Registries Network that holds vital statistics on all residents in the Netherlands.

Results: In this abstract, preliminary results regarding incidence and patient characteristics as they are now, are presented. We currently registered 185 patients, diagnosed in 2017 or in 2018. Of these patients, 60% is male, the median age at diagnosis was 67 years (range, 44-86 years) and 59% of the patients had ≥10% plasma cells. Information regarding diagnostics and first-line treatment will soon be completed for all patients diagnosed in 2017 and 2018.

Conclusions: Many existing amyloidosis registers are hospital-based. Using the population-based systemic AL amyloidosis register, an important tool is available for clinicians to gain more insight in demographic and clinical data since all diagnosed patients are captured.

References:
PT033

NEXT GENERATION GENE SEQUENCING TO MEASURE MINIMAL RESIDUAL DISEASE IN PATIENTS WITH AL AMYLOIDOSIS AND LOW PLASMA CELL BURDEN: A FEASIBILITY STUDY

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1 Boston University Amyloidosis Center.

Keywords: next generation sequencing, minimal residual disease

Background: Achieving minimal residual disease (MRD) negativity by next generation sequencing (NGS) in multiple myeloma is a prognostic factor for both progression free survival and overall survival. In AL amyloidosis achieving MRD negativity by multiparametric flow cytometry (MPF) has been reported to improve progression free survival. Data regarding the potential use and prognostic implications of NGS in AL amyloidosis are limited. We sought to evaluate NGS as a method of isolating a clonal population of plasma cells among patients with systemic AL amyloidosis in this feasibility study.

Methods: Patients were eligible if they had systemic AL amyloidosis and no clinical evidence of concurrent active multiple myeloma. Feasibility was determined in an initial 10 patients and subsequently an additional 27 patients had initial clonal identification by NGS. Approximately five cc’s of peripheral blood and bone marrow aspirate were collected from each patient and processed for CD138 selection and DNA isolation/purification. De-identified samples were sent to Adaptive Biotech Inc. (Seattle, WA) for initial clonal identification using the ClonoSEQ immunoglobulin heavy chain (IGH) assay. Genomic DNA was amplified by implementing consensus primers targeting the IGH complete (IGH-VDJH) locus, IGH incomplete (IGH-DJH) locus, immunoglobulin κ locus (IGK) and immunoglobulin λ locus (IGL). The amplified product was sequenced and a clone identified based on frequency. After achievement of a CR or VGPR samples was sent for measurement of MRD.

Results: In total, 37 patient samples underwent NGS via the ClonoSEQ IGH assay method. All patients had measurable disease prior to treatment based on immunofixation electrophoresis and/or serum free light chains [Table 1]. ClonoSEQ IGH assay identified trackable clones in 31 of 37 patients (84%). Four patients had at least one trackable sequence (range: 1 to 5, median: 2) in the peripheral blood and 29 patients had at least one trackable sequence in the bone marrow aspirate (range: 1 to 7, median: 4). No correlation was seen between the detection of a clone and standard measures of plasma cell tumor burden (SIFE, SPEP, UIFE, UPEP, and sFLCs). Of the 11 patients who have achieved a CR or VGPR and undergone MRD testing, 10 have MRD positivity including 3 patients in a hematologic CR [Table 2].

Conclusion: NGS was successful in identifying an initial clone in 31 of 37 patients with systemic AL amyloidosis, four of which were detectable in the peripheral blood. To date, of those patients with follow-up specimens submitted 91% had MRD positivity, including 3 patients in a hematologic CR. NGS may provide a more sensitive manner of detecting a residual clone in a disease where fibril deposition by a residual plasma cell clone can have detrimental effect on organ outcomes. We are continuing to track MRD status in additional patients after treatment, the results of which will be available for presentation in March 2020.
### Table 1

<table>
<thead>
<tr>
<th>Patient</th>
<th>SIFE</th>
<th>SPEP (g/dL)</th>
<th>UIPEP (mg/day)</th>
<th>dFLC</th>
<th>Plasma cell % on bone marrow biopsy</th>
<th>Peripheral blood clonotype detected? (# of trackable sequences)</th>
<th>Bone marrow clonotype detected? (# of trackable sequences)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>IgG L</td>
<td>0.5</td>
<td>IgG L, neg</td>
<td>26.3</td>
<td>5-10% lambda</td>
<td>no</td>
<td>yes (4)</td>
</tr>
<tr>
<td>2</td>
<td>L</td>
<td>neg</td>
<td>L, neg</td>
<td>904.5</td>
<td>inadequate</td>
<td>no</td>
<td>yes (6)</td>
</tr>
<tr>
<td>3</td>
<td>L</td>
<td>neg</td>
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<td>1064.3</td>
<td>15-20% lambda</td>
<td>yes (1)</td>
<td>yes (7)</td>
</tr>
<tr>
<td>4</td>
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<td>0.3</td>
<td>neg</td>
<td>243</td>
<td>20% lambda</td>
<td>yes (1)</td>
<td>yes (2)</td>
</tr>
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<td>neg</td>
<td>L, 7</td>
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<td>20% lambda</td>
<td>no</td>
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<td>0.44</td>
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<td>10% lambda</td>
<td>no</td>
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</tr>
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<td>7</td>
<td>L</td>
<td>neg</td>
<td>L, neg</td>
<td>1.3</td>
<td>5-10% no predominance</td>
<td>no</td>
<td>no</td>
</tr>
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<td>IgG L, neg</td>
<td>33.6</td>
<td>20-25% lambda</td>
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<td>yes (1)</td>
</tr>
<tr>
<td>9</td>
<td>L</td>
<td>neg</td>
<td>L, neg</td>
<td>80.5</td>
<td>5-10% lambda</td>
<td>no</td>
<td>no</td>
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<tr>
<td>10</td>
<td>IgG K</td>
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<tr>
<td>12</td>
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<td>neg</td>
<td>L, 332</td>
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<tr>
<td>13</td>
<td>IgG L</td>
<td>0.94</td>
<td>L, neg</td>
<td>46</td>
<td>15-20% lambda</td>
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<tr>
<td>14</td>
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<td>81</td>
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<tr>
<td>15</td>
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<td>neg</td>
<td>52</td>
<td>no predominance</td>
<td>no</td>
<td>yes (2)</td>
</tr>
<tr>
<td>16</td>
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<td>yes (2)</td>
</tr>
<tr>
<td>17</td>
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<td>neg</td>
<td>neg</td>
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<tr>
<td>18</td>
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<td>20-40% lambda</td>
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<tr>
<td>19</td>
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<td>neg</td>
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<tr>
<td>20</td>
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<td>L, 59</td>
<td>80.3</td>
<td>30% lambda</td>
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<tr>
<td>21</td>
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<td>0.3</td>
<td>neg</td>
<td>30.3</td>
<td>5% kappa</td>
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<tr>
<td>22</td>
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<td>0.82</td>
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<td>28.9</td>
<td>5-10% lambda, 25% B cells</td>
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</tr>
<tr>
<td>23</td>
<td>IgG L</td>
<td>1.03</td>
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<td>20-25% lambda</td>
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<td>L</td>
<td>neg</td>
<td>L, 35.8</td>
<td>61.8</td>
<td>5% no predominance</td>
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<td>no</td>
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<tr>
<td>26</td>
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<td>neg</td>
<td>L, 200</td>
<td>297.6</td>
<td>15-20% lambda</td>
<td>no</td>
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<tr>
<td>27</td>
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<td>neg</td>
<td>93.1</td>
<td>10-15% kappa</td>
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<td>no</td>
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<tr>
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<td>IgG K, 220</td>
<td>22.3</td>
<td>30% kappa</td>
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<td>yes (1)</td>
</tr>
<tr>
<td>29</td>
<td>neg</td>
<td>neg</td>
<td>neg</td>
<td>72.3</td>
<td>5-10% no predominance</td>
<td>no</td>
<td>yes (1)</td>
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<tr>
<td>30</td>
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<td>neg</td>
<td>L, neg</td>
<td>157.5</td>
<td>25% lambda</td>
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<tr>
<td>31</td>
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<td>neg</td>
<td>L, 77</td>
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<td>30-40% lambda</td>
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<tr>
<td>32</td>
<td>IgG L,</td>
<td>0.1</td>
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<td>153.9</td>
<td>5-10% no predominance</td>
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<td>no</td>
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<tr>
<td>33</td>
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<td>IgG K, neg</td>
<td>82.4</td>
<td>25% kappa</td>
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<tr>
<td>34</td>
<td>neg</td>
<td>neg</td>
<td>L, 99.8</td>
<td>146.2</td>
<td>20% lambda</td>
<td>no</td>
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<tr>
<td>35</td>
<td>neg</td>
<td>neg</td>
<td>L, 49.2</td>
<td>5% no predominance</td>
<td>no</td>
<td>no</td>
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<tr>
<td>36</td>
<td>IgG K</td>
<td>neg</td>
<td>L, 216.9</td>
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<td>10-15% lambda</td>
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</tr>
<tr>
<td>37</td>
<td>IgG K</td>
<td>l</td>
<td>neg, neg</td>
<td>170.1</td>
<td>10-15% lambda</td>
<td>no</td>
<td>yes (4)</td>
</tr>
</tbody>
</table>

### Table 2

<table>
<thead>
<tr>
<th>Patient</th>
<th>Hematologic status at follow-up (abnormal hematologic parameters listed)</th>
<th>Trackable clonotype on follow-up sample?</th>
<th>Peripheral blood clonotype detected at follow-up?</th>
<th>Bone marrow clonotype detected at follow-up?</th>
</tr>
</thead>
<tbody>
<tr>
<td>3</td>
<td>VGPR (+SIFE)</td>
<td>yes</td>
<td>yes</td>
<td>yes</td>
</tr>
<tr>
<td>4</td>
<td>VGPR (+SIFE, 5% lambda plasma cells in marrow)</td>
<td>yes</td>
<td>yes</td>
<td>yes</td>
</tr>
<tr>
<td>5</td>
<td>VGPR (+UIFE)</td>
<td>no</td>
<td>no</td>
<td>no</td>
</tr>
<tr>
<td>8</td>
<td>CR</td>
<td>yes</td>
<td>no</td>
<td>yes</td>
</tr>
<tr>
<td>10</td>
<td>VGPR (+SIFE)</td>
<td>yes</td>
<td>yes</td>
<td>yes</td>
</tr>
<tr>
<td>11</td>
<td>VGPR (+SIFE, 5-10% lambda plasma cells in marrow)</td>
<td>yes</td>
<td>yes</td>
<td>yes</td>
</tr>
<tr>
<td>12</td>
<td>CR</td>
<td>yes</td>
<td>no</td>
<td>yes</td>
</tr>
<tr>
<td>13</td>
<td>VGPR (+SIFE, +UIFE, 15% lambda plasma cells in marrow)</td>
<td>yes</td>
<td>no</td>
<td>yes</td>
</tr>
<tr>
<td>16</td>
<td>VGPR (+SIFE)</td>
<td>yes</td>
<td>no</td>
<td>yes</td>
</tr>
<tr>
<td>28</td>
<td>VGPR (+SIFE, 5% kappa plasma cells in marrow)</td>
<td>yes</td>
<td>no</td>
<td>yes</td>
</tr>
<tr>
<td>30</td>
<td>CR (5% lambda plasma cells in marrow)</td>
<td>yes</td>
<td>no</td>
<td>yes</td>
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</tbody>
</table>
QUANTIFICATION OF CARDIAC [18F]FLUTEMETAMOL UPTAKE IN PATIENTS WITH HEREDITARY TRANSTHYRETIN AMYLOIDOSIS

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Background: Hereditary transthyretin amyloid (ATTRv) is a systemic amyloidosis with mainly neurological and cardiac symptoms. Cardiac DPD-scintigraphy is an accurate method to detect presence of amyloid in patients with amyloid cardiomyopathy and the type A fibrils (full-length and fragmented ATTR). Patients with ATTRv amyloidosis with mainly neurological symptoms and type B fibrils (only full-length ATTR) usually present with a negative DPD-scintigraphy.

Objective: The aim of this pilot study was to evaluate the outcome of [18F]Flutemetamol PET/CT-scan of the heart in long-term survivors with ATTRv amyloidosis.

Patients and Methods: Twenty-one patients with ATTRv amyloidosis and predominantly neurological symptoms, mainly negative on cardiac 99mtechnetium-3,3-diphosphono-1,2-propanodicarboxylic acid (DPD)-scintigraphy, were examined with a dynamic [18F]Flutemetamol PET/CT-scan. Five patients suffering from Alzheimer’s disease and one healthy individual served as controls. Volumes of interests were drawn over the intraventricular septum, lateral wall of the left ventricle and free wall of the right ventricle. Clinical records were reviewed for data from previous completed DPD-scintigraphy of the heart and echocardiography.

Results: Patients with ATTRv amyloidosis had a higher cardiac uptake than the control-group in all analyzed regions of the heart (p < 0.01) and could be identified with a high accuracy (sensitivity 88%, specificity 100%) in static images acquired at 30 or 60 minutes. We found no correlation between cardiac [18F]Flutemetamol uptake and clinical variables. Conclusion: [18F]Flutemetamol PET/CT of the heart can, with high sensitivity and specificity, differentiate between healthy individuals and patients with ATTRv amyloidosis. A following up with [18F]Flutemetamol PET/CT Imaging of amyloidosis in patients with a negative DPD-scintigraphy might be useful in future diagnostics.

Keywords: ATTRv, Flutemetamol, PET
NEXT GENERATION FLOW CYTOMETRY FOR EVALUATION OF MINIMAL RESIDUAL DISEASE IN PATIENTS WITH AL AMYLOIDOSIS

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¹ National and Kapodistrian University of Athens.

Introduction: Complete hematologic response (hemCR) is the goal of therapy in patients with AL amyloidosis in order to improve organ function and overall survival; however, hematologic relapses still occur and organ function may continue to deteriorate even among those in hemCR due to small residual clones that produce toxic light chains, undetectable by conventional techniques. Next generation flow cytometry (NGF) is a very sensitive method to detect the presence minimal residual disease (MRD).

Aim: to evaluate the presence of MRD by NGF in patients with AL at hemCR and its clinical implications.

Methods: We evaluated MRD in 41 patients with AL amyloidosis in hemCR. MRD was assessed in BM samples according to the Euroflow guidelines, using two independent 8-color panels, both containing CD19-PC7, CD27-BV510, CD38-FITC, CD45-PerCPCy5.5, CD56-PE, CD138-BV421, and additionally CD117-APC and CD81-APC only in the surface tube or Cyligκ-APC and Cyligλ-APCC750 only in the intracytoplasmic tube. A median number of 5 million events (range 3.9x10⁶-6.1x10⁶) were acquired for each tube in a BD FACSCantoII cytometer and data analysis was conducted with Infinicyt software. Median sensitivity level was 2.3x10⁻⁶ (range 2x10⁻⁶-3.1x10⁻⁶).

Results: The median age of the patients at diagnosis was 60 years (range 42-75), 82% had λ-light chain, 74% had renal, 18% liver and 46% cardiac involvement; 32% were Mayo stage-1, 50% stage-2 and 18% stage-3. At diagnosis median dFLC was 128 mg/L (range 17-879) and median BM infiltration by clonal plasma cells was 9%. Primary treatment was bortezomib-based in 88% and MDex in 12%, while 17.5% received ASCT. MRD evaluation was feasible in 40/41 (97.5%) tested patients and was not detectable in 18/40 (45%) (MRDneg). An organ response occurred in 72.5% of patients: in 78% of those tested MRDneg vs 67% in MRDpos. Among those with kidney involvement, 20/25 (80%) achieved a renal response, in 11/13 (85%) of MRDneg vs in 9/11 (82%) of MRDpos patients. Among those with cardiac involvement, 13/18 (72%) had a cardiac response, 6/8 (75%) of MRDneg vs 7/10 (70%) of MRDpos. Median time to organ response was similar (p=0.65) and there was no difference in the lower level of proteinuria or NTproBNP among MRDneg vs MRDpos patients that achieved organ responses. After a median follow up of 24 months (range 2-42) post MRD testing, an organ progression (renal progression in all cases) occurred in 1/18 of MRDneg patients vs in 3/22 of MRDpos patients (p=0.435); however, a hematologic relapse has not occurred in any MRDneg while 4/22 (18%) MRDpos patients had hematologic progression (p=0.057).

Conclusions: Among patients with AL amyloidosis in hemCR, 45% were MRD⁰⁰⁰, as assessed with high sensitivity NGF. Organ response rates are high among patients in hemCR, independently of detectable MRD, but, MRD⁰⁰⁰ patients have a significant reduction in the risk of hematologic relapse, indicating that this should be the goal of primary therapy.
IMPACT OF EARLY AND DEEP HEMATOLOGICAL RESPONSE TO FIRST LINE TREATMENT IN AL AMYLOIDOSIS PATIENTS

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Introduction/Background: AL amyloidosis is a rare plasma cell related disease associated with organs damage. High disease burden correlates with advanced disease stages and risk of early death, especially for patients with heart involvement. First line treatments aim to rapidly reduce FLC levels, thus preventing organ damage progression. End treatment deep hematological response (VGPR or better) correlates with better OS and PFS in these patients, but little is known about the impact of an early achievement of this status.

Methods: We retrospectively searched our institute database for non-IgM AL amyloidosis patients with complete baseline characteristics. Those with available hematological assessment after first and second cycle of treatment were included in the analysis, despite risk category, organ involvement and first line treatment choice. Hematological response was assessed according to previous published criteria (Comenzo et al 2012). Kaplan Meier survival analysis and regression cox hazard model were developed to address impact of early deep hematological response on OS.

Results: Forty-nine patients (30 male and 19 female) referred to our center between 2008 and 2018 were included in the analysis. Median age at diagnosis was 64y (44-83), 22 were transplant eligible while 14 had reversible exclusion criteria. Involved light chain was k in 9 (19%) and λ in 40 (81%) patients, associated to heavy chain (21, 63%) or not (18, 37%). Median number of organ involved was 1 (1-3) with heart (75,5%) being the most frequent. According to MAYO 2004 staging criteria 31 patients were low/intermediate (11 stage I, 20 stage II) while 18 patients were high risk (15 stage IIIa, 3 stage IIIb). All patients received bortezomib based induction regimen (5 BMDex, 29 CVD, 6 MelDex, 6 VD, 3 VTD). Median follow-up time was 20 months (4 – 132). After first cycle 11 patients (22%) reached a deep hematological response, while 19 (38%) after second cycle. Patients who achieved at least a VGPR after second cycle of therapy have a 5-year overall survival of 88,5% compared to 14,8% (log-rank p < 0.001), while hematological response after first cycle was non-predictive of long-term survival. Cox regression model confirm that hematological status after two courses of treatment was an independent predictor of long-term survival (TAB.1).

Conclusions: our study showed that an early and deep hematological response (VGPR and better) strongly correlates with improved OS. A rapidly decrease of FLC appears to offer an advantage even in advanced risk patients. Despite the retrospective nature of this study and the limited number of cases involved, this analysis suggested that those without a deep hematological response after the second cycle of treatment should be considered as very high-risk patients with urgent need of salvage treatment. These data need to be confirmed in a larger cohort of patients in order to develop a modern and dynamic treatment algorithm.
**TAB.1** Cox regression model on OS.

<table>
<thead>
<tr>
<th>Predictor</th>
<th>p</th>
<th>95% CI</th>
<th>Hazard Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>age</td>
<td>0.046</td>
<td>(1 - 1.1)</td>
<td></td>
</tr>
<tr>
<td>NTproBNP (pg/L)</td>
<td>0.143</td>
<td>(0.99 - 1.003)</td>
<td></td>
</tr>
<tr>
<td>troponin</td>
<td>0.593</td>
<td>(0.043 - 5.99)</td>
<td></td>
</tr>
<tr>
<td>cardiac involvement</td>
<td>0.757</td>
<td>(0.36 - 4.04)</td>
<td></td>
</tr>
<tr>
<td>renal involvement</td>
<td>0.782</td>
<td>(0.27 - 2.67)</td>
<td></td>
</tr>
<tr>
<td>deep response (VGPR or better) after first cycle</td>
<td>0.87</td>
<td>(0.072 - 22.041)</td>
<td>0.863</td>
</tr>
<tr>
<td>any response (PR or better) after first cycle</td>
<td>0.095</td>
<td>(0.121 - 1.186)</td>
<td>0.092</td>
</tr>
<tr>
<td>deep response (VGPR or better) after second cycle</td>
<td>0.039</td>
<td>(0.010 - 0.888)</td>
<td>0.028**</td>
</tr>
</tbody>
</table>
COMBINED SUBCUTANEOUS FAT ASPIRATE AND SKIN TRU-CUT BIOPSY FOR AMYLOID SCREENING IN PATIENTS WITH SUSPECTED SYSTEMIC AMYLOIDOSIS


1 Odense University Hospital

Conflicts of Interest: The authors report no conflicts of interest.

Background: Screening for systemic amyloidosis is typically done by fat aspirates from the abdominal wall with reported sensitivities of about 70-80% for identifying AL amyloidosis. The reported sensitivity of fat aspirates in ATTR is lower. Fat aspirates are preferred as primary screening instead of organ biopsies because they are less invasive and thereby reduce the potential risk of organ bleeding and other complications when performing organ biopsies.

Objectives: At Odense Amyloidosis Centre we performed a prospective study of whether the combined use of fat aspirates and a simultaneously performed skin tru-cut (2 mm core diameter) biopsy could increase the diagnostic sensitivity.

Methods: Both fat aspirates and skin biopsies were screened with Congo Red staining, and positive biopsies were subsequently subtyped using immunoelectron microscopy (IEM) and mass spectrometry (MS).

Results: Seventy-seven patients were screened for systemic amyloidosis with paired fat aspiration and skin tru-cut biopsy. Twenty-four patients had systemic amyloidosis (2 AL-kappa, 9 AL-lambda, 12 wtATTR, 1 AA), and 6 patients had localized amyloidosis. Thus, in 46 patients the diagnostic work-up did not reveal an amyloid diagnosis. Of 24 patients with systemic amyloidosis, the combined use of fat aspirate and skin biopsy were Congo Red positive in 15 patients (overall sensitivity, 62.5%), and further diagnostic work-up by IEM and MS revealed the subtype. Of the 15 positive fat aspirates and skin biopsies, the fat aspirates were Congo Red positive in 14 patients (overall sensitivity, 58.3%), and the skin biopsies were positive in 5 patients (overall sensitivity, 20.8%). Thus, in only one patient the skin biopsy added extra diagnostic information because the fat aspirate in this patient was negative. This patient was diagnosed with AL-kappa.

The sensitivity of fat aspiration and skin biopsies was different for AL and ATTR. Of the 11 patients who turned out to have AL amyloidosis, the combined use of both fat aspirate and skin biopsy had a sensitivity of 81.8%; fat aspirate a sensitivity of 72.7%, and skin biopsy a sensitivity of 36.4%. Out of the 12 patients with ATTR, the fat aspirate had a sensitivity of 41.7%, whereas none of the skin biopsies identified amyloid. In the patient with AA, both fat aspirate and skin biopsy were positive.

Conclusion: The combined use of fat aspirate and skin biopsy for amyloid screening showed an overall sensitivity of 62.5% with a higher 81.8 % sensitivity in AL patients and lower 41.7% sensitivity in ATTR. The skin biopsy added diagnostic information in only one of 30 patients that were diagnosed with local or systemic amyloidosis.

Key words: Amyloid screening; fat aspirates; skin biopsies
TRANSDISCIPLINARITY IN THE APPROACH TO AMYLOIDOSIS

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D. Kohan¹, M. Valeo Chulbi¹, A. Alfonsin¹

¹Hospital Italiano de Buenos Aires.

Introduction: The growing complexity in diseases approach, challenges the development of a new care paradigm that crosses disciplinary boundaries. Transdiscipline represents a form of professional practice in which professionals from different disciplines address a problem by applying their expertise, respecting the knowledge of the other and contributing to the global view of the problem.

Amyloidosis is a complex pathology that merits this transdisciplinary approach. At the Hospital Italiano de Buenos Aires (HIBA), the Grupo de Estudio de Amiloidosis (GEA) was created in 2010 and, adopting the transdisciplinarity trend, in August 2018, summoned the HIBA Dentistry Unit to incorporate minor salivary gland biopsies (MSG) as a diagnostic tool.

Objectives: Describe the first transdisciplinary experiences of MSG biopsies as a diagnostic tool in the detection of amyloidosis and the intra and postoperative complications of the procedure.

Methods: Patients referred to dentistry by the GEA at the HIBA were included. Period: August 2018-December 2019. Dental procedures: Preparation of dental records, take of informed consent, biopsy: internal mucosal surgical marking, lower lip, decontamination, infiltrative anesthesia, incision, MSG biopsy taking and placement in formalin. Processing by the Pathology Department of HIBA: Congo red and thioflavin stain. Following week: Patient postoperative dental control. Micro and macroscopic results are shared and discussed with the GEA.

Results: 16 biopsies were performed with minimally invasive procedures. There were no intra or postoperative complications.

Conclusions: The first experiences of MSG biopsy as a diagnostic tool of amyloidosis and its complications were described. Due to their absence and minimal aggression to the patient, the GEA decided to continue working in a transdisciplinary way together with the HIBA Dentistry Unit.
PT039

PREDICTIVE FACTORS INFLUENCING OVERALL OUTCOME OF AL PATIENTS

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² Fundeni Clinical Institute, Gastroenterology Department.
³ Emergency Institute for Cardiovascular Diseases, Prof. Dr. C.C. Iliescu.

Background: It is well known that cardiac AL involvement is associated with the worst outcome. We have identified that a cut-off level of liver stiffness (LS) over 10.8kPa is suggestive of multiple organs AL deposits. There is increasing recognition that NLR (neutrophil to lymphocyte ratio) and PLR (platelet to lymphocyte ratio) are markers of systemic inflammation and the elevated values of them might lead to a poor prognosis in various solid tumors.

Aim: To evaluate factors influencing overall survival of patients with AL.

Methods: 72 patients with AL diagnosed, treated and followed-up in the Hematology Department during the time period 2005-2019, have been included in our analysis. Survival rates were calculated using Kaplan-Meier method and compared using log-rank test. Univariate and multivariate Cox proportional hazards analysis were performed to identify factors associated with the worse outcome.

Results: Median survival of patients with AL was 14 months; 6-month, 1 and 3-year mortality were 35.7%, 42.9% and 64.2% respectively. 6-month survival rate was significantly lower in patients with AL and baseline LS>10.8kPa (45.7% vs 82.8%, p=0.01); as well as in AL patients with NLR>2 (56.6% vs 87.5%, p=0.009) and interventricular septum (IVS) >14mm (44% vs 72.7%, p=0.003). PLR>150 did not influenced survival. Significant predictive factors for death of patients with AL were: alkaline phosphatase level, LS value at diagnosis, NLR and PLR values, IVS thickness and administration of chemotherapy. Independent risk factor for death in AL patients was the thickness of the IVS (p=0.03), while administration of chemotherapy influenced positive the overall survival (p=0.0003).

Conclusions: Cardiac, but also liver involvement, as well as easy to use inflammatory markers negatively affects overall survival of AL patients. However, administration of chemotherapy improves their survival.
SCINTIGRAPHY SCAN WITH PLANAR AND SPECT IMAGING OF THE CHEST USING TECHNETIUM 99M PYROPHOSPHATE (740MBQ) FOR THE ASSESSMENT OF AL AMYLOIDOSIS

V. H. Jimenez\(^1\), D. Chan\(^1\), H. Lee\(^1\), N. Fine\(^1\), S. McCulloch\(^1\), J. Tay\(^1\), P. Duggan\(^1\), P. Neri\(^1\), N. Bahlis\(^1\)

\(^1\) Tom Baker Cancer Center

**Introduction:** AL amyloidosis (AL) is a low tumor burden plasma cell disorder characterized by deposition of insoluble fibrils composed of immunoglobulin light chains. Nuclear Scintigraphy is a non-invasive tool with diagnostic potential in the assessment of cardiac amyloidosis (CA). Various tracers have been used in the past for scintigraphic assessment of CA. Based on the above mentioned, we aimed to assess the role of Pyrophosphate (PYP) scan with planar and SPECT imaging of the chest using Technetium 99M PYP in recently diagnosed AL amyloidosis patients treated with Bortezomib-containing regimens (BCR) at our Institution.

**Objectives and methods:** Patients with newly/recently diagnosed AL amyloidosis treated at our Institution with BCR were identified. From these, patients undergoing PYP Scintigraphy for the assessment of cardiac amyloidosis were evaluated. Two-sided Fisher exact test was used to test for differences between categorical variables. A p-value of <0.05 was considered significant.

**Results:** Fifty-one consecutive patients with AL amyloidosis treated with BCR from 01/2012 to 11/2019 were identified. From these, 20 patients underwent PYP Scintigraphy assessment. Clinical characteristics are seen in Table 1. Median age at diagnosis was 68 years and 65% were male. According to the Mayo Clinic staging criteria (2012): 4 patients were classified as stage I (20%), 5 Stage II (25%), 3 Stage III (15%), and 8 Stage IV (40%). Cardiac involvement and renal involvement were the most commonly seen (70% both). PYP scintigraphy was reported as score 0 in 5 (25%), score 1 in 9 (45%), score 2 in 5 (25%) and score 3 in 1 patient (5%). Median heart to contralateral chest ratio was 1.2. Further, median NTproBNP was 204 ng/L for score 0, 1413 ng/L for score 1, 2366 ng/L for score 2 and 5318 ng/L for score 3 (p=0.031). Most of cases with PYP score 2-3 were classified as stage III and IV (83.3% vs 35.7% compared to those with PYP score 0-1, p=<0.05). With a median follow-up of 9 months, no differences on overall survival (OS) are seen among the groups with different scores in the PYP scintigraphy.

**IN CONCLUSION:** PYP scintigraphy is a useful tool to assess cardiac involvement by amyloidosis in patients with AL. Patients with advanced stage disease appeared to have a higher degree of uptake. PYP should potentially be used for the diagnosis and monitoring of AL amyloidosis. In contrast to ATTR, patients with score 2-3 are usually those with severe heart involvement and represented a minority of AL cases (score 2, 25% and score 3, 5%, respectively).

**Key Words:** Pyrophosphate, AL amyloid, Scintigraphy

**Category:** Diagnosis and Prognosis of AL amyloidosis
Table 1. Clinical Characteristics of patients with AL amyloidosis assessed by PYP Scintigraphy at our Institution.

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>N=20</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (median)</td>
<td>68</td>
</tr>
<tr>
<td>Gender</td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>13 (65%)</td>
</tr>
<tr>
<td>Female</td>
<td>7 (35%)</td>
</tr>
<tr>
<td>Hb (g/L)</td>
<td>123</td>
</tr>
<tr>
<td>Creatinine (µmol/L)</td>
<td>92.5</td>
</tr>
<tr>
<td>B2microglobulin (µmol/L)</td>
<td>2.4</td>
</tr>
<tr>
<td>Albumin (g/L)</td>
<td>32.5</td>
</tr>
<tr>
<td>Stage I</td>
<td>4 (20%)</td>
</tr>
<tr>
<td>Stage II</td>
<td>5 (25%)</td>
</tr>
<tr>
<td>Stage III</td>
<td>3 (15%)</td>
</tr>
<tr>
<td>Stage IV</td>
<td>8 (40%)</td>
</tr>
<tr>
<td>LDH (IU/L)</td>
<td>191.5</td>
</tr>
<tr>
<td>BMPC (%)</td>
<td>8%</td>
</tr>
<tr>
<td>Light chain:</td>
<td></td>
</tr>
<tr>
<td>Kappa</td>
<td>6 (30%)</td>
</tr>
<tr>
<td>Lambda</td>
<td>14 (70%)</td>
</tr>
<tr>
<td>Cardiac involvement</td>
<td>70%</td>
</tr>
<tr>
<td>Kidney involvement</td>
<td>70%</td>
</tr>
<tr>
<td>Liver involvement</td>
<td>15%</td>
</tr>
<tr>
<td>Nerve involvement</td>
<td>30%</td>
</tr>
<tr>
<td>GI involvement</td>
<td>20%</td>
</tr>
<tr>
<td>Lung involvement</td>
<td>5%</td>
</tr>
</tbody>
</table>

**BMPC:** Bone marrow plasma cells; **FLC:** Free-light chains only.
PT041

LIVING WITH AMYLOIDOSIS- PERSPECTIVES FROM PATIENTS LIVING WITH LIGHT CHAIN (AL) AMYLOIDOSIS FOR OVER A YEAR

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Introduction: Patients with light chain (AL) amyloidosis often experience challenges with diagnosis and have high symptom burden in the early period after diagnosis and initial treatment. Patient-reported symptoms and the impact on quality of life in AL amyloidosis patients who have lived for one year or longer are not well described.

Objective: To understand, from the patient’s perspective, the most important symptoms impacting daily life after overcoming the early mortality stage (first year) of AL amyloidosis.

Methods: We conducted a qualitative study using semi-structured interviews in patients with a diagnosis of AL amyloidosis identified from a U.S. cancer center and through Amyloidosis Support Groups, Inc. Subjects were sampled to represent diversity in gender and amyloid organ involvement. Interviews were audiotaped, transcribed, coded, and analyzed using NVivo software.

Results: Among 20 subjects interviewed between July to October 2019, 15 had lived with a diagnosis of systemic AL amyloidosis for longer than 1 year (range, 1.5 - 13 years). Seven patients had cardiac, 6 patients had renal, 2 patients had neurologic, and 1 patient had gastrointestinal (GI) involvement. Five patients had 3 or more organs involved, 5 were on active chemotherapy treatment, and 8 had received a prior stem cell transplant. The main patient themes that emerged regarding symptoms included: 1) lack of resolution of symptoms/symptom burden and 2) uncertainties about the symptoms they might have to face. Participants described a wide range of symptoms (Table). The most common symptoms reported were fatigue, pain, GI symptoms, swelling, and dyspnea. Several patients discussed the emotional impact of living with amyloidosis and their apprehension about their symptoms which were secondary to the disease itself as well as from the treatment thereof.

Conclusions: Our study identified the important symptoms that patients living with AL amyloidosis have after overcoming the first and critical year after diagnosis when early mortality and symptom burden is highest. These findings highlight the experience of chronic symptom burden for patients with AL amyloidosis and suggest a need for better recognition of symptoms and symptom management. Future research is needed to describe patient experience of the typical disease course after the first year to inform patient education.

Table

<table>
<thead>
<tr>
<th>System</th>
<th>Symptoms</th>
</tr>
</thead>
<tbody>
<tr>
<td>General</td>
<td>Fatigue, Weight changes, Cold intolerance, Sleep disturbance, Bruising, Speech/voice change</td>
</tr>
<tr>
<td>Cardiac</td>
<td>Swelling, Dizziness, Shortness of breath</td>
</tr>
<tr>
<td>Gastrointestinal</td>
<td>Chewing difficulties, Taste, Dysphagia, Nausea, Anorexia, Diarrhea, Constipation</td>
</tr>
<tr>
<td>Neuromuscular</td>
<td>Pain, Gait imbalance, Falls</td>
</tr>
<tr>
<td>Mental</td>
<td>Anxiety, Anger, Depression, Memory issues, Forgetfulness, Social withdrawal</td>
</tr>
<tr>
<td>Other</td>
<td>Foamy urine, Bruising, Brittle nails</td>
</tr>
</tbody>
</table>
PT042

COMPARISON BETWEEN THREE ASSAYS FOR THE MEASUREMENT OF CIRCULATING FREE LIGHT CHAIN IN 86 CONSECUTIVE NEWLY DIAGNOSED PATIENTS WITH AL AMYLOIDOSIS

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Background: The availability of assays for the measurement of circulating Free Light Chain (FLC) has been an important advance for diagnosis, prognostic stratification and response monitoring in patients with AL amyloidosis. Several assays are commercially available but there is no consensus on their respective performance in AL amyloidosis.

Methods: In this study, we assessed the diagnosis, prognosis and hematologic response sensitivity for routine use of three assays for FLC quantification in 85 consecutive newly-diagnosed patients with AL amyloidosis. Serum FLC concentrations were measured by polyclonal freelite assay from Binding Site (BS), monoclonal N-latex immunoassay from Siemens (Sie) and new polyclonal immunoassay from Sebia (Seb). Hematologic response was assessed at a median of 3 months after the beginning of treatment, based on dFLC defined as the difference between the involved amyloid-forming FLC and the non-involved. A response was defined as at least 50% decrease in dFLC level if dFLC>20mg/L. Survival was estimated by Kaplan-Meyer and risk factors were identify using cox proportional hazard model.

Results: The isotype of the monoclonal FLC was kappa in 22 patients and lambda in 63. The median value of involved FLC was lower with the Seb assay compared to the BS and Sie assay with 66.5 mg/L, 196 mg/L and 177 mg/L, respectively. The three tests are not equivalent for the quantification of absolute FLC concentration. However, they seem to have similar sensibility to detect a monoclonal light chain with 70 patients having both abnormal involved FLC and K/L ratio with the Seb assay, 68 patients with the BS assay and 72 patients with Sie assay. Additionally, 71 patients had an elevated FLC over 20 mg/L with the Seb assay, 74 patients with the BS assay and 72 patients with Sie assay. The concordance between the assays was 84 to 86% with 5 to 9% abnormalities detected by only one test and not by the others. Of note, all tested patients had the involved FLC detected as out of range with at least one assay. Hematological response was assessed at 3 months using dFLC. A median dFLC decrease of 59% with Seb assay was observed, similar to BS (61%) and Sie (62%) assays. In patients with dFLC>20mg/L, a response of at least 50% to front line therapy resulted in significant higher and similar survival rate with all assays; 82 to 88% survival was observed if dFLC<10mg/L at follow-up. Consensus criteria does not allow comparison between the three tests.

Conclusion: The three assays tested in this study were inequivalent to quantify FLC level in patient sera. Nevertheless, they all offer similar clinical performance in the management of AL amyloidosis. Considering the 6 to 10% discrepancy between the assays, parallel testing may be useful to avoid misdetection. Further studies are needed to define new response criteria.

Keywords: AL amyloidosis, free light chain quantification, management of AL amyloidosis.
ROLE OF DEXAMETHASONE IN EARLY CARDIAC MORTALITY OF LIGHT CHAIN CARDIAC AMYLOIDOSIS PATIENTS

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3 Cardiology department, Henri Mondor teaching hospital, Creteil, France.
4 Pathology department, Henri Mondor teaching hospital, Creteil, France.
5 Immunology Department, Henri Mondor Teaching Hospital, Creteil France.

Background: Light chain (AL) cardiac amyloidosis patients often die within three months of starting chemotherapy. In non IgM AL, chemotherapy frequently includes a bortezomib (Bor), cyclophosphamide (Cy), and dexamethasone (D). We reported that NT-ProBNP levels can double within 24 h of administering dexamethasone, suggesting cardio toxicity.

Objectives: To evaluate the role of dexamethasone in the early cardiovascular mortality observed while treating patients with AL cardiac amyloidosis.

Methods: Inclusion of de novo AL cardiac amyloidosis patients treated at our institute between 2009 and August 2018 with successively three different types of chemotherapy regimens: CyBorDComb (all treatments combined on day1), DCyBorSeq (sequential administration of dexamethasone initiated on day 1, cyclophosphamide on day 8, and bortezomib on day 15), and CyBorDSeq (sequential administration of cyclophosphamide initiated on day 1, bortezomib on day 8, and dexamethasone on day 15). The primary endpoint was cardiovascular mortality and cardiac transplantation at day22 and day455.

Results: 100 patients were included in the retrospective analysis; the mean age was 68 years old and mainly males (62%). In terms of cardiac severity, 56 patients were Mayo clinic stage IIIa and 33 were stage IIIb. 34 patients were treated with CyBorDComb, 17 with DCyBorSeq and 49 with CyBorDSeq. At day-22, mortality was 20.6% with CyBorDComb, 23.5% with DCyBorSeq, and 0% with CyBorDSeq(p=0.003). At day- 455, mortality was not significantly different with the regimens (p=0.195). In the multivariate cox regression, dexamethasone was significantly associate with risk of mortality: Hazard ratio was 43.04[5.93; 312.58], p<0.001).

Conclusion: Our study shows that delaying dexamethasone during the first cycle reduces the number of early deaths without extending survival as all patients received dexamethasone at the end. The efficacy/safety balance of dexamethasone for treating patients with AL cardiac amyloidosis needs further investigation.
PT044

SEEKING LIGHT-CHAIN AMYLOIDOSIS (AL) VERY EARLY: THE SAVE TRIAL – IDENTIFYING AL IN PATIENTS WITH SMOLDERING MYELOMA (SMM) OR MGUS

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2 Laboratory of Personalized Genomic Medicine, Columbia University Medical Center, New York, NY, USA.
3 Multiple Myeloma and Amyloidosis Service, Columbia University Medical Center (CUMC).

Background: In systemic AL amyloidosis (AL), the causative free light chains (FLC) are produced in 75% of cases by λ clones. Because patients present with advanced organ damage, earlier diagnosis is a critical unmet need. Nine λ IGLV germline genes account for 80% of patients with AL λ-type, including 3 genes highly associated with AL and not myeloma (MM): IGLV6-57, IGLV2-14 and IGLV1-44. Progression to AL from SMM or MGUS occurs but is not well appreciated (JCO 2014; 32:2679). A laboratory developed test (LDT) is needed to identify those at risk of AL. We also measured soluble B-cell maturation antigen (sBCMA). We hypothesized that the level of sBCMA divided by the FLC may differ in SMM and MGUS patients with evolving AL because of FLC deposition.

Methods: SAVE is a trial (NCT02741999) for λ SMM or MGUS patients with a κ:λ FLC ratio < 0.26 and dFLC > 23mg/L (JCO 2014;32:2679). Eligible patients ship peripheral blood (PB) or marrow (BM) samples to us for IGLV gene identification by RT-PCR with cDNA from CD138+ cells (Blood 2001;98:714) and for sBCMA measurements by ELISA (R&D Systems, Minneapolis, MN, USA). PCR amplicons are sequenced and the IGLV germline genes identified in IMGT (ImMunoGene-Tics, www.imgt.org). If the germline gene is AL-related, further evaluation is pursued. In addition, in an attempt to develop an LDT in a CLIA-certified facility, blinded IGLV gene identification is performed by NGS on the Illumina platform at CUMC.

Results: Twenty asymptomatic λ patients were enrolled by 6/1/2019 (3M, 17F) and 23 PB/4 BM specimens obtained. Medians of months from diagnosis, λ FLC, κ:λ ratios, MNC and CD138-selected cells were 20.5 months, 113mg/L, κ:λ 0.06, 8.1x10⁶ (0.8-24) and 3x10⁵ (0-30), respectively. Seventeen patients had IGLV genes identified by RT-PCR, 12 with the first and 5 with additional specimens. Increased risk of AL was identified in 7 patients, 2 of whom were diagnosed with AL after IGLV2-14 germline gene identification; both had SMM (diagnosed in 2009 and in 2016) and were SCT candidates. Results of attempted identification of the first 10 cases by NGS are shown in Table 1. In 6 cases for which IGLV genes were identified by both methods, including 1 patient with undiagnosed AL, agreement was 83%. sBCMA levels were a median 86.4ng/mL (15-168) and correlated with λ FLC (r=0.49, P=0.05). Ratios can be calculated.

Conclusions: The SAVE trial enables early diagnosis of AL λ-type based on the λ IGLV gene used by the clonal plasma cells. By RT-PCR in this pilot study we identified the clonal λ gene 85% of the time. Measurement of sBCMA levels was also feasible and may be useful. An NGS platform to identify IGLV genes in a CLIA-certified laboratory will extend this research effort into an LDT for widespread use. Patients with SMM and MGUS involving κ clones who meet eligibility thresholds should be studied as well. Earlier diagnosis will enable treatment with effective therapy such as MEL 200 SCT.

Table 1. Comparison of Results with NGS and RT-PCR

<table>
<thead>
<tr>
<th>Spec</th>
<th>BM/PB</th>
<th>NGS</th>
<th>% Reads Assigned</th>
<th>RT-PCR</th>
<th>CDR3 Clonotype</th>
</tr>
</thead>
<tbody>
<tr>
<td>2485</td>
<td>BM</td>
<td>IGLV3-21</td>
<td>90</td>
<td>IGLV3-21</td>
<td>Same</td>
</tr>
<tr>
<td>2453</td>
<td>PB</td>
<td>IGLV7-46</td>
<td>19</td>
<td>IGLV2-14</td>
<td>Different</td>
</tr>
<tr>
<td>2472</td>
<td>BM</td>
<td>IGLV2-14</td>
<td>87</td>
<td>IGLV2-14</td>
<td>Same</td>
</tr>
<tr>
<td>2498</td>
<td>PB</td>
<td>FAILURE</td>
<td></td>
<td>IGLV1-44</td>
<td></td>
</tr>
<tr>
<td>2563</td>
<td>PB</td>
<td>IGLV1-47</td>
<td>19</td>
<td>IGLV1-47</td>
<td>Same</td>
</tr>
<tr>
<td>2497</td>
<td>PB</td>
<td>FAILURE</td>
<td></td>
<td>IGLV1-44</td>
<td></td>
</tr>
<tr>
<td>2492</td>
<td>PB</td>
<td>IGLV2-14</td>
<td>47</td>
<td>IGLV2-14</td>
<td>Same</td>
</tr>
<tr>
<td>2570</td>
<td>PB</td>
<td>IGLV2-11</td>
<td>80</td>
<td>IGLV2-11</td>
<td>Same</td>
</tr>
<tr>
<td>2505</td>
<td>PB</td>
<td>FAILURE</td>
<td></td>
<td>IGLV1-44</td>
<td></td>
</tr>
<tr>
<td>2762</td>
<td>BM</td>
<td>IGLV3-19</td>
<td>87</td>
<td>FAILURE</td>
<td></td>
</tr>
</tbody>
</table>

AL patient identified on SAVE
CHARACTERIZATION OF AMYLOIDOGENIC CLONE IN IGM LIGHT CHAIN AMYLOIDOSIS AND ANALYSIS OF PROGNOSTIC FACTORS

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Background: IgM-AL amyloidosis is a rare condition that represents approximately 6% to 10% of all cases of systemic AL amyloidosis. It is characterized by a lymphoid clone (lymphC) in the majority of cases instead of a plasma cell clone (pcC). IgM-AL amyloidosis has been already studied and prognostic factors have been previously described (Sachchithanantham et al; 2016). However, less is known about how the clonal characteristics (pcC vs. lymphC) influence the clinical features and the outcome.

Objective: Characterization of IgM patients, the underlying clonal disease and identification of prognostic factors for survival in univariate analyses.

Methods: Retrospective data were collected from 91 patients with IgM-AL amyloidosis evaluated at our center. The data were collected at first diagnosis of amyloidosis and then during the follow-up for hematologic or organ response assessment. Clonal disease was evaluated with bone marrow cytology, FACS, histology and MYD88 PCR.

Results: A plasma cellular clone was found in 22 (24%) patients, while in 56 (62%) cases a B-cell lymphoma was present in bone marrow. In 13 (14%) subjects the underlying clonal disease could not be characterized. Patient characteristics are reported in Table 1. Interestingly almost all patients with pcC expressed lambda light chain phenotype (19/22; 86%) and in 55% of the lymphC (31/56) (P=0,01). Another observed feature was that patients with pcC had typically heart and kidney involvement, whereas patients with lymphC had more involvement of liver, nerve system and lung. MYD88 mutation was found in 73% of the patients with lymphC. Thirteen patients were treated for the clonal disease before diagnosis of amyloidosis. They were mainly cases of lymphC (12 patients). Administered therapy after diagnosis of amyloidosis was different, mostly Rituximab based in lymphC and bortezomib or autologous stem cell transplant in pcC. Overall survival (Table 2) rates from the date of first diagnosis of IgM amyloidosis did not differ considerably in lymphC and pcC. Other factors that were found prognostic in the study population are listed in Table 2 (Cardiac staging, FLC and Albumin in serum). MYD88, renal staging, age, PNS involvement, number of involved organs and level of IgM in serum exhibited no correlation with survival rates.

Summary & Conclusion: We confirm that in IgM amyloidosis the underlying clonal disease is mostly lymphatic which interestingly has influence on amyloidogenic light chain and organ involvement. However, in this study a difference in outcome between pcC and lymphC patients is not evident. Finally, we confirmed the prognostic factors that were found in IgM-amyloidosis (Milani et al; 2016).
Table 1: patient characteristics.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Total N. Pts 91</th>
<th>pC N. Pts 22</th>
<th>lymphC N. Pts 56</th>
<th>Unknown N. Pts 13</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, years</td>
<td>71 (50-90)</td>
<td>69 (53-80)</td>
<td>71 (50-90)</td>
<td>75 (58-80)</td>
</tr>
<tr>
<td>Sex, male</td>
<td>66 (70%)</td>
<td>10 (45%)</td>
<td>46 (81%)</td>
<td>11 (85%)</td>
</tr>
<tr>
<td>Organ involvement</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Heart</td>
<td>2 (1-6)</td>
<td>2 (1-6)</td>
<td>2 (1-5)</td>
<td>2 (1-6)</td>
</tr>
<tr>
<td>Kidney</td>
<td>56 (60%)</td>
<td>18 (82%)</td>
<td>32 (56%)</td>
<td>6 (46%)</td>
</tr>
<tr>
<td>Liver</td>
<td>45 (49,5%)</td>
<td>14 (64%)</td>
<td>24 (42%)</td>
<td>7 (54%)</td>
</tr>
<tr>
<td>Lung</td>
<td>5 (6,5%)</td>
<td>0 (0%)</td>
<td>3 (7%)</td>
<td>2 (15%)</td>
</tr>
<tr>
<td>GI</td>
<td>26 (28%)</td>
<td>6 (27%)</td>
<td>15 (26%)</td>
<td>5 (38%)</td>
</tr>
<tr>
<td>ST</td>
<td>45 (48%)</td>
<td>9 (41%)</td>
<td>28 (49%)</td>
<td>8 (62%)</td>
</tr>
<tr>
<td>PNS</td>
<td>20 (21,5%)</td>
<td>4 (18%)</td>
<td>14 (24,5%)</td>
<td>2 (15%)</td>
</tr>
<tr>
<td>ANS</td>
<td>14 (15%)</td>
<td>2 (9%)</td>
<td>12 (21%)</td>
<td>0 (0%)</td>
</tr>
<tr>
<td>Mayo stage</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>I</td>
<td>18 (23,4%)</td>
<td>3 (15%)</td>
<td>12 (24,5%)</td>
<td>3 (37,5%)</td>
</tr>
<tr>
<td>II</td>
<td>31 (40%)</td>
<td>9 (45%)</td>
<td>19 (39%)</td>
<td>3 (37,5%)</td>
</tr>
<tr>
<td>Illa</td>
<td>20 (26%)</td>
<td>7 (35%)</td>
<td>12 (24,5%)</td>
<td>1 (12,5%)</td>
</tr>
<tr>
<td>Illb</td>
<td>8 (10%)</td>
<td>1 (5%)</td>
<td>6 (12%)</td>
<td>1 (12,5%)</td>
</tr>
<tr>
<td>Renal Staging</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>I</td>
<td>49 (71%)</td>
<td>10 (59%)</td>
<td>34 (77%)</td>
<td>5 (63%)</td>
</tr>
<tr>
<td>II</td>
<td>14 (20%)</td>
<td>5 (29%)</td>
<td>9 (20%)</td>
<td>0 (0%)</td>
</tr>
<tr>
<td>Ill</td>
<td>6 (9%)</td>
<td>2 (12%)</td>
<td>1 (3%)</td>
<td>3 (57%)</td>
</tr>
<tr>
<td>Proteinuria, g/24h</td>
<td>1 (0-13)</td>
<td>2 (0.06-10,52)</td>
<td>0.38 (0-13,14)</td>
<td>3 (0.09-10,42)</td>
</tr>
<tr>
<td>eGFR, ml/min x 1.73 m2</td>
<td>70(9.7-137)</td>
<td>69.3(9.7-128,7)</td>
<td>72.4(12-137)</td>
<td>55.8(16-124,3)</td>
</tr>
<tr>
<td>dFLC, mg/L</td>
<td>70(0-3741)</td>
<td>61(0-1413)</td>
<td>85(0.2-3741)</td>
<td>60(8.8-409)</td>
</tr>
<tr>
<td>dFLC&lt;50 mg/L</td>
<td>34(41%)</td>
<td>9(41%)</td>
<td>18(33%)</td>
<td>5(50%)</td>
</tr>
<tr>
<td>Treatment</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HSCT</td>
<td>7(8%)</td>
<td>4(18%)</td>
<td>1(2%)</td>
<td>2(16%)</td>
</tr>
<tr>
<td>RB</td>
<td>27(30%)</td>
<td>0(0%)</td>
<td>27(48%)</td>
<td>0(0%)</td>
</tr>
<tr>
<td>RX*</td>
<td>13(14%)</td>
<td>1(5%)</td>
<td>9(16%)</td>
<td>3(23%)</td>
</tr>
<tr>
<td>Bortezomib(w/o Rituximab)</td>
<td>12(13%)</td>
<td>9(41%)</td>
<td>2(4%)</td>
<td>1(23%)</td>
</tr>
<tr>
<td>Melphalan</td>
<td>6(7%)</td>
<td>3(14%)</td>
<td>1(2%)</td>
<td>2(16%)</td>
</tr>
<tr>
<td>Other**</td>
<td>5(5%)</td>
<td>0(0%)</td>
<td>5(9%)</td>
<td>0(0%)</td>
</tr>
<tr>
<td>Treatment of clonal disease</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>before amyloid diagnosis</td>
<td>13(14%)</td>
<td>1(5%)</td>
<td>12(21%)</td>
<td>0(0%)</td>
</tr>
</tbody>
</table>

**Abbreviations:** ANS: Autonomic nervous system, dFLC: Difference between involved and uninvolved free light chains, eGFR: Estimated glomerular filtration rate, GI: Gastrointestinal tract, HSCT: Hematopoitic stem cell transplantation, PNS: peripheral nervous system, Pts: Patients, RB: Rituximab-Bendamustine, RX: Rituximab based regimens, ST: Soft tissue, w/o: Without.

*RX include: Bortezomib-Rituximab-Bendamustine in 1 patient, Bortezomib-Rituximab-Cyclophosphamide in 2 patients, Bortezomib-Rituximab in 4 patients, Rituximab alone in 3 patients, Rituximab-Methothrexate in 1 patient, Rituximab-Cyclophosphamide in 1 patient, and Rituximab-Rivilamid in 1 patient.

**Other include: Cyclophosphamide in 1 patient, Chlorambucil in 1 patient, Ibrutinib in 1 patient and Pentastatin C in 1 patient.
Table 2: Prognostic factors correlated with survival.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Overall survival (weeks)</th>
<th>Progression free survival (weeks)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Median</td>
<td>P-value</td>
</tr>
<tr>
<td>Heart</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Involved</td>
<td>128</td>
<td>0.002</td>
</tr>
<tr>
<td>Not involved</td>
<td>341</td>
<td></td>
</tr>
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<td>Mayo staging</td>
<td></td>
<td></td>
</tr>
<tr>
<td>I</td>
<td>717</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>II</td>
<td>337</td>
<td></td>
</tr>
<tr>
<td>IIIa</td>
<td>157</td>
<td></td>
</tr>
<tr>
<td>IIIb</td>
<td>146</td>
<td></td>
</tr>
<tr>
<td>dFLC</td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;50 mg/L</td>
<td>341</td>
<td>0.02</td>
</tr>
<tr>
<td>≥50 mg/L</td>
<td>122</td>
<td></td>
</tr>
<tr>
<td>dFLC</td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;180 mg/L</td>
<td>248</td>
<td>0.001</td>
</tr>
<tr>
<td>≥180 mg/L</td>
<td>70</td>
<td></td>
</tr>
<tr>
<td>Albumin i.s</td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;30 g/L</td>
<td>154</td>
<td>0.012</td>
</tr>
<tr>
<td>≥30 g/L</td>
<td>276</td>
<td></td>
</tr>
<tr>
<td>Amyloidogenic clone lymphC</td>
<td></td>
<td></td>
</tr>
<tr>
<td>pcC</td>
<td>412</td>
<td>0.769</td>
</tr>
<tr>
<td>Unknown</td>
<td>490</td>
<td></td>
</tr>
<tr>
<td></td>
<td>447</td>
<td></td>
</tr>
</tbody>
</table>
DYNAMIC BIODISTRIBUTION OF 124I-P5+14 IN PATIENTS WITH AL AMYLOIDOSIS

A. Stuckey 1, E. B. Martin 1, D. Powell 2, Y. Fu 2, M. Besozzi 2, S. Hall 3, S. J. Kennel 1, J. S. Wall 1

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2 Department of Radiology, University of Tennessee Medical Center.
3 University of Tennessee.

Introduction: Clearance of tissue amyloid remains an important, unmet need for patients with systemic amyloidosis. Despite the non-native structure of amyloid, phagocytic cells of the innate immune system do not effectively eliminate this material in patients without appropriate immunoglobulin (Ig) opsonization. This has been shown in vitro, in mouse models, and in patients with hepatic AL amyloidosis (Richards, D.B. et al. [2015] NEJM, 373, 12). Ideally, a single opsonin could target diverse amyloids.

We have reported on synthetic peptides (p5 and p5+14) that adopt an α-helical structure in the presence of polycationic ligands and bind many forms of amyloid in vitro and in vivo, as seen in animal models and in patients with AL-, ATTR- and ALECT2-associated amyloidosis.

Herein, we describe the generation of a novel peptide-Ig fusion where peptide p5 is fused to an Ig light chain (LC), which when associated with an Ig heavy chain (HC) yields a multi amyloid-reactive opsonin.

Objectives: Our aim was to create and characterize a new opsonin for targeting many amyloid types and enhancing macrophage-mediated phagocytosis.

Methods: The peptide-Ig fusion comprised an Ig LC sequence fused to peptide p5. The IgHC sequence comprised IgG1 variable, IgG1 CH1, and IgG2a CH2 and CH3 domains expressed in a second vector system. Vectors were cotransfected into HEK 293T/17 cells and the Ig product isolated using Protein A-conjugated beads. Igp5 and Ig control were radiolabeled with 125I and analyzed by gel electrophoresis. Binding of 125I-Igp5 and Ig control with synthetic amyloid fibrils and amyloid extracts was assessed using pulldown assays. 125I-Igp5 was injected IV into healthy (WT) mice and those with severe systemic AA amyloidosis (H2/IL-6 transgenic). Tissue localization was assessed by SPECT/CT imaging, biodistribution and microautoradiography. Opsonization of synthetic fibrils and amyloid extracts (20 µg) was evaluated ex vivo using pHrodo-red labeled substrates and human THP-1 cells.

Results: Purified Igp5 appeared as an intact Ig with both IgH and IgL peptides following gel electrophoresis. 125I-Igp5 bound rVλ6Wil and Aβ(1-40) amyloid-like fibrils at ~64% (21 fold higher than the control Ig, murine 11-1F4). Binding to amyloid extracts was also 10 – 30 fold higher and correlated positively with that of 125I-p5 (r = 0.9, p = 0.01). When injected into AA mice, 125I-Igp5 was detected in the liver and spleen by SPECT/CT imaging with >15% injected dose per gram (20 h post injection). In WT mice, the value was > 3-fold lower and associated with blood pool as seen in autoradiographs. Phagocytosis of rVλ6Wil fibrils by THP-1 cells was 10-fold higher than an irrelevant Ig (MOPC-31c) and twice that of the control Ig.

Conclusions: Opsonization of amyloid and clearance by cells of the innate immune system remains an important treatment goal. We have developed a novel peptide-Ig fusion that binds many forms of amyloid and is capable of enhancing phagocytosis.
PT047

PREDICTORS OF RENAL OUTCOME IN ALAMYLOID PATIENTS TREATED WITH CHEMOTHERAPY AND/OR BONE MARROW TRANSPLANT IN CLEVELAND CLINIC FOUNDATION: USING A BASELINE STAGING SYSTEM

T. Ashour 1, A. Mehdi1, S. Arrigain1, J. Schold1, M. Hanna1, J. Valent1, G. Nakhoul1, R. Fatica1

1 Cleveland Clinic Foundation

Introduction/Background: Light chain amyloidosis (AL) is the most common form of renal amyloidosis. Renal involvement greatly impacts morbidity and predicting renal outcome is crucial for management. A recent multicenter retrospective study was done by Palladini et al. on 461 patients with AL amyloidosis and renal involvement, and it showed that baseline proteinuria and eGFR predict risk of dialysis at 2 years while censoring mortality.

Objectives: The goal of the study was to evaluate the staging system from Palladini et al. on patients with AL amyloid with renal involvement who received chemotherapy and/or bone marrow transplant at the Cleveland Clinic Foundation from 2007 to 2018.

Methods: We studied 64 patients with newly diagnosed renal AL amyloidosis. All had renal biopsy at the time of diagnosis. We calculated the baseline eGFR using the MDRD equation and proteinuria was measured in a 24 hours urinary collection or a spot urine protein creatinine ratio. We categorized patients as described by Palladini et al.: stage I eGFR (≥50 ml/min/1.73 m2) and proteinuria (≤5 g/day), stage II, eGFR (<50 ml/min/1.73 m2) or proteinuria (>5 g/day) and stage III eGFR (<50 ml/min/1.73 m2) and proteinuria (>5 g/day). We estimated Kaplan-Meier ESRD-free survival censored at death for each stage and compared the groups with the Log-Rank test.

Results: Patients' average age at diagnosis was 61.6 (+9.8) and 44% were male. Average serum albumin was 2.7 (0.76), 34% had cardiac involvement, and 22% multiple myeloma. 16 (25%), 28 (44%) and 20 (31%) of patients were stages 1-3 respectively. Table 1 shows Kaplan-Meier ESRD-free survival estimates (censored at death) at different time points. At 2 years, 91.7% (95%CI: 77.3, 100), 80.8% (67.0, 97.5), and 58.2% (38.3, 88.4) of stage 1, 2 and 3 respectively were ESRD-free.

Table 1. Kaplan Meier ESRD-free Survival Estimates by stage (censored at death)

<table>
<thead>
<tr>
<th>Stage</th>
<th>KM ESRD-free (95% CI)</th>
<th>n=16</th>
<th>KM ESRD-free (95% CI)</th>
<th>n=28</th>
<th>KM ESRD-free (95% CI)</th>
<th>n=20</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 Years</td>
<td>100.0(100.0,100.0)</td>
<td>88.7(77.5,100.0)</td>
<td>68.8(50.9,93.0)</td>
<td></td>
<td>0.025</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2 Years</td>
<td>91.7(77.3,100.0)</td>
<td>80.8(67.0,97.5)</td>
<td>55.1(35.7,84.8)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3 Years</td>
<td>91.7(77.3,100.0)</td>
<td>80.8(67.0,97.5)</td>
<td>55.1(35.7,84.8)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4 Years</td>
<td>91.7(77.3,100.0)</td>
<td>67.8(49.9,92.3)</td>
<td>48.2(29.1,79.9)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5 Years</td>
<td>91.7(77.3,100.0)</td>
<td>67.8(49.9,92.3)</td>
<td>48.2(29.1,79.9)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Conclusions: Baseline eGFR and 24 hours proteinuria are useful for predicting the risk of progression to ESKD in patients with renal AL amyloidosis. Our results validate the baseline staging system proposed by Palladini et al. Future prospective studies are needed to confirm our results.
AMYLOID DETECTION IN SUBCUTANEOUS FAT ASPIRATES USING THE FLUORESCENT DYE FSB: A PROSPECTIVE STUDY IN 451 PATIENTS

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Introduction: In clinical routine, tissue amyloid deposits are typically detected by Congo red (CR) staining visualized under polarized light (CR-PL). Interpretation of CR results is critically affected by factors including quality of staining and equipment, and pathologist’s expertise. In a recent pilot study [1] on abdominal fat aspirates, we reported that the fluorescent CR analogue FSB (1-fluoro-2,5-bis[(E)-3-carboxy-4-hydroxystyryl]benzene) has diagnostic sensitivity comparable to CR-PL, but presents favorable features (strong fluorescence, easiness of staining implementation and interpretation, no interference from collagen, blood and other tissue structures), that make it an attractive candidate as a possible complement or alternative to CR.

Objectives: In this study we evaluated the performances of FSB staining for detecting amyloid in fat aspirates in a large series of 451 consecutive individuals with suspected systemic amyloidosis, through comparison of FSB results against those of CR-PL.

Methods: Consecutive individuals referred to our Center (December 2017 through June 2019) were included. Patients underwent clinical, instrumental and genetic examination. In cases without evidence of amyloid deposits in fat but persistent clinical suspicion of amyloidosis, biopsy of affected organs was performed. Subcutaneous periumbilical fat was acquired by fine-needle aspiration; samples were split into 3 comparable parts to be examined by immuno-electron microscopy (IEM), FSB and CR-PL [1]. FSB staining fluorescence was visualized by confocal microscopy, as described [1]. FSB and CR-PL results were independently interpreted by 2 expert physicians.

Results: Overall, CR-PL and FSB provided concordant results in 443 of 451 cases (98%, 95% confidence interval 96.5%-99%). Of these, 296 were classified as negative and 147 as positive by both. Amyloid was typed by IEM in all CR+ cases; diagnoses included: 5 reactive (AA), 5 wild-type (ATTRwt), and 5 variant (ATTRv) ATTR amyloidosis, 2 apolipoprotein A-I (AApo-AI) and 130 light-chain (AL) amyloidoses (24 ALκ, 106 ALλ). In 2 of the 3 CR+/FSB- cases, systemic amyloidosis was excluded; 1case had ATTRwt with heart involvement. In all the 5 CR-/FSB+ cases, systemic amyloidosis was confirmed (2 had ATTRwt with heart involvement; 3 had AL with kidney involvement).

Conclusions: This study shows, in a large prospective series, that FSB staining has diagnostic sensitivity comparable with that of CR-PL. The optimal signal-to-background ratio of FSB and the easiness of staining and results interpretation make it a promising method for allowing rapid, sensitive and reliable amyloid detection on fat aspirates. This could translate into faster diagnosis and more appropriate referral to specialized centers for typing and treatment.


Keywords: Amyloid detection, diagnosis, amyloid-specific dyes
DIAGNOSTIC DELAY AND EARLY MORTALITY IN SYSTEMIC AL AMYLOIDOSIS. A REAL-WORLD SPANISH MULTICENTER STUDY

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2 University Hospital Virgen de la Victoria.
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Background: Light chain (AL)-amyloidosis is the most common type of systemic amyloidosis (Vaxman I et al.2019). Cardiac (CA) involvement occurs in 60-75% of AL patients (pts) and is the main prognostic factor. Most patients are still diagnosed late when advanced organ damage has already ensued (Merlini G et al.2018). Overall survival (OS) is improving but early mortality (EM) (<6 months) remains high (24% in the period 2010-2014, Muchtar E et al.2017). Real-life data about diagnostic delay (DD) and EM are still lacking.

Objectives: To know the current EM and DD in a Spanish retrospective and real-world multi-center study.

Methods: Data were gathered from all consecutive newly diagnosed AL pts (2013-2019) in four Spanish centers: 1. HPH, 2. HVV, 3. HVN, 4. HCS. Common clinical and lab baseline variables were analyzed. CA was confirmed by endomyocardial biopsy, positive CA criteria on echocardiogram or cardiac MR and other organ biopsy, or an amino-terminal fragment of type B natriuretic peptide (NT-proBNP) >332 ng/L (in pts without renal failure or atrial fibrillation). EM as well as very EM at 2 months (VEM) were analyzed. Comparisons for categorical variables were made with the $\chi^2$-test, using Fisher’s exact test when appropriate. Comparisons of means of quantitative continuous variables between two groups were made with the t-test. OS curves were estimated using the Kaplan-Meier method, and comparisons among groups were carried out with the log-rank test.

Results: One hundred and six pts were included in the study: 1 (n=69), 2 (n=17), 3 (n=14), 4 (n=6). Median age was 65 years (43-87), 55 were men and 51 women. At the time of the analysis, 50 pts have died. Baseline CA was confirmed in 81.1%, ECOG 3 was shown in 32.1% and autologous stem cell transplant was performed in 16%. Median OS for the whole series was 44 m (95% confidence interval 26.1-61.9), without statistically significant difference among the four centers. OS for pts with CA was significantly poorer ($P=0.035$). Median DD was 6 (0-18), 6.5 (1-54), 7.2 (2-36), and 4 (3-5) m, respectively. VEM and EM were 16% and 29.2%, respectively. EM as well as VEM were associated with higher BNP or NT-proBNP (both $P<0.001$), older age ($P=0.028$ and $P=0.002$), and impaired renal function ($P=0.051$, $P=ns$ for VEM). DD was not statistically different for pts with or without EM, but it was associated with shorter time for pts with VEM (5.4 vs 8 m, $P=0.043$).

Conclusions: The median DD in our series is 6 m (this is higher than the one commonly reported for multiple myeloma). Early diagnosis to avoid irreversible CA remains a major aim; EM in our study is 29.2% and is associated with older age, CA, and impaired renal function; A shorter DD is shown in pts who had VEM; Key common warnings should be shared for all caregivers involved, in order to improve diagnostic performance.
RENAL PATHOLOGY IN PATIENTS WITH A PRIOR DIAGNOSIS OF MONOCLONAL GAMMOPATHY OR MULTIPLE MYELOMA: MONOCLONAL IMMUNOGLOBULINS ARE NOT ALWAYS THE CAUSE

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Background: Renal disease in patients with monoclonal gammopathies (MG) has been associated with a broad pathology spectrum, including cast nephropathy and monoclonal gammopathy of renal significance (MGRS); however, unrelated causes (hypertension, diabetes) may also be the cause of renal dysfunction.

Aim: To evaluate the frequency of MG-unrelated renal pathologies in patients with known MGs presenting with renal dysfunction.

Methods: We reviewed data of 79 patients with known MGs that had renal biopsy for evaluation of renal dysfunction, after the diagnosis of MG in a single center (Department of Clinical Therapeutics, Athens).

Results: at the time of renal biopsy, median age was 69 years (39-84); co-morbidities included hypertension (72%), diabetes (28%), CAD (7%); 36 (45%) had symptomatic MM with disease features beyond renal dysfunction (26/36 were on therapy at the time of biopsy, 19/36 in remission). Median eGFR was 33 ml/min/1.73 m2, 15% required dialysis, median proteinuria was 3.1 gr/d, abnormal FLCratio was found in 69.5%, median dFLC level was 85 mg/L. In urine protein electrophoresis (UPEP), median albumin proportion was 40% (3-100%) and median urine monoclonal protein (UMP) was 2% (0-97%). Renal pathology showed a MG-related diagnosis in 68% (cast nephropathy:13%, MIDD: 25%, AL amyloidosis:25%, other MGRS: 5%) and was unrelated in 32% (diabetes:5%, hypertension:14%, drug related:8%, single cases of IgA nephropathy, chinese herb nephropathy, obesity-related GN). In 11/19 (58%) of MM patients in remission, pathology was MG-unrelated vs in 6/17 (35%) of those not in remission. Factors associated with MG-related pathology included serum albumin <3 gr/dl (88% vs 60%, p=0.007), proteinuria >1.7 gr/d (82% vs 48%, p=0.004), positive UIFE (75% vs 33%, p=0.039), UMP>100 mg/d (68% vs 25%, p=0.003), dFLC >50 mg/L (76% vs 37%, p=0.003) and abnormal FLCratio (78% vs 53%, p=0.044). The presence of other co-morbidities, hematuria or the reason leading to renal biopsy were not associated with non MG-related renal pathology. In multivariate analysis, only UMP>100 mg/d (HR: 7.95, p=0.024) was independently associated with a diagnosis of MG-related pathology. If UMP and UIFE were not included, then total proteinuria >1.7 gr/d (HR:4.5, p=0.036) and dFLC>50 mg/L (HR:5.8, p=0.033) were independent predictors. By using these two parameters 7% of those without any vs 30% with any of the two vs 63% with both factors had a MG-related renal pathology.

Conclusions: Among patients with known monoclonal gammopathies and renal dysfunction, 32% had a non-MG-related diagnosis, which had implications in their management. Urine electrophoresis should be evaluated in all patients with MG; otherwise, dFLC>50 mg/L and proteinuria >1.7 gr/d can be used. Renal dysfunction should not be attributed to the underlying MG without careful consideration of the other parameters and of a renal biopsy.
PITFALLS OF SUBTYPING AA AMYLOID USING COMMERCIAL ANTIBODIES

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Background and Introduction: Immunohistochemistry (IHC) is a widely used technique for characterisation of amyloid fibril type. Antisera to all known amyloidogenic proteins are commercially available, and most are reliable in identifying the fibril type in FFPE tissue. Here we present a review of 5 cases from the UK National Amyloidosis Centre (NAC), highlighting differences of amyloid typing using a commercial antibody for AA amyloid in comparison with both an R&D anti-human AA antibody and proteomic analysis.

Methods: IHC was performed on 5 selected cases containing amyloid using a commercially available monoclonal mouse anti human AA antibody, (clone mc1, Agilent), dilution 1:3200, and an R&D anti-human AA antibody (Reu 86.1, supplied by Bouke Hazenberg), dilution 1:100. Staining was carried out on a manual platform, using Impress™ detection kits and a metal enhanced DAB substrate kit for visualizing the immuno compound. Amyloid was laser captured for proteomic analysis from duplicate slides. Proteomic analysis was performed on the Thermo Scientific™ Q-Exactive Plus Orbitrap. Data was analysed using Mascot software and the Swiss-Prot human database. All IHC slides were interpreted blind to any clinical details by two independent reporters. Data obtained from proteomic analysis was analysed at a weekly MDT with an experienced panel of interpreters. IHC results were compared with data obtained from proteomic analysis in particular with respect to positive identification of the amyloid fibril protein.

Results: The amyloid in 4 of 5 cases stained positively with the commercial anti-human AA antibody; however, proteomic analysis indicated the amyloid fibril protein to be apolipoprotein A-IV (apoAIV) in 2, lambda light chain in 1, and kappa light chain in 1. The amyloid failed to stain in any of these four cases using the R&D anti-human AA antibody. The amyloid in the remaining case did not stain with the commercial anti-human AA antibody but proteomic analysis and the R&D antibody indicated AA amyloid. The biochemical results, clinical phenotype and SAP scintigraphic findings were entirely consistent with AA amyloidosis in this final case.

Discussions and Conclusions: This commercially available antibody was chosen according to its specification, listed as an antibody that specifically labels AA amyloid in tissues and shows no reactivity with a host of other known tissue antigens including other known amyloid fibril proteins. However, in our hands a comparison of the IHC findings using this commercial antibody with proteomic analysis and the R&D anti-AA antibody showed that it gave both false positive and false negatives results. This commercial anti-human AA antibody is not reliable for determining the amyloid fibril type and should not be included in routine IHC panels for the subtyping of amyloid. This study will be expanded for further validation and verification.

Keywords: AA amyloid, Immunohistochemistry, Proteomic analysis
EARLY DETECTION OF SYSTEMIC AMYLOIDOSIS USING AUTOMATED INTELLIGENCE WITHIN AN ELECTRONIC MEDICAL RECORD (PROJECT SAFEGUARD)

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1 UMCG

Introduction: As new medications emerge to provide means to delay or halt disease progression in Systemic Amyloidosis (SA), it is becoming increasingly important to decrease the too great diagnostic delay. A solution could be found by using Artificial Intelligence. Unfortunately, its great potential is still hampered by seeming complexity, often narrow application and relatively high costs. However, an Automated Intelligence (AmI) could provide a quick fix by merging acquired human knowledge with computing power.

Objective: To evaluate whether using an AmI based on known ‘red flags’ for SA can be a useful tool for early detection.

Methods: An AmI was created by using the Best Practice Advisory application within the Epic® Electronic Medical Record (EMR). The AmI was configured to trigger on the presence of Carpal Tunnel Syndrome or Polyneuropathy combined with Heart failure or Atrium fibrillation in the absence of the diagnosis SA or Diabetes Mellitus in a patient’s EMR with the age over 50 years. The AmI was active during a period of 2½ month. When entering a patient’s digital chart when the condition was met, a ‘diagnostic warning’ was displayed to the physician with a suggestion for further follow-up trough the Amyloidosis Expert Center. The referred patients underwent diagnostic tests to evaluate the presence of SA. All non-referred patients would later be anonymously evaluated for their chance of having SA.

Results: During the period of 2½ month there were 110,000 unique patient encounters at our center and the AmI identified a total of 141 patients who met the criteria. Of those only 16 were referred, and 10 proved alleageable for further diagnostic follow-up (figure 1). Of those 10 patients, two patients showed some evidence of amyloidosis. In one patient the abdominal subcutaneous fat aspirate showed amyloid (1+) and in the other patient amyloid was suspected because the bone scintigraphy showed grade 1 cardiac tracer uptake. However, in both patients the type of amyloid could not be established. Of the remaining 125 non-referred patients around 64% had additional information within their EMR which suggested that SA could be a likely diagnosis and further evaluation would be warranted (figure 2). The pilot met with some negative reactions from clinicians, as they found the ‘diagnostic warning’ too prescriptive, and some patients did not want to be confronted with a potential disease. Also, the AmI logic lacked specificity due to an inability to access certain non-structurally stored data within in the EMR; e.g. ECG and TTE findings.

Conclusion: This study shows that using an AmI based on ‘red flags’ has potential of being a useful tool for the early detection of SA. Further fine-tuning is needed to make the system more specific and more subservient to the clinicians. Moreover, legal and ethical issues should be addressed.
Figure 1: Diagnostic follow-up of patients identified by Automated Intelligence who were referred to the Amyloidosis Expert Center.

Figure 2: Evaluation of likelihood of Systemic Amyloidosis of non-referred, by Automated Intelligence identified, patients.

Keywords: Early detection, Artificial Intelligence, Systemic Amyloidosis
STANNIOCALCIN-1 AS A POTENTIAL CIRCULATING BIOMARKER OF TOXICITY IN LIGHT CHAIN CARDIAC AMYLOIDOSIS

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Introduction: Light chain amyloidosis (AL) is a life-threatening disease in which misfolded immunoglobulin free light chains aggregate and form amyloid fibrils in the heart and other organs. Increasing evidence has revealed that direct light chain-mediated cardiotoxicity, in addition to amyloid deposition, is a crucial component of pathogenesis in AL amyloidosis [1]. A mechanism by which amyloidogenic free light chains exert cardiotoxicity is upregulation of stanniocalcin-1 (STC-1), which leads to cell death due to mitochondrial oxidative stress and impaired lysosomal function [2].

Objective: We hypothesized that STC-1 may be a specific marker of cytotoxicity in patients with active AL cardiomyopathy when compared to those in remission and to those with another form of cardiac amyloidosis [transthyretin (ATTR) amyloidosis].

Methods: Plasma samples were collected from AL cardiomyopathy patients with active disease and then again 6 months later in the same patients while in remission. Plasma was also collected from ATTR cardiomyopathy patients and asymptomatic transthyretin (TTR) variant carriers. Plasma STC-1 concentrations were measured by western blot. Statistical analysis was performed using analysis of variance.

Results: In patients with active AL cardiomyopathy, plasma STC-1 levels decreased by 43% after 6 months of treatment and achieving a complete remission (n=4 pairs of patient samples, p=0.035). STC-1 levels were not significantly different between patients with ATTR cardiomyopathy and asymptomatic TTR variant carriers (n=8 and n=4, respectively). Furthermore, active AL cardiomyopathy patients had the highest STC-1 level among the study groups.

Conclusions: Plasma STC-1 concentration was significantly elevated in AL cardiomyopathy patients with active disease and decreased 6 months later in remission. As the amount of amyloid deposition is not expected to significantly decrease over this short period, the decrease in STC-1 may be due to eliminating light chain-mediated cardiotoxicity. There was no difference in STC-1 level between ATTR patients and TTR variant carriers, suggesting that STC-1 may be a specific marker of disease activity in AL amyloidosis. Further validation in larger cohorts is needed.

VARIABLE CLINICAL PRESENTATION AND RESPONSE TO THERAPY IN PATIENTS WITH IGM RELATED AL AMYLOIDOSIS

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Introduction: Immunoglobulin light chain (AL) amyloidosis associated with an IgM monoclonal gammopathy is rare accounting for approximately 5% of AL amyloidosis. The clinicopathological features are distinct compared to non-IgM AL amyloidosis.

Methods: We conducted a retrospective review of patients with IgM AL amyloidosis treated at two tertiary institutions in Australia. We describe the clinical presentation, disease features and outcomes in this cohort of patients.

Results: 13 patients with IgM AL amyloidosis were identified. Median age at diagnosis was 68 (range 60-86 years) and 62% (n=8) were female. Organ involvement included kidney in 40% (n=5), neurological in 31% (n=4) and heart in 15% (n=2). Lymph node involvement was present in 46% (n=6). Two patients presented with a retroperitoneal perinephric mass and two presented with pleural effusions. Of 9 patients with data on bone marrow biopsy, 89% (n=8) had a lymphoplasmacytic infiltrate. Of four patients tested, 3 had a MYD88 L265P mutation detected. Light chain type was lambda in 58% (n=12).

Therapy was instituted in 11 patients, with 2 patients remaining untreated with asymptomatic lymphadenopathy as the only disease manifestation. A variety of treatment regimens were utilised including rituximab, cyclophosphamide and dexamethasone (RCD) in 4, bendamustine and rituximab (BR) in 4, bortezomib based therapy in 1, melphalan and dexamethasone in 1 and single agent ibrutinib in 1 patient. One patient received in autologous stem cell transplant after induction with RCD. Of 10 evaluable patients the overall response rate to initial therapy was 70%, complete response in 10%, very good partial response in 20% and partial response in 40%. The 3 patients achieving at least a VGPR all received therapy with BR. The patient receiving ibrutinib achieved a 50% reduction in IgM monoclonal protein level after 3 months of therapy, although the lambda light chain level remained unchanged. 3 patients with neuropathy received therapy with improvement in symptoms seen in 2 patients. After a median follow up of 39 months, 3 patients have died with a corresponding 5 year overall survival of 66%.

Conclusion: Patients with IgM related AL amyloidosis typically present with renal and lymph node involvement and a bone marrow biopsy revealing a lymphoplasmacytic infiltrate. Cardiac involvement appears to be less common than seen in non-IgM AL amyloidosis. Patients treated with regimens directed at a lymphoplasmacytic clone appear to do well. Incorporating novel agents such as ibrutinib to therapy in this population requires further study.

Keywords: AL amyloidosis, IgM
PT055

TYING OF AMYLOIDOSIS USING IMMUNOHISTOCHEMISTRY WITH AMYLOID TYPE-SPECIFIC ANTIBODIES AND THE DIFFERENCE TO MASS SPECTROMETRY

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Introduction: The clinically different amyloidoses are distinguished by the chemical identity of the pathogenic amyloid protein. Amyloid is diagnosed on biopsies of affected patients with Congo red and classified with respect to therapy. For routine classification (or typing) of amyloidosis, two basically different methods have been applied to either immunohistochemistry (IHC) with amyloid-type specific antibodies or mass spectroscopy (MS and LD-MS) in order to learn more about the value and quality of the typing methods.

Material and methods: The tissue samples were either biopitic or autoptic formalin-fixed paraffin section for both methods. The histological diagnosis of amyloid was carried out on formalin-fixed paraffin tissue sections following the use of Congo red. The classification of the amyloids was performed on formalin-fixed paraffin sections using indirect immunohistochemistry (IHC) and then by mass spectrometry (MS) in two varieties as MS and LD-MS. The study was done blinded. It has three sections: Ia, 53 biopitic samples, comparison IHC versus MS. Ib, 38 autoptic sample IHC versus MS and II 60 samples taken from Ia and Ib, IHC versus LD-MS.

Results: The results are expressed as correct (cor), not identical (nid) and data missed (mi). The numbers are in percent for the three sections.

For Ia, IHC cor 89.5 – nid 0 – mi 10.5; MS cor 35.9 – nid 2.6- mi 61.5.
For Ib, IHC cor 92.5 – nid 0 – mi 7.5; MS cor 53.7 – nid 3.7- mi 42.6
For II, IHC cor 91.2 – nid 0 – mi 9.8; LD-MS cor 53.0 – nid 5.4- mi 42.4

The comparison shows that this IHC is highly significant and more sensitive in contrast to MS/LD-MS. In addition, the sensitivity of this kind of IHC in 581 patients, with the recognition of 15 different amyloids, was 97.9% and its sensitivity was 99.3%.

Discussion and conclusions: This reliable performance of this IHC is based on homologous antibodies that are amyloid-type specific, thereby circumventing most of the unspecific proteins contained in the histologic region of amyloid. In addition, proof that both the IHC and MS/LD MS methods are equally valid for amyloid typing has been derived from the fact that all amyloids which could be typed by MS/LD MS showed identical amyloid types. Moreover, this type of IHC provides an independent marker independent of any additional information, including clinical data provided by the built-in controls. IHC showed no unexpected (possibly incorrect) amyloid type due to the possibility of microscopical control of the specimen which is now also possible with the new method of MALDI-MS.
EARLY CARDIAC RESPONSE IS POSSIBLE IN PATIENTS WITH IMMUNOGLOBULIN LIGHT CHAIN AMYLOIDOSIS AND IS ASSOCIATED WITH PROLONGED SURVIVAL

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Introduction: despite the introduction of novel therapeutic agents, patients with cardiac stage IIIb immunoglobulin light chain (AL) amyloidosis still have a dismal prognosis. Achieving a rapid profound reduction of dFLC results in an improvement of survival even in this subset of patients (Manwani, et al. Hematologica 2018). However, the possibility and the prognostic relevance of cardiac biomarker response has not been evaluated so far.

Objective: evaluate the prognostic impact of cardiac response at 90 days from treatment initiation

Methods: our prospectively maintained database of 1378 patients with AL amyloidosis newly diagnosed between 2004 and 2018 was searched for cardiac stage IIIb patients. Hematologic and cardiac responses were assessed by intent to treat at 30 and 90 days from starting chemotherapy. Survival curves were plotted according to Kaplan Meier with a landmark at 30 and 90 days, and differences in survival were tested for significance with the log-rank test.

Results: we identified 249 patients with stage IIIb AL amyloidosis, representing 18% of the whole cohort. Patients’ characteristics are reported in Table 1. Two-hundred nine patients (84%) died. Median overall survival was 4 months. Hematologic response was observed in 50 (20%) patients (8% at least very good partial response [VGPR]) at 30 days and in 53 (22%) subjects (14% at least VGPR) at 90 days after starting chemotherapy. Achieving at least a VGPR at 30 and 90 days was associated with a better overall survival (51 vs. 3 months; P<0.001 and 51 vs. 6 months; P<0.001, respectively). Cardiac response at 90 days was observed in 19 (8%) subjects. Overall survival was significantly better among cardiac responders (54 vs. 20 months; P<0.001). Cardiac progression was observed in 197 (80%) patients and was associated with a shorter survival (20 vs. 3 months; P<0.001), also in subjects who achieved at least a VGPR at 90 days (50 vs. 20 months; P=0.02). A difference between involved and uninvolved light chains (dFLC) >500 mg/L and troponin I >0.5 ng/mL were identified as independent prognostic factors at baseline. Patients with either of these biomarkers above the cutoff had a worse prognosis (6 vs. 3 months; P<0.001). Having a dFLC >500 mg/L was associated with lower rates of high-quality hematologic response at 90 days (at least VGPR in 5% vs. 18%; P=0.002). However, there was no significant difference in the rate of cardiac response in patients with dFLC (25% vs. 20%; P=0.687) or troponin I (19% vs. 20%; P=0.796) above or below the cutoffs.

Conclusions: Achieving an early cardiac response is rare but possible in patients with advanced cardiac AL amyloidosis, and is associated with a dramatically longer survival. However, most stage IIIb patients have a rapid cardiac progression which is associated with a dismal outcome. Changes in NT-proBNP remain robust predictors of survival also in patients with advanced cardiac disease.

Keywords: amyloidosis, response, prognosis
### Table 1. Patients’s characteristics

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>N (%) – mean (IQR)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, years</td>
<td>68 (60-74)</td>
</tr>
<tr>
<td>Sex, male</td>
<td>145 (58)</td>
</tr>
<tr>
<td>Organ involvement</td>
<td>129 (51) / 43 (17) / 51 (20) / 6 (2) / 23 (9) / 21 (8)</td>
</tr>
<tr>
<td>Isolated heart involvement</td>
<td>75 (30)</td>
</tr>
<tr>
<td>Organ involved &gt;2</td>
<td>76 (31)</td>
</tr>
<tr>
<td>NT, proBNP, ng/L</td>
<td>17089 (12179-25014)</td>
</tr>
<tr>
<td>Troponin I, ng/mL</td>
<td>0.265 (0.169-0.500)</td>
</tr>
<tr>
<td>Troponin I &gt;0.5 ng/mL</td>
<td>63 (25)</td>
</tr>
<tr>
<td>Renal stage: I / II / III</td>
<td>108 (43) / 111 (45) / 26 (10)</td>
</tr>
<tr>
<td>Intact MC : LC only</td>
<td>121 (49) : 128 (51)</td>
</tr>
<tr>
<td>Kappa : lambda</td>
<td>53 (21) : 196 (79)</td>
</tr>
<tr>
<td>dFLC, mg/L</td>
<td>259 (142-543)</td>
</tr>
<tr>
<td>dFLC &lt;50 mg/L</td>
<td>13 (5)</td>
</tr>
<tr>
<td>dFLC &gt;180 mg/L</td>
<td>167 (67)</td>
</tr>
<tr>
<td>dFLC &gt;500 mg/L</td>
<td>69 (28)</td>
</tr>
<tr>
<td>BMPC, %</td>
<td>12 (8-20)</td>
</tr>
<tr>
<td>Treatment</td>
<td>118 (47) / 95 (38)</td>
</tr>
<tr>
<td>BMPC, %</td>
<td>28 (11) / 5 (1)</td>
</tr>
</tbody>
</table>

ANS, autonomic nervous system; BMPC, bone marrow plasma cells; dFLC, difference between involved and uninvolved free light chain; GI, gastrointestinal; LC, light chain; MC, monoclonal component; MDex, melphalan and dexamethasone; PNS, peripheral nervous system;
CORRELATION BETWEEN TWENTY-FOUR HOUR PROTEINURIA AND RANDOM ALBUMIN TO CREATININE RATIO IN SYSTEMIC AL AMYLOIDOSIS

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Introduction: The evaluation of proteinuria is essential for diagnosis and monitoring of renal involvement in AL amyloidosis. A 24 hour protein collection (24h UP) is the gold standard for proteinuria assessment however it is cumbersome and can be inaccurate due to under or over collection. A spot urine albumin to creatinine ratio (uACR) has been proposed as a convenient method to estimate 24h UP. We aimed to validate the correlation between uACR and 24h UP in a large cohort of patients.

Methods: We retrospectively analyzed data from biopsy proven systemic AL amyloidosis patients evaluated during their disease course at Mayo clinic, between January 1, 2010 and September 26, 2019. Patients with a random spot urine and 24h UP collected less than 7 days apart were included. The correlation between spot urine albumin to creatinine ratio (uACR) and 24 hour proteinuria was assessed using a Pearson r test. Linear regression analysis was used to construct a model to predict the 24h UP using uACR as the primary predictor. Possible confounders (age, gender, body mass index, morning versus afternoon spot urine collection, estimated glomerular filtration rate) for the primary relationship between uACR and 24h UP were evaluated in the model. Receiver operating characteristic (ROC) analysis was used to identify the best uACR cutoff to predict significant proteinuria (defined as a 24h UP >500mg). A Chi-square test was used to test categorical data.

Results: A total of 665 patients were included, with a median age of 66 years (IQR 59-72). The spot urine was collected in the morning (before 1200 hours) in 382 (57%) patients, and in the afternoon in 283 (43%) patients. The median 24h UP was 321mg (IQR 129-2512.5mg), median uACR was 107 mg/g (IQR 13.5-1845 mg/g), and median serum creatinine was 1.2 mg/dL (IQR 1-1.8 mg/dL). The uACR correlated well with 24h UP (Pearson’s r = 0.83, 95% CI 0.80-0.85). Linear regression showed that E (24h UPi) = 362 + 1.05(uACRi), and this model was statistically and clinically significant (p<0.001 and R2 of 0.68, respectively). Age, gender, body mass index, eGFR, and time of day of random urine collection did not confound the primary relationship between uACR and 24h UP, and no collinearity was observed. A uACR cutoff of > 280 mg/g was the best predictor of a 24h UP >500 mg (area under the ROC curve 0.98, sensitivity 92%, specificity 97%). For simplicity, we assessed the predictive value of uACR >300 mg/g for 24h UP >500 mg. Among patients with 24h uACR > 300 mg/g 264 (96%) had a 24h UP >500 mg, and 31 (7%) of patients with uACR <300 mg/g had a 24h UP >500 mg (p<0.001).

Conclusions: In systemic AL amyloid patients, we showed that uACR on a random urine sample correlated well with 24h UP, and can be used to estimate proteinuria with a linear regression model. Based on these findings, and the convenience of uACR testing for patients, we propose that uACR should be used to monitor renal response to AL amyloidosis therapy.
MULTIPARAMETRIC FLOW CYTOMETRY FOR THE ASSESSMENT OF AL AMYLOIDOSIS


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Introduction: AL amyloidosis is a plasma cell disorder characterized by the presence of clonal plasma cells in bone marrow able to produce monoclonal light chains that deposit into extracellular tissues, leading to organ dysfunction. Multiparametric flow cytometry (MFC) has been recently used for the diagnosis and disease monitoring of AL amyloidosis. Furthermore, it was recently proposed as a tool for fast screening and risk-assessment of this entity.

Objectives: Our study aims to assess the impact of MFC over clinical outcomes in a series of patients with systemic AL amyloidosis seen at our institution.

Methods: All consecutive newly-diagnosed patients with biopsy-proven systemic AL amyloidosis, who underwent bone marrow MFC immunophenotyping between 09/2009 and 10/2018, were included. Clinical data were extracted from a prospectively maintained database. MFC was performed as per Institutional guidelines.

Results: 151 patients with AL amyloidosis were included in the present study. Median age was 66 years and 62.3% were male. Heart and kidney involvement was seen in 74.8% and 66.2% of cases, respectively. The median percentage of monoclonal plasma cells detected by MFC was 1.2%. CD38 expression was positive in 98.6%, CD 19 in 28.6%, CD20 in 29.9%, CD56 in 56.5%, and CD117 in 23.8% (Table 1). In addition, 5 patients were found to have another B-cell disorder associated to AL amyloidosis (3.4%). Median overall survival (OS) for the entire group has not been reached but an estimated survival is 56.7 months. Median OS was similar according to the expression (positive vs negative) of CD20 (NR vs 59 months, p=0.6), CD56 (NR vs 56.7 months, p=0.6) and CD117 (NR vs 56 months, p=0.7). Median OS was longer in the group with <2.5% Monoclonal Plasma Cells (MPC) (NR vs 21.8 months, p=0.001). The median percentage of polyclonal plasma cells was 0.4% (range 0.1-4%). Median OS was similar among patients with ≥1% or <1% polyclonal plasma cells (p=0.4). Furthermore, median OS was longer in those cases with BMPC’s of <10% (NR vs 24.2 months, p=0.01). A ratio between MPC and Bone Marrow Plasma Cells’s (BMPC) was created. Median BMPC/MPC ratio was 4.3. 46.8% of cases had a ratio ≥5. Median OS for patients with a MPC/BMPC’s ratio ≥5% or <5% was NR (not-reached) in both groups (p=0.1). No differences on the degree of response were seen among these cases based on the BMPC/BMPC ratio (<5% vs ≥5%, p=0.1) or the number of MPC (≥2.5% VS <2.5%, p=0.2).

In conclusion, MPF is a useful tool for the assessment of AL amyloidosis. The present study is in agreement with previous reports of predictive value of MPF while evaluating survival outcomes. As reported by others, MFC could be used for disease monitoring, prognostication and even for minimal residual disease assessment. Further studies using MFC are warranted, especially to evaluate its impact in the clinical decision making process in patients with relapsed disease.

Keywords: AL amyloidosis, MFC and Survival
Category: Diagnosis and prognosis of AL amyloidosis
LIGHT CHAIN AMYLOIDOSIS – DIAGNOSTICS AND TREATMENT CHALLENGE: SINGLE CENTER EXPERIENCE

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Introduction: Light chain amyloidosis (AL) represents a rare plasma cell neoplasia characterized by organ damage caused by deposition of amyloid fibrils. Often unrecognized, resulting with advanced clinical presentation, represents challenge for diagnostics and treatment.

Aim: The goal of this study was to analyze clinical and laboratory characteristics patients (pts) with AL Amyloidosis, and course of disease in the view of applied treatment.

Patients, diagnostics and treatment: The retrospective analysis included 32 newly diagnosed pts (male 21pts, female 11pts; median age 58yrs, range 39-77yrs) with AL amyloidosis, identified at the Hematopathology Unit, Clinic of Hematology, Clinical Center of Serbia, from January 2014 - October 2019. Histopathological diagnosis was made by the demonstration of amyloid deposits in a tissue biopsy using a Congo red stain, and immunohistohemistry. The staging and treatment were conducted according to the current recommendations of International Society of Amyloidosis.

Results: Organ involvement, regarding amyloid deposition, was as follows: Abdominal fat (14pts, 43.8%), kidney (13pts, 40.6%), bone marrow (13pts, 40.6%), heart (12pts, 37.5%), liver (5pts, 15.6%), skin (4pts, 12.5%), and lung (1pts, 3.1%). Four patients (12.5%) had ≥2 organ affected. The most frequent isotype of monoclonal protein was lambda light chain (22pts, 68.7%). The elevation of biomarkers of cardiac involvement (BNP, NT-proBNP, troponin) was identified in 18pts (56.3%). Elevation of LDH was found in 9pts (28.1%). According to the risk stratification, the distribution was as follows: Low 4pts (12.5%); Intermediate 16pts (50.0%); and High risk 12pts (37.5%). Due to the advanced organ affection at presentation, mainly cardiac involvement at presentation, majority of patients were treated with standard alkylating agents based combinations (Cyclo/MelDex HT, 26pts, 81.3%), whereas bortezomib based chemotherapy (CyBorD) was applied in 6pts (18.7%). High-dose treatment (HDT, Melphalan 200mg/m2 ) with autologous stem cell transplantation (ASCT) was performed in 3pts (9.4%). Overall treatment response (ORR, ≥PR) was achieved in 23pts (71.8%). Median duration of PFS in patients treated with standard chemotherapy was 32m (range 3-45m), while median OS was 40m (range 6-51m) indicating lack of availability of adequate second line treatment. Complete remission (CR) was achieved in all of 3pts treated with HDT+ASCT, with PFS1 36m; PFS2 48m; and PFS3 72m; while OS was as follows: OS1 36m; OS2 62m; and OS3 90m.

In conclusion, early recognition of disease and avoidance of organ involvement is of essential importance for the course of disease and treatment outcome, with necessity of further analyses of optimal treatment approach in relapse of disease.

Keywords: Amyloidosis, Treatment, Prognosis
PROTEOMIC AND MICROSCOPIC ANALYSIS OF SUBCUTANEOUS FAT ASPIRATES – SINGLE CENTER EXPERIENCE

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3 Institute of Molecular and Translational Medicine, Faculty of Medicine and Dentistry, Palacky University Olomouc, Olomouc, Czech Republic.

Introduction: The diagnostic algorithm in patients with suspected systemic amyloidosis commonly starts with a non-targeted biopsy and an abdominal fat aspiration has become an ideal diagnostic tool. Examination of Congo red-stained smears is simple and low-cost diagnostic approach. The proteomic analysis of fat tissue samples has a goal to reduce the organ biopsies while obtaining relevant information on the proteomic profile of the aspirated sample.

Objectives: The purpose of the study was to compare two diagnostic approaches to subcutaneous fat aspiration samples (namely microscopic examination of Congo red-stained smears and proteomic analysis) and the question was whether the microscopic method is of satisfactory quality for identification of systemic amyloidosis in standard equipped pathology departments.

Methods: 35 patients underwent FNAB from subcutaneous fat as a part of routine examination for suspected amyloidosis. The obtained tissue was divided into two parts and processed as follows: one part into Congo red stained smears and second part was sent to proteomic analysis.

Results: The studied group consisted of 19 patients with systemic amyloidosis and 16 negative cases; 17 patients with lambda light chain amyloidosis and two patients with the kappa subtype. In comparison of both methods, we can see an identical sensitivity of 95%. The microscopic examination showed 100% sensitivity and the proteomic analysis demonstrated a sensitivity of only 75%. Both the positive predictive value (82%) and the negative predictive value (92%) were lower in the proteomic analysis than in the microscopic method (100% and 94%, respectively). Ten samples showed blood contamination, which brought difficulties in proteomic analysis in five patients (four false positive results and one sample inaccurately identified as kappa AL although the patient suffered from lambda AL amyloidosis).

Conclusions: Based on this study we summarize, that microscopic examination of Congo red-stained smears provides reliable information about the presence of amyloid in the tissue, when it is adequately performed in experienced hands. By contrast, proteomic analysis allows amyloid diagnosis and typing at the same time and specification of the entire protein spectrum. Therefore, proteomic analysis plays a unique role in complicated cases, e.g. in cases of biopsy samples where standard diagnostic methods failed.

Supported by grant from the Ministry of Health of the Czech Republic (16-31156A).
GLYCOSYLATION OF IMMUNOGLOBULIN LIGHT CHAINS IS ENRICHED IN PATIENTS WITH IGM LIGHT CHAIN AMYLOIDOSIS

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2 Mayo Clinic, Rochester, MN, USA.

Introduction: The primary structure of light chains (LCs), including sequence and post-translational modifications, is thought to be important for their amyloidogenic potential. Post-translational glycosylation is seen more often in patients with light chain amyloidosis (AL) compared with other plasma cell disorders (17.5% vs. 4.1%, p<0.001). (Kumar S et al. Leukemia 2019 and Milani P et al, American Journal of Hematology 2017). It has been observed in one-third of κ-AL and 10% of λ-AL patients. This study evaluates LC glycosylation in patients with IgM amyloidosis to characterize the role of glycosylation in IgM amyloidogenesis.

Methods: Patients with newly diagnosed IgM AL seen at our institution from 01/2006 to 12/2015 with cryopreserved serum samples in an IRB approved biobank were included (N=71). Glycosylation was assessed on cryopreserved serum samples by immuno-enrichment-based matrix-assisted laser desorption ionization time-of-flight mass spectrometry (MALDI-TOF-MS), termed MASS-FIX. Glycosylated LCs were identified by the shift in their molecular mass due to the added molecular mass of the carbohydrates. Consecutive patients with IgM gammopathies other than AL amyloidosis, who underwent MASS-FIX from 07/23/2018 to 05/14/2019 served as a control group.

Results: The median age at diagnosis was 69 years. Involved LC was λ in 68% and κ in 32% of patients. The median dFLC was 12.5 g/dL. The median NTProBNP was 1576 pg/mL. The distribution per 2012 Mayo stage I-IV was (%): 36/22/20/22. Organ involvement was as follows (%), heart: 58%, kidney: 49%, soft tissue: 39%, peripheral nerve: 34%, liver: 10%, and lung: 7%. (Table 1) Glycosylation was present in 21% (N=15) of patients with IgM amyloidosis; in 26% (6/23) of κ-IgM AL patients and in 19% (9/48) of λ-IgM AL patients (p=0.5). There was no statistically significant difference in baseline laboratory characteristics or organ involvement in patients with and without glycosylated LCs. (Table 1), including involved LC type, BM clonal cell percentage, dFLC levels, cardiac biomarker levels, presence of MYD88 mutation or organ involvement. In the control group of patients with non-amyloid IgM gammopathies, glycosylation was seen in 7% (31/418) of patients compared with 21% in the IgM amyloid cohort (p=0.001). The primary diagnoses in the control group included: MGUS (49%), Waldenstrom’s Macroglobulinemia (31%), chronic lymphocytic lymphoma (4%), multiple myeloma (2%) and others (13%).

Conclusions: Glycosylation of LCs was significantly more common in IgM amyloid patients compared to patients with non-AL IgM gammopathies (21% vs. 7%, p=0.001). This is similar to observations in all patients with AL amyloidosis. This suggests that glycosylation plays a similar role in IgM AL amyloidogenesis as it does in non IgM amyloidosis patients. The presence of glycosylation on mass spectrometry in patients with IgM monoclonal gammopathies should alert physicians to the possibility of amyloidosis.
Table 1: Baseline characteristics of IgM AL amyloidosis patients with and without light chain glycosylation.

<table>
<thead>
<tr>
<th></th>
<th>All IgM AL patients, N=71</th>
<th>Glycosylated LC, N=15</th>
<th>Non-glycosylated LC, N=56</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age, years</strong></td>
<td></td>
<td></td>
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<td></td>
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<tr>
<td></td>
<td>69 (63-73)</td>
<td>69 (65-71)</td>
<td>69 (63-74)</td>
<td>0.8</td>
</tr>
<tr>
<td><strong>Sex, males</strong></td>
<td>53 (75)</td>
<td>13 (87)</td>
<td>40 (71)</td>
<td>0.2</td>
</tr>
<tr>
<td><strong>iFLC- Lambda</strong></td>
<td>48 (68)</td>
<td>9 (60)</td>
<td>39 (70)</td>
<td>0.5</td>
</tr>
<tr>
<td><strong>BM clonal cells %</strong></td>
<td>10 (5-25)</td>
<td>16 (5-53)</td>
<td>10 (5-22)</td>
<td>0.3</td>
</tr>
<tr>
<td><strong>Serum M-protein, g/dL</strong></td>
<td>1.0 (0.6-1.5)</td>
<td>1 (0.7-1.5)</td>
<td>1 (0.5-1.6)</td>
<td>&gt;0.99</td>
</tr>
<tr>
<td><strong>dFLC, mg/dL</strong></td>
<td>12.5 (3.7-36.2)</td>
<td>12.5 (3.6-49.6)</td>
<td>12.5 (3.4-26.6)</td>
<td>0.5</td>
</tr>
<tr>
<td><strong>Troponin-T, ng/mL</strong></td>
<td>0.01 (0.01-0.06)</td>
<td>0.02 (0.01-0.06)</td>
<td>0.01 (0.01-0.07)</td>
<td>0.9</td>
</tr>
<tr>
<td><strong>NTProBNP, pg/mL</strong></td>
<td>1576 (221-4137)</td>
<td>1696 (241-4200)</td>
<td>1576 (221-4137)</td>
<td>0.8</td>
</tr>
<tr>
<td><strong>GFR, ml/min/m²</strong></td>
<td>72 (43-84)</td>
<td>69 (45-83)</td>
<td>73 (42-85)</td>
<td>0.6</td>
</tr>
<tr>
<td><strong>24 hour urine protein, mg</strong></td>
<td>660 (144-4031)</td>
<td>438 (127-817)</td>
<td>879 (144-5206)</td>
<td>0.3</td>
</tr>
<tr>
<td><strong>Alkaline phosphatase IU/mL</strong></td>
<td>87 (72-113)</td>
<td>97 (68-131)</td>
<td>86 (73-111)</td>
<td>0.7</td>
</tr>
<tr>
<td><strong>Mayo 2012 stage (%)</strong></td>
<td>21/13/12/13, N=59</td>
<td>4/2/4, N=12</td>
<td>17/11/10/9, N=47</td>
<td>0.8</td>
</tr>
<tr>
<td></td>
<td>(36/22/20/22)</td>
<td>(33/17/17/33)</td>
<td>(36/23/21/19)</td>
<td></td>
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<tr>
<td><strong>Mayo 2004 Stage (%)</strong></td>
<td>18/20/21, N=59</td>
<td>5/3/4, N=12</td>
<td>13/17/17, N=47</td>
<td>0.6</td>
</tr>
<tr>
<td></td>
<td>(31/34/36)</td>
<td>(42/25/33)</td>
<td>(28/36/36)</td>
<td></td>
</tr>
<tr>
<td><strong>Renal stage, 1/2/3</strong></td>
<td>36/4/10, N=60</td>
<td>8/3/1, N=12</td>
<td>28/11/9, N=48</td>
<td>0.7</td>
</tr>
<tr>
<td></td>
<td>(60/23/17)</td>
<td>(67/25/8)</td>
<td>(58/23/19)</td>
<td></td>
</tr>
<tr>
<td><strong>FISH, trisomy/tetrasomy</strong></td>
<td>4/32 (13)</td>
<td>2/7 (29)</td>
<td>2/25 (8)</td>
<td>0.2</td>
</tr>
<tr>
<td><strong>FISH, t(11;14)</strong></td>
<td>8/32 (25)</td>
<td>2/6 (33)</td>
<td>6/26 (23)</td>
<td>0.6</td>
</tr>
<tr>
<td><strong>MYD88&lt;sup&gt;L265P&lt;/sup&gt; mutation</strong></td>
<td>28 /47 (60)</td>
<td>8/10 (80)</td>
<td>20/37 (54)</td>
<td>0.1</td>
</tr>
</tbody>
</table>

**Organ involvement**

<p>| | | | | |</p>
<table>
<thead>
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<tbody>
<tr>
<td><strong>Heart</strong></td>
<td>41 (58)</td>
<td>7 (47)</td>
<td>34 (61)</td>
<td>0.3</td>
</tr>
<tr>
<td><strong>Renal</strong></td>
<td>35 (49)</td>
<td>5 (33)</td>
<td>30 (54)</td>
<td>0.2</td>
</tr>
<tr>
<td><strong>Liver</strong></td>
<td>7 (10)</td>
<td>2 (13)</td>
<td>5 (9)</td>
<td>0.6</td>
</tr>
<tr>
<td><strong>Gastrointestinal</strong></td>
<td>14 (20)</td>
<td>5 (33)</td>
<td>9 (16)</td>
<td>0.1</td>
</tr>
<tr>
<td><strong>Autonomic nerves</strong></td>
<td>9 (13)</td>
<td>3 (20)</td>
<td>6 (11)</td>
<td>0.4</td>
</tr>
<tr>
<td><strong>Peripheral nerve</strong></td>
<td>24 (34)</td>
<td>3 (20)</td>
<td>21 (38)</td>
<td>0.2</td>
</tr>
<tr>
<td><strong>Soft tissue</strong></td>
<td>28 (39)</td>
<td>7 (47)</td>
<td>21 (38)</td>
<td>0.5</td>
</tr>
<tr>
<td><strong>Lung</strong></td>
<td>5 (7)</td>
<td>0 (0)</td>
<td>5 (9)</td>
<td>0.6</td>
</tr>
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</table>
A NOVEL MASS SPECTROMETRY-BASED METHOD FOR THE IDENTIFICATION OF SUBTYPE SPECIFIC AMYLOIDOGENIC PROTEINS

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Background: Mass spectrometry-based proteomics (MS) is a powerful technology for large scale analysis of proteins and has – in combination with laser dissection microscopy (LDM) – become an indispensable tool for the identification of the amyloidogenic protein in a quantitative manner. LDM-MS does not only identify the subtype-specific protein but also measures an amyloid protein signature that is shared across all amyloidosis subtypes in various tissues. This protein signature consists of serum amyloid P, and the apolipoproteins A4, and E. Identification of the subtype-specific protein from MS-data and evaluation of the amyloid protein signature relies on manual inspection of MS-data. In clear-cut cases with extreme levels of the amyloidogenic protein, this is not of concern. Oftentimes, the MS-data are ambiguous, and a certain degree of subjective interpretation by an MS expert is required. This subjective interpretation is un-desirable as it may bias the result leading to wrong subtype diagnosis. In the presented work we therefore build a classification model for the objective evaluation of MS-data from the analysis of amyloid deposits.

Methods: 85 Congo Red (CR) positive biopsies and 55 CR negative biopsies from various tissues were subjected to LDM-MS and validated by IEM analysis as previously described (Abildgaard et al, 2019, Amyloid). Protein data was randomly divided into a training and test set. 42 prospectively collected samples were used as a validation set. Statistical analysis of the data from the LDM-MS analysis was done in R statistical software (version 3.6.0). Identification of novel amyloid signature proteins was done by the Boruta feature selection method whereas a Support Vector Machine algorithm (SVM) was used for building classifiers to differentiate between CR positive and negative biopsies, as well as to identify subtype proteins in CR positive biopsies based on the amyloidogenic proteins Ig-K, Ig-L, SAA and TTR.

Results: The Boruta algorithm estimates the capacity each of the 1866 proteins of the merged dataset to differentiate between the CR-positive and CR-negative samples. Among the top six most differentiating proteins, the algorithm identified ApoA4, ApoE, SAP and Vitronectin, as well as 2 proteins that have not previously been associated with amyloid deposits, Clusterin and Complement component C9. The classifiers build using the SVM method demonstrated that this method correctly identified the 42 amyloidosis patients from the validation cohort when training the classifier with the top 6 proteins as identified with the Boruta algorithm. When training the classifier with the four most common amyloidogenic proteins 40 out of the 42 patients where correctly subtyped.

Conclusions: In present study we identified 2 novel amyloid deposit signature proteins, Clusterin and Complement component C9. Moreover, we developed a statistical approach that objectively identified the correct subtype in 40 out of 42 cases.
THE VALUE OF PROTEOMIC ANALYSIS OF SCANTY AMYLOID DEPOSITS IN BONE MARROW TREPINES AND BONE

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Introduction: Bone marrow trephines (BMTs) are commonly analysed histologically for the identification, typing and diagnosis of amyloid. Usually bone samples are decalcified, formalin-fixed and paraffin-embedded (FFPE) for amyloid identification and typing using Congo red (CR) and immunohistochemistry (IHC). When present in bone, amyloid deposits are usually scanty and are often ‘cut through’ on serial sections making fibril typing by IHC difficult. Interpretation of CR staining in cortical bone can be problematic, where the prevalence of collagen can masquerade as amyloid. Some BMTs are processed in resin medium, which avoids decalcification and preserves tissue morphology well, but does not always give conclusive immuno-typing.

Objectives: To assess the value of proteomic analysis in the identification and typing of scanty amyloid deposits in BMTs and bone specimens received at our centre as either FFPE or resin-embedded samples and in confirming whether or not CR-staining in cortical bone represents true amyloid by looking for amyloid signature proteins.

Methods: Over a 3-year period, 95 BMTs/bone biopsies were chosen for proteomic analysis when CR staining was positive but IHC failed to definitively type the amyloid fibril, and in cases where CR staining was equivocal. Liquid Chromatography Mass Spectrometry was performed on the Thermo Scientific™ Q-Exactive Plus Orbitrap and data was interpreted using MASCOT software and the Swiss-Prot human database. Analyses were interpreted by two experienced operators, blind to clinical details.

Results: Of 95 samples, 86 were CR positive and 9 were equivocal for the presence of amyloid by CR. CR positivity was supported by the presence of at least 2 amyloid signature proteins (serum amyloid P component, apolipoprotein A-IV or apolipoprotein E) in 69/86 (80%) of CR positive samples, but not in 4/86 (5%) of CR positive samples. 13/86 (15%) of CR positive samples gave inadequate results by proteomic analysis. 7/9 (78%) of samples that were equivocal by CR did not demonstrate an amyloid signature and 2/9 (22%) showed the amyloid signature. In 21/86 (24%) CR positive cases, fibril type was suggestive by IHC. Fibril types were confirmed by proteomics in 15/21 (71%) of these cases. Fibril typing was not achieved by IHC in 65/86 (76%) CR positive samples, 49/65 (75%) of which went on to be typed by proteomic analysis. 64/86 (74%) of all CR positive samples were typed by proteomics. Six samples were processed in resin; 1/6 of these was typed by IHC and 6/6 were typed by proteomics. All other samples were FFPE.

Proteomics confirmed the presence of amyloid in 29/36 (81%) of cortical bone samples that stained positively with CR.

Conclusions: When used alongside CR and IHC, proteomic analysis is a useful tool in the diagnosis of amyloid in BMT and bone samples and should be interpreted in the context of the overall clinical picture.

Keywords: Bone marrow trephines, Immunohistochemistry, Proteomic analysis
PHYSIOLOGIC ANALYSIS OF SERUM AMYLOID A (SAA) GENES AND PROTEINS

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Introduction: SAA proteins, evolutionarily well-conserved, have been implicated in multiple physiologic pathways. However, detailed understanding of function(s) for these molecules has been elusive.

Objectives: Clarify specific SAA species in human physiology and cellular interaction(s) involved


Results: Human colostrum specimens contain full-length SAA proteins corresponding uniquely to products of the SAA1 gene; there was no evidence for the SAA3 protein. DNA sequence comparison of all reported mammalian SAA3 gene sequences shows a single nucleotide (A) insertion that is limited to humans and bonobos predicting a shortened reading frame and a premature stop codon. Although the human SAA3 gene is transcribed the location of the insertion relative to established splice junctions likely leads to nonsense-mediated mRNA decay; no protein corresponding to an altered and/or shortened transcript has been detected. SAA1 production in colostrum is not apparently related to acute phase stimulus(i) that is(are) well-established in other systems (e.g. liver). SAA1 interaction with the gastrointestinal tract of newborns may provide an early protective response, consistent with the relative absence of SAA species among the proteins in mature milk. Further studies of SAA interactions with more cell types is justified by these observations and is currently the focus of study.

Conclusions: SAA1 is the only member of the SAA family found in human colostrum. Comparing evolution of SAA3 gene nucleotide sequences shows a single nucleotide insertion in human and bonobos. This predicts nonsense-mediated mRNA decay for nascent SAA3 transcripts, consistent with the absence of SAA3 protein in our specimens. Control of SAA1 transcription in human mammary epithelium likely is mediated by hormonal exposure (e.g. progesterone) rather than acute phase factors, implying additional complexity in the control of this locus. A protective effect of SAA1 on the gastrointestinal tract of the newborn implies interaction of this protein with cell types that have not been evaluated previously. The importance of SAA1 as a participant in survival and protective physiology is consistent with these observations and implies that both control of SAA1 production as well as cellular responses to the protein itself are subject to a wider array of stimuli than hitherto recognized.

Keywords: Serum Amyloid A (SAA); gene structure; hormone; nonsense-mediated mRNA decay; neonatology
PT065

GATEWAY AND JOURNEY OF PATIENTS WITH CARDIAC AMYLOIDOSIS

O. Lairez

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Background: The high variability in clinical manifestations of cardiac amyloidosis (CA) can lead to chaotic care pathway and delays between symptom onset and correct diagnosis.

Objectives: To describe the journey to CA diagnosis from initial clinical and to analyze time to diagnosis. Methods: Between January 2001 and May 2019, 270 consecutive patients with CA diagnosed at Toulouse University Hospital were retrospectively included in this cross-sectional study: 111 (41%) light chain amyloidosis (AL), 122 (45%) wild-type transthyretin amyloidosis (ATTRwt) and 37 (14%) hereditary transthyretin amyloidosis (ATTRv). Journey to diagnosis including symptoms, first-line specialist, number of specialists consulted and tests performed before diagnosis were investigated.

Results: CA onset occurred mostly with dyspnea (50%) or systematic follow-up (10%). Cardiologist was the first-line specialist in 68% of patients, followed by nephrologist (9%) and neurologist (8%). Patients encountered a median (minimum-maximum) number of 2 (1-7) physician specialists and performed a median (minimum-maximum) number of 3 (1-8) tests before diagnosis. Median delay between symptom onset and CA diagnosis was 8 [IQR 5-14], 10 [IQR 3-34] and 18 [IQR 4-49] months, respectively in AL, ATTRwt and ATTRv subgroups, p=.060). Having performed an electromyography or a spirometry was associated with a longer delay in diagnosis in the overall population: odds ratio - OR=1.13; 95% confidence interval,1.02 to 1.24 and OR=1.13; 1.03 to 1.24, respectively, probably due to aspecific initial symptoms.

Conclusion: CA is a protean disease with various first-line specialists causing a diagnostic wandering despite increasing medical community awareness. It requires a multidisciplinary specialist care networks to educate and manage symptoms and therapies.
PREVALENCE, DETERMINANTS AND PROGNOSTIC VALUE OF ECHOCARDIOGRAPHIC LEFT VENTRICULAR DIASTOLIC FUNCTION PATTERNS IN CARDIAC AMYLOIDOSIS

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Introduction: Cardiac amyloidosis (CA) is an increasingly recognized cause of restrictive cardiomyopathy but its prognosis remains pejorative because of late recognition. The ASE/EACVI guidelines for the evaluation of left ventricular diastolic function (LVDF) have never been applied to CA.

Objectives: To assess the pattern of LVDF in CA according to these new recommendations, to identify their associated phenotypes and determinants.

Methods: We conducted a monocentric, observational study on a large cohort of confirmed CA. We analysed their diastolic function by standard echocardiography and collected their clinical, biological and survival parameters.

Results: 464 patients with CA balanced between the three main types (AL, hereditary or wildtype TTR) were included; 41% had a restrictive mitral pattern (diastolic dysfunction grade III), 25% had diastolic function grade II, and 25% had diastolic dysfunction grade I; 9% were unclassified. No difference was found between the main types of CA. After multivariate analyses, grades II and III (increase of LV filling pressures) were independently associated with dyspnoea, higher NT-proBNP level, cardiac infiltration (interventricular septal thickness) and systolic dysfunction (assessed by global longitudinal strain). Grade I patients had a better prognosis than grades II and III despite an early impairment of systolic function especially showed by mitral s’ velocity (< 9cm/s).

Conclusions: CA cannot be ruled out in patients with mild impairment of diastolic function. S’ velocity being the most constantly impaired parameter, could be useful for considering diagnosis of CA in the absence of restrictive pattern.
PREVALENCE AND CHARACTERISTICS OF TRANSTHYRETIN AMYLOID CARDIOMYOPATHY IN HOSPITALIZED PATIENTS REFERRED FOR TECHNETIUM PYROPHOSPHATE SCAN

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Background: Understanding the characteristics of patients with a final diagnosis of transthyretin amyloid cardiomyopathy (ATTR-CA) may help optimally triage patients for Tc99m pyrophosphate (PYP) scanning.

Objective: The study was aimed to investigate the prevalence as well as the clinical features along with the electrocardiographic and echocardiographic characteristics of patients with ATTR-CA, who were referred for PYP scan after being hospitalized for decompensated heart failure.

Methods: We included all the patients who were referred for PYP scan after heart failure-related hospitalization at our center between 06/2015 and 06/2019. ATTR-CA was diagnosed by positive PYP scan and negative serum studies for AL amyloidosis; gene testing results were used to differentiate between wild-type ATTR-CA and hereditary ATTR-CA. The transthoracic echocardiogram and electrocardiogram were included which were performed around the time of PYP scan. The echocardiographic measurements were based on the American Society of Echocardiography guidelines. The medical history was obtained through chart review.

Results: Of the 155 patients who underwent PYP scan, 45 (29%) were PYP +ve. All PYP +ve patients were wild-type ATTR-CA. In the PYP negative group, 8 patients (7.2%) had biopsy-proven AL amyloidosis. Compared to the PYP -ve group, the PYP +ve group had similar age (mean 79.5 vs 78.1 years, p=0.9), BMI (28.2 kg/m2 vs 30.1 kg/m2, p=0.7) and a higher prevalence of carpal tunnel syndrome (62% vs 15%, p< 0.01). PYP +ve patients also had lower left atrial volume index (LA VI) (43 mL/m2 vs 59 mL/m2, p=0.001), greater left ventricular septal thickness (1.67cm vs 1.5cm, p=0.01), increased posterior wall thickness (1.5cm vs 1.3cm, p=0.02) and worse diastology (2.6±0.5 vs 1.7± 0.8, p< 0.001). There was no difference in ejection fraction (51% vs 50%, p=0.7) or pulmonary arterial systolic pressure (39 mmHg vs 37.5 mmHg, p=0.9).

Conclusion: At our center, almost one-third of the hospitalized patients who were clinically suspected to have cardiac amyloidosis had a final diagnosis of wild-type ATTR-CA. A history of carpal tunnel syndrome, ECG findings of low QRS voltage, AVB and LAFB, and echocardiographic features of left ventricular hypertrophy and high-grade diastolic dysfunction were prevalent in wild-type ATTR-CA.
NON-INVASIVE DIAGNOSIS OF CARDIAC AMYLOIDOSIS USING AMYLOID IMAGING

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Introduction: Cardiac amyloidosis commonly occurs in systemic amyloidosis patients and is the most serious clinical complication. Immunoglobulin light chain (AL), hereditary transthyretin (ATTRv), and wild-type transthyretin (ATTRwt) amyloidosis are major systemic amyloidosis that involve the heart, and have each different pathological background, prognosis, and therapeutic approaches. Therefore, early detection of amyloidosis and early determination of its subtype is critically important to provide better outcome.

Objectives: The aim of this study is to investigate the utility of combination use of 11C-Pittsburgh compound B (11C-PiB) positron emission tomography (PET) imaging and 99mTc-Pyrophosphate (99mTc-PYP) scintigraphy for detecting and differentiating three major types of cardiac amyloidosis, AL, ATTRv, and ATTRwt amyloidosis.

Methods: Whole body 11C-PiB PET and 99mTc-PYP scintigraphy were performed in 17 AL amyloidosis, 22 ATTRv, and 8 ATTRwt amyloidosis patients. Correlation between organ involvement and uptake of 11C-PiB and 99mTc-PYP were analyzed in each patient.

Results: Cardiac amyloidosis was detectable by using 99mTc-PYP scintigraphy or 11C-PiB PET in all systemic amyloidosis patients with cardiac involvement. 99mTc-PYP scintigraphy and 11C-PiB PET showed an interesting complementary relation. Strict combination of positive 11C-PiB and negative 99mTc-PYP uptake (PiB pattern) was observed in all AL amyloidosis patients with cardiac involvement. In contrast, strict combination of positive 99mTc-PYP and negative 11C-PiB uptake (PYP pattern) was observed in all ATTRwt amyloidosis patients with cardiac involvement. ATTRv amyloidosis patients with cardiac involvement were divided into two groups; PiB pattern or PYP pattern. All the early-onset (disease onset age <50) V30M (p.V50M) ATTRv patients showed PiB pattern, whereas all the late-onset (disease onset age ≥50) V30M and non-V30M ATTRv patients showed PYP pattern.

Conclusions: All three major types of cardiac amyloidosis can be detected and differentiated non-invasively by combined use of the two amyloid imaging methods and TTR gene test.

Key words: amyloid, Pyrophosphate scintigraphy, Pittsburgh compound B PET.
DISCORDANT RESPONSE OF SERUM BIOMARKERS IN PATIENTS WITH CARDIAC AL AMYLOIDOSIS NOT RECEIVING IMIDS. SHOULD ORGAN RESPONSE DEFINITION BE REVISITED?

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Background: Staging systems in AL amyloidosis are mostly based on cardiac biomarkers (NT-ProBNP and Troponin). In addition, cardiac response is defined by a NT-ProBNP decrease >30% and 300 mg/L (in subjects with baseline NT-ProBNP>650 ng/L) or NYHA improvement of at least 2 classes. In patients treated with immunomodulatory drugs (IMIDs) a paradoxical initial increase in NT-ProBNP levels has been described, making difficult the evaluation of cardiac organ response. However, in some patients not receiving IMIDs a discordant evolution of serum cardiac biomarkers (troponin decrease but stable NTproBNP) can be observed. These patients do not comply with current accepted criteria for organ response.

Methods: The evolution of 5 patients with cardiac AL amyloidosis, not receiving IMIDs, who displayed discordant response between high-sensitivity troponin T (TnT) and NTProBNP is presented.

Results: Three male and 2 female patients (age: 41-70 years) are presented. Four of them were patients with stage IIIa newly-diagnosed cardiac AL amyloidosis who started treatment with CyBorD (followed by autologous stem cell transplant [ASCT] in 2 cases and by daratumumab in one patient) while the fifth patient suffered a stage IIIb relapse after BMDex and ASCT and started treatment with daratumumab. In 3 patients, a complete hematologic response (with positive minimal residual disease) was obtained and in the other 2 a very good partial response was achieved. Although NT-ProBNP remained stable during follow-up (range, 8-24 months), a decrease in TnT ranging between 30% to 85% was observed (Table 1). No relevant changes in creatinine clearance during treatment could justify these findings in any case. In four of the five patients, an improvement in the functional NYHA class was associated.

Conclusions: A discordant response of cardiac biomarkers with important decrease of troponin without significant NT-proBNP changes can be observed in some patients with cardiac AL amyloidosis not receiving IMIDs. Whether the current criteria for cardiac response should consider this phenomenon should be evaluated in larger series of patients.

Keywords: AL amyloidosis, cardiac biomarkers, organ response.
A NOVEL METHOD FOR AMYLOID DETECTION IN HUMAN TISSUES

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Fluorescent probes identifying amyloid deposits are of great interest as such deposits are the pathological hallmark of many diseases. In this work, we present a novel method for the detection of amyloid in human tissues based on the use of a new fluorescent dye – disodium salt of 2,7-(1-amino-4-sulfo-2-naphthylazo) fluorene (DSNAF). Synthesis of DSNAF was performed by diazotization of 2,7-diaminofluorene in a stream of argon, followed by azo coupling with naphthionic acid. Structural characterization of DSNAF was performed using MALDI mass spectrometry. Myocardial autopsy samples from 11 random individuals (males and females, >85 years old) with cardiac amyloidosis diagnosis were the material for histological study. Paraffin sections of the myocardium were stained with a 0.1% aqueous solution of Congo red or with an aqueous solution (0.1% or 0.034%) of DSNAF under the same conditions. Both, spectral characteristics of the dyes and fluorescence intensity of Congo red/DSNAF upon binding to amyloid were analyzed using confocal laser scanning microscopy. We have demonstrated for the first time that new fluorene-based analogue of Congo red, DSNAF, can be successfully used to identify amyloid deposits in histological sections of human myocardium. In terms of the specificity and intensity of amyloid staining, DSNAF is comparable to Congo red, which is the gold standard for amyloid detection. The fluorescence intensity of DSNAF upon binding to amyloid fibrils is significantly higher than the intensity of Congo red fluorescence, and the background fluorescence intensity of cardiac muscle tissue is lower in the case of DSNAF. This makes it possible to detect small amyloid deposits in the tissues with high accuracy. The found advantages of using DSNAF allow us to consider the developed technology for the detection of amyloid as a promising new method for the identification of amyloid deposits in human tissues.

Keywords: amyloid imaging, fluorescent dye, Congo red

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CLINICAL CHARACTERISTICS, NATURAL HISTORY AND PROGNOSTIC FACTORS IN PATIENTS WITH LIGHT CHAIN AND TRANSTHYRETIN CARDIAC AMYLOIDOSIS

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Introduction and objectives: Light-chain amyloidosis (AL) and transthyretin amyloidosis (ATTR) are the most common types of cardiac amyloidosis (CA). We sought to study the clinical characteristics and prognosis of both diseases.

Methods: We conducted a single-center, retrospective review of all patients diagnosed from CA between 1998 and 2018. Clinical characteristics, complementary tests, survival and other adverse clinical events were studied.

Results: We identified 105 patients with CA, 65 ATTR-CA and 40 AL-CA. Mean age was 74.4 years; 24.8% were women. In both groups, heart failure was the most frequent clinical presentation (55.2%). The most prevalent electrocardiographic findings were the pseudoinfarct pattern (68.5%) and a Sokolow-Lyon index <1.5mV (67.7%), with no differences between the two subtypes of CA. One-year, 3-year, and 5-year survival was 43.3%, 40.4% y 35.4%, respectively, in AC-AL patients, and 85.1%, 57.3% and 31.4% in AC-ATTR patients (p=0.004).

AL-CA subtype (HR 3.41, CI95% 1.45-8.06, p=0.005), previous admission for heart failure (HR 4.25, CI95% 1.63-11.09, p=0.003) and a NYHA class III-IV (HR 2.76, CI95% 1.09-7.03, p=0.033) were independent predictors of mortality, while beta-blocker therapy was associated with longer survival (HR 0.23, CI95% 0.09-0.59, p=0.002).

Conclusions: There are significant differences with regard to the clinical presentation of AL-CA and ATTR-CA. Both entities are associated to poor prognosis.

Keywords: cardiac amyloidosis, transthyretin, light chain.
IN CARDIAC AL AMYLOID A HIGHER BODY MASS INDEX IS ASSOCIATED WITH A LOWER RATE OF CARDIAC RESPONSE

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Introduction: Light-chain amyloidosis (AL) is a clonal plasma cell disorder in which Ig light chains cause organ-specific disease due to toxic misfolded light-chain aggregates and extracellular deposition of amyloid fibrils. Approximately half of AL amyloid patients present with cardiac involvement and survival is largely driven by the extent of heart failure. In the general heart failure population, overweight and mild/moderate obesity is associated with lower mortality, termed the obesity survival paradox (Vest et al 2015). Conversely for patients with multiple myeloma, a disease similar in pathophysiology to AL, obesity is associated with increased mortality (Teras et al 2014).

Objective: We hypothesized that patients with cardiac amyloidosis would exhibit an obesity survival paradox and sought to determine the impact of body mass index (BMI) on mortality, hematological, and cardiac responses to anti-plasma cell treatment.

Methods: We conducted a single tertiary center retrospective study of consecutive patients with cardiac AL amyloidosis, referred between 1/1/2009 and 09/30/2018. We collected demographics and BMI prior to treatment. We recorded the date of diagnosis and subsequent dates of hematological and/or cardiac response, mortality or end of follow-up. We constructed a Cox proportional hazards model examining the association between BMI and mortality with a restricted cubic spline function curve. Logistic regression models were constructed to examine the association between high BMI (≥ 25 kg/m2) and cardiac or hematological response. Models were adjusted for age, sex and cardiac stage at time of diagnosis.

Results: Of 79 patients, 17 patients had BMI of 17-22.5, 19 a BMI of 22.6-25, 23 a BMI of 25.1-29.7, and 20 a BMI of ≥ 30 kg/m2. Crude mortality was 31/79 (39%). There was no relationship between BMI as a continuous variable and mortality (adjusted HR 0.98, 95% CI 0.91-1.06, p=0.625), although a survival paradox trend was suggested by the cubic spline curve. While there was no relationship between high BMI and hematological response (adjusted OR 1.00, 0.37-2.75, p=0.996), there was a relationship between high BMI and lower likelihood of achieving cardiac response (adjusted OR 0.23, 0.07-0.71, p=0.011).

Conclusions: In this small cohort of patients with AL cardiac amyloidosis, there was no significant relationship between BMI and mortality. Hematological response was unrelated to BMI, but patients with a higher BMI were significantly less likely to achieve a cardiac response. These findings suggest that patients with higher BMI might be associated with poorer cardiac outcomes in AL amyloidosis, highlighting the importance of a multidisciplinary approach involving oncologists, cardiologists, and nutritionists in the treatment of this very complex multi-organ disease.
QUANTITATIVE AND FUNCTIONAL EVALUATION OF ENDOTHELIAL PROGENITOR CELLS IN PATIENTS WITH CARDIAC AMYLOIDOSIS

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Background: Endothelial progenitor cells (EPCs) have an important role in the process of vascular repair by promoting re-endothelialization following endothelial injury. Endothelial microvascular dysfunction is a known mechanism of injury of cardiac amyloidosis (CA), but evidence regarding EPCs’ levels and function in patients with CA is lacking.

Objectives: We aimed to define EPCs’s quantitative and functional profile in patients with CA and to explore a possible effect of amyloid-targeted therapies.

Methods: Study population included patients with light-chain or transthyretin (TTR) CA. Patients with diagnosed heart failure and preserved ejection fraction (HFrEF) with negative monoclonal gammopathy and TTR-incompatible 99mTc-DPD scans were used as controls. Blood circulating EPCs were assessed quantitatively by the expression of VEGFR-2, CD34 and CD133 using flow cytometry, and functionally by the formation of colony forming units (CFUs) and MTT assay. Tests were repeated 3 months following the initiation of targeted therapies (either tafamidis or chemotherapy) in CA patients.

Results: Our preliminary cohort include 13 CA patients (median age 74 years, 62% TTR CA). Patients with CA vs. patients with HFrEF demonstrated diminished expression of CD34(+)/VEGFR-2(+) [0.51% (IQR 0.4, 1.1) vs. 1.18% (IQR 0.9, 1.6), P=0.025] and CD133(+)/VEGFR-2(+) [0.35% (IQR 0.23, 0.61) vs. 1.34% (IQR 1.1, 1.5), P=0.014]. Functionally, no differences were noted between groups. Following the initiation of amyloid-targeted therapies in CA patients, we observed increased expression of CD34(+)/VEGFR-2(+) [2.47% (IQR 2.1, 2.7), P<0.001] and CD133(+)/VEGFR-2(+) [1.38% (IQR 1.1, 1.7), P=0.003]. Moreover, functionally, proliferation of EPCs was also increased microscopically (from 0.5 CFUs [IQR 0, 1.5] to 2 CFUs [IQR 1, 3.5], P=0.023), and when assessed by an MTT assay [0.12 (IQR 0.04, 0.12) to 0.24 (IQR 0.16, 0.3), p=0.014].

Conclusions: These preliminary results demonstrate reduced EPCs levels in CA amyloidosis patients indicating significant microvascular impairment. Amyloid-targeted therapies induce the activation of EPCs, thus possibly promoting endothelial regeneration. These findings may represent a novel mechanism of action of amyloid-targeted therapies.
PRIOR CARPAL TUNNEL SYNDROME AND EARLY CONCOMITANT ECHOCARDIOGRAPHIC FINDINGS AMONG PATIENTS WITH SYSTEMIC AMYLOIDOSIS

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Background: Amyloid deposits are found in the carpal tunnel of the hand prior to the diagnosis of immunoglobulin light chain (AL) or transthyretin (TTR) systemic amyloidosis, thus raising the potential for early diagnosis.

Objectives: To describe early echocardiographic parameters coincident with the diagnosis of carpal tunnel syndrome (CTS).

Methods: Single-center retrospective analysis of diagnosed AL and TTR patients. Echocardiographic examinations performed at the time of CTS diagnosis were reviewed for parameters suggestive of cardiac amyloidosis. Patients with known or suspected amyloidosis at the time of the echocardiography exam were excluded from the cohort.

Results: Included were 108 patients with confirmed AL (n=82) and TTR (n=26) amyloidosis in whom CTS was previously diagnosed in 25% and 62%, respectively. The median age at the diagnosis of CTS was 63 (IQR 56, 73) years, approximately 5 (IQR 2.9, 7) years before the diagnosis of systemic amyloidosis. Echocardiographic findings at the time of CTS diagnosis showed increased thickness of the intraventricular septum [1.3 (IQR 1.1, 1.5) cm], increased relative wall thickness [0.46 (IQR 0.40, 0.50)] and increased left ventricular mass [234 (IQR 177, 292) grams]. Of the 11 patients who underwent pulsed wave Doppler mitral flow evaluation, 55% had Doppler data supportive of abnormal left ventricular diastolic function.

Conclusion: Early echocardiographic findings coincident with CTS diagnosis and preceding the diagnosis of systemic amyloidosis are suggestive of concentric hypertrophy and diastolic dysfunction. Larger-scale prospective studies are warranted to define screening algorithms.
USE OF CHRONIC AMBULATORY INOTROPIC SUPPORT IN PATIENTS WITH END-STAGE CARDIAC AMYLOIDOSIS

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Introduction: Chronic ambulatory inotropic support is offered to selected patients with end-stage heart failure as palliative therapy. The tolerability and benefits of this approach for patients with end-stage heart failure secondary to cardiac amyloidosis are unknown.

Objectives: Describe the characteristics, duration of therapy, and outcomes of patients with end-stage cardiac amyloidosis treated with chronic ambulatory inotropic support.

Methods: Retrospective case reviews of patients with end-stage cardiac amyloidosis who were discharged home on either Milrinone or Dobutamine and continued to follow in our center. Our multidisciplinary amyloidosis center, uses the following criteria to initiate chronic ambulatory inotropic support in patients with end-stage cardiac amyloidosis: at least two hospitalizations for heart failure in 3 months, invasive hemodynamics demonstrating elevated filling pressures, cardiac index <2.0 ml/kg/m², inability to tolerate standard heart failure medical therapies, and ineligibility for advanced therapies (heart transplantation or durable mechanical support).

Results: A total of seven patients with end-stage heart failure were treated with chronic ambulatory inotropic support, 4 with wild-type ATTR amyloidosis, 2 with light chain amyloidosis and 1 with hereditary ATTR amyloidosis. The mean duration of therapy was 219 days. Six had systolic heart failure with left ventricular ejection fraction < 50%. The average number of hospitalizations 12 months before starting inotropes was 3.28. The average number of hospitalizations after inotropes was 1.14. Four patients are still alive and on therapy.

Conclusion: Research of outpatient inotrope use in patients with end stage cardiac amyloidosis has not previously been described. Inotropic support is well tolerated in amyloidosis and can be a long term treatment modality, specifically to reduce hospital readmission rates. The use of palliative outpatient inotrope support should be considered as a treatment strategy in patients with end stage heart failure from cardiac amyloidosis.

Keywords: Inotropes, Amyloidosis, Heart Failure
NATURAL HISTORY OF TRANSTHYRETIN AMYLOID CARDIOMYOPATHY: INSIGHTS FROM THE TAFAMIDIS IN TRANSTHYRETIN CARDIOMYOPATHY CLINICAL TRIAL (ATTR-ACT)

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Introduction: Transthyretin amyloid cardiomyopathy (ATTR-CM) is a progressive, fatal disorder that remains underdiagnosed. A greater understanding of its presentation and progression could aid in disease awareness, earlier diagnosis and treatment. The Tafamidis in Transthyretin Cardiomyopathy Clinical Trial (ATTR-ACT) was the first large clinical trial to include both wild-type (ATTRwt) and hereditary (ATTRv) patients and represents one of the largest, and longest, collections of natural history data in patients with ATTR-CM.

Objective: To describe the natural history of ATTR-CM utilizing data from placebo-treated patients in ATTR-ACT.

Methods: Clinical evaluations in ATTR-ACT included: mortality, cardiovascular (CV)-related hospitalizations, functional capacity (assessed by 6-minute walk test [6MWT] distance) and health-related quality of life (assessed by Kansas City Cardiomyopathy Questionnaire overall summary ([KCCQ-OS] score). This is an analysis of the changes in clinical endpoints from baseline to Month 30 in patients treated with placebo, all values are mean (SD) unless stated otherwise.

Results: A total of 177 patients received placebo; 134 ATTRwt, 43 ATTRv. At baseline, ATTRwt patients tended to have less severe disease as shown by proportion of NYHA class III patients (ATTRwt, 32.8%; ATTRv 44.2%) and mean 6MWT distance (ATTRwt, 366.7 [126.2]; ATTRv, 311.2 [117.1]). KCCQ-OS scores (ATTRwt, 65.1 [21.3]; ATTRv, 68.4 [23.1]) and NT-proBNP levels (3826.3 [2840.2] ATTRwt vs 3905.4 [3384.2] ATTRv) were similar.

Over the duration of the study, there were 76 (42.9%) all-cause deaths, of which 63 (35.6%) were CV-related, and 107 (60.5%) patients had a CV-related hospitalization. There was a lower proportion of all-cause deaths in ATTRwt (49, 36.6%) than ATTRv (27, 62.8%) patients. This difference was evident both in patients with baseline NYHA Class I or II (ATTRwt, 25 events [27.8%]; ATTRv 12 events [50.0%]) and those with baseline NYHA Class III (ATTRwt, 24 events [54.5%]; ATTRv, 15 events [78.9%]). Mean 6MWT distance declined from baseline to Month 30, with a change of 89.7 (105.2) m; this was similar in ATTRwt (93.9 [93.7] m) and ATTRv (89.1 [107.2] m) patients. KCCQ-OS score also declined with a mean change of 14.6 (21.4); the decline was less severe in ATTRwt (13.8 [20.7]) than ATTRv (21.0 [26.4]) patients.

Conclusions: Patients with ATTR-CM experience a severe, progressive disease. In ATTR-ACT, placebo-treated ATTRv patients, compared with ATTRwt, had more severe disease at baseline, as measured by NYHA and functional capacity (6MWT), and their disease progressed more rapidly as shown by mortality, hospitalizations, and health-related quality of life (KCCQ-OS) over time. Understanding the presentation and progression of ATTR-CM, and the differences between ATTRwt and ATTRv, emphasizes the need for earlier diagnosis and will help guide physicians treating these patients.
A DIAGNOSTIC SCORING MODEL TO DIFFERENTIATE BETWEEN CARDIAC AMYLOIDOSIS AND OTHER HYPERTROPHIC CARDIOMYOPATHIES BASED ON COMMON CARDIAC MAGNETIC RESONANCE PARAMETERS

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Background: Cardiac magnetic resonance (CMR) is nowadays a widely available tool to distinguish between different forms of cardiac hypertrophy. Especially novel techniques like T1 mapping increased the sensitivity and specificity for detection of cardiac involvement in amyloidosis. However, those techniques are often only available at large and specialized centers.

Objectives: The aim of this study was to create a diagnostic scoring model that enables accurate diagnosis of cardiac amyloidosis using parameters within CMR that are simple, easy to measure and do not require special sequences or software.

Methods: We measured 25 different parameters in standard CMR images of 295 patients with cardiac amyloidosis treated at our center (152 AL and 143 TTR). 107 CMRs were performed at our center and 188 CMRs were requested from external hospitals and radiological practices. 76 patients served as a control group (47 hypertrophic cardiomyopathy, 17 hypertensive heart disease, 12 non-amyloid restrictive cardiomyopathy). A point-system based scoring model was calculated, allowing each individual to obtain a summed score between 0 and 5. Using ROC statistics and multivariate binary logistic regression analysis, 5 CMR parameters were identified and optimal cut-off values were determined.

Results: The following five parameters were included within the model: right atrial diameter, atrial septal thickness, left ventricular end-diastolic diameter, tricuspid annular plane systolic excursion and any form of late gadolinium enhancement. The AUC of the summed score of the patient cohort vs. the control cohort was found to be 0.93 (CI 0.90-0.96) with a specificity of 0.84 and sensitivity of 0.91 respectively at a score of three from possible five parameters reaching the cut-off.

Conclusions: The proposed scoring model, based on CMR parameters easy to measure and not requiring special sequences or software, allows a differentiation between cardiac amyloidosis and common differential diagnoses (hypertensive heart disease, hypertrophic cardiomyopathy, non-amyloidotic restrictive cardiomyopathy) with high specificity and sensitivity and might be useful for clinical routine.
CHADS-VASC SCORE IN TRANSTHYRETIN CARDIAC AMYLOIDOSIS

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Introduction: Embolic events are a frequent severe complication in transthyretin cardiac amyloidosis (ATTR). CHADS-VASC score has shown to be useful in predicting embolic events in patients with nonvalvular atrial fibrillation (AF) but its performance in ATTR patients with and without AF is unknown.

Objectives: We sought to analyze if CHADS-VASC score is accurate in predicting embolic events in patients with TTR cardiac amyloidosis.

Methods: Clinical characteristics at initial evaluation and embolic events (stroke, transient ischemic attack or peripheral embolism) from TTR cardiac amyloidosis patients evaluated at 5 centers: Columbia University Hospital (New York, US), University of Bologna (Italy), University of Pavia (Italy), National Amyloid Center (London, UK) and Hospital Universitario Puerta de Hierro (Madrid, Spain) were retrospectively collected. Patients were stratified according to CHADS-VASC score in 4 groups: 0 (low risk), 1-2 (intermediate risk), 3-5 (high risk) and 6 or more (very high risk).

Results: 1157 patients with TTR cardiac amyloidosis (87.4% male, mean age 75.7±8.6 years, 83% ATTRwt and 17% ATTRh) were included in this analysis. 43 patients (3.78%; CI95% 2.70-4.97%) had an embolic event during a median follow-up of 23.2 months (IQR 12.2-39.2). Among them, 20 (46.5%) patients were under anticoagulation (either with AVK or DOAC) at the time of the event. Rate of embolic events was similar among patients with baseline AF (24/574, 4.18%) and those without baseline AF (19/583, 3.26%) (p=0.5). Baseline CHADS-VASC score had a strong correlation with the rate of embolic events in patients with baseline AF (p trend <0.001), but this correlation was not found in patients without baseline AF (p trend=0.71) (Table 1). Interestingly, only patients with baseline AF who had a CHADS-VASC Score ≥3 points had embolic events.

Conclusions: Higher values of CHADS-VASC score are associated with a higher risk of embolic events in TTR cardiac amyloidosis with AF. In contrast, it does not accurately predict embolic events in patients without AF.

Table 1:

<table>
<thead>
<tr>
<th>CHADS-VASC</th>
<th>Embolic events (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Baseline AF (N=574)</td>
</tr>
<tr>
<td>0</td>
<td>0/5</td>
</tr>
<tr>
<td>1-2</td>
<td>0/169</td>
</tr>
<tr>
<td>3-5</td>
<td>15/362 (4.1%)</td>
</tr>
<tr>
<td>≥6</td>
<td>9/38 (23.7%)</td>
</tr>
<tr>
<td>P trend</td>
<td>P &lt; 0.001</td>
</tr>
</tbody>
</table>
PT080

THE PRESENCE OF TRANSTHYRETIN CARDIAC AMYLOIDOSIS DOES NOT AFFECT MORTALITY OUTCOMES IN COEXISTING SEVERE AORTIC STENOSIS

H. Rosenblum 1, D. Narotsky 1, J. Goldsmith 1, N. Hamid 1, R. Hahn 1, S. Kodali 1, T. Vahl 1, T. Nazif 1, O. Khalique 1, S. Bokhari 1, A. Castano 1, M. Maurer 1

1 Columbia University Irving Medical Center

Background: Advances in noninvasive diagnostic imaging have increased the recognition of transthyretin cardiac amyloidosis (ATTR-CA) and severe symptomatic AS. However, the implications of ATTR-CA on outcomes after TAVR remain undetermined.

Methods: We screened patients undergoing TAVR for ATTR-CA using Technetium-99m pyrophosphate scintigraphy as described previously. Using Kaplan Meier analysis, we compared the primary endpoint of death and a combined endpoint of death and heart failure (HF) hospitalization in patients with and without ATTR-CA. Cox proportional hazards modeling was used to determine the association of ATTR-CA with these endpoints.

Results: 167 patients (mean 83 yrs, 62% male, Society of Thoracic Surgeons (STS) score 6.4%, 71% NYHA class III/IV) were included: 24 had ATTR-CA (14.4%). In Kaplan Meier analysis, there was no difference in mortality at 3 years between patients with and without ATTR-CA (log rank, p = 0.73). Similarly, there was no significant difference in Kaplan Meier analysis for the combined endpoint (log rank, p = 0.70). In Cox proportional hazard modeling, the presence of ATTR-CA was not associated with death (HR 1.2, 95% CI 0.5-2.6 p=0.726). However, patients with ATTR-CA had increased rates of HF hospitalization at both 1 year (0.316 vs 0.092 events/person year, RR 3.4, p=0.009) and 3 years (0.144 vs 0.084 events/person year, RR 2.1, p=0.063) following TAVR.

Conclusions: In this prospective study of moderate-risk patients with severe AS undergoing TAVR, evaluation for coexistence of ATTR-CA revealed a high prevalence (14.4%) which did not affect mortality after TAVR. However, we observed an increased burden of HF hospitalization following TAVR in those with ATTR-CA, suggesting the consequences of the underlying myopathic process.
PT081

BIOMARKERS AND PROGNOSIS PREDICTION IN TRANSTHYRETIN RELATED CARDIAC AMYLOIDOSIS: A DIRECT COMPARISON OF TWO STAGING SYSTEMS.

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2 Tuscan Regional Amyloidosis Centre, Careggi University Hospital, Florence, Italy, Division of Interventional Structural Cardiology, Cardiothoracovascular Department, Careggi University Hospital, Florence, Italy.
3 Tuscan Regional Amyloidosis Centre, Careggi University Hospital, Florence, Italy, Cardiology Department, Azienda Sanitaria Firenze (ASF) Florence Italy.
4 Division of Interventional Structural Cardiology, Cardiothoracovascular Department, Careggi University Hospital, Florence, Italy.

Background: Severity of heart disease at presentation varies widely among patients with transthyretin-related cardiac amyloidosis (ATTR-CA) and availability of tools able to predict prognosis is essential for clinical and research purposes. Currently, two biomarker-based staging systems are available. The aim of this work is to compare their predictive performance.

Methods and Results: 175 patients diagnosed with ATTR-CA (133 wild-type and 42 hereditary) were stratified into different disease stages on the basis of two systems: one including N-terminal pro-B-type natriuretic peptide (NT-proBNP) and estimated glomerular filtration rate (eGFR) and one including NT-proBNP and Troponin I (TnI). Survival estimates and age-adjusted survival for all-cause mortality were analysed over a median follow-up of 27 months [IQR 16-43]. Kappa analysis showed that a significant but modest concordance between scores was present with a kappa of 0.541 and 70% of patients were classified in the same stage using the two different staging systems. Predictive performance was more accurate when NT-proBNP and eGFR were applied, resulting in effective survival stratification: 64.4 months for stage 1, 44.6 months for stage 2, and 20.5 months for stage 3 (p<0.01 for stage 1 vs 2; p<0.0001 for stage 1 vs 3; p<0.0001 for stage 2 vs 3). The combination of NT-proBNP and TnI was unable to effectively differentiate survival: 64.5 months for stage 1, 50.9 months for stage 2, and 27.3 months for stage 3 (p= 0.223 for stage 1 vs 2; p<0.0001 for stage 1 vs 3 and p<0.0001 for stage 2 vs 3). Cox proportional hazards regression analysis, adjusted for age and including comparison of staging systems in the whole population and in the wtATTR subgroup were performed (see table).

<table>
<thead>
<tr>
<th>Group</th>
<th>Model item</th>
<th>Result</th>
<th>Gillmore Score</th>
<th>Modified Grogan score</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cardiac ATTR whole population</td>
<td>Stage 2 vs 1</td>
<td>HR (95% CI) 2.071 (1.157-3.707)</td>
<td>1,444 (0.731-2.853)</td>
<td>0.290</td>
</tr>
<tr>
<td></td>
<td></td>
<td>P value 0.014</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Stage 3 vs 1</td>
<td>HR (95% CI) 7.898 (3.896-16.011)</td>
<td>4,040 (2.263-7.213)</td>
<td>0.0001</td>
</tr>
<tr>
<td></td>
<td></td>
<td>P value 0.0001</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Stage 2 vs 3</td>
<td>HR (95% CI) 3.435 (1.847-6.390)</td>
<td>2,686 (1.431-5.029)</td>
<td>0.002</td>
</tr>
<tr>
<td></td>
<td></td>
<td>P value 0.0001</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Age</td>
<td>HR (95% CI) 1.042 (0.998-1.088)</td>
<td>1,043 (0.999-1.088)</td>
<td>0.052</td>
</tr>
<tr>
<td></td>
<td></td>
<td>P value 0.065</td>
<td></td>
<td></td>
</tr>
<tr>
<td>wtATTR subgroup</td>
<td>Stage 2 vs 1</td>
<td>HR (95% CI) 2.736 (1.274-5.875)</td>
<td>1,507 (0.630-3.602)</td>
<td>0.357</td>
</tr>
<tr>
<td></td>
<td></td>
<td>P value 0.010</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Stage 3 vs 1</td>
<td>HR (95% CI) 7.681 (3.182-1.541)</td>
<td>3,493 (1.710-7.134)</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td></td>
<td>P value 0.0001</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Stage 2 vs 3</td>
<td>HR (95% CI) 2.818 (1.371-5.792)</td>
<td>2,168 (0.986-4.768)</td>
<td>0.054</td>
</tr>
<tr>
<td></td>
<td></td>
<td>P value 0.005</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Age</td>
<td>HR (95% CI) 1.064 (1.005-1.125)</td>
<td>1,068 (1.011-1.128)</td>
<td>0.019</td>
</tr>
<tr>
<td></td>
<td></td>
<td>P value 0.032</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Conclusion: A staging system including Nt-proBNP and eGFR should be the preferred staging system in predicting prognosis in ATTR-CA given its better prognostic accuracy.
PT082

PREDICTORS OF ATRIAL FIBRILLATION DEVELOPMENT IN TRANSTHYRETIN CARDIAC AMYLOIDOSIS.

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1 Hospital Universitario Puerta de Hierro (Madrid, Spain).
2 National Amyloid Center (London, UK).
3 Columbia University Hospital (New York, US).
4 University of Bologna (Italy).
5 University of Pavia (Italy)

Introduction: Atrial fibrillation (AF) is the most common arrhythmia found in cardiac transthyretin amyloidosis (ATTR) and is associated with high risk of embolic events. It is unknown which factors are associated with AF onset in ATTR.

Objectives: The primary aim of this study was to identify which factors are associated with the development of AF among cardiac TTR amyloid patients in sinus rhythm in a large multicentre cohort of patients with cardiac ATTR.

Methods: Clinical characteristics at initial evaluation and AF onset data from cardiac ATTR patients evaluated at 5 international amyloidosis centers: Columbia University Hospital (New York, US), University of Bologna (Italy), University of Pavia (Italy), National Amyloid Center (London, UK) and Hospital Puerta de Hierro (Madrid, Spain) were retrospectively collected. A multivariable stepwise Cox regression model fitted with predictors associated with AF onset in univariate analysis was constructed.

Results: 583 patients with cardiac ATTR (84.2% male, mean age 74.4±9.4 years, 74.9% ATTRwt and 25.1% ATTRh) and no previous history of AF were included in this analysis. During a median follow-up of 26.2 months (IQR 13.8-40.8), 187 (32.1%) patients developed AF with 66 (11.3%) patients during the initial 12 months since the baseline evaluation. Predictors of AF appearance during follow-up at univariate analysis were male sex, age, wild-type ATTR, NYHA class, hypertension, renal function (eGFR) and left atrium diameter. We performed a multivariate analysis with predictors that were statistically significant in univariate analysis. As left atrium diameter was not statistically associated with AF appearance on initial multivariate analysis and was available only in 249 (43.7%) patients, this parameter was excluded from the final multivariate model. Predictors that remained independently associated with AF appearance in the final multivariate analysis were male sex, age ≥65 years, wild-type ATTR, NYHA class III/IV, eGFR and hypertension (Table). The model including the abovementioned predictors exhibited a Harrell’s C index of 0.68.

Conclusions: This study shows that patients with cardiac ATTR who are at risk of AF development can be identified using readily available clinical parameters like age, sex, NYHA class, renal function, hypertension and ATTR subtype.

Table:

<table>
<thead>
<tr>
<th>Predictors of AF development</th>
<th>Univariate analysis</th>
<th>Multivariate analysis</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex (male)</td>
<td>2.36 (1.44-3.87)</td>
<td>2.48 (1.41-4.37)</td>
</tr>
<tr>
<td>Age (≥65 years)</td>
<td>2.46 (1.59-3.80)</td>
<td>1.66 (1.03-2.69)</td>
</tr>
<tr>
<td>Wild type ATTR</td>
<td>2.23 (1.55-3.22)</td>
<td>1.81 (1.18-2.78)</td>
</tr>
<tr>
<td>Diabetes</td>
<td>1.05 (0.71-1.54)</td>
<td></td>
</tr>
<tr>
<td>Hypertension</td>
<td>1.53 (1.15-2.04)</td>
<td>1.41 (1.04-1.93)</td>
</tr>
<tr>
<td>BMI (per Kg/m²)</td>
<td>1.02 (0.98-1.05)</td>
<td></td>
</tr>
<tr>
<td>eGFR &gt;45 ml/min/1.73 m²</td>
<td>2.06 (1.43-2.96)</td>
<td>1.64 (1.09-2.46)</td>
</tr>
<tr>
<td>NT-proBNP ≥3000 ng/L</td>
<td>1.50 (1.06-2.11)</td>
<td>1.19 (0.80-1.76)</td>
</tr>
<tr>
<td>NYHA Class III/IV</td>
<td>1.88 (1.30-2.72)</td>
<td>1.93 (1.29-2.89)</td>
</tr>
<tr>
<td>LVEF ≥45%</td>
<td>1.14 (0.81-1.61)</td>
<td></td>
</tr>
<tr>
<td>Left atrium diameter (per mm)</td>
<td>1.05 (1.02-1.08)</td>
<td>1.02 (0.98-1.06)*</td>
</tr>
</tbody>
</table>

1Body Mass Index
2estimated Glomerular Filtration Rate
3Left Ventricular Ejection Fraction
EMBOLIC EVENTS IN PATIENTS WITH TRANSTHYRETIN CARDIAC AMYLOIDOSIS WITHOUT ATRIAL FIBRILLATION.

A. Rivas-Pérez 1, S. Gnanasampanthan 2, L. Mitrani 3, G. Saturi 4, R. Mussinelli 5, F. Domínguez 1, M. Renju 2, C. Gagliardi 4, A. Sabena 5, E. González-López 1, M. Fontana 2, A. Giuseppe Caponetti 4, G. Palladini 5, S. Perlini 5, C. Rapezzi 4, M. Maurer 3, J. D. Gillmore 2, P. García-Pavía 1

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2 National Amyloid Center (London, UK).
3 Columbia University Hospital (New York, US).
4 University of Bologna (Italy).
5 University of Pavia (Italy)

Background: Embolic events are a potential complication in transthyretin (TTR) cardiac amyloidosis. Recently, some concerns have emerged about high incidence of intracardiac thrombus in patients with TTR cardiac amyloidosis, even in the absence of atrial fibrillation (AF).

Objectives: We sought to describe incidence of embolic events in a large international cohort of patients with wild type and hereditary TTR cardiac amyloidosis and no history of AF.

Methods: Clinical characteristics at initial evaluation and embolic events (stroke, transient ischemic attack or peripheral embolism) from TTR cardiac amyloidosis patients without history of AF evaluated at 5 centers: Columbia University Hospital (New York, US), University of Bologna (Italy), University of Pavia (Italy), National Amyloid Center (London, UK) and Hospital Universitario Puerta de Hierro (Madrid, Spain) were retrospectively collected. Patients were censored from this analysis at the time when AF appeared during follow-up.

Results: 583 patients with cardiac TTR amyloidosis (84.2% male, mean age 74.4±9.4 years, 74.9% ATTRwt and 25.1% ATTRh) were included in this analysis. During a median follow-up of 20.4 months (IQR 8.9-33.7), 16 patients (2.74%; IC95%: 1.69-4.44) had an embolic event without previous history of AF. As a consequence of the embolic event, AF was diagnosed in 3 individuals. Incidence rate of embolic events in ATTR cardiac patients without history of AF was 1.16 embolic events per 1000 person-months. Patients who had an embolic event without previous history of AF were more frequently women (37.5% vs 15.2%; p=0.016). No other factors associated with embolisms were identified (Table 1).

Conclusions: Patients with TTR cardiac amyloidosis and no history of AF had a non-negligible number of embolic events during follow-up. Women had a higher number of embolic events than men in this setting, and no other factors associated with embolisms were identified.

Table 1:

<table>
<thead>
<tr>
<th>Baseline characteristics</th>
<th>Embolism (N=16)</th>
<th>No embolism (N=567)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>75.2±6.0</td>
<td>74.4±9.5</td>
<td>0.735</td>
</tr>
<tr>
<td>Female (n, %)</td>
<td>6 (37.5)</td>
<td>86 (15.2)</td>
<td>0.016</td>
</tr>
<tr>
<td>ATTRh (n, %)</td>
<td>6 (37.5)</td>
<td>140 (24.7)</td>
<td>0.245</td>
</tr>
<tr>
<td>Hypertension (n, %)</td>
<td>6 (37.5)</td>
<td>235 (41.5)</td>
<td>0.752</td>
</tr>
<tr>
<td>Diabetes (n, %)</td>
<td>2 (12.5)</td>
<td>88 (15.6)</td>
<td>0.737</td>
</tr>
<tr>
<td>Previous embolism (n, %)</td>
<td>0 (0)</td>
<td>61 (10.8)</td>
<td>0.165</td>
</tr>
<tr>
<td>Previous heart failure (n, %)</td>
<td>4 (25.0)</td>
<td>166 (30.4)</td>
<td>0.643</td>
</tr>
<tr>
<td>Peripheral vascular disease (n, %)</td>
<td>1 (6.3)</td>
<td>94 (16.7)</td>
<td>0.267</td>
</tr>
<tr>
<td>Left atrium diameter (mm)</td>
<td>42.6±4.7</td>
<td>44.6±6.8</td>
<td>0.293</td>
</tr>
<tr>
<td>Oral anticoagulation (n, %)</td>
<td>0 (0)</td>
<td>37 (6.6)</td>
<td>0.304</td>
</tr>
<tr>
<td>CHADS-VASC Score ≥2 (n, %)</td>
<td>16 (100)</td>
<td>511 (94.6)</td>
<td>0.341</td>
</tr>
<tr>
<td>Body Mass Index (kg/m²)</td>
<td>25.7±2.5</td>
<td>26.3±4.0</td>
<td>0.566</td>
</tr>
</tbody>
</table>
EMBOLIC EVENTS IN TRANSTHYRETIN CARDIAC AMYLOIDOSIS. ANALYSIS FROM A LARGE INTERNATIONAL COHORT.

A. Rivas-Pérez 1, S. Gnanasampanthan 2, L. Mitrani 3, G. Saturi 4, A. Sabena 5, E. González-López 1, M. Renju 2, S. Longhi 4, R. Mussinelli 1, A. Hernández 1, M. Fontana 2, A. Giuseppe Caponetti 4, L. Obici 5, S. Perlini 5, C. Rapezzi 4, M. Maurer 3, J.D. Gillmore 2, P. García-Pavía 1

1 Hospital Universitario Puerta de Hierro (Madrid, Spain).
2 National Amyloid Center (London, UK).
3 Columbia University Hospital (New York, US).
4 University of Bologna (Italy).
5 University of Pavia (Italy).

Background: Embolic events are a potential complication in transthyretin (TTR) cardiac amyloidosis but data on prevalence and incidence of this severe complication are scarce. Recently, some concerns have emerged about high incidence of atrial thrombus in patients with TTR cardiac amyloidosis, mainly in the context of atrial fibrillation (AF) and planned electrical cardioversion.

Objectives: We sought to describe the prevalence and the incidence of embolic events in a large international cohort of patients with TTR cardiac amyloidosis and to identify risk factors for embolic occurrence.

Methods: Clinical characteristics at initial evaluation and embolic events (stroke, transient ischemic attack or peripheral embolism) from cardiac TTR amyloidosis patients evaluated at 5 centers: Columbia University Hospital (New York, US), University of Bologna (Italy), University of Pavia (Italy), National Amyloid Center (London, UK) and Hospital Universitario Puerta de Hierro (Madrid, Spain) were retrospectively collected. Patients were classified according to the presence of baseline AF or not.

Results 1157 patients with cardiac ATTR amyloidosis (87.4% male, mean age 75.7±8.6 years, 83% ATTRwt and 17% ATTRh) were included in this analysis. 155 (13.4%) had already had an embolic event prior to initial evaluation (94 (60.7%) were on AF at baseline and 61 were not) and 300 (25.9%) were receiving anticoagulants (12.3% of those with no previous history of AF). During a median follow-up of 23.2 months (IQR: 12.2-39.2), 43 patients (3.72%; CI95% 2.77-4.96%) had an embolic event (25 strokes, 13 transient ischemic attacks and 7 peripheral embolisms). Among them, 20 (46.5%) patients were under anticoagulation (either with AVK or DOAC) at the time of the event. Rate of embolic events was similar (p=0.427) among patients with baseline AF (24/574, 4.18%) and those without baseline AF (19/583, 3.12%). Among those individuals who had embolic events and did not show AF at baseline, 6 had AF at the time of the embolic event (3/6 were receiving anticoagulation) Predictors of embolic events in the overall cohort were female sex, hypertension and peripheral vascular disease. Age and previous embolism were also close to significance. Among patients with baseline AF, age (OR 1.08 per year; CI95% 1.01-1.16), hypertension (OR 3.28; CI95% 1.24-8.70), previous embolism (OR 4.70; CI95% 1.92-11.49) and peripheral vascular disease (OR 1.94; CI95% 1.24-6.98) were associated with embolic events during follow-up in multivariate analysis. We were not able to identify factors associated with embolic events in patients without AF at baseline.

Conclusions: Embolism is a frequent complication of ATTR cardiac amyloidosis that affects up to 16.3% of patients. Rate of embolic events remains high despite oral anticoagulation and patients without AF have a considerable number of events. Risk factors associated with embolic events are female sex, hypertension and peripheral vascular disease.
TEMPORAL ANALYSIS OF WILD-TYPE TRANSTHYRETIN AMYLOID CARDIOMYOPATHY AND ASSOCIATED DIAGNOSES USING A NOVEL MACHINE LEARNING MODEL

A. Castaño 1, A. Huda 1, M. Bruno 1, M. B. Sultan 1, R. Deo 2, S. J. Shah 3

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2 Brigham and Women’s Hospital, Boston, MA, USA.
3 Northwestern University Feinberg School of Medicine, Chicago, IL, USA.

Introduction: Wild-type transthyretin amyloid cardiomyopathy (ATTRwt) is an under-recognized cause of heart failure (HF) that is associated with other non-cardiac and cardiac diagnoses. Better understanding of the timing of these associated diagnoses relative to ATTRwt diagnosis may help raise suspicion and lead to earlier ATTRwt diagnosis.

Objectives: To assess temporal relationships among non-cardiac and cardiac diagnoses associated with ATTRwt (ICD10 E85.82) diagnosis using a validated machine learning model developed to facilitate identification of patients (pts) at risk of ATTRwt.

Methods: Pts with HF were identified from US medical claims data (IQVIA: n>300 million pts with >10 y of medical history) using ICD codes; pts with HF+ATTRwt codes (n=1071) were compared with pts with HF but no amyloidosis codes (controls; n=1071). Model output revealed 40 clinical diagnoses (20 non-cardiac+20 cardiac) significantly associated with ATTRwt. Time differences were calculated between earliest date of each associated clinical diagnosis and dates of ATTRwt and HF diagnoses. (Date of ATTRwt diagnosis was the earliest date of amyloidosis or ATTRwt ICD code designation, tc-99m pyrophosphate scintigraphy, or endomyocardial biopsy.)

Results: On average, HF diagnosis preceded ATTRwt diagnosis by a median (interquartile range [IQR]) of 9.5 (32.9, 0.2) months. Non-cardiac diagnoses occurred earlier than cardiac diagnoses associated with ATTRwt (9.6 [39.9, -2.9] vs. 1.1 [19.0, -2.2] months, respectively; p<0.0001). The earliest non-cardiac and cardiac diagnoses to occur before diagnosis of ATTRwt were rotator cuff strain (55.6 [69.7, 38.5] months) and abnormal electrocardiogram (14.1 [43.1, 0.0] months), respectively (Table).

Conclusions: This temporality analysis confirmed clinical observations that non-cardiac, largely orthopedic manifestations of cardiac amyloidosis appear significantly earlier than cardiac manifestations. Awareness of these temporal relationships may help support earlier identification, diagnosis, and treatment of this life-threatening disease.

Temporal Relationships Among ATTRwt-Associated Diagnoses and ATTRwt and HF Diagnoses

<table>
<thead>
<tr>
<th>Associated Diagnoses*</th>
<th>Pts with ATTRwt n (%)</th>
<th>Time Difference†, months (median [IQR])</th>
<th>ATTRwt‡ (n=1071)</th>
<th>HF (n=1071)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Non-Cardiac</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rotator cuff strain</td>
<td>73 (7)</td>
<td>55.6 (69.7, 38.5)</td>
<td>11.0 (32.3, 0.7)</td>
<td></td>
</tr>
<tr>
<td>Synovitis/tenosynovitis</td>
<td>224 (21)</td>
<td>46.8 (74.3, 19.8)</td>
<td>8.3 (29.8, 0)</td>
<td></td>
</tr>
<tr>
<td>Tendon rupture</td>
<td>95 (9)</td>
<td>44.9 (64.7, 13.5)</td>
<td>7.7 (31.3, 0)</td>
<td></td>
</tr>
<tr>
<td>Carpal tunnel syndrome</td>
<td>342 (32)</td>
<td>41.4 (70.4, 4.8)</td>
<td>7.4 (27.8, 0)</td>
<td></td>
</tr>
<tr>
<td>Seborrheic keratosis</td>
<td>313 (29)</td>
<td>38.3 (71.6, 7.8)</td>
<td>6.1 (35.0, 0)</td>
<td></td>
</tr>
<tr>
<td><strong>Cardiac</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Abnormal ECG</td>
<td>588 (55)</td>
<td>14.1 (43.1, 0)</td>
<td>8.9 (32.8, 0.1)</td>
<td></td>
</tr>
<tr>
<td>Atrial fibrillation</td>
<td>773 (72)</td>
<td>12.3 (39.8, 0)</td>
<td>9.3 (32.2, 0.1)</td>
<td></td>
</tr>
<tr>
<td>Cardiomegaly</td>
<td>697 (65)</td>
<td>4.9 (27.9, 0)</td>
<td>9.8 (33.3, 0.3)</td>
<td></td>
</tr>
<tr>
<td>Pleural effusion</td>
<td>509 (48)</td>
<td>3.5 (19.9, -1.2)</td>
<td>13.9 (36.9, 0.9)</td>
<td></td>
</tr>
<tr>
<td>Long-term anticoagulation</td>
<td>569 (53)</td>
<td>3.4 (26.1, -4.4)</td>
<td>10.2 (35.1, 0.3)</td>
<td></td>
</tr>
</tbody>
</table>

*5 earliest relative to ATTRwt.
†Between earliest date of associated diagnosis and date of ATTRwt and HF diagnoses.
‡p<0.0001, ATTRwt vs HF (all).
THE INCIDENCE AND TEMPORAL RELATIONSHIP OF EARLY CLINICAL MANIFESTATIONS AND TRANSTHYRETIN AMYLOID CARDIOMYOPATHY IN TAFAMIDIS CLINICAL TRIALS

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Introduction: Transthyretin amyloid cardiomyopathy (ATTR-CM) is a progressive, life-threatening disease and an often overlooked cause of heart failure. Diagnosis and treatment of ATTR-CM in its early symptomatic stage is critical to slow amyloid deposition and disease progression and may be facilitated by education and screening for initial cardiac and non-cardiac manifestations. However, there is a limited understanding of initial ‘red-flag’ manifestations and their timing. The Tafamidis in Transthyretin Cardiomyopathy Clinical Trial (ATTR-ACT), a double-blind, placebo-controlled, randomized trial, and an ongoing long-term extension (LTE) trial provide a large data set for evaluation of the natural history of the disease in ATTR-CM patients.

Objective: To explore the incidence of early clinical manifestations and their temporal relationship to ATTR-CM diagnosis in patients participating in ATTR-ACT and the LTE.

Methods: In ATTR-ACT, tafamidis efficacy and safety were assessed in patients with variant (ATTRv) and wild-type (ATTRwt) disease. Patients completing ATTR-ACT were eligible to enrol in an LTE, which was subsequently expanded to also include patients with ATTR-CM not previously enrolled in ATTR-ACT. The incidence of clinical manifestations of the disease (based on medical history at enrolment) was assessed in both populations. In the ATTR-ACT population, the time from symptom onset to ATTR-CM diagnosis was analysed by genotype.

Results: The most common clinical manifestations in patients in ATTR-ACT (n=441) and the LTE (n=924) included atrial fibrillation (60% and 58%), carpal tunnel syndrome (41% and 50%), osteoarthritis (25% and 32%), and lumbar spinal stenosis (8% and 11%). In ATTR-ACT, the onset of atrial fibrillation and carpal tunnel syndrome was earlier relative to ATTR-CM diagnosis in ATTRwt versus ATTRv patients (Table).

Conclusions: Clinical manifestations such as atrial fibrillation and carpal tunnel syndrome are early signs of ATTR-CM that may be useful for screening/diagnosis. These cardiac and non-cardiac ‘red flags’ appear earlier, but not exclusively, in patients with ATTRwt compared with ATTRv.

Time from Symptom Onset to ATTR-CM Diagnosis in ATTR-ACT*

<table>
<thead>
<tr>
<th>Symptom</th>
<th>ATTRv (n=106)</th>
<th></th>
<th>ATTRwt (n=335)</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n (%)</td>
<td>Mean (SD)</td>
<td>Median (IQR)</td>
<td>n (%)</td>
</tr>
<tr>
<td>Atrial fibrillation</td>
<td>39 (37)</td>
<td>0.5 (3.0)</td>
<td>0.2 (-0.5, 1.3)</td>
<td>155 (46)</td>
</tr>
<tr>
<td>Carpal tunnel syndrome</td>
<td>29 (27)</td>
<td>5.7 (5.2)</td>
<td>3.6 (1.7, 9.9)</td>
<td>133 (40)</td>
</tr>
<tr>
<td>Osteoarthritis</td>
<td>10 (9)</td>
<td>10.4 (8.1)</td>
<td>9.8 (5.4, 14.0)</td>
<td>51 (15)</td>
</tr>
<tr>
<td>Lumbar spinal stenosis</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>36 (11)</td>
</tr>
</tbody>
</table>

*For potential ‘red-flag’ clinical manifestations with symptom onset data available in ≥10 patients.

IQR, interquartile range
PT087

PREDICTORS OF SURVIVAL IN POLISH PATIENTS WITH ADVANCED CARDIAC LIGHT-CHAIN AMYLOIDOSIS – CARDIOLOGY DEPARTMENT EXPERIENCE

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**Background:** Cardiac light-chain amyloidosis (AL amyloidosis) is the most common cause of restrictive cardiomyopathy in Poland. Although it has a hematological background, the presentation of the disease is mainly cardiological. Thus identification of survival predictors among parameters obtained during cardiological assessment may help to plan hematological treatment.

**Objectives:** The aim of the study was to assess clinical data, vital signs and echocardiographic parameters as predictors of overall survival in cardiac AL amyloidosis.

**Methods:** The analysis covered all consecutive patients (pts) with AL amyloidosis diagnosed in the cardiology department from August 2011 to August 2019. Clinical data including weight loss and systolic and diastolic blood pressure (SBP and DBP, respectively) in sitting position was collected. NT-proBNP and high-sensitivity troponin T (hs-TnT) concentrations were measured. The following echocardiographic parameters were assessed: left ventricular end-diastolic diameter (LVEDD) and ejection fraction (LVEF), tricuspid annular plane systolic excursion (TAPSE), longitudinal myocardial velocities of left and right ventricles (LV s’ and RV s’, respectively) estimated by tissue Doppler imaging (TDI), and the ratio of the transmitral early peak velocity (E) estimated by pulsed wave Doppler over the early mitral annulus velocity (e’) estimated by TDI (E/e’ ratio).

**Results:** Sixty-four pts (median age 61.5 yrs) with symptomatic heart failure were enrolled. Forty-seven pts had advanced heart failure (NYHA functional class III or IV). According to the Mayo 2012 risk stratification system, two pts had Stage I, 7 pts had Stage II, and 26 and 29 pts respectively had Stage III and IV. Median (interquartile range) NT-proBNP and hs-TnT concentrations were 4947.5 (2144.5-11017) pg/ml and 77 (31.3-144.4) ng/l, respectively. Forty pts underwent chemotherapy, 9 pts met the criteria for autologous stem cell transplant. With median follow-up of 8 months, 39 pts died with median survival 3.5 months. The Univariate Cox proportional hazard model indicated that weight loss >10 kilos within one year, DBP, SBP, NT-proBNP and hs-TnT, E/e’ ratio, LVEDD, LVEF, LV s’, RV s’ and TAPSE were significant predictors of survival (p<0.05). The Multivariate Cox proportional hazard model included DBP and RV s’ with hazard ratios (95% confidence intervals, CIs) of 0.93 (0.89-0.97) and 0.78 (0.69-0.89), respectively. The Kaplan-Meier curve analysis showed that LV s’<5 cm/sec and RV s’<10 cm/sec predicted worse survival. Comparison of ROC curves revealed that RV s’ (AUC=0.842, 95% CI: 0.728-0.922) and TAPSE (0.829, 0.714-0.913) were as good as hs-TnT (0.849, 0.736-0.927) and NT-proBNP (0.800, 0.681-0.890) in death prediction.

**Conclusions:** Right ventricular function assessment using RV s’ and TAPSE, in addition to biomarkers, helps to predict prognosis in advanced cardiac AL amyloidosis. Efforts to diagnose the disease early are greatly needed.
Background: Nowadays, direct biochemical analysis of amyloid fibrils has been dependent on their isolation, requiring large amounts of material and on complex extraction time-consuming procedures.

AIM of this study is to set up a new methodological approach, based on Western Blot analysis (WB), comprising the research of appropriate extraction solvents and separation strategies, for small-scale isolation, direct detection and characterization of amyloid proteins from both fresh and formalin-fixed (FFPE) cardiac tissues for diagnostic purpose.

Methods: Seventy seven (77) cases were evaluated and divided in two sets: 40 cases for set up the protocol and 37 cases for validation. For the first set 20 amyloid negative and 20 positive samples were selected on the base of Congo red staining and electron microscopy (EM). For the validation set, 37 consecutive samples were selected among the cases referred to our center with suspected amyloidosis: 29 endomyocardial biopsies (EMBs) and 8 autopsy hearts. Samples are homogenized and solubilized under different conditions of lysis buffer composition, temperature and additives, followed by SDS-PAGE and WB. The results were compared with those of immuno-electron microscopy (IEM) and mass spectrometry (MS), which we consider as reference.

Results: K and \( \lambda \) proteins are not modified by temperature, buffer composition, pH and additives, when subjected to SDS-PAGE and WB analysis. On the contrary, TTR proteins (trimer, dimer and monomer) show great modifications in protein band fragments under different conditions, leading also to false positive monomer band. Only high alkaline pH preserves correctly TTR monomer formation, leading to identification of only true TTR positive amyloid cases. This new approach allows a direct proteins biochemical analysis just after extraction procedure and demonstrate its concordance with IEM and MS.

In the validation study, this new proteomic technique showed a sensitivity of 97% and a specificity of 100%. Discordant outcome results in just one case. In three of eight TTR cases (37%), WB shows a TTR-monomer electrophoretic double band, underlining the presence of mutation or post-translational TTR-protein modification. MS analysis confirms the presence of post-translational TTR/Cys10-Sulfonate modification.

Conclusions: We develop and standardize an easy and reliable diagnostic technique for direct chemical characterization of the amyloidogenic proteins, which is able to correctly identify specific types of amyloidosis. To date this is the only technique that can recognize simultaneously the presence of both amyloid proteins and theirs post-translational modification.

Key Words: amyloid fibrils micro-extraction, amyloid typing, SDS-PAGE and Western Blot
PT089

CARDIAC MAGNETIC RESONANCE TO ASSESS TREATMENT RESPONSE IN CARDIAC IMMUNOGLOBULIN LIGHT CHAIN AMYLOIDOSIS – RESULTS AT 1 YEAR POST-CHEMOTHERAPY

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Introduction: Cardiac involvement in immunoglobulin light chain amyloidosis (AL) is the major determinant of survival. Cardiac response to chemotherapy is conventionally assessed by serum brain natriuretic peptide (NT-proBNP) and echocardiography, but neither quantify amyloid burden.

Objectives: We assessed the hypothesis that CMR with T1 mapping and extracellular volume (ECV) can evaluate amyloid burden in cardiac AL amyloidosis tracking changes over time at 3 months, 6 months and 1 year after the initiation of chemotherapy.

Methods: 94 patients with cardiac AL amyloidosis were studied serially using CMR with T1 mapping and ECV at baseline, 3 months, 6 months and 1 year post-chemotherapy.

Results: At 6 months, 62% of patients achieved a complete response (CR) or very good partial haematological response (VGPR), and 38% a partial response (PR) or no response (NR). Amyloid regression was detectable in only 1 patient, however, there was amyloid progression in 34% of patients (Figure, panel A). Although this occurred in the PR group, it also occurred in the CR and VGPR groups (63%).

At one year, 64% of patients achieved a CR or VGPR. Regression of amyloid was seen in 30% of patients (Figure, panel B), all with CR or VGPR and 0 patients in PR or NR (p<0.05). However, not every patient with CR or VGPR had amyloid regression by CMR, having 4 of these patients amyloid progression. Interestingly, these 4 patients achieved the good haematological response late (after the first 6 months of chemotherapy). 46% of patients with changes in ECV consistent with amyloid regression had also visual changes in the pattern of LGE. Amyloid regression was associated with significant reduction in LV mass and NT-pro BNP (p<0.05) and in native T1 (p<0.01).

Conclusion: In newly diagnosed and treated AL amyloidosis, CMR demonstrates the dynamic biology of infiltration: increasing rapidly, particularly if chemotherapy fails to switch off light chain production promptly; regressing more slowly if effective. Serial monitoring of myocardial infiltration has the potential for new AL amyloidosis therapeutic regimes based on myocardial organ response.

Keywords: AL amyloidosis, cardiac response, CMR
POOR RIGHT VENTRICULAR FUNCTION IS ASSOCIATED WITH IMPAIRED EXERCISE CAPACITY AND VENTILATORY EFFICIENCY IN TRANSTHYRETIN CARDIAC AMYLOID PATIENTS


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4 Cardiac Rehabilitation Unit, Department of Experimental & Clinical Medicine, University of Florence Italy.
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Background: Cardio-Pulmonary Exercise Test (CPET) is the gold standard to evaluate functional capacity in patients at high risk of heart failure (HF). Few studies, with a limited number of subjects and conflicting results, analyzed the role of CPET in patients with systemic amyloidosis.

Objectives: Aims of our study were the assessment of the response to exercise in patients with Transthyretin amyloid (ATTR) cardiomyopathy (CA) and the correlation of clinical, biohumoral and echocardiographic parameters with CPET parameters, such as oxygen consumption (VO2) peak and ventilator equivalent for carbon dioxide (VE/VCO2) slope.

Methods: From February 2018 to March 2019, 72 cardiac ATTR patients were prospectively enrolled and underwent a complete clinical, biohumoral, echocardiographic and CPET assessment.

Results: All patients completed the exercise stress test protocol without any adverse event. At CPET, they achieved a mean VO2 peak of 14 mL/Kg/min and a mean VE/VCO2 slope of 31. The blood pressure response to exercise was inadequate in 26 (36%) patients (flat in 25 and hypotensive in 1), while 49/72 patients (69%) showed an inadequate heart rate recovery. At multivariate analysis, tricuspid peak systolic tissue velocity (s′) was the only independent predictor of VO2 peak, while both tricuspid annular plane systolic excursion (TAPSE) and tricuspid s’ – assessed in two different models to avoid collinearity – were independently associated with the VE/VCO2 slope.

Conclusion: Our data demonstrate the role of right ventricular function as an independent predictor of exercise capacity and ventilator efficiency in ATTR. At CPET evaluation, a significant proportion of patients presented an abnormal arterial pressure response and heart rate variation to exercise.
SEVERE WEIGHT LOSS INDICATES MORE ADVANCED RESTRICTIVE PATHOPHYSIOLOGY OF HEART DISEASE IN POLISH PATIENTS WITH LIGHT-CHAIN CARDIAC AMYLOIDOsis


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Background: Unintentional weight loss is a sign of light-chain amyloidosis (AL amyloidosis) with complex etiology. Intestinal involvement causing diarrhea, loss of appetite resulting from the neoplastic character of the disease or abdominal organs congestion are considered.

Objectives: The aim of the study was to assess the frequency of severe weight loss (SWL) in Polish patients (pts) with cardiac AL amyloidosis and to investigate if this sign indicates more advanced cardiac involvement and worse prognosis.

Methods: The analysis covered all consecutive pts with cardiac AL amyloidosis diagnosed in the cardiology department from August 2011 to August 2019. SWL was defined as weight loss >10 kilos within one year. The duration of heart disease symptoms such as effort dyspnea, chest pain or orthopnea was assessed. Serum concentrations of free light-chains, NT-proBNP, high-sensitivity troponin T (hs-TnT), creatinine, albumin, total protein, bilirubin and activity of alanine aminotransferase, γ-glutamyltransferase and alkaline phosphatase were measured. Pulmonary circulation and the presence of pleural effusion were assessed in chest X-ray. During echocardiography, tissue Doppler imaging was used to assess an average early mitral annulus velocity (e’) and longitudinal myocardial velocities of left (LV s’) and right (RV s’) ventricles. Standard parameters were measured including the ratio of the transmitral early peak velocity (E) estimated by pulsed wave Doppler over the average e’ (E/e’ ratio).

Results: Sixty-four pts (median age 61.5 yrs) were enrolled. Median (interquartile range, IQR) duration of heart disease symptoms was 11 (7-16) months. Median (IQR) left ventricular ejection fraction (LVEF) was 53% (40-65). SWL and diarrhea were reported by 50% and 13% of pts, respectively. There were significant differences (p<0.05) between the SWL(+) and the SWL(-) groups in regard to: NT-proBNP (median, 11017 vs. 2823 pg/ml), hs-TnT (134 vs. 37 ng/l), left ventricular posterior wall (16.7 vs. 15 mm), end-diastolic diameter (LVEDD, 42.5 vs. 45 mm) and systolic volume (SV, 33 vs. 52 ml), tricuspid annular plane systolic excursion (TAPSE, 13.7 vs. 16.5 mm), LV s’ (4 vs. 5 cm/sec), RV s’ (9 vs. 11 cm/sec), e’ (4 vs. 5.4 cm/sec) and E/e’ ratio (20 vs. 17). Pleural but not pericardial effusion was more frequent in the SWL(+) group (59% vs. 34 %, p=0.045). There were no differences in regard to the duration of heart disease symptoms, age, other laboratory tests, other echocardiographic parameters and frequency of diarrhea or pulmonary congestion. Median (IQR) survivals for the SWL(+) and the SWL (-) groups were 3 (2-5.3) and 16 (7-23) months, respectively (p=0.005). The Kaplan-Meier curves analysis showed that SWL predicted worse survival (Log rank test, p<0.0001).

Conclusions: Severe weight loss is a frequent sign of cardiac AL amyloidosis in Polish patients. Cardiologists should be aware that this sign is connected with more advanced disease and higher mortality.
INCIDENTAL HIGH MYOCARDIAL UPTAKE ON DPD SCINTIGRAPHY.

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Introduction: Tc-DPD (diphosphono-propanodicarboxylic acid) is first and foremost a scintigraphic agent used in bone pathology, particularly for detection of bone metastases in breast and prostate cancer. Incidental myocardial uptake is extremely rare with another bone scintigraphic agent MDP (methylene diphosphonate), and in our experience, uncommon with HDP (hydroxymethylene diphosphonate). We have addressed incidentally found high myocardial uptake in a patient population referred for bone scintigraphy, recording also relevant comorbidity and life expectancy.

Materials and methods: In a cohort of 1195 patients, examined between 2009-2013 high myocardial uptake, Perugini grade 2 or 3, was found in 10 patients, 9 males and one female, mean age 79 years. Nine of the patients were accessible for follow up. In two of these patients an HDP scintigraphy was also performed.

Results: At follow up, all nine patients with available results were deceased. Median survival was 2 years, but three of the patients were dead within 7 months.

Additionally it was mentioned in the referral that 2 patients had carpal tunnel syndrome and 8 patients had various cardiac conditions.

More anecdotally, the two cases of HDP scintigraphy in patients examined with DPD, one two years prior to DPD, the other two years after DPD, were both described as negative. Retrospectively the patient re-examined after two years would be classified as a Perugini 1. He then also presented with numerous bone metastasis.

Conclusion: In an ongoing clinical trial we have found a high mortality rate which lead us to review patients with incidental high cardiac uptake. Incidental cardiac uptake was found in 1% of the patients referred for bone scintigraphy. In this small cohort of 9 patients, 3 were deceased within 7 months. This results is, however, biased by comorbidity, as all these patients had malignancies.

Keywords: ATTRwt, cardiac amyloidosis, DPD.
Predictive Factors for Non-Invasive Detection of Transthyretin Cardiac Amyloidosis in 756 Patients Undergoing Technetium-99m Pyrophosphate Scanning

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Introduction/Background: The diagnosis and management of transthyretin cardiac amyloidosis (ATTR-CA) continues to advance as non-invasive testing pathways and new therapies become widely available.

Objectives: The aim of this study was to evaluate how non-invasive testing pathways are being used and to identify predictive factors for ATTR-CA diagnosis.

Methods: We analyzed the results of 99m-technetium pyrophosphate (PYP) nuclear scanning in 756 consecutive patients scanned from December 22, 2010 to October 3, 2019 at a single medical center which serves as a primary care center for a multi-ethnic urban population as well as a quaternary amyloidosis referral site. Demographics, echocardiography, laboratory, and biopsy results were abstracted from the medical record. PYP scans were graded using visual scoring from 0-3 and quantitative scoring using the counts over the heart to the contralateral chest ratio (H:CL). Logistic regression was used for multivariable analysis.

Results: 516 patients (68%) were male, mean age was 77 years, 14% were black, and 7% were Hispanic. 312 had visual score of 0 (41%), 174 of 1 (23%), 45 of 2 (6%), and 225 of 3 (30%). H:CL ratio was calculated in 753 patients with a median 1.27 (IQR 1.10-1.64). 257 (34%) had H/CL ≥1.5. 170 of 581 (29%) patients who underwent PYP testing for clinical (non-research) indications did not receive both serum protein electrophoresis and serum light chain testing. In analysis of 105 patients undergoing cardiac biopsy, grade 2 or 3 PYP scan had sensitivity of 91% and specificity 90% in distinguishing between patients with and without ATTR-CA on biopsy. Through the study period, PYP scanning frequency increased from a mean of 15 per year to 136 per year from 2010-2014 to 2015-2019, respectively. Race/ethnicity were not significantly associated with a grade 2-3 scan. Median left ventricular posterior wall thickness (LVPWT), measured by echocardiography in 738 patients, was 11 and 15 mm in patients with visual score 0-1 and 2-3, respectively. Biopsy-diagnosed patients had a median LVPWT of 16 mm whereas patients diagnosed with grade 2-3 PYP without biopsy had a median of 14 mm. LVPWT was significantly associated with grade 2-3 PYP (C-statistic 0.82, 95% CI 0.79-0.85). In multivariable analysis, LVPWT (OR 1.42/mm), interventricular septal thickness (OR 1.12/mm), older age (OR 1.06/year), and male sex (OR 2.16) were significantly associated with grade 2-3 scan.

Conclusions: Non-invasive testing pathways are frequently used to evaluate for ATTR-CA, and there is some suggestion that patients diagnosed non-invasively are more likely to be found at an earlier stage of disease. However, patients undergoing PYP scanning frequently do not undergo monoclonal protein testing to rule out AL amyloidosis as per guidelines. LV wall thickness, increased age, and male gender are predictive of a grade 2-3 PYP scan and inclusion of these factors in risk models may identify patients most likely to have ATTR-CA.

Disclosures: TJP owns stock in Abbott Laboratories and Abbvie, Inc. AJE serves as a consultant to W. L. Gore and Associates; his institution receives funding from NIH, Canon Medical Systems USA, Roche Medical Systems, and W. L. Gore and Associates. MSM’s institution receives funding from NIH/NIA, Pfizer, Inc., Novartis, Ionis Pharmaceuticals, Prothena Biosciences Limited, EIDOS Therapeutics, Inc.

Keywords: Transthyretin amyloidosis, Cardiac amyloidosis, Bone scintigraphy.
ASSOCIATION OF CARDIAC BIOMARKERS WITH OXYGEN CONSUMPTION IN AL AND TRANSTHYRETIN CARDIAC AMYLOIDOSIS: FURTHER EVIDENCE OF LIGHT CHAIN TOXICITY

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Background: Cardiac involvement and dysfunction are common in patients presenting with AL and ATTR cardiac amyloidosis. Cardiopulmonary exercise testing (CPET) performance is the gold standard to measure functional capacity. While ejection fraction poorly correlates with exercise capacity, the relationship with other echocardiographic parameters or biomarkers in cardiac amyloidosis is not known.

Objectives: To explore correlations between biomarkers and echocardiographic parameters with CPET performance in AL and TTR cardiomyopathy and assess the predictive value of CPET for survival in patients with AL and ATTR amyloidosis.

Methods: CPET results, echocardiographic dimensions and biomarkers for 17 patients with AL and 24 patients with TTR amyloidosis (63.7 ± 7.8 years, 72.5% male) were collected. We estimated end systolic, diastolic volumes, LV mass and volume from M-mode echo to calculate EF and myocardial contraction fraction (MCF). Correlations were evaluated using Pearson correlation coefficients between percent predicted VO2 max, volumes, MCF and cardiac biomarkers. We used Cox regression analysis with age, Percent predicted VO2 max, VE/VCO2, NT-proBNP, Troponin T, and serum creatinine to study predictors of mortality.

Results: Percent predicted peak VO2 strongly correlated with NT-proBNP (r = -0.57, P=0.006), Troponin T (r = -0.70, p <0.001) and revised Mayo stage (r = -0.67, p = <0.001) in the AL subgroup. In ATTR, similar although weaker association of Percent predicted peak VO2 with NT-proBNP (r = -0.4, P = 0.04) and Troponin T (r = -0.57, P = 0.002) were observed. Ejection fraction (63% vs. 54%, p<0.001), MCF (0.31 vs. 0.15, p<0.001) and resting stroke volume (61 vs. 45 mL, p=0.002) were significantly higher in the AL amyloidosis, whereas left ventricular mass was significantly greater in TTR amyloidosis (331.9 vs. 215.0 g, p=0.001). EF and MCF were not associated with peak VO2 (r=0.110, p=0.492 and r=-0.01, p=0.934, respectively. Long term survival data was available in 37/41 patients and with a median follow-up of 40 months (range 6.3-119 months); 8/37 patients died. On univariable cox regression analysis, percent predicted peak VO2 (HR 0.95, P=0.03) and Troponin T (HR 14.8, P=0.03) were significantly associated with mortality and on multivariable analysis only age (HR 1.15, P=0.01) was associated with mortality.

Conclusion: NT-proBNP and Troponin T are associated with percent predicted peak VO2, whereas EF and MCF are not. The association of cardiac biomarkers with peak VO2 is stronger for AL-CA than ATTR-CA despite lower LVM in the former, suggesting further evidence for light chain toxicity. Furthermore, there is an association of Percent predicted peak VO2 with survival of patients with Cardiac amyloidosis, which should be evaluated in prospective manner.
MIRCROCALCIFICATIONS ASSOCIATED WITH TRANSTHYRETIN AMYLOIDOSIS: AN AUTOPSY STUDY

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Introduction: Amyloid precursor proteins, such as ATTR and AL, are the major types of amyloidogenic proteins in systemic amyloidosis, in which patients develop cardiac, neurological, and other organ dysfunction. Bone scintigraphy utilizing either pyrophosphate (PYP) or 3,3-diphosphono-1,2-propanodicarboxylic acid (DPD) has become a more common tool to detect cardiac ATTR amyloidosis. PYP and DPD reportedly bond to calcium, and microcalcifications associated with amyloid deposits were reportedly more frequently found in ATTR amyloidosis in comparison with AL amyloidosis. However, precise histopathological findings of microcalcifications remain to be elucidated in both type of amyloidosis.

Materials and Methods: We employed 35 cases (20 cases with hereditary ATTR (ATTRv) amyloidosis, 3 cases with wild-type ATTR (ATTRwt) amyloidosis, 7 cases with AL amyloidosis, and 5 cases without amyloidosis) autopsied at Kumamoto University from 1990 to 2018. We investigated cardiac microcalcifications in those autopsy cases by means of modified von Kossa staining (Calcium Stain Kit, ScyTek Laboratories, West Logan, UT, USA). Using those cardiac tissue samples, we also performed immunoblotting with antibodies against TTR 49-127 to detect C-terminal fragments of TTR from amyloid deposits and performed mass spectrometric analyses to determine molecules associated with cardiac microcalcifications in ATTR amyloidosis.

Results: Cardiac microcalcifications were found in 12 (60%) of 20 cases with ATTRv amyloidosis, 3 (100%) of 3 cases with ATTRwt amyloidosis, 7 cases with AL amyloidosis, and 5 cases without amyloidosis. Cardiac microcalcifications were more frequently found in ATTRv amyloidosis cases with C-terminal fragments of TTR in amyloid deposits (type-A) in comparison to cases without C-terminal fragments of TTR in amyloid deposits (type-B). Mass spectrometric analyses revealed that TTR, lipopolysaccharide-binding protein, prolargin were more frequently found in amyloid deposits of cardiac tissue in comparison to cardiac tissue areas outside of amyloid deposits in ATTR amyloidosis.

Conclusion: Cardiac microcalcifications may be associated with ATTR amyloidosis.
TECHNETIUM PYROPHOSPHATE SCANNING FOR DIAGNOSING CARDIAC TRANSTHYRETIN AMYLOIDOSIS IN A COMMUNITY SETTING

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Boston Healthcare System

Background: The amyloid community has long maintained that low familiarity in the medical community with cardiac transthyretin (TTR) amyloidosis (ATTR) leads to its underdiagnosis. The diagnosis of ATTR until recently depended on a biopsy containing amyloidosis. Technetium pyrophosphate (TcPyP) scanning now offers a simpler diagnostic test for cardiac ATTR. With new ATTR treatments available, clinicians might increasingly appreciate the importance of diagnosing ATTR.

Objectives: To evaluate the use of TcPyP scanning in a community (non referral) setting.

Methods: The USA Department of Veterans Affairs (VA) consists 23 networks that care for armed forces veterans. Nuclear medicine records from one VA network were reviewed to identify patients who have had a TcPyP scan ordered to evaluate for amyloidosis. For patients with multiple scans, only the first was included. Scans were evaluated using standard scoring (Perrugini score comparing left ventricular uptake: bone uptake and the ratio of cardiac to contralateral thorax uptake). Scans with a ratio ≥1.5 were considered positive.

Results: From 2013 through October 2019, 108 patients had at least one scan. From 2015-2019, the use of TcPyP scanning has nearly doubled annually (table 1). The 108 scans were ordered by 74 practitioners and read by 22 radiologists at four sites. 45 scans were positive (ratio ≥1.5, including all 39 with score=3 and 6/10 with score=2; table 2). The 59 negative scans were done in patients with an average age of 73 (range 51-93, including 24 patients ≤ age 70). The 45 positive scans were done in patients with an average age of 84 (range 66-98). Of the positives, only two were under age 74: one TTR V122I heterozygote, age 66, with a score = 3, and one patient age 68 of unknown TTR genotype with a borderline positive scan (score 2, ratio 1.59). Three patients known to carry TTR V122I all had a score of 3. No other patients are known to carry a variant.

Conclusions: Orders for TcPyP scanning have been nearly doubling annually recently in a community setting (non amyloidosis referral center). In patients under age 74, nearly all scans were negative, suggesting overuse of scans in patients with a low pre-test probability of a positive scan. While continued education about the prevalence of cardiac ATTR remains warranted, it is also important to educate the medical community about typical clinical features of cardiac ATTR, to avoid unnecessary testing unlikely to be clinically useful. The long-held view among amyloid specialists that the wider medical community is failing to consider cardiac amyloidosis appears, finally, no longer to apply.
NEW ORAL ANTICOAGULANTS VERSUS VITAMIN K ANTAGONISTS AMONG PATIENTS WITH CARDIAC AMYLOIDOSIS: A WARNING

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Introduction: Atrial fibrillation (AF) is common among patients with cardiac amyloidosis (CA), who have an increased risk of intracardiac thrombus. The aim of this study was to explore the effectiveness and safety of vitamin K-antagonists (VKA) and direct oral anticoagulants (DOAC) in patients with CA.

Methods and Results: Between January 2008 and April 2019, 163 patients with CA and history of AF receiving oral anticoagulant were enrolled in the study. There were 115 (71%) and 48 (29%) patients with VKA and DOAC, respectively. As compared to patients in the DOAC group, patients with VKA had more frequently light chain amyloidosis (39 versus 10%; P<0.001), with a decreased renal function (estimated glomerular filtration rate 39±22 ml/min versus 57±16 ml/min; P<0.001) and higher NT-proBNP level (4881 ng/L [2640-14124] versus 1829 ng/L [3460-5621]; P=0.02). A total of 93 (57%) patients met the primary endpoint of all-cause mortality: 77 (67%) and 16 (33%) among patients with VKAs and DOACs, respectively (P<0.0001). After multivariate analysis including age and renal function, VKA was no longer associated with all-cause mortality HR 0.85 (95M confident interval 0.38 - 1.87; P = 0.678). There was no difference between groups for thrombo-embolic events (17 versus 15%; P= 0.819) and no difference for bleeding complications (22 versus 21%; P= 1.000)

Conclusions: Among patients with CA and history of AF receiving oral anticoagulant, VKA are associated with an increased mortality, which disappears after adjustment for age and renal function. Finally, DOAC do not show difference for effectiveness and safety than VKAs.
TRANSTHYRETIN CARDIAC AMYLOIDOSIS: IMPROVING DIAGNOSTIC PERFORMANCE OF TECHNETIUM-99M PYROPHOSPHATE IMAGING

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1 Mayo Clinic

Introduction: Technetium-99m pyrophosphate (99mTc PYP) scintigraphy is considered reliable for the non-invasive diagnosis of transthyretin cardiac amyloidosis (TTR-CA). Conventional planar heart-to-contralateral uptake ratios (H/CL) may be confounded by blood pool/extra-cardiac isotope uptake reducing the diagnostic accuracy of planar imaging.

Objectives: To determine if single-photon emission computed tomography-computed tomography (SPECT-CT) acquisition improves the performance of 99mTc PYP planar scintigraphy for the diagnosis of TTR-CA.

Methods: We performed a retrospective analysis of all patients undergoing simultaneous 99mTc PYP planar and SPECT-CT scintigraphy for suspected TTR-CA at Mayo Clinic from 2014 to 2019 (n=1011). At 3 hours, a planar H/CL > 1.3 was considered positive (POS) and ≤ 1.3 was negative (NEG). SPECT-CT images were assessed for myocardial uptake by visual inspection and coded POS or NEG. Clinical data were categorized by planar/SPECT-CT concordance.

Results: See Table 1. A total of 584 patients were NEG by both planar and SPECT-CT, while 427 patients were POS by planar imaging. Of these planar POS patients, 71 (16.6%) were SPECT-CT NEG. Compared to patients POS by both modalities, planar POS/SPECT-CT NEG patients had lower H/CL values, were more commonly female, and had received higher 99mTc PYP doses. Further, they had lower NTproBNP and troponin T levels, and less severe perturbations in cardiac structure and function on echocardiography. Additionally, 25 (4.3%) patients were POS by SPECT-CT but not by planar imaging. Their clinical/echo characteristics were similar to planar POS/SPECT-CT POS patients (p>0.18 for all) except they had thinner walls and greater troponin T levels (p≤0.01 for both). Of these planar NEG/SPECT-CT POS patients, 3 had an endomyocardial biopsy all of which were positive for ATTR on tissue mass spectrometry.

Conclusions: One in six patients with POS planar imaging had no evidence of myocardial 99mTc PYP uptake on SPECT-CT. These patients have a distinct phenotype which suggests the absence of TTR-CA. Of planar NEG patients, ~4% had myocardial 99mTc PYP uptake on SPECT-CT with a similar clinical phenotype to planar POS/SPECT-CT POS patients. These cases may represent early myocardial ATTR deposition that is elusive to planar imaging, and in whom endomyocardial biopsy is necessary to definitively establish the diagnosis of TTR-CA. These data support combined use of planar and SPECT-based methods for diagnosis of TTR-CA as stipulated by recently published expert consensus.
<table>
<thead>
<tr>
<th>Table 1</th>
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<tr>
<td><strong>Mean ± SD, Median (IQR) or n (% values shown)</strong></td>
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<tr>
<td>---------------------------------</td>
</tr>
<tr>
<td><strong>Age (yrs)</strong></td>
</tr>
<tr>
<td><strong>Male</strong></td>
</tr>
<tr>
<td><strong>99mTc PYP Dose (mCi)</strong></td>
</tr>
<tr>
<td><strong>HtCl Ratio</strong></td>
</tr>
<tr>
<td><strong>eGFR by Creatinine (mL/min/m²)</strong></td>
</tr>
<tr>
<td><strong>Troponin T (ng/mL)</strong></td>
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<tr>
<td><strong>High Sensitivity Troponin T (ng/L)</strong></td>
</tr>
<tr>
<td><strong>NT-proBNP (pg/mL)</strong></td>
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<tr>
<td><strong>LVEF (%)</strong></td>
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<tr>
<td><strong>LVEDd (mm)</strong></td>
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<tr>
<td><strong>SV/BSA (mL/m²)</strong></td>
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<tr>
<td><strong>Septal Wall Thickness (mm)</strong></td>
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<tr>
<td><strong>Relative Wall Thickness (%)</strong></td>
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<tr>
<td><strong>LV Mass/BSA (g/m²)</strong></td>
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<tr>
<td><strong>LA Volume (mL)</strong></td>
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<tr>
<td><strong>LV Global Strain (%)</strong></td>
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<tr>
<td><strong>RV Function (TAPSE, mm)</strong></td>
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<tr>
<td><strong>Mitral e’ (m/sec)</strong></td>
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<td><strong>E/E’</strong></td>
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</table>

**Abbreviations:** Per text; otherwise, eGFR, estimated glomerular filtration rate; BSA, body surface area; NT-proBNP, N-terminal pro-brain natriuretic peptide; LV, left ventricular; LVEF, LV ejection fraction; SV, stroke volume; LVEDd, LV end-diastolic dimension; RV, right ventricular; TAPSE, tricuspid annular plane systolic excursion. * Chi Squared p value.
PT099

PREVALENCE AND CLINICAL SIGNIFICANCE OF ORTHOPEDIC RED FLAGS IN CARDIAC AMYLOIDOSIS

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Introduction: Orthopedic manifestations have been described as preceding symptomatic cardiac disease in transthyretin cardiac amyloidosis (ATTR-CA). The prevalence and predictive accuracy of individual and combinations of orthopedic red flags (RFs) in wild-type and hereditary ATTR-CA (ATTRwt and ATTRh) genotypes has not been evaluated. We hypothesize that a constellation of orthopedic RFs would be more predictive of ATTR-CA compared to orthopedic RFs individually.

Objectives: Better characterization of constellations of orthopedic manifestations in cardiac amyloidosis may help clinicians identify this disease earlier in its course so that patients can benefit from recent diagnostic and therapeutic innovations.

Methods: We studied 397 patients with ATTR-CA (74.1±9.67 years, range 31-95 years; male/female 84%/16%, ATTRwt /ATTRh 70%/30%) and 57 patients with HF of a non-amyloid etiology served as controls. A chart review was conducted by a single observer (RK) for the presence of 4 RFs in the clinical history: Carpal Tunnel Syndrome (CTS), Spinal Stenosis (SS), Tendinopathies including tendon tear and tendon rupture (T), and Joint Replacement (JR). Prevalence of individual RFs as well as common combinations of two and three RFs (RF clusters) were described. The sensitivity (Se), specificity (Sp), negative predictive value (NPV), and positive predictive value (PPV) of orthopedic RFs for ATTR-CA were analyzed for individual RFs and for RF clusters.

Results: Subjects with ATTR-CA differed in age from controls (74±10 vs. 88±11 years, p < 0.001), and were more often males (84.8% vs 29.8%, p<0.001). The prevalence of at least one RF was 68.3% in ATTR-CA and did not differ by genotype, and 56% of subjects had greater than one RF. The most common single RF was CTS, which had the highest Sn and Sp for ATTR-CA. Sp for ATTR-CA was higher for RF clusters than for individual RFs, with the highest Sp being achieved for the combination of CTS + SS + JR (Table 1).

Conclusions: The majority of patients with ATTR-CA had greater than one orthopedic RF, and prevalence of individual RFs and RF clusters did not vary by genotype. Sp of orthopedic RFs for ATTR-CA as an etiology for HF is higher for RF clusters than for individual RFs, with combinations of 3 RFs achieving Sp as high as 96.4%.

Table 1. Prevalence, Sn, Sp, PPV, and NPV value of RFs for ATTR-CA.

<table>
<thead>
<tr>
<th>RF</th>
<th>ATTR-CA N (%)</th>
<th>Controls N (%)</th>
<th>Sn</th>
<th>Sp</th>
<th>PPV</th>
<th>NPV</th>
</tr>
</thead>
<tbody>
<tr>
<td>CTS</td>
<td>204 (51.4)</td>
<td>6 (51.4)</td>
<td>51.4</td>
<td>89.3</td>
<td>97.1</td>
<td>20.6</td>
</tr>
<tr>
<td>SS</td>
<td>101 (25.4)</td>
<td>17 (25.4)</td>
<td>25.4</td>
<td>69.6</td>
<td>85.6</td>
<td>11.6</td>
</tr>
<tr>
<td>JR</td>
<td>94 (23.7)</td>
<td>11 (23.7)</td>
<td>23.7</td>
<td>80.4</td>
<td>89.5</td>
<td>12.9</td>
</tr>
<tr>
<td>T</td>
<td>69 (17.4)</td>
<td>15 (17.4)</td>
<td>17.4</td>
<td>73.2</td>
<td>82.1</td>
<td>11.1</td>
</tr>
<tr>
<td>CT+SS</td>
<td>68 (17.1)</td>
<td>4 (12.3)</td>
<td>17.1</td>
<td>92.9</td>
<td>94.4</td>
<td>13.6</td>
</tr>
<tr>
<td>CT+JR</td>
<td>60 (15.1)</td>
<td>4 (15.1)</td>
<td>15.1</td>
<td>92.9</td>
<td>93.8</td>
<td>13.4</td>
</tr>
<tr>
<td>CT+T</td>
<td>49 (12.3)</td>
<td>5 (12.3)</td>
<td>12.3</td>
<td>91.0</td>
<td>90.7</td>
<td>12.8</td>
</tr>
<tr>
<td>SS+T</td>
<td>35 (8.8)</td>
<td>9 (8.8)</td>
<td>8.8</td>
<td>83.9</td>
<td>79.6</td>
<td>11.5</td>
</tr>
<tr>
<td>SS+JR</td>
<td>33 (8.3)</td>
<td>5 (8.3)</td>
<td>8.3</td>
<td>90.0</td>
<td>86.8</td>
<td>12.3</td>
</tr>
<tr>
<td>T+JR</td>
<td>18 (4.5)</td>
<td>7 (4.5)</td>
<td>4.5</td>
<td>87.5</td>
<td>72</td>
<td>11.5</td>
</tr>
<tr>
<td>CT+SS+T</td>
<td>29 (7.3)</td>
<td>4 (7.3)</td>
<td>7.3</td>
<td>92.9</td>
<td>87.9</td>
<td>12.4</td>
</tr>
<tr>
<td>CT+SS+JR</td>
<td>22 (5.5)</td>
<td>2 (5.5)</td>
<td>5.5</td>
<td>96.4</td>
<td>91.7</td>
<td>12.6</td>
</tr>
<tr>
<td>CT+T+JR</td>
<td>12 (3.0)</td>
<td>3 (3.0)</td>
<td>3.0</td>
<td>94.6</td>
<td>80.0</td>
<td>12.1</td>
</tr>
<tr>
<td>SS+T+JR</td>
<td>12 (3.0)</td>
<td>4 (3.0)</td>
<td>3.0</td>
<td>92.9</td>
<td>75.0</td>
<td>11.9</td>
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</table>
PT100

IDENTIFYING CARDIAC AMYLOIDOSIS PATIENTS FROM A LARGE CROSS-DATABASE (ELECTRONIC HEALTH RECORD AND INSTITUTIONAL REGISTRY OF AMYLOIDOSIS) IN BUENOS AIRES, ARGENTINA

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1 IMTIB

Introduction: Amyloidosis is an underdiagnosed disease with an estimated global incidence of fewer than 10 cases per million patient-years. The Hospital Italiano de Buenos Aires (HIBA) in Argentina has an institutional registry of amyloidosis (IRA) and electronic health records (EHR). This project has cross-database references (EHR-IRA) to identify amyloidosis patients and improve understanding of the disease. Currently, diagnosis requires a tissue biopsy to confirm the disease. Alternatively, it would be highly beneficial to use EHR to analyze the frequency of amyloidosis and validate models that will allow its early detection. We sought to build a predictive model using supervised machine learning tools to accurately predict patients with cardiac amyloidosis that consult our medical institution.

Objective: To design, develop and evaluate a predictive model to identify patients with cardiac amyloidosis.

Design: We performed a dynamic retrospective cohort study of all adult patients (>17 years old) admitted (outpatient or inpatient) at several medical units at the HIBA from January to December 2018. We represented each patient as a vector using information from EHR. Each feature vector included structured demographics (such as gender, age, BMI), diagnosis, blood pressure, biomarkers, echocardiographic data, laboratory data and number of consultations with different professionals (such as neurology, cardiology, hematology, among others) per year. A supervised predictive model was developed and evaluated through machine learning tools and over-sampling technique. A treebag predictive model was used to identify incident cases with cardiac amyloidosis from the database based on the HIBA population.

Results: We selected 4700 patients. A total of 25 amyloidosis patients were identified with a cross-database search (IRA-EHR) and these were used as positive cases (gold standard). The remaining cases were considered as negative cases. Splitting was used with 70/30% for training and testing of the model. After training, the model predicted the presence of cardiac amyloidosis in 5 patients that were used for testing. The model had an AUC of 0.7, accuracy of 0.8 and F1-score of 0.4. The most important predictive variables were left ventricular ejection fraction, tissue doppler velocities, age, carpal tunnel syndrome, BMI.

Conclusion: This model identifies cardiac amyloidosis patients from a large cross-database, we believe that the project can contribute to enrolling patients candidates for screening. Early detection allows for treatments that will improve health outcomes.
PT101

ECHOCARDIOGRAPHIC THRESHOLDS FOR EARLY DIAGNOSIS OF CARDIAC INVOLVEMENT IN HEREDITARY TRANSTHYRETIN AMYLOIDOSIS

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Background: Early detection of cardiac involvement in patients with transthyretin hereditary amyloidosis (ATTRv) using routine tools such as transthoracic echocardiography is particularly important since the introduction of treatments reducing mortality in this indication.

Objectives: To determine transthoracic echocardiography (TTE) thresholds to predict early cardiac involvement in patients with ATTRv on the basis of technetium-labeled bone scintigraphy (TcBS) cardiac uptake (CU).

Methods: We carried out a bicentric study on ATTRv patients who underwent TTE and TcBS as part of their standard diagnostic workup. Ninety-four patients with ATTRv were included and divided according to the Perugini score.

Results: Patients with CU were older, more symptomatic and were more likely to have atrial fibrillation. Low voltage prevalence and biomarkers levels were not different between scores. Patients with CU had lower left ventricular (LV) ejection fraction and global longitudinal strain (LS), thicker LV wall and more LV diastolic dysfunction than patients without CU. None of these parameters was different between scores. Finally, considering only patients in the early stage of the disease (scores 0 and 1), a mean LV wall thickness of 11 mm [area under the curve (AUC) 0.95; 95% confidence interval (CI) 0.88–1.00], a lateral Ea wave of 8 cm/s [AUC 0.96; 95% CI 0.90–1.00], a E/Ea ratio of 11 [AUC 0.93; 95% CI 0.81–1.00] or a global LS of -17% [AUC 0.93; 95% CI 0.82–1.00] were sensitive (90%) and specific (>85%) for detecting cardiac involvement.

Conclusion: Among patients with ATTRv, mean LV wall thickness ≥ 11 mm, global LS ≥ -17%, lateral Ea wave > 8 cm/s and E/Ea ratio > 11 should lead to TcBS to detect early stage of cardiac involvement.
VALUE OF LONGITUDINAL STRAIN TO IDENTIFY WILD TYPE TRANSTHYRETIN AMYLOIDOSIS IN PATIENTS WITH AORTIC STENOSIS

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Toulouse University Hospital.

Background: Wild-type transthyretin amyloidosis (ATTRwt) and degenerative aortic stenosis (AS) are both age-related. Diagnostic of cardiac amyloidosis (CA) among patients with AS may be difficult due to overlapping morphological and functional criteria.

Objectives: To study echocardiographic longitudinal strain (LS) pattern among patients with AS with and without ATTRwt.

Methods: Patients with AS with ATTRwt (n=30), AS without ATTRwt (n=50) and ATTRwt without AS (n=31) underwent two-dimensional speckle-tracking echocardiography. Transthyretin CA was based on positive bone scintigraphy without monoclonal gammopathy.

Results: All patients showed a gradual decrease in LS from the base to the apex resulting in a decrease of the global LS. A cut-off value of 1.0 for relative apical LS (average apical LS/(average basal LS + mid-LS)) was sensitive (88%) but less specific (68%) in differentiating ATTRwt among patients with severe AS (area under the curve 0.74). The best cut-off value for relative apical LS for identifying patients with ATTRwt among the whole population was 0.9 (sensitivity 74%, specificity 66%, area under the curve 0.70). Furthermore, 35%, 25% and 11% of patients with ATTRwt without AS, with moderate AS and with severe AS, respectively, did not reach this cut-off value.

Conclusions: Decrease of global and relative apical LS are common in patients with AS, even in absence of ATTRwt. ATTRwt cardiac amyloidosis can be present even in absence of relative apical sparing of LS.
FEASIBILITY OF HEART TRANSPLANTATION FOR PATIENTS WITH AMYLOID CARDIOMYOPATHY ACCOMPANYING EXTRACARDIAC ORGAN INVOLVEMENT


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2 Department of Cardiology, University of Ulsan College of Medicine, Asan Medical Center, Seoul, South Korea.
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4 Department of Laboratory Medicine, University of Ulsan College of Medicine, Asan Medical Center, Seoul, South Korea.

Background: Cardiac involvement is associated with poor prognosis in immunoglobulin light-chain amyloidosis (AL amyloidosis), with a median survival of 6 months without heart transplantation (HT) from the onset of symptomatic heart failure. Although HT is an efficacious treatment option for patients with amyloid cardiomyopathy, consensus on its indication is not fully established because of limited experience.

Method: We reviewed our prospective AL amyloidosis database to identify all patients who received HT for their amyloid cardiomyopathy. Data regarding baseline characteristics, treatment and survival outcomes were obtained by reviewing the database.

Results: Out of 71 patients diagnosed with amyloid cardiomyopathy between Oct 2004 and Oct 2018, 11 patients undergoing HT were identified. There was no significant difference between those who underwent HT and not in terms of baseline characteristics including sex (female): 45.5% vs. 41.7%, ECOG performance status (0-1): 81.8% vs. 60.0%, number of extra-cardiac organ involvement (≥3) organs: 9.1% vs. 8.3%, Boston university (BU) cardiac stage (≥3): 63.7% vs. 66.7%, left ventricle (LV) dysfunction (ejection fraction < 50%): 54.5% vs. 36.7%, and high LV filling pressure measured by E/E’ [E/E’ >14]: 90.9% vs. 85.0%. Median time from diagnosis to HT was 4.1 months (range, 0.6-15.3). With a median follow-up of 56.5 months, 1-year and 2-year post-transplant survival rates were 90.9% and 72.7%. In comparison, 1-year and 2-year survival rates in those who did not undergo HT were 41.7% and 39.8%. Four transplant recipients died of herpetic encephalopathy and pneumonia while there was no death related to disease progression or heart failure. On the other hand, 34 patients (56.7%) among those who did not undergo HT died due to progressive heart failure. In the multivariate analysis, OS was significantly associated with HT (hazard ratio (HR) 0.2, [95% Confidential interval (CI), 0.1-0.6, p=0.006]), and bone marrow involvement of plasma cells >20% (HR 2.6, [95% CI, 1.3-5.3, p=0.009]). Interestingly, all patients with extra-cardiac organ involvement except two with 80% and 70% of clonal plasma cells in bone marrow were alive at 2 years from HT when hematologic response was achieved (5 patients with CR, and 1 with VGPR). Those two patients with heavy infiltration of plasma cells in bone marrow also had kidney involvement and did not achieve renal response before HT. On the other hand, prolonged overall survival of median 70.2 months was observed in patients achieving renal response before HT despite kidney involvement at the time of diagnosis.

Conclusion: HT might be a feasible therapeutic option even for patients with amyloid cardiomyopathy accompanying extracardiac organ involvement as long as hematologic and extra-cardiac organ response is achieved and plasma cell count in bone marrow is less than 20%.

Keywords: AL amyloidosis, Amyloid cardiomyopathy, Heart transplantation
THE EFFICACY, AFFORDABILITY AND LIMITATIONS OF DIFLUNISAL THERAPY IN TRANSTHYRETIN CARDIAC AMYLOIDOSIS (ATTR-CA)

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Introduction: Awareness of ATTR-CA has significantly increased with growing appreciation of non-invasive diagnostic techniques and the increasing number of a new generation of disease modifying therapies. Diflunisal is a transthyretin stabiliser that has been showed in several small studies to be associated with improved patient outcomes. However, there is limited evidence regarding the efficacy of long-term diflunisal use. Toxicity concerns in heart failure patients with ATTR-CA include gastrointestinal adverse effects and renal impairment. We aim to evaluate the current use and tolerance of diflunisal in a large Australian Amyloidosis Service.

Methods: All ATTR-CA patients who were treated with diflunisal therapy between 2014 and 2019 were identified from our institution’s dedicated amyloidosis database. Clinicopathological data were retrospectively collected and analysed. Patients who discontinued diflunisal therapy due to poor tolerance were compared to those who continued therapy with no adverse events.

Results: A total of 136 patients with ATTR-CA were identified. Sixty-two (45.6%) patients were treated with diflunisal. Cost to diflunisal was approximately USD $1/day. 30 patients (49%) discontinued diflunisal due to adverse side effects. Common reasons for discontinuation of therapy were renal impairment (12; 19.7%) gastrointestinal toxicity (8; 13.1%). In a 6 months landmark analysis of 95 patients, those who were never treated with diflunisal (45 patients) or ceased therapy early due to toxicity (15 patients) had an inferior median estimated survival (40.9 months) vs the 35 patients who continued diflunisal for >6 months (52.8 months; p=0.013). Median follow up was 18 months (range 6-50 months). However, patients intolerant or not treated with diflunisal were older and had more advanced stage at diagnosis.

<table>
<thead>
<tr>
<th></th>
<th>Tolerant (n = 32)</th>
<th>Intolerant (n = 30)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>Median (IQR)</td>
<td>76 (68-81)</td>
<td>82 (75-84)</td>
</tr>
<tr>
<td>Gender</td>
<td>Male</td>
<td>30 (96.8%)</td>
<td>25 (83.3%)</td>
</tr>
<tr>
<td>Gillmore Stage</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>24 (77.4%)</td>
<td>11 (36.7%)</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>4 (12.9%)</td>
<td>13 (43.3%)</td>
<td>0.005</td>
</tr>
<tr>
<td>3</td>
<td>3 (9.7%)</td>
<td>6 (20.0%)</td>
<td></td>
</tr>
<tr>
<td>Follow up (Months)</td>
<td>Median (IQR)</td>
<td>23 (12-32)</td>
<td>16 (8-32)</td>
</tr>
<tr>
<td>Duration of diflunisal therapy (Months)</td>
<td>Median (IQR)</td>
<td>17 (11-30)</td>
<td>2 (1-7)</td>
</tr>
<tr>
<td>Atrial Fibrillation</td>
<td>17 (54.8%)</td>
<td>21 (70.0%)</td>
<td>0.222</td>
</tr>
<tr>
<td>Intraventricular septum diameter (mm)</td>
<td>Median (IQR)</td>
<td>17 (15-19)</td>
<td>15 (14-17)</td>
</tr>
</tbody>
</table>

Table 1. Comparison of baseline characteristics.

Conclusions: In this retrospective study, Diflunisal appears to be affordable and possibly highly effective therapy in ATTR-CA in those patients who can tolerate it. However, discontinuation rates are high, especially in patients >75 years old and with advanced stage disease. We believe that diflunisal remains a reasonable therapy to consider in patients with no access to more novel therapies due to cost or licensing issues. Further prospective trials are desperately needed to confirm survival advantages with this stabiliser, identify patients who would benefit most from this therapy, and to assess if combination of diflunisal with a gene silencer are feasible and beneficial.
SUDOSCAN: A NEW TOOL TO EVALUATE SMALL FIBER NEUROPATHY IN PATIENTS WITH WILD TYPE CARDIAC AMYLOIDOSIS

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Introduction: The evaluation of the neurological involvement affecting small fibers in patients with Wild-Type Cardiac Amyloidosis (WT-TTR-CA) is very often misdiagnosed by cardiologists. Several studies have evidenced presence of small fiber neuropathy (SFN) in patients with familial TTR- amyloidosis but never in patients with WT-TTR-CA.

Objective: We aimed to evaluate Sudoscan, a simple tool, to diagnose small fiber neuropathy in patients with WT-TTR-CA.

Methods: We enrolled 74 patients with WT-TTR-CA during 2 years (2018-2019) admitted to the French referral center for cardiac amyloidosis. In addition to usual cardiac evaluation, each patient had a neurological evaluation of the feet and hands by Sudoscan, a quick, non-invasive and quantitative method to assess function of the small C fibers innervating sweat glands. The patients also completed a neurological questionnaire of quality of life to obtain their feelings about the presence or absence of neurological symptoms. Presence of SFN was defined according to feet electrochemical skin conductance (FESC): ≥ 70 µS no SFN, < 70 µS SFN.

Results: Among the 74 patients, 50.4% of patients had SFN as defined by Sudoscan results. Demography and cardiac parameters of patients classified in two groups according to presence/absence of SFN are displayed in table. In the 12 patients with diabetes 67% had SFN vs 48% in the patients without diabetes. When looking at cardiac decompensation and/or mortality in the 62 patients without diabetes using Kaplan-Meier analysis to compare patients with or without SFN the log-rank test p value was 0.03.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Whole population (n=74)</th>
<th>FESC≥70 (n=36)</th>
<th>FESC &lt; 70 (n=38)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age years</td>
<td>78.3 (76.5-80.1)</td>
<td>78.4 (76.1-80.7)</td>
<td>78.2 (75.4-81.1)</td>
<td>0.8973</td>
</tr>
<tr>
<td>Males n (%)</td>
<td>68 (91.9)</td>
<td>35 (97.2)</td>
<td>33 (86.8)</td>
<td>0.1020</td>
</tr>
<tr>
<td>NYHA class III-IV vs I-II, n (%)</td>
<td>24 (32.4)</td>
<td>11 (30.6)</td>
<td>13 (34.2)</td>
<td>0.7371</td>
</tr>
<tr>
<td>Heart rate,beats/min</td>
<td>76.5 (73.6-79.3)</td>
<td>77.7 (73.3-82.0)</td>
<td>75.2 (71.4-79.1)</td>
<td>0.3906</td>
</tr>
<tr>
<td>Pacemaker, n (%)</td>
<td>33 (44.6)</td>
<td>15 (41.7)</td>
<td>18 (47.4)</td>
<td>0.6221</td>
</tr>
<tr>
<td>ICD, n (%)</td>
<td>12 (16.2)</td>
<td>3 (8.3)</td>
<td>9 (23.7)</td>
<td>0.0734</td>
</tr>
<tr>
<td>Diabetes, n (%)</td>
<td>12 (16.2)</td>
<td>4 (11.1)</td>
<td>8 (21.1)</td>
<td>0.2462</td>
</tr>
<tr>
<td>NT-proBNP, pg/ml</td>
<td>4163 (3230-5095)</td>
<td>3098 (1957-4239)</td>
<td>5227 (3783-6672)</td>
<td>0.0216</td>
</tr>
<tr>
<td>Troponin T HS, ng/ml</td>
<td>78.3 (53.69-102.9)</td>
<td>57.5 (43.6-71.4)</td>
<td>98.4 (51.6-145.3)</td>
<td>0.0967</td>
</tr>
<tr>
<td>Creatinin, µmol/L</td>
<td>117.4 (107.1-127.7)</td>
<td>104.2 (93.2-115.2)</td>
<td>131.0 (114.2-147.8)</td>
<td>0.0089</td>
</tr>
<tr>
<td>Atrial fibrilation, n (%)</td>
<td>9 (13.2)</td>
<td>4 (11.1)</td>
<td>5 (13.2)</td>
<td>0.9672</td>
</tr>
<tr>
<td>LVEF, n (%)</td>
<td>47.5 (44.6-50.4)</td>
<td>49.2 (44.7-53.7)</td>
<td>45.8 (42.1-49.5)</td>
<td>0.2425</td>
</tr>
<tr>
<td>LVTTD, mm</td>
<td>44.2 (42.6-45.7)</td>
<td>45.5 (42.8-47.7)</td>
<td>43.2 (41.2-45.2)</td>
<td>0.1760</td>
</tr>
<tr>
<td>LVTSAD, mm</td>
<td>32.6 (30.6-34.7)</td>
<td>33.2 (29.4-37.0)</td>
<td>32.2 (30.0-34.4)</td>
<td>0.6591</td>
</tr>
<tr>
<td>IVST, mm</td>
<td>18.3 (17.5-19.1)</td>
<td>17.6 (16.4-18.8)</td>
<td>18.9 (17.9-20.0)</td>
<td>0.0941</td>
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<tr>
<td>Global Strain, n (%)</td>
<td>9.5 (8.6-10.4)</td>
<td>10.0 (8.6-11.4)</td>
<td>9.2 (7.9-10.4)</td>
<td>0.3861</td>
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</table>

Conclusion: Sudoscan a quick, reliable, objective and quantitative method to evaluate small fibers could be easily used by cardiologists to screen their patients with WT-TTR-CA for small fibers neuropathy involvement and adapt their management beyond cardiac damages.
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CLINICAL, LABORATORY AND IMAGING FEATURES IN A COHORT OF PATIENTS WITH AMYLOIDOSIS. STILL A MISDIAGNOSED DISEASE?

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¹Instituto do Coração da Faculdade de Medicina da Universidade de São Paulo.

Background: Cardiac amyloidosis is a progressive cardiomyopathy and the diagnosis is often missed or delayed resulting in an advanced disease by the time treatment is contemplated.

Objective: To disclose clinical factors and complementary methods in attaining the diagnosis of cardiac amyloidosis

Methods: We describe the clinical, electrocardiographic, magnetic resonance and echocardiographic data of 60 patients in whom amyloidosis was diagnosed until July 2019.

Results: Most patients were men, with a mean age of 59 years. The most common subtype was transthyretin amyloidosis (75%) and 22% wild type. The most prevalent genetic mutation was Val50Met (53%). Almost half (46%) of patients have cardiac involvement. The most common initial clinical picture was HFrEF (49%) and polyneuropathy (19%). The median time to diagnosis was 3.4 years.

Conclusion: Amyloidosis is diagnosed when the clinical, ECG, and echocardiogram patterns are “typical”, but most of the cases fail to be diagnosed, especially in elderly people, due to the association with other cardiac diseases, lack of diastolic dysfunction at the echocardiogram and only a slightly thickened ventricular wall. In our service these patients are taking too long to be diagnosed, as seen in the literature, and therefore the first step to improve is to know better our patient’s characteristics.
HOSPITALIZATIONS AND MORTALITY AMONG CARDIAC AMYLOIDOSIS PATIENTS COVERED BY MEDICARE

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2 Pfizer Inc, New York City, NY.
3 Pfizer Inc, Collegeville, PA.
4 Medical University of South Carolina, Charleston SC.

Background: Transthyretin amyloid cardiomyopathy (ATTR-CM) is a rare disease caused by the deposition of amyloid fibrils in the myocardium with typical clinical presentation as heart failure. The epidemiology and burden of ATTR-CM is not well described in the literature.

Objective: Assess hospitalizations and mortality of Medicare beneficiaries with cardiac amyloidosis in contrast to matched patients with heart failure.

Methods: A retrospective cohort study was conducted using Medicare fee-for-service claims available from January 2010 until April 2019. Cohort 1 of cardiac amyloidosis was comprised of (a) confirmed ATTR-CM wild type patients (ICD10 code E85.82) and (b) patients with combined diagnoses of amyloidosis and HF or CM (ICD9 codes 277.30 and 277.39, ICD10 codes E85.0, E85.2, E85.82, E85.89, E85.9). Cohort 2 was comprised of heart failure patients without cardiac amyloidosis. Patients had at least 6 months of continuous Medicare enrollment prior and 12 months of enrollment or enrollment interrupted by death after index date, defined as the first amyloidosis diagnosis. Patients who had organ or stem cell transplants, light chain amyloidosis or dementia (cerebral amyloid angiopathy) were excluded. Patients in each cohort were matched 1:1 based on demographic characteristics. Hospitalizations (all-cause and cardio-vascular-related) and mortality were described for 12-month follow-up.

Results: A total of 26,274 patients with cardiac amyloidosis (cohort 1) and 7,952,403 with non-amyloid heart failure (cohort 2) were identified. Mean age was comparable, at 79.81 years ± 7.67 and 79.78 years ± 8.52 in cohorts 1 and 2, respectively (p<0.0001). Males constituted 58% and 45% of each cohort (p<.0001). Compared to matched heart failure patients (N=26,271), more patients with cardiac amyloidosis had all-cause hospital admissions (72.61% vs 63.42%, p < 0.0001), cardiovascular-related admissions (67.36% vs 58.92%, p<.0001), heart failure readmissions in 30 days (50.38% vs 33.65%, p<.0001) and mortality (34.25% vs 12.29%, p<.0001).

Conclusions: In a large retrospective analysis across more than 9 years, we observed increased burden of all-cause and cardiovascular hospitalizations and mortality among cardiac amyloidosis patients compared to non-amyloid heart failure patients. Additional research is needed to describe the overall burden of disease to patients, caregivers and health care systems. Disease awareness and access to new effective therapies are likely to decrease burden of disease.
MYOCARDIAL PERFUSION MAPPING IN CARDIAC AMYLOIDOSIS, MORE THAN MEETS THE EYE?

L. Chacko 1, T. Kotecha 1, A. Martinez-Naharro 1, J. Brown 1, R. Martone 1, C. Little 1, D. Knight 1, S. Anderson 1, J. Moon 2, P. Hawkins 1, J. Gillmore 1, R. Rakhit 3, T. Lockie 3, N. Patel 3, H. Xue 4, P. Kellman 4, M. Fontana 1

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2 Barts Heart Centre, St Bartholomew’s Hospital, London, UK.
3 Royal Free Hospital, London, UK.
4 National Heart, Lung, and Blood Institute, National Institutes of Health, Bethesda, USA.

Background: Cardiac involvement is the main driver of outcome in systemic amyloidosis but the relationship between amyloid deposits and outcomes is poorly understood.

Objectives: We assess the presence of myocardial ischaemia in patients with cardiac amyloidosis (CA), compare to patients who have undergone invasive coronary angiography (ICA), and assess correlation of perfusion mapping parameters to markers of disease severity and prognosis.

Methods: 86 patients and 20 healthy volunteers (HV) underwent CMR at 1.5T (Siemens) with standard cine imaging, Phase Sensitive Inversion Recovery Late Gadolinium Enhancement, T1, T2, Extracellular Volume (ECV) and adenosine stress with myocardial blood flow (MBF) mapping. Thirty-eight patients also underwent ICA with 3 vessel assessment of Index of Microcirculatory Resistance and Fractional Flow Reserve: 7 had CA, 8 had normal coronary physiology (NCP), 15 had microvascular dysfunction (MVD) and 8 had triple vessel disease (3VD).

Results: CA patients had severe reduction in stress MBF and myocardial perfusion reserve (MPR)–(1.22ml/g/min±0.70 and1.62±0.63 respectively) compared to HV (3.21ml/g/min±0.64,p<0.001 and 4.17±0.78,p<0.001 respectively), NCP (2.66± 0.56, p<0.001 and 2.51±0.43, p=0.036) and MVD (2.10 ±0.31, p<0.001 and 2.29 ± 0.87, p=0.014) with the degree of reduction similar only to patients with 3VD (1.44±0.54, p=1.000 and 1.64± 0.68, p=1.000). CA stress MBF (and MPR) inversely correlated with myocardial amyloid burden (measured as ECV, r=-0.715, p<0.001); transmurality of LGE (no LGE 2.24ml/min/g, subendocardial LGE 1.16ml/min/g and transmural LGE 0.81 ml/min/g, p<0.01); systolic dysfunction (EF, r=0.405, p<0.01) and blood biomarkers (NT-proBNP, r=-0.678, p<0.001 and Troponin T, r=-0.628, p<0.001). There was a correlation between stress MBF and native T1 (r=-0.588, p<0.001) but not with T2 values (p=0.591).

Conclusions: Stress MBF and MPR were found to be early disease markers, elevated in patients with early cardiac infiltration. Myocardial ischaemia is common in CA, highlighting the potential role of myocardial ischaemia as a key mechanism in the pathophysiology of CA.

Keywords: Perfusion, Amyloidosis, CMR
CMR DERIVED EXTRACELLULAR VOLUME CALCULATION IN THE QUANTIFICATION OF EXTRA CARDIAC AMYLOID BURDEN

L. Chacko 1, M. Boldrini 1, R. Martone 2, A. Martinez-Naharro 1, T. Kotecha 1, J. Brown 1, H. Lachmann 1, D. Knight 1, J. Moon 3, Hui Xue 4, P. Kellman 4, J. Gillmore 1, P. Hawkins 1, M. Fontana 1

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3 Barts Heart Centre, St Bartholomew's Hospital, London, UK.
4 National Heart, Lung, and Blood Institute, National Institutes of Health, Bethesda, USA.

Background: Systemic amyloidosis is a disease characterized by amyloid infiltration in different organs and tissues. SAP scintigraphy is the reference standard to assess extra cardiac amyloidosis however lacks the provision of quantitative information. CMR is the reference standard to assess cardiac amyloid infiltration, that can be visualized with LGE and measured with ECV. The utility of ECV in detecting and quantifying amyloid in other organs has not previously been investigated.

Objectives: The objectives are to assess the diagnostic accuracy of ECV mapping using a bolus only approach in the liver and spleen against the current gold standard SAP scintigraphy and to assess the ability of ECV mapping to track the amyloid load as measured by SAP scintigraphy.

Methods: 732 consecutive patients with suspected systemic amyloidosis were prospectively enrolled into the study between 2015 and 2017. All patients underwent scintigraphy with 123I-labeled serum amyloid P component (SAP scan), the reference standard for assessment of amyloid burden in the spleen and liver. Uptake in the spleen and liver was classified into four categories (no amyloid, small, medium, large). All patients underwent a standard cardiac MR protocol including acquisition of three short axis slices with pre and post contrast T1 mapping (by Modified Look-Locker Inversion recovery, MOLLI) and online ECV mapping reconstruction. The liver and spleen ECV was measured on the ECV maps acquired for the left ventricular short axis.

Results: There was good correlation between liver ECV and amyloid burden in the liver as assessed by SAP scintigraphy (Spearman’s coefficient of rank correlation: 0.409, p<0.001) and similarly between spleen ECV and amyloid burden in the spleen (Spearman’s coefficient of rank correlation: 0.647, p<0.001). This was also confirmed when patients with systemic AL amyloidosis were analyzed separately, both in the liver (Spearman’s coefficient of rank correlation: 0.409, p<0.001) and in the spleen (Spearman’s coefficient of rank correlation: 0.647, p<0.001)

Conclusions: CMR has a high sensitivity for the detection and quantification of amyloid in the spleen and liver. This does not require any additional image acquisition and can therefore be easily integrated into routine clinical practice. ECV measurement of the liver and spleen may significantly improve the diagnostic pathway for extra-cardiac amyloid, and importantly, carries potential as a tool to monitor treatment response.

Keywords: Amyloidosis, ECV, SAP
CARPAL TUNNEL SYNDROME RELATED TO CARDIAC AMYLOIDOSIS

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Background: Carpal tunnel syndrome (CTS) is often an early manifestation of systemic amyloidosis. A recent study showed that 10.2% of men older than 50 years and women older than 60 years undergoing surgery for idiopathic CTS had amyloid deposition in the tenosynovial biopsy. Patients often present with the cardiac disease several years after undergoing surgical management for CTS.2,3

Methods: This is a retrospective cohort study using the 2016 National Inpatient Sample (NIS) of adults hospitalized for Cardiac Amyloidosis (CA) as the admitting diagnosis and carpal tunnel syndrome (CTS) as a secondary diagnosis based on ICD-10 codes. Multivariate linear regression adjusting for confounders of age, gender, race, hypothyroidism, rheumatoid arthritis (RA), osteoarthritis (OA), wrist fracture and obesity was performed using STATA 15 for data analysis.

Results: 12,745 patients were admitted with CA in 2016, of which 60 patients had carpal tunnel syndrome (0.47%), 25 were females (41.66%) and 35 males (58.33%). In terms of race, 50% of patients were Caucasian (n=30); 42%, African American (n=25); and 8%, Hispanic (n=5). The mean age was 68.5 years for those with CA and CTS, compared to 73.3 years for CA alone. Multivariate linear regression adjustment for confounders of age, gender, race, hypothyroidism and diabetes mellitus showed a significant relationship between CA and CTS (OR: 4.31; 95%-CI 2.46-7.56; p-value: <0.001). No comorbid RA or OA was found using ICD-10 coding.

Conclusion: This study demonstrates the significant relationship that exists between CA and CTS, when adjusting for other causes of the latter. This reinforces the idea that physicians should have a high index of suspicion of CA in patients with history of CTS. More population based studies are needed to necessitate early screening for CA in patients with CTS given more availability of therapies to change the natural history of CA, including stabilizers and RNA silencers for transthyrein disease and monoclonal antibodies for light chain disease.

WHEN TO LOOK FOR TRANSTHYRETIN AMYLOIDOSIS IN HEART FAILURE: INCREASING CHANCES OF A POSITIVE GAMMAGRAPHIC STUDY


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Introduction: Transthyretin amyloidosis (ATTR) has become increasingly recognized as a major cause for heart failure (HF). Diagnosis requires complex work up such as DPD scintigraphy (DPDs). Taking into account the huge number of patients with heart failure who might suffer from TTRA availability of DPDs is limited, urging to identify factors to increase its diagnostic rentability.

Methods: Transversal study of HF patients between 2013 and 2019 with suspected ATTR and DPDs was performed. Baseline characteristics, biomarkers, EKG findings, transtorathic echocardiogram parameters [left ventricular (LV) ejection fraction, diastolic function, TAPSE, diastolic interventricular septum (IVS), LV mass (LVM), relative LV wall thickness (RWT), indexed LV telediastolic volume and diameter, indexed left atrium diameter (iAPLAD)] and DPDs results were studied. Two groups were created according to DPDs (SP those with positive results for amyloidosis and SN those with negative results). For statistical SPSS v.25 was used.

Results: 37 patients were studied. In our population 54% had positive DPDs for amyloidosis. Among SP patients 50% were classified as grade 2 of Perugini classification meanwhile 50% were grade 3; mean value of heart to contralateral ratio was 2,73±0,8. There were no differences in NYHA classification. Differences among SP and SN patients are reflected in Table 1.

Conclusions: In our population SP was found to have higher biomarkers values and higher IVS, LVM, RWT and iAPLAD with poorer RV function. These factors may increase the pre-test probability of a positive gammagraphic test. Further investigation is needed in order to confirm our result and, hopefully, they may be used to develop a mathematical model that precisely predicts the pre-test probability in this patient profile.

Table 1. Results

<table>
<thead>
<tr>
<th></th>
<th>Total (37)</th>
<th>SP (20)</th>
<th>SN (17)</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>78.0 ± 7.9</td>
<td>80.0 ± 6.5</td>
<td>75.3 ± 8.7</td>
<td>&lt; 0.05</td>
</tr>
<tr>
<td>Gender (male) (%)</td>
<td>30 (8)</td>
<td>19 (95)</td>
<td>11 (64)</td>
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<tr>
<td>Carpal tunnel sdr (%)</td>
<td>3 (8)</td>
<td>3 (15)</td>
<td>0 (0)</td>
<td>0.09</td>
</tr>
<tr>
<td>Systolic blood pressure (mmHg) (x ± sd)</td>
<td>127.0 ± 21.0</td>
<td>118.0 ± 18.0</td>
<td>138.5 ± 19.0</td>
<td>~ 0.01</td>
</tr>
<tr>
<td>ProBNP (pg/mL) (x ± sd)</td>
<td>3596.0 ± 4002.0</td>
<td>4615.0 ± 4538.0</td>
<td>1761.0 ± 1927.0</td>
<td>~0.05</td>
</tr>
<tr>
<td>Troponin T (x ± sd)</td>
<td>132.0 ± 360.0</td>
<td>66.4 ± 35.0</td>
<td>43.0 ± 39.0</td>
<td>0.09</td>
</tr>
<tr>
<td>Pseudoinfarction patter in EKG (%)</td>
<td>26 (70)</td>
<td>17 (85)</td>
<td>9 (52)</td>
<td>&lt; 0.05</td>
</tr>
<tr>
<td>Diastolic interventricular septum (mm) (x ± sd)</td>
<td>14.6 ± 4.5</td>
<td>16.6 ± 5.3</td>
<td>13.3 ± 3.0</td>
<td>~ 0.01</td>
</tr>
<tr>
<td>Indexed left ventricular mass (g/m2) (x ± sd)</td>
<td>142.5 ± 68.0</td>
<td>180.0 ± 80.0</td>
<td>111.0 ± 22.0</td>
<td>~ 0.01</td>
</tr>
<tr>
<td>Relative wall thickness (x ± sd)</td>
<td>0.68 ± 0.42</td>
<td>0.8 ± 0.24</td>
<td>0.56 ± 0.5</td>
<td>~ 0.01</td>
</tr>
<tr>
<td>Left ventricular ejection fraction (%) (x ± sd)</td>
<td>57.4 ± 11.0</td>
<td>54.0 ± 12.0</td>
<td>60.0 ± 10.0</td>
<td>0.12</td>
</tr>
<tr>
<td>Indexed left atrium anteroposterior diameter (mm/m2) (x ± sd)</td>
<td>25.4 ± 6.0</td>
<td>28.7 ± 6.0</td>
<td>22.7 ± 4.1</td>
<td>~0.01</td>
</tr>
<tr>
<td>TAPSE (mm) (x ± sd)</td>
<td>18.6 ± 5</td>
<td>16.1 ± 4</td>
<td>20.6 ± 5</td>
<td>&lt; 0.05</td>
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</tbody>
</table>

Keywords: Cardiac amyloidosis; Transthyretin amyloidosis; Diagnosis
TRANSTHYRETIN CARDIAC AMYLOIDOSIS IN THE AFRICAN AMERICAN POPULATION

A. Gabrovsek 1, J. Donnelly 1, L. Sul 1, K. Bischel 1, B. Hansen 1, D. Clements 1, C. Rigelsky 1, M. Hanna 1

1 Cleveland Clinic

Introduction/Background: Approximately 4% of African Americans (AA) carry the Val122Ile variant for transthyretin (TTR) which can increase the risk of developing hereditary TTR cardiac amyloidosis (ATTRv-CA). Age-related wild-type (ATTRwt) is the most common cause of ATTR-CA overall, seen most often in white males. We present the racial distribution of ATTR-CA, focusing on the AA subpopulation.

Objectives: To outline the prevalence of ATTRwt-CA in AA patients compared to ATTRv-CA due to Val122Ile

Methods: This is a retrospective review of patients with ATTR-CA seen between 2008 and 2018 at the Cleveland Clinic. Diagnosis of ATTR-CA was established via endomyocardial biopsy or with advanced cardiac imaging with appropriate lab testing to rule out light chain amyloidosis. TTR genetic sequencing was obtained when possible.

Results: Of 382 patients with ATTR-CA, 118 (31%) identified racially as AA. In the AA patients with available genetic testing (n = 104), 75% of cases were due to Val122Ile ATTRv and 23% were due to ATTRwt. Variants Asp18Asn and Glu54Gln were also represented with one case each. The clinical, imaging, and laboratory parameters between AA with Val122Ile ATTRv versus ATTRwt were overall similar, although the number of patients in the ATTRwt group was too small to make a valid comparison.

Conclusions: In the AA patients with ATTR-CA, the majority had ATTRv due to the Val122Ile mutation, but nearly one quarter had ATTRwt. Current estimates of the prevalence of ATTR-CA in the AA population focus on those with the Val122Ile variant at risk, but do not account for AA who may develop ATTRwt, potentially underestimating the true prevalence.

Keywords: Transthyretin, African American, wild-type
HIGH RIGHT VENTRICULAR AMYLOID BURDEN IS ASSOCIATED WITH WORSE FUNCTIONAL LIMITATION IN PATIENTS WITH AL AMYLOIDOSIS: A 18F-FLORBETAPIR PET/CT STUDY

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Introduction: In light chain amyloidosis, infiltration of the myocardium by misfolded light chains results in a highly fatal cardiomyopathy (AL-CMP). Right ventricular dilatation1 and dysfunction2 are known poor prognostic markers in AL-CMP, but, the impact of right ventricular AL deposits on heart failure and functional limitation are not known.

Objectives: To quantify right ventricular amyloid burden by 18F-florbetapir PET/CT and determine its relation to severity of heart failure and functional impairment in patients with systemic AL amyloidosis.

Methods: We studied 79 subjects with biopsy-proven systemic AL amyloidosis enrolled in a prospective study: Molecular Imaging of Primary Amyloid Cardiomyopathy (clinical trials.gov NCT: 02641145). All subjects underwent a cardiac 18F-florbetapir PET scan. Right ventricular free wall retention index (RI) of 18F-florbetapir was calculated. Functional limitation of the subjects was assessed by Karnofsky Performance Status Score (score ranging from 0-100, with lower scores indicating worse status), Minnesota Living With Heart Failure Questionnaire (higher scores indicating greater impairment), 6-minute walk test distance, and New York Heart Association heart failure class (NYHA, I to IV, higher indicating worse heart failure status).

Results: The study cohort (mean age 62 ± 7 years) included 42% women; 66% subjects had NYHA Class I/II heart failure. Right ventricular 18F-florbetapir RI was moderately negatively correlated with Karnofsky scale (r = -0.36, p = 0.002). Subjects who were unable to perform 6-minute walk test had significantly higher 18F-florbetapir RI compared to those who performed the 6-minute walk test (0.089 ± 0.035 /min vs. 0.051 ± 0.024/min, p < 0.0001). 18F-florbetapir RI was significantly higher in subjects with moderate to severe heart failure (NYHA class III/IV) compared to subjects with minimal or no heart failure (NYHA class I/II) (0.072 ± 0.039 vs. 0.049 ± 0.018, p < 0.001).

Conclusions: 18F-florbetapir PET is emerging as a highly specific and quantitative tool for assessment of cardiac amyloidosis. Our results suggest that increased right ventricular myocardial 18F-florbetapir uptake is associated with more severe functional impairment as assessed by Karnofsky performance status, 6-minute walk test, and by NYHA heart failure class. Further studies are warranted to better understand the prognostic value of right ventricular 18F-florbetapir PET in AL cardiomyopathy.

RESIDUAL MYOCARDIAL AMYLOID BURDEN AND PERSISTENT FUNCTIONAL LIMITATION IN PATIENTS WITH AL AMYLOIDOSIS AND LONG-TERM HEMATOLOGIC RESPONSE

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Introduction: Light chain amyloid cardiomyopathy (AL-CMP), if severe and untreated, is highly fatal with a median survival of < 6-months. Highly-effective plasma cell therapies have substantially improved survival of systemic AL amyloidosis. However, despite successful hematologic response, AL-CMP patients can sometimes be left with significant residual functional and quality of life limitations.1

Objectives: The primary aims of this study were to determine in patients with cardiac AL amyloidosis and long-term hematologic response:(1) the burden of myocardial amyloidosis by 18F-florbetapir PET/CT; and (2) magnitude of limitation from heart failure, functional impairment, and quality of life.

Methods: 79 patients with biopsy-proven systemic AL amyloidosis were enrolled in a prospective study: Molecular Imaging of Primary Amyloid Cardiomyopathy (clinicaltrials.gov NCT: 02641145). Subjects were enrolled into three groups: (1) AL-CMP with complete response or very good partial response to anti plasma cell therapies for at least one year (CMP-HR, N=19); (2) active AL-CMP (CMP, N = 46) and (3) active AL without CMP (pre-CMP, N = 14). All subjects underwent a cardiac PET with 18F-florbetapir and myocardial retention index (RI) was calculated. Quality of life and functional limitation were assessed by Karnofsky Performance Scale score, Minnesota Heart Failure Questionnaire, 6-minute walk test distance, and New York Heart Association heart failure class (NYHA, I to IV , higher indicating worse heart failure status).

Results: The CMP-HR cohort (mean age 63 ± 6 years, 42% men, 63% NYHA Class II, and 11% NYHA Class III) received primarily (79%) bortezomib based therapy and daratumumab or stem cell transplantation as needed. The mean left ventricular 18F-florbetapir RI was 0.078 ± 0.03/min. Functional limitation was lower in CMP-HR compared to CMP, but equal to the Pre-CMP cohort (Table).

<table>
<thead>
<tr>
<th>Comparison of Functional Parameters Between the Three Study Groups</th>
</tr>
</thead>
<tbody>
<tr>
<td>AL-preCMP</td>
</tr>
<tr>
<td>6-minute walk distance (meters)</td>
</tr>
<tr>
<td>Karnofsky Performance Scale (0-100, lower score worse status)</td>
</tr>
<tr>
<td>Minnesota Living with Heart Failure Score (higher score worse status)</td>
</tr>
</tbody>
</table>

AL-CMP and AL-preCMP: prior to chemotherapy initiation; HR: at least 1 year of hematological response. * p < 0.05; **p < 0.0001 vs. Active-AL-CMP.

Conclusions: Functional status, quality of life, and heart failure was better in the AL- long-term HR cohort compared to active AL CMP cohort, despite persistent AL deposits, supporting light chain toxicity. However, persistent limitation was observed, comparable to active AL Pre-CMP cohort, underscoring the need for novel targeted anti-amyloid therapies for AL-CMP to improve symptoms and quality of life after HR.

HEAR: HEALTHCARE EUROPEAN AMYLOIDOSIS REGISTRY.

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Introduction: Amyloidosis is an under-recognized disease. The prognosis of the disease is related to cardiac involvement. Cardiac amyloidosis is often misdiagnosed or diagnosed late. Real world data on cardiac amyloidosis is missing (characteristics, management, and outcomes with and/or without treatment). Clinical trial populations differ from real life population. Phase IV studies on cardiac amyloidosis treatments need adverse events monitoring and incidence of these events.

Objectives: The objectives HEAR (Healthcare European Amyloidosis Registry) are to collect medical and quality of life data from patients with cardiac amyloidosis including patients referred for suspicion of amyloidosis (controls) and every type of amyloidosis. There is three main type of cardiac amyloidosis: AL amyloidosis, TTR amyloidosis included wild type form (wt-TTR) and hereditary form (h-TTR). HEAR will be the first registry including all patients referred for suspicion of cardiac amyloidosis (wt-TTR, h-TTR, AL, AA, ApoA1, …).

Material & Methods: HEAR is a non-interventional study. Two types of data will be collected. Retrospective data from patients included in centers between 2009 and 2019 (= 2000 patients with their follow-up and outcomes). Prospective data: patients newly included (= 3000 patients). 25 centers in France dedicated to cardiac amyloidosis will participate to the registry. Other international centers will be included between 2020 and 2021.

Discussion & Conclusions: Including all these types of patients in our registry will allow us to collect a large amount of data to better understand the disease, the course of care, develop diagnostic and prognostic tools (comparative groups needed (AL and controls…)) and improve the management of patients with amyloidosis. The creation of the Registry HEAR will also offer the opportunity to set up post AMM project and facilitate inclusions in future therapeutic trials by a better knowledge of the population and the disease and also a base of centers and prescreened patients to achieve our goals faster. Our goal is to include more than 5000 patients over a period of 5 years in the 25 centers initiated in France, aims to expand quickly to other countries.
INITIATION OF CARDIOLOGY CARE IN CARDIAC AMYLOIDOSIS: A SINGLE CENTER EXPERIENCE

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Background: Cardiac amyloidosis is an underrecognized cause of heart failure. With new and evolving treatment options, early diagnosis is critical for patients as depth of cardiac involvement carries worst prognosis. Cardiology is always involved in the care of these patients. However, when and why cardiology initiates care for these patients is not well established.

Objective: To determine when and why patients with cardiac amyloidosis established care with cardiology in relationship to their diagnosis.

Methods: We identified 121 patients (average age 69.1 years, 22% female, 69% Caucasian) with cardiac amyloidosis (CA) seen at our institution (1/2008-11/2019): 46 light chain amyloidosis (AL), 36 wild-type amyloidosis (ATTRwt), and 34 hereditary amyloidosis (ATTRm). Of ATTRm, 74% had the p.Val142Ile mutation. Diagnosis was confirmed by cardiac biopsy with mass spectroscopy or technetium pyrophosphate scan. If transthyretin amyloid was present, genetic testing was conducted to assess for familial ATTR. Charts were reviewed for first clinical contact with cardiology.

Results: For AL CA patients, 58.7% patients established cardiology care less than 3 year prior to diagnosis. The median time to diagnosis was 1.5 months (average 5.9 months). The most common chief complaints were dyspnea (70.4%), edema (22.2%) and heart failure (24.8%). Before AL amyloidosis diagnosis, a minority of patients (15.2%) had >4 years (average 7.8 years, range 4-23 years) of established cardiology care for coronary artery disease (6.5%), atrial arrhythmia (2.1%), or valve disease (8.6%). After AL amyloidosis diagnosis, 26.1% of patients established care with cardiology. For ATTRwt CA patients, 52.3% established care with cardiology less than 3 years prior to diagnosis. The median time to diagnosis was 6 months (average 11.3 months). The most common complaint was arrhythmia (36.8%), dyspnea (31.6%), and heart failure (21.1%). Cardiology care was established for greater than 4 years (average 8.6 years, range 4-20 years) for 47.2% of ATTRwt patients prior to diagnosis. The most common complaints were coronary artery disease (47.1%), arrhythmia (29.4%), and dyspnea (17.6%). All patients established care with cardiology prior to diagnosis. For ATTRm CA patients, 64.7% patients established cardiology care less than 3 year prior to diagnosis. The median time to diagnosis was 10.5 months (average 12.4 months). The most common complaints were dyspnea (54.5%), heart failure (27.2%), and edema (18%). Cardiology care was established for greater than 4 years (average 6.5 years, range 5-14) for 17.6% of ATTRm patients with most common complaint of dyspnea (33.3%). After positive genetic testing, 11.8% ATTRm patients established cardiology care.

Conclusion: Most patients establish care with cardiology for months and even years prior to their cardiac amyloidosis diagnosis. This study reveals an opportunity for earlier diagnosis of cardiac amyloidosis by the cardiology field.
HIGH PRICE AND US INSURANCE PRACTICES DELAY AND LIMIT ACCESS TO TAFAMIDIS FOR PATIENTS WITH TRANSTHYRETIN CARDIAC AMYLOIDOSIS: A SINGLE CENTER EXPERIENCE

C. Campbell 1, N. Bumma 2, S. Parikh 3, J. Prosek 3, J. Bittengle 4, S. Almaani 3, K. Pfund 4, S. LoRusso 5, M. Freimer 3, E. Redder 6, Y. Efebera 2, N. Sharma 2, R. Kahwash 1, A. Vallakati 1

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Background: Transthyretin cardiac amyloidosis is an increasingly recognized cause of heart failure. Tafamidis was approved by the United States Food and Drug Administration in May 2019 for treatment of transthyretin cardiac amyloidosis. Pfizer, the manufacturer, priced Tafamidis at $225,000 per year and this drug became available commercially in July 2019.

Objectives: To assess barriers to patients receiving Tafamidis after receiving a physician prescription for the medication.

Methods: We evaluated medical records for patients seen at our clinic between June 2019 and October 2019. We identified 23 patients who were prescribed Tafamidis. Information was primarily compiled from clinic notes, telephone encounters, and interactions with our Medication Assistance Program. The average patient age was 75.3 years, 8.6% (2) patients were female, and 47.8% (11) patients identified their ethnicity as black. The majority of patients, 56.5% (13), had hereditary ATTR amyloidosis with 85% (11) carrying the p.Val142Ile mutation.

Results: Overall, 82.6% (19) of patients who were prescribed Tafamidis obtained the medication. Most patient underwent a multistep process that took an average of 37.6 days (range 7-65 days) including insurance prior authorization and applications for medical financial assistance. Insurance companies required 73.9% (17) patients to obtain a prior authorization. Prior authorization took an average of 20 days (range 7-35 days). Prescriptions were approved for coverage for 3 patients without prior authorization. In addition, one patient was covered through Veteran’s Affairs, and 2 patients had no prescription drug coverage. Financial medication assistance was requested by 69.6% (16) patients for an average monthly co-pay of $2,032 (range $500-5000) and obtained by 47.8% (11) patients. One patient did not complete the necessary paperwork. Four patients were deemed ineligible for financial assistance due to high income. Of these patients, the average monthly co-pay was $987. Three patients chose not to fill their Tafamidis prescription due to price. Seeking financial assistance significantly increased the average time to obtain the medication to 45.2 days compared to 28.9 days (p=0.02). Only 13.1% (3) patients received Tafamidis within two weeks of initial prescription. After prior authorization, their co-pays were $3.80, $30, and $550. No financial assistance was sought.

Conclusions: The high cost of Tafamidis limits access to this therapy for patients with transthyretin cardiac amyloidosis in the United States. Insurance prior authorization and applications for financial assistance due to high co-pays result in delays in obtaining the medication. Patients and providers should be aware of these issues and advocate for changes to improve access.
AL AMYLOIDOSIS CAUSING POSITIVE TECHNETIUM-99M PYROPHOSPHATE IMAGING

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Introduction/Background: The diagnosis of transthyretin (TTR) amyloid cardiomyopathy (ATTR-CM) with 99mTc-pyrophosphate scan (99mTc-PYP) is feasible if light chain (AL) amyloidosis has been effectively ruled out with appropriate laboratory testing. The need to rule out AL arose since light chain amyloid cardiomyopathy (AL-CM) can occasionally lead to significant myocardial uptake on 99mTc-PYP, defined as ≥ grade 2 on qualitative scoring.

Objectives: To describe the prevalence and degree of myocardial uptake on 99mTc-PYP in patients with AL-CM.

Methods: This is a retrospective review of patients with AL-CA seen at our institution between July 2015 and November 2019. We identified those patients with AL-CM who underwent 99mTc-PYP. At our center, planar imaging and SPECT is done 3 hours after injection of nuclear agent. The grading scheme on planar imaging is well described as grade 0 (no myocardial uptake), grade 1 (uptake less than rib), grade 2 (equal to rib), or grade 3 (greater than rib). A heart to contralateral ratio (H/CL) is determined for each patient. Finally SPECT imaging is used to determine if the uptake is in the myocardium (described either as focal or diffuse) or in the blood pool. A positive scan was defined as an H/CL ratio > 1.3 AND ≥ grade 2 myocardial uptake.

Results: A total of 227 99mTc-PYP were positive during this study period, and 26 patients were subsequently diagnosed with AL-CM. Four of the 26 (15%) had ≥ grade 2 myocardial uptake AND H/CL ratio > 1.3 on planar imaging that was confirmed on SPECT imaging (Table 1 in bold). Patient 1 who had grade 3 uptake had very significant amounts of both AL and ATTR amyloid deposits on endomyocardial biopsy (TTR genetic testing negative). Overall, 2.2% of the positive scans were actually due to AL-CM.

Conclusions: In patients with AL-CM who underwent 99mTc-PYP, 15% had significant myocardial uptake. Thus AL-CM must be ruled out in order to make the diagnosis of ATTR-CM when using 99mTc-PYP for noninvasive diagnosis.

Table 1: AL-CA Patients with myocardial uptake on 99mTc-PYP-BS

<table>
<thead>
<tr>
<th>Patient</th>
<th>Amyloid Type</th>
<th>Grade</th>
<th>H/CL Ratio</th>
<th>SPECT Uptake pattern</th>
<th>kappa (mg/L)</th>
<th>lambda (mg/L)</th>
<th>M protein Serum (+/-)</th>
<th>Urine (+/-)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Lambda &amp; Transthyretin</td>
<td>3</td>
<td>1.8</td>
<td>Diffuse</td>
<td>26</td>
<td>365</td>
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<td>+</td>
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<td>Diffuse</td>
<td>17</td>
<td>1289</td>
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<td>-</td>
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<td>+</td>
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<td>+</td>
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</tr>
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<td>1.06</td>
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<td>41</td>
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<td>-</td>
</tr>
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<td>Persistent Blood Pool</td>
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<td>1279</td>
<td>+</td>
<td>+</td>
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</tbody>
</table>

Keywords: AL, Technecium-99m, Imaging
ORTHOPAEDICS HISTORY PRECEDING DIAGNOSIS OF CARDIAC AMYLOIDOSIS: TIMING AND VARIATION BY SUBTYPE

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Background: Cardiac amyloidosis is a progressive, underrecognized cause of cardiomyopathy characterized by the deposition of amyloid fibrils within the myocardium. Early manifestations of this systemic disease can include bilateral carpal tunnel syndrome and lumbar spinal stenosis. The frequency of other musculoskeletal disease has not been assessed.

Objectives: To determine frequency and timing of musculoskeletal disease in patients diagnosed with cardiac amyloidosis.

Methods: We identified 121 patients (average age 69.1 years, 22% female, 69% Caucasian) with cardiac amyloidosis (CA) seen at our institution 1/2008-11/2019: 46 light chain amyloidosis (AL), 36 wild-type amyloidosis (ATTRwt), and 34 hereditary amyloidosis (ATTRm). Of ATTRm, 74% had the p.Val142Ile mutation. Diagnosis was confirmed by cardiac biopsy with mass spectroscopy or technetium pyrophosphate scan. If transthyretin amyloid was present, genetic testing was conducted to assess for familial ATTR. Charts were reviewed for all documented orthopedic diagnoses and procedures.

Results: The majority of CA patients had orthopedic injuries (74%) and underwent surgical procedures (67%). The most common diagnoses were carpal tunnel syndrome (47.9% with 25.6% bilateral), spinal stenosis (37.2%), and rotator cuff tear (28.1%). The most common procedures were joint replacement (33.1%), carpal tunnel release (31.4%), spinal laminectomy (14.9%), and rotator cuff repair (14.9%). Achilles or biceps tendon rupture occurred in 7.4% of cardiac amyloidosis patients. For AL CA patients, spinal stenosis occurred most frequently (37.2%; 10.9% laminectomy) followed by rotator cuff injury (23.9%; 4.3% repair), carpal tunnel syndrome (21.7%; 13.0% release), and joint replacement (19.7%). Procedures preceded AL diagnosis by 14.3 years (laminectomy), 9.89 years (joint replacement), 7.0 years (carpal tunnel release), and 1 year (rotator cuff repair). For ATTRwt CA patients, joint replacement was most common (63.8%), followed by carpal tunnel syndrome (52.8%; 44.4% release), spinal stenosis (44.4%; 33.3% laminectomy), and rotator cuff repair (30.6%; 27.8% repair). Procedures preceded ATTRwt diagnosis by an average of 14 years (carpal tunnel release), 10.3 yrs (rotator cuff repair), 8.9 year (joint replacement), and 6.3 years (laminectomy). For ATTRm CA patients, carpal tunnel syndrome was most common (85.3%; 47.1% repair) followed by spinal stenosis (41.2%; 2.9% laminectomy), rotator cuff injury (35.3%; 17.6% repair), and joint replacement (17.6%). Procedures preceded ATTRm diagnosis by an average of 7.4 years (rotator cuff repair), 7.0 years (joint replacement), 6.7 years (carpal tunnel release), and 4 years (laminectomy).

Conclusions: Patients with cardiac amyloidosis frequently have orthopedic injuries and procedures in the 5-15 years prior to diagnosis. Understanding this association and evaluating for amyloidosis with biopsy during these procedures may prompt earlier diagnosis of cardiac amyloidosis.
LONGER-TERM SURVIVORS OF LIGHT CHAIN CARDIAC AMYLOIDOSIS

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Introduction/Background: Heart failure (HF) due to light chain cardiac amyloidosis (AL-CA) is considered to have a bleak prognosis with a median untreated survival of 6 months. A prognostic staging system has been developed by the Mayo Clinic utilizing NT-proBNP, troponin T, and the difference in serum free light chains. Median survival of the two most advanced Mayo stages are 14 months for stage III, and 5.8 months for Stage IV. However, advances in anti-plasma cell therapies have improved outcomes.

Objectives: To demonstrate that some patients with AL-CA can experience longer-term survival.

Methods: We identified pts with AL-CA seen between July 1997 and May 2016 at the Cleveland Clinic. Diagnosis of AL-CA was established with endomyocardial biopsy or extra-cardiac biopsy plus diagnostic cardiac imaging and/or biomarkers. We arbitrarily set a survival of ≥ 3 years (yrs) as “longer-term” and determined the proportion of patients who achieved this. We calculated Mayo Stage in those pts who had the necessary data available and determined survival over 5 years according to each stage.

Results: Of 316 pts with AL-CA, 144 had data available to calculate Mayo Stage. The number and survival for each stage over 5 yrs is shown in Table 1. A total of 81 pts (26%) survived for ≥ 3 yrs. Of these 81 pts, the average age at diagnosis was 61 ±11 years, 57% were male, and 69% had multi-organ involvement. Of the 41 out of 81 patients who had data available for Mayo staging, 4 were Stage I, 9 were Stage II, 17 were Stage III, and 11 were Stage IV. 16 patients (5% of the overall cohort of 316) experienced exceptionally remarkable long-term survival of ≥ 8 years.

Conclusions: The survival in patients with AL-CM is limited, but even in those who present with more advanced disease there is a cohort that can achieve survival ≥ 3 yrs. Patients with AL-CA should be promptly treated with the help of an experienced multidisciplinary team.

Table 1

<table>
<thead>
<tr>
<th>Time (yrs)</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.78 (9)</td>
<td>0.92 (13)</td>
<td>0.48 (56)</td>
<td>0.29 (66)</td>
</tr>
<tr>
<td>2</td>
<td>0.78 (7)</td>
<td>0.85 (12)</td>
<td>0.37 (26)</td>
<td>0.21 (18)</td>
</tr>
<tr>
<td>3</td>
<td>0.54 (7)</td>
<td>0.69 (11)</td>
<td>0.35 (19)</td>
<td>0.19 (12)</td>
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<tr>
<td>4</td>
<td>0.54 (4)</td>
<td>0.61 (9)</td>
<td>0.32 (17)</td>
<td>0.19 (11)</td>
</tr>
<tr>
<td>5</td>
<td>0.54 (3)</td>
<td>0.34 (7)</td>
<td>0.13 (12)</td>
<td>0.11 (7)</td>
</tr>
</tbody>
</table>

Values represent survival (patients) at start of interval.

Keywords: Cardiac Amyloidosis, survival
NATURAL HISTORY OF TRANSTHYRETIN CARDIAC AMYLOIDOSIS WITHOUT HEART FAILURE


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Introduction: Cardiac transthyretin (ATTR) amyloidosis is an increasingly recognized, progressive, and fatal cardiomyopathy in which the natural history remains unclear. Patients without heart failure (HF) have not been included in clinical trials and it is mostly unknown how would them evolve during follow-up and if it is worth considering early initiation of specific treatments.

Objectives: We sought to describe natural history and prognosis of patients with ATTR cardiomyopathy without HF.

Methods: Clinical characteristics and events from cardiac ATTR amyloidosis patients without HF (NYHA I and Nt-proBNP <600pg/ml) were retrospectively collected at 4 international amyloidosis centers: Columbia University Hospital (New York, US), University of Bologna (Italy), University of Pavia (Italy) and Hospital Puerta de Hierro (Madrid, Spain).

Results: 100 patients with cardiac ATTR amyloidosis without HF (mean age 63.4±14.4 years at diagnosis, 76% males and 63% hereditary TTR) were included in this analysis. Mean ejection fraction at baseline was 60±12% and interventricular septum was 15.1±3.6mm. During a median follow-up of 4.2 years (IQR: 1.8-7.0), 32% patients progressed to NYHA class ≥II and 5% died due to HF or required heart transplantation (2 and 3 patients, respectively). 2 patients died due to other cardiovascular conditions. 14 patients, a pacemaker was implanted due to rhythm disturbances, 16 developed atrial fibrillation, and 1 had a stroke. At last follow-up, NYHA class was still I in 68%, II in 19%, III in 9% and IV in 4%. Incidence of worsening in NYHA class was 6.5 patients in 100 patients-year of follow-up. Median time to develop HF was 2.8 years (IQR 1.9-3.7). Follow-up was longer in patients who developed HF (6.5 vs 4.1 years; p=0.007). Presence of atrial fibrillation at baseline was more frequent in patients who developed symptoms than in those who did not (25% vs 6%; p=0.03). There were not statistical differences in other baseline characteristics. At last follow-up, patients with NYHA class ≥ II had lower left ventricular ejection fraction (52.5% vs 61.2%, P<0.001).

Conclusions: Almost one third of cardiac ATTR patients without HF develop HF during a median follow-up of 4 years. The rate of other events, namely pacemaker implantation is also remarkable. Further studies are necessary in order to clarify what are the most appropriate therapies for ATTR patients at initial stages of the disease.
THE ROLE OF MULTIPARAMETRIC ECHOCARDIOGRAPHY IN REFINING THE DIAGNOSIS OF CARDIAC AMYLOIDOSIS

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Background: Cardiac amyloidosis (CA) is a serious yet increasingly treatable cause of heart failure once identified. Although the diagnosis of CA is challenging and frequently unclear at echocardiography, this modality remains the most accessible and widely used first line form of imaging.

Objectives: We aimed to study the accuracy of a broad range of echocardiographic variables to develop a multiparametric approach for the diagnosis of CA in patients with proven light chain (AL) amyloidosis and those with increased heart wall thickness (IWT) in whom amyloid was suspected. We also aimed to further characterise structural and functional changes associated with amyloid infiltration.

Methods: We studied 1187 patients referred to three centres with a specialist interest in amyloidosis and analysed morphological, functional and strain-derived echo parameters with the aim of developing a score-based diagnostic algorithm. Cardiac amyloid burden was quantified using cardiac magnetic resonance derived extracellular volume measurements.

Results: 332 AL amyloidosis patients were confirmed to have CA and 339 patients had transthyretin (ATTR) CA. Concentric remodelling and strain derived parameters had the best diagnostic performance. A multivariable logistic regression model incorporating relative wall thickness, E wave/e' wave, longitudinal strain and tricuspid annular plane systolic excursion had the greatest diagnostic performance in AL amyloidosis (area under the curve AUC 0.90 [95%CI [0.87-0.92]], whilst the addition of septal apical to base ratio yielded the best diagnostic accuracy in the IWT group (AUC 0.87 [0.85-0.90]).

Conclusions: Specific functional and structural parameters characterize different burdens of CA deposition with different diagnostic performances, hence enabling a score-based algorithm that provides a sensitive and specific tool for the diagnosis or exclusion of CA.

Keywords: Amyloidosis, Echocardiography, Multiparametric
NATURAL PROGRESSION OF TRANSTHYRETIN CARDIAC AMYLOIDOSIS

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Introduction: Data about the rate of natural progression of transthyretin cardiac amyloidosis (ATTR-CM) are scarce and criteria to establish ATTR-CM progression are not available. Appropriate definition of disease progression would be crucial in the future to determine appropriate or absence of response to new specific therapies.

Objective: We sought to describe clinical, analytical and echocardiographic parameters associated with progression of ATTR-CM.

Methods: 97 consecutive patients with ATTR-CM who had not received any anti-ATTR therapy and who were followed at a referral cardiac amyloidosis center were analyzed. Baseline and 24-months follow-up clinical, analytical, electrocardiographic (EKG) and echocardiographic parameters were evaluated.

Results: A total of 97 patients with ATTR-CM (mean age: 81±9.6 years-old, 72.2% male, 14.7% with hereditary ATTR, 86.6% with heart failure, 53.1% as first manifestation) were included in this analysis. Mean follow-up was 23.4±17.7 months, 50 patients (52.1%) died during follow-up. Among the 37 patients (38%) with 2 years follow-up data, NYHA class had deteriorated in 15 (41%) patients, mean NT-proBNP raised from 4727±6575pg/L to 6518±8808pg/L (p=0.2); mean troponin I levels increased from 0.11±0.1µg/L to 0.12±0.1µg/L (p=0.26) and mean eGFR progressed 58±16 mL/min/1.73m² to 54.9±19 mL/min/1.73m² (p=0.19). Regarding EKG parameters: mean PR and QRS intervals widened from 178.8 ±47.1 to 181.7±59.6ms (p=0.027) and 110±32 to 125±38 ms (p=0.004), respectively. In terms of echo parameters, mean left ventricular (LV) thickness increased from 16.7±2.5 to 17.7±1.8 mm (p=0.005) and left atrium volume changed from 47.7±10.4 to 56.9±20.1 ml/m² (p=0.013). LV S wave by Tissue Doppler imaging (TDI) decreased from 8.1±2.4 to 6±2 cm/s (p=0.014), while no significant changes were observed in LV ejection fraction (58.2±12.9% vs 53.9±11.9% (= 0.1)), right ventricular longitudinal shortening parameters (TAPSE 16 ± 4.6 vs 14.8 ± 3.6 mm (p=0.3) and TDI S wave 8.5 vs 8.4 cm/s (p=0.9).

Conclusions: PR and QRS intervals, LV wall thickness, LA volume and LV longitudinal shortening by TDI S are parameters that show progression in untreated ATTR-CM patients at 24 months.
CLINICAL AND SCIENTIFIC CONCEPT OF A COMPREHENSIVE INTERDISCIPLINARY AMYLOIDOSIS CENTER

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Background: Amyloidosis represents a rare, but life-threatening condition. Diagnosis is frequently delayed and extremely complex requiring a multidisciplinary team, board decisions, and SOPs. Treatment options are on the horizon, but the structured follow-up of these patients is not appropriately addressed by currently available structures. Therefore, a center-based approach is needed.

Objectives: Overarching goal was the deep integration of routine care into clinical research and vice versa. We thus set up an Interdisciplinary Amyloidosis Center facilitating a) accelerated access for patients with suspected amyloidosis to specific diagnostic work-up and treatment, and b) interdisciplinary collaborative research.

Methods: Based on field research and input by selected major players, we designed a structure that both optimally suits the regional demands and fits our health care system.

Results: Outpatient activities started in Nov 2017. The Center was officially founded in Jan 2018, with by-laws acknowledged by the University Hospital. All patients undergo a standardized, harmonized, interdisciplinary detailed work-up including common and evolving diagnostic tools. Participating disciplines are hematology, cardiology, nephrology, gastroenterology, hepatology, rheumatology, neurology, dermatology, ophthalmology, and pathology. In case of proven amyloidosis, treatment and standardized follow-up will be initiated, monitored by the Board of the Center. Participation in treatment studies is systematically offered to eligible patients. Complementary scientific concepts focus on non-invasive characterization of organ involvement, assessment of early organ response, disease burden, and psychological comorbidities. We include all patients with suspected and proven amyloidosis in a prospective cohort study allowing data collection and biomaterial sampling. Since Nov 2017, 175 patients have been assessed. N=150 patients were recruited into the cohort study since Aug 2018. The mean age is 67±12 years and 61% are male. Diagnoses include: suspected amyloidosis n=35; systemic AL amyloidosis n=33; localized AL amyloidosis n=10; ATTR amyloidosis n=34 (ATTRv 9%); AA amyloidosis n=8; ApoC2 amyloidosis n=1; carrier n=1; excluded amyloidosis n=9, ongoing or incomplete diagnostics n=14. Pattern of organ involvement showed cardiac involvement in n=72 (65%), renal manifestation in n=27 (25%), gastrointestinal manifestation in n=13 (12%) and neurologic manifestation in n=34 patients (31%).

Conclusion: Well-defined and harmonized pathways for the work-up and follow-up of patients with suspected and proven amyloidosis resulted in immediate supra-regional acceptance and visibility. Building on the above described structure, we expect the Center to become an attractive crystallization point for national and international joint research with the scientific community, patient groups, and the industry alike.

Keywords: amyloidosis center, diagnostic work-up, cohort study
International Symposium on Amyloidosis

PT125

POTENTIAL INTERACTION OF TTR STABILIZERS WITH ATORVASTATIN; A CASE SERIES

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Introduction: The TTR stabilizer, tafamidis, showed an excellent safety profile in the ATTR-ACT trial, with adverse effects no greater than placebo. AG10 has also shown no safety signals of concern in Phase-1 and 2 studies. Statins can rarely produce rhabdomyolysis; this is a potentially dose-related side-effect and drugs interacting with statins need to be used with caution. We have noted 2 cases of rhabdomyolysis associated with TTR stabilization therapy when co-administered with high dose atorvastatin use, 1 occurring with tafamidis and 1 in a trial of AG10.

Cases: A 74-year-old woman with ATTR-CMP due to isoleucine 122 (Ile 122) mutation in a trial of AG10 complained of malaise, weakness and difficulty walking for >1 week. Medications included AG10, atorvastatin 80 mg, amiodarone and apixaban. Lab tests showed markedly elevated serum transaminases, initially attributed to hepatic dysfunction. However, she described diffuse muscle soreness, and creatinine phosphokinase (CPK) was 12,303 U/L, consistent with rhabdomyolysis. AG10, atorvastatin and amiodarone were held. CPK down trended. Atorvastatin was discontinued and AG10 was restarted after two weeks and CPK remained normal.

An 85-year-old woman with Ile122 ATTR-CMP, atrial flutter, chronic kidney disease, coronary disease and type II diabetes complained of weakness and with difficulty walking one week after starting tafamidis 61 mg daily. Medications included apixaban, atorvastatin 80 mg, allopurinol, amiodarone. Serum transaminases were 5-fold higher than her baseline, initially attributed to a congested liver. She was discharged to a rehab facility for physical therapy, where transaminases continued to rise. CPK was 18, 569 U/L, consistent with rhabdomyolysis. Tafamidis and atorvastatin were held and her CPK down trended.

Discussion: Both patients developed rhabdomyolysis while on stabilizer therapy and high-dose statin. The package insert for tafamidis notes that it inhibits breast cancer resistant protein (BCRP) and may increase exposure of substrates of this transporter, including rosuvastatin.1). These cases suggest that this may also be the case for atorvastatin and that AG10 may have similar inhibition of BCRP. This has important implications, especially for patients that may have been excluded from clinical studies due to complex comorbidities. An unexpected elevation of serum transaminase in a patient on concomitant TTR stabilizer and statin should prompt an exclusion of a muscle source due to statin-induces rhabdomyolysis.
PT126

IMPACT OF TRANSCATHETER AORTIC VALVE IMPLANTATION IN CARDIAC AMYLOIDOSIS PATIENT- A CASE REPORT

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Introduction: Amyloidosis is characterized by tissue deposition of insoluble amyloid fibrils leading to progressive organ failure. Heart is the commonest organ involved and the main cause of morbidity and mortality. We report here the marked improvement after transcatheter aortic valve implantation (TAVI) in a patient with cardiac AL amyloidosis.

Case description: A 66-year-old patient presented with heart failure, fatigue and bone pain. He was found to have thick walled heart with diastolic dysfunction, anaemia and stage II chronic kidney disease. There was lambda light chain excess (437.7mg/L), bone marrow showed 30% lambda restricted plasma cells and trephine biopsy confirmed amyloid deposits of AL type by immunohistochemistry. 99mTc-DPD scan showed no cardiac uptake ruling out ATTR amyloidosis and cardiac MRI was characteristic of amyloid deposition. NT-proBNP was 3041ng/L and cTroponin T was 40ng/L. An FDG-PET CT showed lytic bone disease. Patient was diagnosed as Multiple Myeloma with systemic AL amyloidosis and stage IIIa cardiac involvement. He remained symptomatic with NYHA grade 3-4 dyspnoea, oedema and persistent supine hypotension <90mmHg on midodrine but no postural hypotension. He started treatment with Cyclophosphamide/Bortezomib/Dexamethasone (CyBorD) plus Daratumumab. which was complicated by recurrent fluid overload, pulmonary oedema and difficult to manage hypotension. A repeat echocardiogram showed features of amyloid cardiomyopathy and severe aortic stenosis with elevated blood pressure in pulmonary artery was of 56mmHg. He completed a further cycle of CyBorD-Dara and achieved light chain complete haematologic response (dFLC 29mg/L). He remained an inpatient for 90 days as he was unable to be weaned off a furosemide infusion. He underwent an uncomplicated TAVI in July 2019. After a week, his systolic BP increased to >100 mmHg, the furosemide infusion and midodrine were discontinued and he was discharged on oral diuretics. He completed 5 cycles of CyBorD-Dara and within 3 months, the NT-proBNP was 904ng/L. His symptoms were significantly improved and he did not require any regular diuretics. In October 2019, his echocardiogram showed: TAVI valve in situ and well seated. Peak velocity of 2.58m/s and mean gradient of 13mmHg. Trace of paravalvular regurgitation. Ejection fraction of 69%. Long axis function was impaired with mild diastolic dysfunction.

Conclusion: This case illustrated the impact of severe aortic stenosis on cardiac symptoms in AL amyloidosis. This can give a false impression of very advanced cardiac AL when a substantial contribution may be due to the valvular disease. TAVI should be considered in these patients to allow for adequate treatment delivery and potential to improve outcomes. More systematic studies are needed to confirm the safety and impact of this procedure in cardiac AL with severe aortic stenosis.
CARDIAC AMYLOID ASSESSMENT WITH (TECHNETIUM-3-3DIPHOSPHONO-1-2-PROPANODICARBOXYLIC ACID) TC-99M DPD SPECT/CT SEGMENTATION IMPROVES DETECTION OF LOW GRADE UPTAKE AND EARLY DISEASE

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Introduction: Low grade uptake in the myocardium may indicate early amyloid infiltration e.g. transthyretin related (ATTR), which Perugini analysis routinely underestimates. Single Photon Emission Computer Tomography / Computed Tomography (SPECT/CT) is a superior method of imaging in nuclear medicine which is underutilized in diagnosis of cardiac amyloid. The CT in used for organ localization and for correction of the images for scatter and attenuation so the resulting SPECT is a more correct measurement of uptake within the patient.

Objectives: Assessment of the utility of myocardial uptake compared a reference region (myocardial uptake compared to blood pool expressed as a ratio called Heart matrix) and to evaluate SPECT/CT segmentation to quantify organ uptake for assessment of cardiac amyloidosis and the application in diagnosis of ATTR disease

Methods: A retrospective analysis of 90 patients (64M, 26F Avg 66.5 Yrs.) was performed on the 3 hour SPECT/CT of patients undergoing assessment for suspected cardiac amyloidosis. Regions of interest for bone, lung and soft tissues are generated from the CT scan using tissue segmentation methodology. Additional regions are identified on the liver and the heart chambers using scan features to identify the valve planes. Region of interest on the aortic arch is used as a blood pool reference for uptake calculations. Uptake 10% greater than bloodpool identified abnormal areas of DPD in the heart. Heart matrix was calculated by multiplying each chamber/reference region value. Standard Perugini analysis was performed from the standard planar imaging. Relevant measures of cardiac dysfunction and amyloid typing were evaluated where available.

Results: In 40 of 90 patients the amyloid gene mutation was present (14 Wild Type TTR, 23 ATTR, 3 Non ATTR). Planar Perugini analysis was abnormal in 26/90 cases. Heart matrix identified abnormal uptake in an additional 11 cases (37/90) at levels which were undetectable on the planar scan. Wild Type TTR patients demonstrated increased soft tissue deposition and decreased bone uptake with increasing myocardial uptake. Elevated cardiac dysfunction levels (NTProBNP) was detected in 42/90 patients. Abnormal NTProBNP and Heart Matrix were detected in 26 of 42 cases. Wild Type TTR demonstrated a reduction of eGFR and increased troponin levels with NTProBNP level greater than 10 times normal.

Conclusions: Heart matrix calculation identified abnormal uptake in the myocardium at lower levels than planar analysis. Earlier detection with Heart matrix can identify patients who may benefit from treatment at a lower disease level burden than with Perugini analysis and also allows for monitoring of changes with treatment.
PT128

TOTAL ELECTRICAL ACTIVITY ACROSS AN ELECTROCARDIOGRAM AND LEFT VENTRICULAR WALL THICKNESS FROM AN ECHOCARDIOGRAM ACCURATELY PREDICT TECHNETIUM-99M PYROPHOSPHATE (PYP) SCANNING RESULTS

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Introduction/Background: A hallmark of transthyretin cardiac amyloidosis (ATT-CA) an infiltrative/restrictive cause of heart failure is a discordance between the wall thickness caused by deposits of protein in the myocardium and the voltage on the electrocardiogram (EKG). Prior studies have shown an ability to discriminate between hypertensive heart disease, hypertrophic cardiomyopathy, and CA by evaluating QRS amplitude compared to left ventricular (LV) wall thickness.

Objectives: The aim of the study was to determine if automated measurement of EKG amplitudes combined with left ventricular wall thickness from echocardiography (echo) could predict which patients would have a Grade 0 or 1 PYP scan (considered negative) versus Grade 2 or 3 (considered positive).

Methods: We analyzed the PYP scan results of 756 serial patients evaluated at a single medical center from 2010 to October 3, 2019 which included 591 patients who also had at least one EKG and echo in our system prior to their PYP scan. Each EKG leads’ peak P, QRS, and T-wave amplitudes were generated and summed. This data was then divided by LV dimensions. These values were then indexed to gender, age, and height. Chi-square, logistic regression, and receiver operating characteristic (ROC) analyses were conducted to determine predictive capacity.

Results: Summing peak amplitudes across a 12-lead EKG (e.g. total P, QRS, and T voltage) and dividing by LV posterior wall thickness plus age generated the Screening Positive for Amyloid (SPA) score. There was a significant difference between median SPA scores for both women (86.7 vs 50.5, p<0.001) and men (71.5 vs 52.3, p<0.001) in predicting negative versus positive PYP scans, respectively. ROC curves for SPA were found to have a c-statistic of 0.82 (CI 0.79-0.85, p<0.001) in women and 0.8 (CI 0.78-0.82, p<0.001) in men.

Conclusions: Summing peak amplitudes across a 12-lead EKG and dividing by LV posterior wall thickness plus age is predictive of PYP scan results and may serve as a potential screening mechanism.

Keywords: Screening, Risk Score
Category: Cardiac amyloidosis
DIGOXIN USE IN CARDIAC AMYLOIDOSIS

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Introduction/Background: There are limited options for rate control of atrial fribillation (a fib) and for low-output heart failure (HF) in cardiac amyloidosis (CA). Digoxin (dig) could be of potential use in these situations as it does not cause hypotension and in fact can increase cardiac output, however its use is discouraged due to a reported increased risk of sensitivity and toxicity in CA.

Objectives: To describe our institution’s experience with dig use in patients (pts) with transthyretin (ATTR) and light chain (AL) CA and to determine the event rate of suspected dig-related arrhythmias and toxicity.

Methods: Pts seen at our institution with a diagnosis of CA between November 1995 and October 2018 were screened for a history of ≥7 days of continuous dig use. Medical records were used to gather demographic information and to identify suspected dig-related arrhythmias and toxicity events.

Results: 69 pts with CA (42 ATTR, 27 AL) used dig for a median duration of 6 months (IQR, 1-16). Indication for use was rate control of a fib in 64% and symptomatic HF in 36%. Dig-related arrhythmias and toxicity events were suspected in 8 pts which was 12% of the total cohort (see Table 1). There were 2 ventricular tachycardia (V tach), 3 bradycardia (brady), 2 junctional (1 accelerated and 1 tach), and one dig toxicity event with mental status changes. Eleven pts died while on dig, however none of these deaths were attributed to dig use; in most cases the mode of death was due to progressive HF in the setting of CA.

Conclusions: In selected cases of ATTR- and AL-CA, dig can be a therapeutic option for rate control of a fib or for symptoms of low output HF. Careful pt selection and close monitoring are recommended. The risk/benefit ratio of using dig should be considered on a case by-case basis.

Keywords (3): Digoxin, Cardiac Amyloidosis

Table 1. Suspected Dig-related Arrhythmia and Toxicity Events

<table>
<thead>
<tr>
<th>Patient #</th>
<th>Gender, Age (years), Amyloid Type</th>
<th>Event Description</th>
<th>Notes</th>
<th>Outcome, Months from Event</th>
</tr>
</thead>
<tbody>
<tr>
<td>1*</td>
<td>M, 73, ATTRwt</td>
<td>V tach</td>
<td>ICD interrogation revealed 5 sudden-onset V tach detections terminated with ATP and one shock.</td>
<td>Death, 22</td>
</tr>
<tr>
<td>2*</td>
<td>M, 68, ATTR V122I</td>
<td>V tach</td>
<td>V tach storm, started on palliative dobutamine and IV diuretics, discharged to hospice.</td>
<td>Death, 0.5</td>
</tr>
<tr>
<td>3</td>
<td>F, 66, AL-k</td>
<td>Dig toxicity</td>
<td>Confusions and hallucinations (dig level 3.8ng/mL); stopped dig and received Digibind, HR improved over the next 48h.</td>
<td>Death, 0.3</td>
</tr>
<tr>
<td>4</td>
<td>F, 81, ATTR V122I</td>
<td>Junctional tach</td>
<td>Noted during admission for ADHF; dig discontinued upon discharge to hospice.</td>
<td>Death, 4</td>
</tr>
<tr>
<td>5</td>
<td>F, 76, AL-κ</td>
<td>Accelerated junction rhythm</td>
<td>Stopped dig 9 days later upon bortezomib initiation</td>
<td>Death, 0.6</td>
</tr>
<tr>
<td>6</td>
<td>F, 58, AL-κ</td>
<td>Junctional brady</td>
<td>Junctional bradycardia with dig level of 1.8 during admission for ADHF, digoxin stopped.</td>
<td>Death, 11</td>
</tr>
<tr>
<td>7</td>
<td>M, 83, ATTR V122I</td>
<td>Symptomatic brady</td>
<td>Symptomatic bradycardia with occasional dizziness at rest</td>
<td>Alive, 54</td>
</tr>
<tr>
<td>8</td>
<td>M, 63, ATTRwt</td>
<td>Brady</td>
<td>Brady and altered mental status in setting of enterococcal sepsis</td>
<td>Death, 0.3</td>
</tr>
</tbody>
</table>

*Pt died while on dig
MITRA-CLIP IN TRANSTHYRETIN AMYLOIDOSIS CARDIOMYOPATHY; A CASE SERIES

S. Cuddy 1, R. Falk 1

1 Brigham & Women’s Hospital.

Introduction: Non-invasive valvular interventions have dramatically changed the management of patients with multiple complex comorbidities. Patients with cardiac amyloidosis (CA) are often considered high-risk for surgical intervention. Trans-catheter aortic valve replacement (TAVR) is tolerated in this population, however, little is known about the safety and efficacy of per-cutaneous mitral valve interventions, such as the MitraClip procedure. We present two patients with CA and mitral regurgitation (MR) who have undergone this procedure.

Cases: A 75-year-old man with wild-type transthyretin cardiac amyloidosis (wtATTR), atrial fibrillation with prior ablations, a Watchman device in the left atrial appendage, resection of a malignant lung nodule presented with recurrent right pleural effusions. On exam, he had a loud MR murmur, which had been present for many years. Transthoracic echocardiogram (TTE) showed mild to moderate MR, which was largely unchanged compared with studies from several years prior. Despite the TTE findings, it was hypothesized that MR may be playing a role in his recurrent effusions. A transesophageal echocardiogram (TEE) revealed a partial flail of the posterior leaflet of the mitral valve resulting in moderate to severe eccentric anteriorly directed MR, amenable to the placement of 2 MitraClips. This procedure led to marked improvement in his dyspnea and a decrease in the frequency of thoracenteses for recurrent pleural effusions.

A 71-year-old man with wtATTR and a permanent pacemaker was seen in follow-up with marked dyspnea on exertion and fatigue, despite being euvoletic on exam; jugular venous pressure was not elevated but there was a Kussmaul’s sign. First and second heart sounds normal and no MR murmur was heard, even in the left lateral position. TTE revealed moderate MR. His symptoms appeared disproportionate to the severity of his amyloid heart disease by exam as he had no evidence of right-sided congestion. Given his symptoms it was suspected that the degree of MR was underestimated by TTE and likely hemodynamically significant. TEE revealed mild diffuse thickening of the mitral valve leaflets with severe, functional MR that was directed centrally. The valve anatomy was suitable for MitraClip, and he underwent successful placement of 2 MitraClips.

Discussion: Both patients had wtATTR and mitral regurgitation that by TTE did not appear to warrant intervention. On further evaluation, both had severe MR. It is likely that the stiffness of the left atrium, due to amyloid infiltration, contributes to the hemodynamic significance. It is important to consider other cardiac pathologies that co-exist in wtATTR, which can contribute to patients’ symptoms and are amenable to intervention. These cases demonstrate that MR can be under appreciated on TTE in patients with CA and that MitraClip is feasible with careful patient selection.
CLINICAL AND HISTOLOGICAL EVIDENCE OF CONCURRENT TRANSTHYRETIN (ATTR) AND LIGHT CHAIN (AL) CARDIAC AMYLOIDOSIS IN TWO PATIENTS

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1 The Scripps Research Institute.

Introduction/Background: Transthyretin (ATTR) and light chain (AL) amyloidosis account for >95% of cases of cardiac amyloidosis (CA). Rarely, both types can occur simultaneously in the same patient causing CA. In cases where there is both significant myocardial uptake on ⁹⁹ᵐTc technetium pyrophosphate bone scintigraphy (⁹⁹ᵐTc-PYP) AND the presence of monoclonal protein on laboratory testing, endomyocardial biopsy is crucial to determine the accurate type of CA.

Objectives: To describe the rare occurrence of both ATTR and AL simultaneously causing CA, and to highlight the importance of a thorough diagnostic workup for CA with endomyocardial biopsy when there is any conflicting data.

Methods: Two cases of endomyocardial biopsy-proven concurrent ATTR-CA and AL-CA were identified retrospectively from our cardiac amyloid database and were reviewed via medical records.

Results: Case 1 was an 89-year-old female who was admitted with renal failure in the setting of heart failure. Case 2 was an 85-year-old male who was seen in the outpatient clinic for dyspnea on exertion. The clinical and imaging parameters in each case were highly suggestive of CA. Both patients had very significant grade 3 myocardial uptake on ⁹⁹ᵐTc-PYP and were initially suspected to have ATTR-CA. However, both also had very high serum free lambda levels and a lambda M protein on serum and urine immunofixation. This prompted endomyocardial biopsy in both cases confirming CA. Immunohistochemistry revealed significant amounts of both AL and ATTR amyloid deposits. Liquid chromatography tandem mass spectrometry (LC-MS) confirmed the immunohistochemistry results – codeposition of both ATTR and AL amyloid fibrils (Table 2 for Case 2).

Conclusions: The diagnosis of both AL-CA and ATTR-CA in the same patient is a rare occurrence. In the two cases described, there was clinical and histologic evidence that both types were contributing significantly to CA. When CA is suspected, a complete diagnostic workup is essential to ensure the proper subtype, with the recognition that both types can occur concurrently.

Keywords (3): Cardiac Amyloidosis, Codeposition.

Table 1: Monoclonal lab testing

<table>
<thead>
<tr>
<th></th>
<th>Kappa (mg/L)</th>
<th>Lambda (mg/L)</th>
<th>Ratio</th>
<th>Serum IFE</th>
<th>Urine IFE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Case 1</td>
<td>20.6</td>
<td>531.3</td>
<td>0.04</td>
<td>M-protein</td>
<td>M-protein</td>
</tr>
<tr>
<td>Case 2</td>
<td>26.3</td>
<td>365.5</td>
<td>0.07</td>
<td>M-protein</td>
<td>M-protein</td>
</tr>
</tbody>
</table>

Table 2: Liquid Chromatography Tandem Mass Spectrometry (LC/MS) Analysis for Case 2

<table>
<thead>
<tr>
<th>Protein</th>
<th>Protein Spectrum Matches (PSM)</th>
<th>PSM Duplicate 2</th>
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</thead>
<tbody>
<tr>
<td>Serum amyloid P-component</td>
<td>35</td>
<td>51</td>
</tr>
<tr>
<td>Apolipoprotein E</td>
<td>31</td>
<td>28</td>
</tr>
<tr>
<td>Vitronectin</td>
<td>30</td>
<td>34</td>
</tr>
<tr>
<td>Apolipoprotein A-IV</td>
<td>29</td>
<td>40</td>
</tr>
<tr>
<td>Lambda</td>
<td>25</td>
<td>30</td>
</tr>
<tr>
<td>Transthyretin</td>
<td>23</td>
<td>23</td>
</tr>
<tr>
<td>IGH</td>
<td>23</td>
<td>23</td>
</tr>
<tr>
<td>Lambda</td>
<td>22</td>
<td>28</td>
</tr>
<tr>
<td>Apolipoprotein A-I</td>
<td>18</td>
<td>27</td>
</tr>
<tr>
<td>Kappa</td>
<td>16</td>
<td>25</td>
</tr>
<tr>
<td>IGH</td>
<td>15</td>
<td>23</td>
</tr>
</tbody>
</table>

Constant Components of Amyloid
Immunoglobulins
Transthyretin

*Other proteins are not represented in the table.

TTR & lambda chains similar or more abundant than constant components of amyloid.
PT132

CAUTION!!! NOT ALL THAT GLITTERS IS TRANSTHYRETIN AMYLOID! AORTIC REGURGITATION, YET ANOTHER MIMICKER

N. Akhiyat 1, D. Davies 1, A. Dispenzieri 1, M. Grogan 1, O. A. Ezzeddine 1

1 Mayo Clinic, Rochester.

Background: 99mTechnetium-pyrophosphate cardiac scintigraphy (PYP) is reliable for the diagnosis of Transthyretin cardiac amyloidosis (TTR-CA) and has triggered a paradigm shift towards non-invasive diagnosis. A risk to this paradigm shift is that other pathologies may cause significant blood pool radiotracer uptake, confounding results and reducing the diagnostic accuracy of PYP.

Case: 68 y/o male presented with exertional dyspnea and peripheral edema. History was pertinent for paroxysmal atrial fibrillation/flutter and renal insufficiency (GFR 40 mL/min/BSA). He had no history of carpal tunnel syndrome, spinal stenosis, peripheral neuropathy and no family history of amyloidosis. ECG revealed sinus rhythm without low voltage or a pseudo-infarct pattern. NTproBNP was elevated (1,844 pg/mL) but HS troponin T (13 ng/L) and serum free light chains were normal. Echo demonstrated moderate LV enlargement, LVEF 49%, normal wall thickness (9mm), mildly elevated filling pressures (E/e’ 12), borderline abnormal left ventricular strain (-17%) without typical amyloid pattern. A bicuspid aortic valve with an eccentric jet of moderate aortic regurgitation (AR) was noted. Ischemic workup was unremarkable. PYP showed Perugini grade 2 uptake on planar imaging but single-photon emission computed tomography (SPECT) was not performed. TTR DNA sequencing showed no mutations. He was diagnosed with wild-type TTR-CA and prescribed Tafamidis.

He presented to our institution for a 2nd opinion. A high-pitched diastolic murmur was auscultated and raised suspicion for severe AR. Transthoracic echo showed anterior aortic cusp prolapse. PYP was repeated to ensure myocardial uptake. While this confirmed grade 2 uptake on planar imaging, SPECT showed increased radiotracer activity in the blood pool without evidence of myocardial uptake. Transesophageal echo confirmed flail aortic valve with severe AR. Tafamidis was discontinued and surgical intervention for severe AR was recommended.

Discussion: Expert consensus recommendations stipulate that in cases of unexplained HF (without ischemic or valvular etiologies), a diagnosis of TTR-CA be non-invasively made only in the presence of both “grade 2 or 3 myocardial radiotracer uptake” and “typical cardiac imaging features” for amyloid infiltration. Moreover, they emphasize that in cases showing planar myocardial radiotracer uptake, SPECT-based imaging is necessary to differentiate myocardial uptake from blood pool. Here, we present a patient whose HF was valvular in etiology and in whom typical cardiac imaging features were absent suggesting a low pre-test likelihood for TTR-CA to warrant PYP imaging. Further, this case illustrates AR as yet another cause of false-positive planar imaging- which we hypothesize is- due to LV enlargement and regurgitant flow. Judicious interpretation of clinical/imaging data and adhering to expert consensus recommendations is crucial to avoid misdiagnosis and evade potentially serious consequences.
PT133

PATTERNS OF PSYCHO-EMOTIONAL DISTRESS IN PROVEN AND SUSPECTED AMYLOIDOSIS – RESULTS FROM AMYKOS

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2 Department of Internal Medicine I/Cardiology, University Hospital Würzburg, Würzburg.
3 Department of Neurology, University Hospital Würzburg, Würzburg.
4 Department of Internal Medicine II/Hepatology, University Hospital Würzburg, Würzburg.

Introduction: The diagnosis of amyloidosis is complex and frequently delayed. Therapeutic options range from “watch-and-wait” to invasive treatment such as chemotherapy or organ transplantation. Up to now, there are only few data about the psycho-emotional patterns associated with amyloidosis.

Objectives: We aimed to assess various domains of distress among patients with suspected and established amyloidosis.

Methods: AmyKoS is the regional prospective follow-up study that consecutively includes and comprehensively phenotypes all patients presenting at our Interdisciplinary Amyloidosis Center. Psychometric assessment includes self-rated questionnaires addressing selected domains of psycho-emotional distress via the distress thermometer (DT as visual analogue scale, range 1-10), anxiety (GAD-7: Generalized Anxiety Disorder 7-item scale), depression (PHQ-9: Patient Health Questionnaire 9-item), and fear of progression (PA-F: Fear of Progression Questionnaire-Short Form 12-item).

Results: We here report on the first 110 patients with suspected or proven amyloidosis recruited by AmyKoS (08/18-07/19). Distress was comparatively analyzed in three subgroups at the first contact: ATTRwt (n=21) vs systemic amyloidosis (n=30) vs suspected amyloidosis (n=35). Median age was 74 (61-86) vs 64.5 (46-85) vs 68 (48-85) years; male:female ratio was 19:2 vs 15:15 vs 18:17; median EGOC status was 0.5 (0-2) vs 0.5 (0-2) vs 0 (0-1); median number of involved organs was 2 (1-2) vs 1.5 (1-4) vs suspected in 1 (0-3); cardiac involvement was found in 95% (20/21) vs 70% (21/30) vs suspected in 71% (25/35); median NT-proBNP was 2360 (87-9309) vs 2070 (139-58236) vs 376 (22-41333) pg/ml, and high-sensitive troponine was 39.4 (9-104) vs 56.4 (8-298) vs 15.6 (5-139) pg/ml. Median distress scores assessed by DT were 4.0 (0-9) vs 6.5 (1-9) vs 6.0 (1-10). GAD-7 revealed median scores of 4.5 (0-12) vs 5.0 (0-21) vs 5.0 (0-17). Minimal symptoms of anxiety were found in 50% vs 48% vs 49%, mild symptoms in 40% vs 41% vs 23%, and moderate-to-severe symptoms in 10% vs 10% vs 29%. Median PHQ-9 scores were 5.0 (0-10) vs 7.0 (0-14) vs 5.0 (0-17). Minimal symptoms of depression were measured in 39% vs 29% vs 49%, mild symptoms in 50% vs 39% vs 23%, and moderate-to-severe symptoms in 11% vs 32% vs 29%. PA-F showed median scores of 26.5 (14-51) vs 32.0 (14-51) vs 27.0 (15-52).

Conclusion: Distress is highly prevalent in patients with both proven and suspected amyloidosis. ATTR amyloidosis appears to cause less distress, anxiety and fear of progression despite continuous disease progression and limited approved treatment options. In contrast, depressive mood and fear of progression seems to be more prevalent in AL amyloidosis. These preliminary data await confirmation in larger samples, but indicate an urgent need for professional support.
"THE GIANT AWAKES" – RAPID INCREASES IN THE DIAGNOSIS OF TRANSTHYRETIN (TTR) AMYLOIDOSIS AFTER THE ATTR-ACT TRIAL OF TAFAMIDIS

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Introduction: Historically, TTR cardiac amyloidosis (ATTR-CA) was considered a rare disease with no effective treatment except liver transplantation. However, with the advent of non-invasive diagnostic algorithms and introduction of new disease modifying therapies, there has been a marked increase in awareness of ATTR-CA, leading to enhanced screening and diagnosis. The accurate incidence of the disease remains unknown. Additionally, the long-term efficacy of older and cheaper treatment options such as diflunisal (an old non-steroidal anti-inflammatory drug with TTR stabilising actions) and TTR fibril disrupters (doxycycline and epigallocatechin gallate) have not been thoroughly studied.

Objectives: To update our understanding of the epidemiology and treatment patterns of ATTR-CA in the Australian environment in light of increasing referrals after the release of the ATTR-ACT study results.

Methods: All patients with ATTR-CA who were referred between November 2014 and December 2019 were identified from the patient database of our specialised regional amyloidosis centre. Clinicopathological data and investigative results were retrospectively collected and analysed.

Results: 150 patients with ATTR-CA were referred between 2014 and 2019, 136 have been formally reviewed. Between 2015 and 2019, there has been a 400% increase in referrals of ATTR-CA, with at least one new referral a week since the benefits of tafamidis were reported. Median age was 80 years old (IQR 75-84) and 94% (128 out of 136) were male. Median follow up period was 13 months (IQR 5-26 months). 25 patients (18.4%) deaths were reported during the follow up. 108 patients (79.4%) were commenced on at least one TTR specific therapy.

Conclusions: There was significant increase in the detection of ATTR-CA since non-invasive diagnosis of ATTR-CA was reported and tafamidis was identified as significantly beneficial therapy. Referrals continue to increase, and the accurate assessment of the true incidence of ATTR-CA remains difficult. Assessment of older non-tafamidis therapies in this population is ongoing.

<table>
<thead>
<tr>
<th></th>
<th>ATTR-CA n = 136</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age</strong> (Median IQR)</td>
<td>80 (75-84)</td>
</tr>
<tr>
<td><strong>Gender</strong></td>
<td>Male 128 (94.1%)</td>
</tr>
<tr>
<td><strong>Gillmore Stage</strong></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>64 (47.4%)</td>
</tr>
<tr>
<td>2</td>
<td>49 (36.3%)</td>
</tr>
<tr>
<td>3</td>
<td>22 (16.3%)</td>
</tr>
<tr>
<td><strong>Duration of Follow up (Months)</strong> (Median IQR)</td>
<td>13 (5-26)</td>
</tr>
<tr>
<td><strong>Atrial Fibrillation</strong></td>
<td>Yes 81 (60.0%)</td>
</tr>
<tr>
<td></td>
<td>No 54 (40.0%)</td>
</tr>
<tr>
<td><strong>IVSD (mm)</strong> (Median IQR)</td>
<td>16 (14-18)</td>
</tr>
<tr>
<td><strong>Carpal Tunnel Syndrome</strong></td>
<td>Yes 96 (74.4%)</td>
</tr>
<tr>
<td></td>
<td>No 33 (25.6%)</td>
</tr>
<tr>
<td><strong>Diflunisal</strong></td>
<td>Yes 61 (44.9%)</td>
</tr>
<tr>
<td></td>
<td>No 75 (55.1%)</td>
</tr>
<tr>
<td><strong>Doxycycline</strong></td>
<td>Yes 58 (42.6%)</td>
</tr>
<tr>
<td></td>
<td>No 78 (57.4%)</td>
</tr>
<tr>
<td><strong>Epigallocatechin gallate</strong></td>
<td>Yes 77 (56.6%)</td>
</tr>
<tr>
<td></td>
<td>No 59 (43.4%)</td>
</tr>
<tr>
<td><strong>Death</strong></td>
<td>Yes 25 (18.4%)</td>
</tr>
<tr>
<td></td>
<td>No 111 (81.6%)</td>
</tr>
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</table>

International Symposium on Amyloidosis 363
CARDIAC AMYLOIDOSIS AT MOUNT SINAI: REVIEW OF 164 CASES WITH SPECIAL REFERENCE TO ASSOCIATED ARTHROPATHY, NEUROPATHY, MYOPATHY AND GAMMOPATHY

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Introduction: 11 biochemically distinct forms of amyloidosis affect the heart, two of which (ATTRwt; AANF) are considered to be diseases of aging. Microdeposits of amyloid are also concomitants of aging in articular cartilage/synovial membranes of patients undergoing joint replacement for osteoarthritis, and have been identified in intervertebral discs; recent studies have linked musculoskeletal amyloid to TTR, both descriptively and pathogenically (Matusaki, T et al 2017; Rubin J, 2017).

Objectives: We aimed to assess the prevalence of these disease manifestations, including entrapment and sensorimotor neuropathy, osteoarthritis requiring joint arthroplasty, and gammopathy, among patients with cardiac amyloidosis seen at our institution over the period 1997-2020.

Materials and Method: Amyloid was typed in endomyocardial biopsies/postmortem tissue by immunohistochemistry, direct extraction from formalin fixed samples, or two dimensional gel electrophoresis of frozen material during the first 10 years, and more recently by mass spectroscopy. Full genetic sequencing of TTR was carried out to identify mutations, and to characterize patients as wild-type. Pyrophosphate scanning was utilized both to identify amyloid in heart, but also to image the axial skeleton and major joints. Assessments currently being used include evaluations for both large and small fiber, as well as autonomic, neuropathy. Gammopathy was assessed by serum and urine immunofixation.

Results: Our cohort includes 34 patients with AL (lambda/kappa ratio 8.6:1); 63 (38%) with ATTRwt, 31 (19%) with the Ile122 substitution, 32 (19.5%) with other ATTR mutations, and 4 in which AA or Aβ2m were identified in myocardium. Non-Ile122 cardiac amylloids represented 84% of the total number of familial amyloid polyneuropathy (FAP) patients seen over this period. There has been a significant increase in the percent patients with ATTR compared to AL over the past decade, likely reflecting referral bias and greater appreciation of disease spectrum. Major differences between ATTRwt and ATTRIle122 patients include average age at diagnosis (79.2 versus 69.0), sex (97% versus 77% male; women averaging 11 years older than men among patients with Ile122), and ethnicity (3% versus 97% African-american or Afro-caribbean). In particular, the latter included 10 Caribbean islands, Guyana, and Peru. Among the patients with ATTR wt compared to ATTR with the Ile122 substitution, the incidence of prior Carpal tunnel syndrome/release surgery was 36.5% vs 38.7%; spinal stenosis/laminectomy 23.8% vs 25.8%; severe osteoarthritis/prior joint replacement 25.3% vs 25.8%, sensorimotor neuropathy 12.6% vs 19.4%, and MGUS 15.8% vs 12.9%

Discussion: This retrospective analysis forms a backdrop with which to interpret linkage between the major systemic forms of cardiac predominant amyloidosis and musculoskeletal/neurologic symptomatology that may be directly related to subunit fibril deposition in joints, axial skeleton and muscle. In particular, deposition may precede the early clinical manifestations of atrial arrhythmias, valvular amyloid, or heart failure with preserved ejection fraction by many years, and could serve to identify individuals at risk, including carriers of Ile122 and other ATTR mutations.
PW001

ESTIMATED NUMBER NEEDED TO TREAT (NNT) TO PREVENT ALL-CAUSE MORTALITY WITH TAFAMIDIS FROM THE TAFAMIDIS IN TRANSTHYRETIN CARDIOMYOPATHY CLINICAL TRIAL (ATTR-ACT)


1 Stanford University School of Medicine, Stanford, California, USA.
2 Pfizer, Groton, CT, USA.
3 Pfizer, New York, NY, USA.
4 Division of Cardiology, Department of Medicine, Northwestern University Feinberg School of Medicine, Chicago, IL, USA.
5 Envision Pharma group.

Introduction: Tafamidis reduced all-cause mortality and cardiovascular (CV)-related hospitalizations in patients with transthyretin amyloid cardiomyopathy (ATTR-CM) in the Tafamidis in Transthyretin Cardiomyopathy Clinical Trial (ATTR-ACT). The number needed to treat (NNT; number treated to prevent 1 negative event) can inform therapeutic decisions.

Objective: To compare the NNT for tafamidis from ATTR-ACT with other relevant cardiovascular drug trials.

Methods: ATTR-ACT, a 30-month, double-blind, placebo-controlled, randomized study (n=441), examined tafamidis efficacy and safety in ATTR-CM patients. The NNT from ATTR-ACT was calculated and compared with recent key HF trials with the equation: 1/(control event rate [CER]-experimental event rate [EER]) x 100. NNT calculation was adapted for CV-related hospitalization as a recurring event to calculate patient-years of treatment needed to prevent 1 CV-related hospitalization.

Results: The all-cause mortality NNT was 8 for tafamidis in ATTR-ACT; and ranged from 9-44 for comparators (Table). In ATTR-ACT, 4.4 patient-years of treatment were needed to prevent 1 CV-related hospitalization (ie, to prevent 1 hospitalization: 1 patient treated for 4.4 years; or 4.4 patients treated for 1 year).

Conclusions: While comparisons were made in distinct patient populations with different CV disease states, the NNT for all-cause mortality with tafamidis was lower vs comparators, providing further context for the benefits of tafamidis in ATTR-CM.

Keywords: ATTR; cardiomyopathy; tafamidis

Table: All-cause mortality NNTs for treatments for Heart Failure

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Trial</th>
<th>Patients</th>
<th>Duration (mths)</th>
<th>Patients (n)</th>
<th>Control</th>
<th>CER</th>
<th>EER</th>
<th>NNT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tafamidis</td>
<td>ATTR-ACT</td>
<td>441</td>
<td>30.0</td>
<td>441</td>
<td>Placebo</td>
<td>42.9</td>
<td>29.5</td>
<td>8</td>
</tr>
<tr>
<td>Enalapril</td>
<td>SOLVD</td>
<td>HF</td>
<td>41.4</td>
<td>2569</td>
<td>Placebo</td>
<td>39.7</td>
<td>35.2</td>
<td>22</td>
</tr>
<tr>
<td>Metoprolol</td>
<td>MERIT-HF</td>
<td>HF</td>
<td>12.0</td>
<td>3991</td>
<td>Placebo</td>
<td>10.8</td>
<td>7.3</td>
<td>28</td>
</tr>
<tr>
<td>Spirolactone</td>
<td>RALES</td>
<td>HF</td>
<td>24.0</td>
<td>1663</td>
<td>Placebo</td>
<td>46.0</td>
<td>35.0</td>
<td>9</td>
</tr>
<tr>
<td>Carvedilol</td>
<td>COPERNICUS</td>
<td>HF</td>
<td>10.4</td>
<td>2289</td>
<td>Placebo</td>
<td>16.8</td>
<td>11.2</td>
<td>18</td>
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<tr>
<td>Eplerenone</td>
<td>EPHESUS</td>
<td>HF</td>
<td>16.0</td>
<td>6632</td>
<td>Placebo</td>
<td>16.7</td>
<td>14.4</td>
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<tr>
<td>Candesartan</td>
<td>CHARM</td>
<td>HF</td>
<td>40.0</td>
<td>4576</td>
<td>Placebo</td>
<td>31.0</td>
<td>28.0</td>
<td>33</td>
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<tr>
<td>Sacubitril-valsartan</td>
<td>PARADIGM-HF</td>
<td>HF</td>
<td>27.0</td>
<td>8442</td>
<td>Enalapril</td>
<td>19.8</td>
<td>17.0</td>
<td>36</td>
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<tr>
<td>Dapagliflozin</td>
<td>DAPA-HF</td>
<td>HF with reduced ejection fraction</td>
<td>18.2</td>
<td>4744</td>
<td>Placebo</td>
<td>13.9</td>
<td>11.6</td>
<td>43</td>
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</tbody>
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THE 6-MINUTE WALK TEST IS A PREDICTOR OF SURVIVAL IN THE TAFAMIDIS IN TRANSTHYRETIN CARDIOMYOPATHY CLINICAL TRIAL (ATTR-ACT)

M. S. Maurer 1, C. Rapezzi 2, M. Waddington-Cruz 3, G. Merlini 4, J. H. Schwartz 5, B. Gundapaneni 6, T. A. Patterson 6, M. B Sultan 5, P. Elliott 7

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Introduction: In the Tafamidis in Transthyretin Cardiomyopathy Clinical Trial (ATTRACT), tafamidis was shown to be an effective treatment for patients with transthyretin amyloid cardiomyopathy (ATTR-CM). The reduction in mortality and cardiovascular-related hospitalizations with tafamidis was more pronounced in patients who were New York Heart Association (NYHA) class I or II at baseline than in NYHA class III patients; however, NYHA classification may be an imperfect measure of disease severity. The 6-minute walk test (6MWT) is a simple, easily standardized, and broadly accepted test that has been identified as a predictor of overall survival in patients with chronic heart failure.

Objective: To conduct a post-hoc analysis of data from ATTR-ACT to assess the utility of functional capacity, assessed by 6MWT distance, as a measure of disease severity, and as predictor of ATTR-CM disease progression and response to treatment with tafamidis.

Methods: ATTR-ACT was an international, multi-center, double-blind, placebo-controlled, randomized study. Hereditary (ATTRm) or wild-type (ATTRwt) patients with ATTR-CM (N=441) were randomized to tafamidis (n=264) or placebo (n=177) for 30 months. Patients with NYHA class I (n=37), class II (n=263), and class III (n=141) at baseline were included in the study.

Results: While many patients with higher NYHA class also had a shorter median 6MWT distance at baseline, there was a large overlap in distances recorded across NYHA classes. Median (Q1-Q3) 6MWT distance by NYHA class was: class I, 442 (372-480) m; class II, 390 (317-458) m; class III, 256 (180-336) m. Baseline 6MWT distance correlated strongly with survival in all patients, with mortality at the end of the trial being: 88.9% in patients with a baseline 6MWT distance <0.0001).

Conclusions: In patients with ATTR-CM, 6MWT is a significant predictor of survival and could be used together with other measures such as NYHA class as a measure of disease severity and prognosis. These data further support the importance of early diagnosis and treatment in patients with ATTR-CM.

Keywords: Amyloidosis, cardiac; cardiac amyloidosis; transthyretin; drug therapy.
CAUSES OF CARDIOVASCULAR HOSPITALIZATION AND DEATH IN THE TAFAMIDIS IN TRANSTHYRETIN CARDIOMYOPATHY CLINICAL TRIAL (ATTR-ACT)

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Introduction: In the Tafamidis in Transthyretin Cardiomyopathy Clinical Trial (ATTRACT), tafamidis significantly reduced mortality and cardiovascular (CV)-related hospitalizations compared with placebo in patients with transthyretin amyloid cardiomyopathy (ATTR-CM).

Objective: To assess the causes of CV-related hospitalizations and deaths in ATTR-ACT.

Methods: ATTR-ACT was an international, multi-center, double-blind, placebo-controlled, randomized study. Patients with hereditary or wild-type ATTR-CM were randomized to tafamidis (n=264) or placebo (n=177) for 30 months. The independent Endpoint Adjudication Committee determined whether certain investigator-reported events met the definition of disease-related efficacy endpoints using pre-defined endpoint criteria. Hospitalization reports were reviewed to determine if CV-related (heart failure [HF], arrhythmia, myocardial infarction [MI], transient ischemic attack [TIA]/stroke, and other CV causes) as were deaths (HF, arrhythmia, MI, sudden death, stroke, and other CV causes).

Results: The number of patients with a CV-related hospitalization was 138 (52.3%) with tafamidis and 107 (60.5%) with placebo. The total number of CV-related deaths was 53 (20.1%) with tafamidis and 50 (28.2%) with placebo. All predefined causes of hospitalization or death were less frequent with tafamidis than with placebo (see Table).

Conclusions: The most common cause of hospitalization or death in ATTR-ACT was HF, while there were relatively few instances of CV-related sudden death. These data provide further insight into CV disease progression in patients with ATTR-CM.

<table>
<thead>
<tr>
<th>Hospitalizations*</th>
<th>Deaths</th>
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<tbody>
<tr>
<td></td>
<td>Tafamidis</td>
</tr>
<tr>
<td>(N=264)</td>
<td>(N=177)</td>
</tr>
<tr>
<td>Adjudicated cause, n (%)</td>
<td></td>
</tr>
<tr>
<td>Heart Failure</td>
<td>114 (43.2)</td>
</tr>
<tr>
<td>Arrhythmia</td>
<td>40 (15.2)</td>
</tr>
<tr>
<td>TIA/stroke</td>
<td>7 (2.7)</td>
</tr>
<tr>
<td>Myocardial infarction</td>
<td>2 (0.8)</td>
</tr>
<tr>
<td>Other CV</td>
<td>45 (17.0)</td>
</tr>
<tr>
<td>Sudden death</td>
<td>-</td>
</tr>
</tbody>
</table>

*All new hospitalizations are counted, with multiple occurrences of the same hospitalization reason in a single patient counted once. Keywords: Amyloidosis, cardiac; cardiac amyloidosis; transthyretin
MODELING OF SURVIVAL AND FREQUENCY OF HOSPITALIZATION IN TRANSTHYRETIN CARDIOMYOPATHY WITH TAFA MIDIS

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Introduction: Tafamidis is an oral small molecule that stabilizes transthyretin developed for the treatment of Transthyretin Amyloid Cardiomyopathy (ATTR-CM). A single Phase 3, multicenter, 3-arm, parallel design, placebo-controlled, randomized, 30-month study determined efficacy, safety and tolerability of tafamidis on clinical outcomes in patients with ATTR-CM.

Objective: The primary objectives of this analysis were:
1. To assess relationships between 20 mg or 80 mg tafamidis meglumine and placebo on allcause mortality and frequency of cardiovascular (CV)-related hospitalizations (HO) using a model-based approach;
2. To evaluate effects of predictors on dose or exposure-response relationships, with exposure derived from a population PK model.

Methods: Parametric hazard distributions were developed for: 1. All-cause mortality and 2. Frequency of CV-related HOs. Time-to-event (TTE) models were fitted to survival data, repeated-time-to-events (RTTE) models to the HO data. Disease-specific characteristics, lab values, and echocardiogram parameters were assessed as predictors of death and HO hazards.

Results: The percentage of patients alive at Month 30 across pooled tafamidis and placebo were 70.5% and 57.1% respectively. In total, 154 deaths were reported.

There were 495 CV-related HO visits (0 to 8 visits/patient) with an average of 1.12 over 30 months/patient. About 44.4% did not experience a HO visit during the trial and 21.8% of patients dropped out, the majority under placebo.

A Gompertz time-varying model best described the TTE hazard. The estimated drug effect was a cumulative hazard ratio of 0.583, indicating a 41.7% [95% CI (23.2; 57.3)] decreased cumulative risk of death over 30 months in the pooled tafamidis group over placebo. Significant predictors of improved survival were N-terminal prohormone B-type natriuretic peptide (NT-proBNP), left ventricular ejection fraction (LVEF), 6-minute walk test (6MWT), genotype and blood urea nitrogen (BUN).

A Gompertz model also described the HO hazard, suggesting increased risk of HO over time. The drug effect was a 30.7% reduction in the average cumulative risk across all CV-related HO events. Baseline troponin I (TROP), 6MWT, BUN and LVEF were identified as significant covariates in the RTTE model.

Within the 30-month trial duration, no differentiation between tafamidis meglumine 20 mg and 80 mg doses or exposure was identified on either endpoint.

Conclusions: TTE and RTTE models characterized mortality and CV-related HO risk in ATTR-CM patients on tafamidis or placebo. Both risks were characterized by a time-varying hazard increasing over time. Significant predictors, 6MWT, BUN, LVEF, TROP, NT-proBNP and genotype modified the baseline hazards while tafamidis treatment affected the hazards’ time course. These data support improved cumulative survival probability and reduced cumulative HO risk for patients treated with tafamidis, and suggest that treating ATTR-CM patients earlier may result in a better outcome.

Keywords: tafamidis, transthyretin amyloid cardiomyopathy, mortality & morbidity
SAFETY OF THE UTAH DIFLUNISAL PROTOCOL FOR PATIENTS WITH TRANSTHYRETIN CARDIOMYOPATHY

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Introduction: Diflunisal is a transthyretin stabilizer that slows the progression of polyneuropathy in patients with hereditary transthyretin amyloidosis (ATTR). This is a non-steroidal antiinflammatory with limited utilization in cardiomyopathy because of concerns with the side effects. Before the approval of Tafamidis in 2019, patients with ATTR cardiomyopathy were treated with Diflunisal using the Utah Diflunisal Amyloidosis Protocol in our center.

Objectives: Our goal is to describe the safety of the Utah Diflunisal Amyloidosis Protocol in patients with ATTR cardiomyopathy.

Methodology: The Utah Diflunisal Amyloidosis Protocol was offered to patients with confirmed ATTR cardiomyopathy who were < 90 years, with estimated glomerular filtration rate (GFR) > 30 ml/min/1.73m2 and serum creatinine < 2.0 mg/dl. Patients were treated with Diflunisal 250 mg of twice daily in combination with a proton pump inhibitor. The protocol screened for side effects such as anemia, increase in creatinine, decrease in platelets, decrease in GFR, myocardial infarction (MI), stroke, heart failure (HF) and gastrointestinal (GI) pain. Follow up was done through phone or office visits at 1 week, 1 month, 3 months, 6 months and 12 months.

Results: Thirty patients were treated with Diflunisal and four were excluded because of lack of data. The mean age was 78 years, 88% were white males and 96% had wild-type ATTR. Patients were treated for a mean time of 67 weeks. There was no statistically significant difference in platelets, hemoglobin and creatinine, between baseline to 6 months and 12 months. - Table. Out of 26 patients, five discontinued Diflunisal due to side effects; three patients had GI pain, one patient developed GI bleeding and one discontinued Diflunisal due to heart failure exacerbation. Nine patients developed heart failure exacerbation during the follow-up; of them, four had increasing edema and the five worsening dyspnea. The median time of heart failure was 20 weeks from initial administration of Diflunisal and only two exacerbations happened in the first two months. Eight heart failure exacerbations resolved with titration of diuretics and one needed hospitalization at 38 weeks from Diflunisal starting date.

Table 2.

<table>
<thead>
<tr>
<th>Laboratories during follow-up</th>
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<tr>
<td></td>
</tr>
<tr>
<td>Hemoglobin (g/dl),</td>
</tr>
<tr>
<td>Platelets (k/µl)</td>
</tr>
<tr>
<td>Creatinine (mg/dl)</td>
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</table>

*P value comparing baseline with 6 months
**P value comparing baseline with 12 months

Conclusion: Diflunisal was well tolerated in patients with cardiac ATTR using this protocol and represents a therapeutic alternative to patients who have no access to Tafamidis. The most common adverse event was heart failure exacerbation which was controlled mostly in an outpatient setting.

Key words: amyloidosis, transthyretin, Diflunisal
DESIGN OF CARDIO-TTRANSFORM, A PHASE 3 STUDY TO EVALUATE THE EFFICACY AND SAFETY OF AKCEA-TTR-LRx (ION-682884) IN PATIENTS WITH TRANSTHYRETIN-MEDIATED AMYLOID CARDIOMYOPATHY


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Background: Transthyretin amyloidosis cardiomyopathy (ATTR-CM) is a fatal condition, leading to heart failure (HF) and ultimately death. ATTR-CM is caused by misfolding and aggregation of transthyretin (TTR), a protein produced by liver. Depending on the presence or absence of a destabilizing mutation in the TTR gene, the disease can be classified as hereditary ATTR-CM (hATTR-CM) or wild-type (wtATTR-CM), respectively. Despite the treatment with a TTR stabilizer, tafamidis, recently approved in United States for the treatment of ATTR-CM, disease progression still occurs. AKCEA-TTR-LRx is an antisense oligonucleotide (ASO) that inhibits production of TTR. It has the same sequence as inotersen (the parent compound, TEGSEDI) but is conjugated to N-acetyl galactosamine (GalNAc) which targets the asialoglycoprotein receptors expressed abundantly on hepatocytes. Presence of GalNAc allows the use of a lower dose to achieve identical pharmacodynamic results. In a phase 1 healthy volunteer study, AKCEA-TTR-LRx given at a 45 mg dose by subcutaneous injection (SC) every four weeks (dose 27-fold smaller than inotersen) achieved a median of > 85% serum TTR reduction compared to baseline.

Study Design and Methods: CARDIO-TTRANSFORM (ClinicalTrials.gov NCT04136171) is a Phase 3 global, double-blind, randomized, placebo-controlled study assessing the efficacy and safety of AKCEA-TTR-LRx in hATTR-CM or wtATTR-CM patients receiving available background standard of care (SoC) therapy. Approximately 750 patients with a history of HF due to ATTR-CM will be randomized 1:1 to receive either AKCEA-TTR-LRx (45 mg) or placebo administered by SC injection once every 4 weeks. Key inclusion criteria include confirmed diagnosis of ATTR-CM by tissue biopsy or positive PYP/DPD/HMDP scan, end-diastolic interventricular septum thickness of >12mm, NT-proBNP >600 pg/mL, NYHA class I-III and 6-minute walk distance (6MWD) >150 m. Key exclusion criteria include estimated glomerular filtration rate < 30 mL/min/1.73m2, platelet count below the lower limit of normal and urine protein/creatinine ratio (UPCR) ≥ 750 mg/g. Concomitant treatment with tafamidis as SoC for ATTR-CM is allowed, if locally approved and available. The study consists of a 120-week Treatment Period and a 20-week Post-Treatment Evaluation Period. Primary efficacy endpoint is the composite of cardiovascular (CV) mortality and frequency of CV clinical events at Week 120 study visit, analyzed by the Finkelstein-Schoenfeld method. Secondary endpoints include the change from baseline in the 6MWD, Kansas City Cardiomyopathy Questionnaire score, rate of CV mortality, CV clinical events, and all-cause of mortality at Week 120.

Conclusions: Despite recent advances, more efficacious, safe and convenient treatment options for ATTR-CM are needed. The CARDIO-TTRANSFORM trial is a large Phase 3 trial designed to evaluate the clinical efficacy and safety of AKCEA-TTR-LRx compared to placebo for the treatment of ATTR-CM.
REAL WORLD EXPERIENCE WITH TAFAMIDIS

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Introduction: CEPARM was created in 1984. Since 2005 more than 340 patients with ATTR were seen in our center. Tafamidis was approved in Brazil in 2016 and only recently has been available by the Public Health System. Nevertheless, since its approval in Europe in 2011 a few Brazilian patients have been receiving tafamidis by either lawsuit, compassionate use or other means.

Objective: To present CEPARM’s multidisciplinary real-world experience treating ATTR patients with tafamidis.

Methods: We extracted total Neuropathy Impairment Score (NIS), Polyneuropathy Disability score (PND), Karnofisky Performance Status (KPS), New Your Heart Association classification for heart failure patients (NYHA), intraventricular septum thickness (IVS), heart ejection fraction (EF), Body Mass Index (BMI) and vital status from baseline (first visit) and at last evaluation.

Results: From 35 treated patients (26 men), 14 interrupted treatment. Reasons were: death due to a severe cardiomyopathy (n=1), liver transplantation (n=3), enrollment in a clinical trial (n=3) and dispensation ceased for the remainders. 24 patients were reevaluated (extraction as of October 2018) with at least one-year follow-up (median time of 19.5 months). 30 patients were V30M, median age of 37 years (26-87), 13 were late-onset and median time from disease onset at start of treatment was 4 years. Other than the death above described, 2 nonV30M patients with severe cardiomyopathy died after 5 months, and 1 year and 7 months after starting treatment. The last one refused cardiac transplantation. At last follow-up PND increased in 6 patients (PND 1 at baseline in 3) and did not change in 18. Total NIS were available in 21 patients, of which 10 had an increase of more than 4 points (8 with NIS > 10 at baseline) and 6 patients had an increase of more than 10 points at last evaluation. KPS (n=24) did not change in 13, worsened in 13 and improved in 1. From the 4 patients with heart failure, 2 had no change in NYHA. From 14 echocardiograms, IVS increased > than 2 mm in 2 patients (1 V122I ATTR). No patient had EF less than 50% at last evaluation. BMI was preserved in 21 patients. Only 1 patient changed from obesity to overweight.

Conclusions: This is a single center multidisciplinary effort to ascertain for disease progression and response to treatment in the real-world. We acknowledge the caveats of lack of control for several aspects of the population and evaluations. Final decision about responsiveness remained the expert best judgment although tafamidis proved of benefit for several aspects of the disease in this particular scenario. This is preliminary data from CEPARM. More controlled data will come in the near future with regular dispensation of tafamidis by the Health Authority.

Keywords (proposed): ATTR, tafamidis treatment.

Funding acknowledgements: Some patients received compassionate tafamidis from Pfizer.
RATIONALE AND DESIGN OF NEURO-TTRANSFORM, A PHASE 3 STUDY TO EVALUATE THE EFFICACY AND SAFETY OF AKCEA-TTR-LRX (ION-682884) IN PATIENTS WITH HEREDITARY TRANSTHYRETIN-MEDIATED AMYLOID POLYNEUROPATHY (HATTR-PN)

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2 Mayo Clinic.
3 University of Pennsylvania School of Medicine.
4 Ionis Pharmaceuticals.
5 Akcea Therapeutics.

Background: Hereditary transthyretin-mediated amyloid polyneuropathy (hATTR-PN) is a progressive and fatal axonal sensorimotor and autonomic neuropathy caused by misfolding/aggregation of transthyretin (TTR), a liver produced protein. Inotersen (Tegsedi™) is an antisense oligonucleotide (ASO) that binds to TTR mRNA to inhibit the production of TTR. In a double-blind placebo-controlled study (NEURO-TTR) inotersen slowed disease progression in patients with hATTR-PN. AKCEA-TTR-LRx is an ASO with the same sequence as inotersen conjugated to a triantennary N-acetyl galactosamine (GalNAc3) which targets the asialoglycoprotein receptors expressed abundantly on hepatocytes. This additional chemical moiety (GalNAc) allows the use of a lower dose to achieve identical pharmacodynamic results. In a phase 1 healthy volunteer study, AKCEA-TTR-LRx given at a 45 mg dose by subcutaneous injection (SC) every four weeks (Q4W) achieved a median reduction of > 85% serum TTR compared to baseline.

Objective: NEURO-TTRANSForm (ClinicalTrials.gov number NCT04136184) is a phase 3 global, open-label study that aims to determine if AKCEA-TTR-LRx is effective and safe as compared to historical placebo (placebo arm in the NEURO-TTR trial) for the treatment of hATTR-PN.

Methods: Approximately 140 hATTR-PN patients will be randomized to receive either AKCEA-TTR-LRx (n = 120; 45 mg SC Q4W) or inotersen (n = 20; 300 mg SC weekly). Key inclusion criteria include preserved ambulatory status (Familial Amyloid Polyneuropathy or FAP stage 1 or stage 2), confirmed TTR mutation, and Neuropathy Impairment Score (NIS) between 10 and 130. Key exclusion criteria include estimated glomerular filtration rate < 45 mL/min/1.73 m2, platelets ≤ 125 × 109/L and urine protein/creatinine ratio ≥ 1000 mg/g. Concomitant treatment with tafamidis, inotersen, patisiran, diflunisal, doxycycline and tauroursodeoxycholic acid (TUDCA) are not allowed. If previously treated with tafamidis, diflunisal, doxycycline or TUDCA, patients must have discontinued treatment for at least 2 weeks prior to Study Day 1. Co-primary efficacy endpoints at Week 66 (primary endpoint analysis) are change from baseline in: serum TTR concentration, modified NIS+7 and Norfolk Quality of Life-Diabetic Neuropathy. An interim analysis will be performed at Week 35. Secondary endpoints include the change from baseline in the Neuropathy Symptom and Change Score, Physical Component Summary score of 36-Item Short Form Survey, Polyneuropathy Disability Score and Modified Body Mass Index.

Results: This trial is currently enrolling patients.

Conclusions: Despite recent advances, there is still a need for effective, well-tolerated and convenient treatments for hATTR-PN. NEURO-TTRANSForm is a phase 3 trial designed to evaluate the efficacy and safety of AKCEA-TTR-LRx compared to the placebo arm in NEURO-TTR for the treatment of hATTR-PN.
Efficacy and Safety of Tafamidis 61 mg in Patients with Transthyretin Amyloid Cardiomyopathy

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3 Pfizer, Groton, Connecticut, USA.
4 Pfizer, New York, NY, USA.
5 Medical University of South Carolina, Charleston, South Carolina, USA.

Introduction: Tafamidis was shown to be safe and effective in patients with transthyretin amyloid cardiomyopathy (ATTR-CM) in the Tafamidis in Transthyretin Cardiomyopathy Clinical Trial (ATTR-ACT), where patients received tafamidis meglumine 80 mg (given as four 20 mg capsules) or 20 mg (referred to as tafamidis 80 mg and 20 mg, respectively) daily. For patient convenience, a new formulation bioequivalent (but not interchangeable per mg) to tafamidis meglumine 80 mg was subsequently developed in a single capsule as tafamidis free acid 61 mg (referred to as tafamidis 61 mg). The ongoing, and expanded, long-term extension (LTE) trial utilizes tafamidis 61 mg in patients who transitioned from ATTR-ACT and in new patients not previously enrolled in ATTR-ACT.

Objective: To better understand the efficacy of tafamidis 61 mg/80 mg vs tafamidis 20 mg, and the safety profile of tafamidis 61 mg.

Methods: Patients in ATTR-ACT were randomized (2:1:2) to daily tafamidis 80 mg, 20 mg, or placebo for 30 months. Those completing ATTR-ACT could enrol in the LTE study and either continue on the same dose of tafamidis or, if previously receiving placebo, be randomized (2:1) to tafamidis 80 mg or 20 mg (median follow-up 38 months). The LTE was then expanded to include patients with ATTR-CM not previously enrolled in ATTR-ACT. Patients in the expanded LTE received tafamidis 61 mg (median follow up 6 months); all patients in the initial LTE transitioned to tafamidis 61 mg. In an interim analysis, all-cause mortality was analyzed integrating 61 mg data from the ongoing and expanded LTE (as of 1 Aug 2019) with 80 mg data and compared with 20 mg data from a combined ATTR-ACT and LTE cohort (as of 1 Aug 2018). All-cause mortality, with heart transplant (HT) or cardiac mechanical assist device (CMAD) implantation counted as death, was assessed in this age adjusted analysis using cox proportional hazards model. A sensitivity analysis using all deaths (HT or CMAD implantation not counted as death) adjusted for age was also performed.

Results: There was a significant reduction in the risk of death with tafamidis 61/80 mg group (N=943) vs 20 mg (N=116); with a hazard ratio of 0.628 (95% CI: 0.430-0.917), corresponding to a 37.2% reduction. In the sensitivity analysis, there was also a significant reduction in the risk of death with tafamidis 61/80 mg vs 20 mg (0.582 [95% CI: 0.392-0.866]); a 41.8% reduction. As of 1 Aug 2019, safety data were available for 715 patients treated with tafamidis 61 mg in the LTE (mean treatment duration 184 days). No new safety concerns were identified. The safety profile was consistent with the known tafamidis 80 mg safety profile; previously reported as comparable to placebo.

Conclusions: This analysis of data from ATTR-ACT combined with that from the LTE supports the use of the higher dose of tafamidis (61 mg or 80 mg) in patients with ATTR-CM. The safety profile of tafamidis 61 mg is comparable to placebo; no adverse drug reactions were identified.

Keywords: Amyloidosis, cardiac; cardiac amyloidosis; transthyretin; drug therapy.
INHIBITION OF THE AGGREGATION OF TRANSTHYRETIN BY STRUCTURE-BASED PEPTIDES IN VITRO AND IN VIVO

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⁴ Indiana University.

Transthyretin amyloidosis (ATTR) is a condition caused by the abnormal systemic deposition of transthyretin (TTR), a blood protein synthesized and secreted mainly by the liver. In health, TTR functions as a transporter of retinol through the interaction with retinol binding protein and thyroxin by direct binding (1). TTR deposits are made of elongated amyloid fibrils, which accumulate in the nerves, the heart, and other organs that eventually fail. ATTR develops as cardiomyopathy and/or polyneuropathy, and is found both in inherited (hATTR) or sporadic cases (wtATTR). Current treatments are limited to a specific cohort of hATTR patients and are sometimes found insufficient to halt disease progression. These are liver transplantation that replaces the main production source of mutant TTR, TTR stabilization by compounds such as tafamidis or diflunisal, and gene expression modulation. These lines of treatment show limited efficacy at late stages or in patients with advanced cardiac involvement (2, 3). Sporadic wtATTR has no therapeutic options.

We have discovered that amyloid fibrillar seeds formed during disease catalyze aggregation of wild-type TTR, even in the presence of stabilizers such as tafamidis or diflunisal (4, 5). We have designed a peptide inhibitor (TabFH2) that hinders this process based on the crystal structures of the two amyloid-driving segments of transthyretin (6). We find that TabFH2 inhibits amyloid seeding caused by fibrils extracted from ATTR patients (5). In addition, TabFH2 treatment results in both reduction of TTR deposition and improvement of motor skills in Drosophila polyneuropathic ATTR models (7). We envision ATTR patients to have access to multi-target medications: chemotherapy to temporary reduce TTR production, chemical stabilization of the native form, and an anti-fibril peptide inhibitor to impede amyloid seeding by preform fibril seeds. With the right combination of treatments, ATTR may evolve from a fatal to chronic or even curable disease.

Keywords: transthyretin, seeding, inhibitor
FIRST-IN-HUMAN PHASE 1 INVESTIGATION OF THE LIGAND-CONJUGATED ANTISENSE OLIGONUCLEOTIDE AKCEA-TTR-LRX FOR THE TREATMENT OF TRANSTHYRETIN AMYLOIDOSIS

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Introduction: Hereditary transthyretin amyloidosis (hATTR) is a rare, progressive fatal disease caused by pathogenic variants in the transthyretin (TTR) gene that destabilize the normal tetrameric structure of the TTR oligomers leading to formation of insoluble, extracellular amyloid deposits in multiple organ systems. hATTR patients predominantly develop neuropathy and/or cardiomyopathy. Wild-type TTR can also form amyloid deposits, leading to congestive heart failure. Inotersen is an antisense oligonucleotide (ASO) that inhibits production of both variant and wild-type TTR protein by degradation of the TTR messenger RNA. Inotersen is approved for the treatment of hATTR patients with polyneuropathy. AKCEA-TTR-LRx (ION-682884), an ASO of similar design and identical sequence, is conjugated to a ligand for selective, receptor-mediated delivery to hepatocytes, the principal source of systemically circulating TTR. This delivery approach has yielded an up to 30-fold increase in potency, and improved the safety and tolerability profiles of ASOs in human clinical trials. In preclinical studies, AKCEA-TTR-LRx produced significant dose-dependent reductions of TTR messenger RNA and protein levels, with a marked increase in potency compared to inotersen. AKCEA-TTR-LRx is currently under development for the treatment of both hereditary and wild-type ATTR.

Objective: Based on prior clinical experience with ligand-conjugated ASOs and supporting preclinical results, AKCEA-TTR-LRx is expected to have an increased potency and improved tolerability and safety profile compared to inotersen. A phase 1/2 study was designed to evaluate the safety, tolerability, pharmacokinetics and pharmacodynamics of AKCEA-TTR-LRx initially in healthy volunteers (NCT03728634).

Methods: Eligible subjects were assigned to one of three multiple-dose cohorts (45, 60 and 90 mg, n=12 per cohort) and randomized 10:2 (active:placebo) to receive 4, once monthly, SC doses of study drug. A higher, single-dose cohort (120 mg) was also evaluated.

Results: Monthly subcutaneous doses of AKCEA-TTR-LRx produced sustained decreases in TTR levels over time with maximum mean reductions from baseline to two weeks after the final fourth dose of 86% and 94% at doses of 45 and 90 mg, respectively. This study is ongoing. Assessment of the safety, tolerability, pharmacokinetics and pharmacodynamics of AKCEA-TTR-LRx over the full dose-range will be presented.

Conclusions: These results support once monthly SC administration of AKCEA-TTR-LRx in phase 3 studies in patients with ATTR cardiomyopathy and hATTR polyneuropathy.
A NOVEL RNA INTERFERENCE THERAPY FOR AMYLOIDOSIS UTILIZING MULTI-THERAPEUTIC POTENTIAL OF CYCLODEXTRIN/DENDRIMER CONJUGATE GENE TRANSFER CARRIER

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2 Department of Physical Pharmaceutics, Graduate School of Pharmaceutical Sciences, Kumamoto University.
3 Department of Amyloidosis Research, Nagasaki International University.

Introduction: RNA interference (RNAi) therapy holds enormous potential for intractable disease treatment that can specifically regulate the responsible gene of disease. In various types of amyloidosis, it is well-documented that amyloidosis progress through 3 critical pathological processes, such as, 1. overproduction or mutation of amyloid precursor gene, 2. amyloid fibril formation caused by conformational change, 3. tissue deposition of amyloid fibrils. To suppress all pathological processes simultaneously, we utilized characteristic features of polyamidoamine starburst dendrimer as a multifunctional material & gene transfer carrier, and developed glucuronylglicosyl-aza-CyD/Dendrimer conjugate (GUG-aza-CDE) as a multi-therapeutic gene transfer carrier for RNAi drugs.

Objectives: We used GUG-aza-CDE as a multi-therapeutic gene transfer carrier for RNAi therapy, and evaluated its therapeutic effects (effectively suppressing 3 pathological process in parallel) on various types of amyloidosis.

Methods: Therapeutic effects of GUG-aza-CDE/RNAi drug complex on various amyloid precursor proteins, such as transthyretin (TTR) in TTR amyloidosis and amyloid A (A) in Alzheimer’s disease, were evaluated in this study. Gene silencing effect was examined by using quantitative PCR (qPCR). Inhibitory effects on amyloid fibril formation and dissolving effects on amyloid fibrils were evaluated by Thioflavin-T (Th-T) assay in vitro. In addition, human TTR V30M Transgenic (Tg) rats were treated with GUG-aza-CDE/ RNAi drug for TTR (0.053 mg/kg) complex by tail vain injection, and evaluated their therapeutic effects by determining TTR deposition in colon by immunohistochemical staining and ELISA assay.

Results: GUG-aza-CDE/RNAi drug for TTR complex significantly reduced TTR expression at mRNA levels by assessing qPCR. This complex also exhibited both inhibitory effects on TTR amyloid fibril formation and dissolving effects on TTR amyloid fibrils in vitro. Interestingly, those therapeutic effects were synergistically enhanced by GUG-aza-CDE/RNAi drug complex formation, compared with GUG-aza-CDE treatment only. Consistent with those in vitro results, GUG-aza-CDE/ RNAi drug complex exhibited the synergistic therapeutic effects in TTR V30M Tg rats in vivo, as assessed by significant reduction of TTR deposition in colon. Furthermore, those synergistic effects of GUG-aza-CDE/ RNAi drug complex was also observed in A amyloid formation process.

Conclusions: Our results suggest that GUG-aza-CDE/RNAi drug complex may have potential as a novel RNAi therapy for amyloidosis by suppressing 3 pathological process of amyloidosis simultaneously with single treatment. By selecting suitable gene specific RNAi drugs for amyloid precursor gene, our multi-target drug may also be applicable for various types of amyloidosis.
TAFAMIDIS 80 MG DEMONSTRATES A SURVIVAL BENEFIT OVER 20 MG AFTER AGE ADJUSTMENT IN PATIENTS WITH TRANSTHYRETIN AMYLOID CARDIOMYOPATHY

T. Damy 1, G. Merlini 2, B. Ebede 3, B. Gundapaneni 4, T. A. Patterson 4, S. See Tai 3, M. B. Sultan 5, R. Witteles 6

1 APHP, CHU Henri Mondor, and INSERM U955, Clinical Investigation Center 006, and DHU ATVB all at Creteil, France.
2 Amyloidosis Center, IRCCS Policlinico San Matteo and University of Pavia, Pavia, Italy.
3 Pfizer, Collegeville, Pennsylvania, USA.
4 Pfizer, Groton, Connecticut, USA.
5 Pfizer, New York, USA.
6 Stanford University School of Medicine, Stanford, CA, USA.

Introduction: The Tafamidis in Transthyretin Cardiomyopathy Clinical Trial (ATTR-ACT) demonstrated a reduction in mortality and cardiovascular-related hospitalizations in the primary comparison of pooled tafamidis meglumine (80 mg and 20 mg) with placebo. Separately, both 80 mg and 20 mg improved survival in ATTR-ACT versus placebo, without a significant difference in mortality between the two doses; however, the study was not designed for a definitive assessment by dose. Given age is a prognostic factor for survival outcome, meaningfulness of patient age distribution in treatment groups was explored in ATTR-ACT.

Objective: To evaluate the difference in mortality between tafamidis meglumine 80 mg and 20 mg when adjusted for age using data from ATTR-ACT, alone and combined with the ongoing extension study.

Methods: ATTR-ACT was an international, double-blind, placebo-controlled, randomized study conducted over 30 months. Patients who completed ATTR-ACT could enroll in a long-term, extension study and either continue on the same daily dose of tafamidis meglumine or, if previously treated with placebo, be randomized (2:1) to tafamidis meglumine 80 mg or 20 mg for up to 60 months. All-cause mortality, with heart transplant (HT) or cardiac mechanical assist device (CMAD) implantation counted as death, was assessed adjusted for age using a cox proportional hazards model (as of 1 Aug 2018). In a sensitivity analysis, all deaths (HT or CMAD implantation not counted as death) were analysed adjusted for age.

Results: In ATTR-ACT, a significant difference (p=0.0405) in median age between patients treated with 80 mg (76.0 years; n=176) and 20 mg (73.5 years; n=88) was noted. Adjusted for age, there was a 16.4% reduction in risk of death with 80 mg vs 20 mg (HR [95% CI], 0.836 [0.506, 1.382]). In the sensitivity analysis there was a 28.7% reduction in risk of death with 80 mg vs 20 mg (0.713 [0.422, 1.205]). In ATTR-ACT combined with the extension study (median follow-up of 38 months), there was a 20.8% reduction in the risk of death for patients continuing on 80 mg compared with those continuing on 20 mg (0.792 [0.507, 1.239]); in the sensitivity analysis, there was a 33.0% reduction (0.670 [0.421, 1.068]). When these combined data were expanded to include patients who were on placebo in ATTR-ACT and switched to tafamidis meglumine in the extension study (80 mg, n=230; 20 mg, n=116), there was a 31.5% reduction with 80 mg (0.685 [0.462, 1.017]); 38.7% in the sensitivity analysis (0.613 [0.405, 0.930]).

Conclusions: Age-adjusted mortality analyses that accounted for a baseline age imbalance consistently demonstrated better efficacy with tafamidis meglumine 80 mg. This was further supported by longer-term treatment data and was more evident with the addition of patients who switched from placebo. These analyses contribute to the totality of evidence supporting the use of tafamidis 80 mg as the preferred dose in patients with transthyretin amyloid cardiomyopathy.
**PW015**

**CHANGES IN NT-PROBNP WITH TAFAMIDIS ARE PREDICTIVE OF SURVIVAL IN PATIENTS WITH TRANSTHYRETIN AMYLOID CARDIOMYOPATHY**

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2 Cleveland Clinic, Cleveland, Ohio, USA.
3 Pfizer, Groton, Connecticut, USA.
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5 Pfizer, New York, NY, USA.
6 APHP, CHU Henri Mondor, and INSERM U955, Clinical Investigation Center 006, and DHU ATVB all at Creteil, France.

**Introduction:** In transthyretin amyloid cardiomyopathy (ATTR-CM), a number of biomarkers have been examined for their prognostic value. Increases in NT-proBNP and troponin T, or reductions in eGFR, have been associated with increased mortality. However, there are limited data on how changes in biomarkers as a consequence of treatment impact survival. In the Tafamidis in Transthyretin Cardiomyopathy Clinical Trial (ATTR-ACT), pooled tafamidis meglumine (80 mg and 20 mg) significantly reduced mortality and cardiovascular-related hospitalizations compared with placebo. In addition, the observed increase in NT-proBNP over time was significantly reduced with tafamidis meglumine compared with placebo and this reduction was significantly greater with 80 mg than 20 mg. Analysis of the impact of changes in NT-proBNP on survival may provide context for the observed differences in NT-proBNP with tafamidis meglumine 80 mg compared with 20 mg and help inform decisions on the optimal dose.

**Objective:** To evaluate if changes in NT-proBNP are predictive of survival in patients treated with tafamidis meglumine and to further assess the changes in NT-proBNP by tafamidis meglumine dose in ATTR-ACT.

**Methods:** ATTR-ACT was a double-blind, placebo-controlled, randomized study in which patients were randomized (2:1:2) to tafamidis meglumine 80 mg, 20 mg, or placebo for 30 months. In this post-hoc analysis, tafamidis meglumine-treated patients were grouped into tertiles based on changes from baseline in NT-proBNP at Month 12 of: ≤0 pg/ml; between 0 and 920 pg/ml; and ≥920 pg/ml. Survival over the subsequent 18 months of ATTR-ACT was then assessed with heart transplant or cardiac mechanical assist device implantation counted as death.

**Results:** In ATTR-ACT, a total of 264 patients were treated with tafamidis meglumine and 214 had baseline and Month 12 NT-proBNP data. Patients in the group with change from baseline in NT-proBNP ≤0 pg/ml had a better survival rate at Month 30 than those in other groups (see Table). At Month 12, NT-proBNP was reduced from baseline in 50.7% of patients treated with tafamidis meglumine 80 mg compared with 36.1% with 20 mg; and at Month 30, 45.5% with tafamidis meglumine 80 mg and 23.3% with 20 mg. There was no meaningful difference in safety between the doses.

**Conclusions:** Change in NT-proBNP in patients treated with tafamidis meglumine is associated with patient survival. As shown in this analysis, improvement, or a smaller increase, in NT-proBNP with treatment is associated with improved survival. Tafamidis meglumine 80 mg significantly reduced the increase in NT-proBNP in patients with ATTR-CM compared with tafamidis meglumine 20 mg and a greater proportion of patients experienced reductions in NT-proBNP in the 80 mg group. These data support the use of tafamidis meglumine 80 mg as the recommended dose.

<table>
<thead>
<tr>
<th>Change in NT-proBNP from Baseline to Month 12</th>
</tr>
</thead>
<tbody>
<tr>
<td>≤0 pg/ml</td>
</tr>
<tr>
<td>(N=98)</td>
</tr>
<tr>
<td>Number (%) alive at Month 30</td>
</tr>
</tbody>
</table>
NI006: A HUMAN-DERIVED ANTIBODY DIRECTED AGAINST AMYLOID TRANSTHYRETIN IN CLINICAL DEVELOPMENT FOR THE TREATMENT OF ATTR CARDIOMYOPATHY

A. Michalon 1, B. Combaluzier 1, A. Hagenbuch 1, C. Huy 1, E. Varela 1, T. Damy 2, O. Suhr 3, M. Saraiva 4, C. Hock 1, R. Nitsch 1, J. Grimm 1

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2 Henri Mondor University Hospital, AP-HP, Creteil, France.
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Introduction and objectives: Amyloid transthyretin (ATTR)-cardiomyopathy results from the accumulation of misfolded and aggregated transthyretin in the myocardium. The accumulation of amyloid causes elevated heart tissue rigidity which can severely impact heart function and survival. A reduction in cardiac amyloid load is therefore expected to translate into a regression of clinical symptom in patients. Towards that goal we developed a human antibody that can trigger the elimination ATTR fibrils.

Methods: Human antibodies selective for ATTR were generated by comprehensive immune repertoire analyses of healthy elderly subjects. A clinical antibody candidate was selected based on its biochemical and biophysical properties and its pharmacological effects in in-vitro and ex-vivo assays and in a new animal model. Further studies investigated the developability and safety profile of this antibody for clinical use.

Results: We developed NI006, a recombinant human monoclonal IgG1 antibody that exclusively binds with high affinity the disease-associated amyloid conformation but not physiological forms of transthyretin. NI006 targets both wild-type ATTR and ATTR mutants that are linked to familial forms of ATTR cardiomyopathy and ATTR polyneuropathy. NI006 dose-dependently induced the clearance of pathological cardiac ATTR from patient tissues by human macrophages ex vivo. In a mouse fibril graft model NI006 triggered the dose-dependent phagocytic removal of patient-derived ATTR fibrils with a minimal effective dose of 0.5 mg/kg. NI006 presented a favorable developability profile including high stability and absence of cross-reactivity and was selected for further evaluation in patients.

Conclusion: Human antibody NI006 mediates a rapid removal of human ATTR in preclinical models. NI006 is currently evaluated for safety and tolerability in patients with ATTR cardiomyopathy.

3 keywords: antibody, therapy, cardiomyopathy
THE BIOEQUIVALENCE OF TAFAMIDIS 61 MG FREE ACID CAPSULES AND TAFAMIDIS MEGLUMINE 4 × 20 MG CAPSULES IN HEALTHY VOLUNTEERS

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2 Pfizer, New York, NY, USA.
3 Pfizer, Brussels, Belgium.
4 Pfizer, Beijing City, China.

Introduction: Tafamidis meglumine, available as 20 mg capsules, is widely approved for use in adults with early-stage symptomatic transthyretin amyloid (ATTR) polyneuropathy. Administered as an 80 mg, once-daily dose (4 × 20 mg capsules), this agent is also licensed (Japan, USA) for the treatment of adults with hereditary and wild-type ATTR cardiomyopathy. The tafamidis 61 mg free acid capsule was developed as an alternative single, solid, oral formulation for patient convenience (currently licensed in USA, UAE). In this single-center, open-label, randomized, 2-period, 2-sequence, crossover, multiple-dose phase 1 study, we compared the rate and extent of absorption of the new and original tafamidis formulations.

Materials & Methods: Eligible individuals included healthy volunteers of both sexes, aged 18–55 years, with a total body weight >50 kg and body mass index 17.5–30.5 kg/m². Absorption of tafamidis 61 mg free acid capsules (test) vs. tafamidis meglumine 80 mg (4 × 20 mg) capsules (reference) was assessed after 7 days of repeated oral dosing under fasted conditions. The steady-state area under the concentration–time profile over the 24-hour dosing interval (AUCtau) and maximum observed concentration (Cmax) were estimated for each volunteer and formulation using non-compartmental methods. Geometric means were calculated, with variability based on geometric percentage coefficient of variation (%CV). Based on guidance from the US Food and Drug Administration, bioequivalence was concluded if the 90% CIs of AUCtau and Cmax test/reference adjusted geometric mean ratios fell within the 80% to 125% acceptance range.

Results: In 30 healthy male volunteers (means: age, 39 y; weight, 79 kg; and BMI, 25.2 kg/m²), geometric mean AUCtau and Cmax values for the tafamidis 61 mg free acid and tafamidis meglumine 4 × 20 mg capsules were similar (Table). The ratios of adjusted geometric means (90% CI) for the test/reference formulations for AUCtau (102.3 [98.0, 106.8]) and Cmax (94.1 [89.1, 99.4]) satisfied prespecified bioequivalence acceptance criteria. The incidence of adverse events was similar for the two tafamidis formulations, and no new safety signals were reported.

Discussion & Conclusions: In this phase 1 study, a new formulation of tafamidis (61 mg free acid capsules) was shown to be bioequivalent to the currently marketed formulation (tafamidis meglumine 80 mg [4 × 20 mg] capsules) after 7 days of repeated oral dosing in healthy volunteers. The single solid oral tafamidis 61 mg free acid formulation provides a more convenient dosing option for patients with ATTR cardiomyopathy.

Table. Summary of Tafamidis Treatment Comparison

<table>
<thead>
<tr>
<th>Parameter (units)</th>
<th>Adjusted Geometric Means (Geometric %CV)</th>
<th>Ratio (Test/Reference) of Adjusted Means, % (90% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Tafamidis 61 mg Free Acid Capsules (Test)</td>
<td>Tafamidis Meglumine 4 × 20 mg Capsules (Reference)</td>
</tr>
<tr>
<td>AUCtau* (ng•hr/mL)</td>
<td>170,000 (23)</td>
<td>166,200 (20)</td>
</tr>
<tr>
<td>Cmax (ng/mL)</td>
<td>8,553 (23)</td>
<td>9,087 (18)</td>
</tr>
</tbody>
</table>

*Over 24-hr dosing interval.

Keywords: tafamidis, ATTR; bioequivalence
LONG-TERM, INTEGRATED SAFETY OF PATISIRAN IN PATIENTS WITH HEREDITARY TRANSTHYRETIN-MEDIATED AMYLOIDOSIS WITH POLYNEUROPATHY


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2 Centro Hospitalar Lisboa Norte, Hospital de Santa Maria, Lisboa, Portugal.
3 Johns Hopkins University, Baltimore, USA.
4 National Amyloidosis Centre, London, UK.
5 Alnylam Pharmaceuticals, Cambridge, MA, USA.
6 Amyloidosis Center, Boston University, Boston, MA, USA.

Introduction/Background: Hereditary transthyretin-mediated (hATTR) amyloidosis, also known as ATTRv amyloidosis, is a progressive, life-threatening disease often with a mixed phenotype of polyneuropathy and cardiomyopathy. Patisiran halts or reverses polyneuropathy and improves quality of life from baseline in the majority of patients with hATTR amyloidosis as shown in the Phase 3 APOLLO study. Based on these data, patisiran is approved in certain countries globally for the treatment of hATTR amyloidosis with polyneuropathy. The patisiran clinical development program for hATTR amyloidosis with polyneuropathy comprises the largest clinical trial patient population with a wide spectrum of disease severity and genotypes.

Objectives: To present the long-term, comprehensive, integrated safety data from the patisiran clinical development program in hATTR amyloidosis with polyneuropathy.

Methods: Safety data as of September 24, 2018 from the Phase 2 Open-Label Extension (OLE) (NCT01961921), Phase 3 APOLLO (NCT01960348), and ongoing Global OLE (NCT02510261) studies were analyzed.

Results: Across the three studies, 224 patients received patisiran for a mean (range) of 34.0 (0.7 – 59.3) months, with a cumulative 633.9 patient-years of exposure; 104 (46.4%) patients received patisiran for ≥3 years and 35 (15.6%) patients received patisiran for ≥4 years. In this cohort, 149 (66.5%) had medical histories of cardiac disorders per MedDRA System Organ Class (SOC), consistent with a mixed phenotype. A total of 222 (99.1%) patients experienced at least one adverse event (AE) and 116 (51.8%) patients experienced at least one serious AE. AEs considered to be related to patisiran occurring in >5% of patients included infusion-related reactions (IRRs) (23.7%) and diarrhea (6.3%). IRRs were mild or moderate in severity and resolved over time, with 97.3% and 2.7% of patients experiencing mild or moderate IRRs by maximum severity.

Conclusions: Patisiran continues to demonstrate a positive benefit: risk profile in patients with hATTR amyloidosis with polyneuropathy.

Keywords: hATTR, patisiran, safety
FACTORS ASSOCIATED WITH INCREASES IN SERUM TRANSTHYRETIN LEVELS AFTER TAFAMIDIS IN TRANSTHYRETIN CARDIOMYOPATHY AND ASSOCIATION WITH CLINICAL OUTCOMES

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1 Columbia University Irving Medical Center

Background: Administration of transthyretin (TTR) stabilizers, particularly AG10, has been shown to increase TTR levels. The clinical and demographic factors associated with these changes are not well defined. Additionally, whether these changes are associated with clinical outcomes are unknown. Additionally, with clinical outcomes are unknown.

Objectives: To evaluate associated factors and major adverse cardiovascular events (MACE) with serum TTR changes on tafamidis.

Methods: Serum TTR was measured before and after tafamidis in 74 subjects with ATTR-CA (77.8 ± 5.9 years, 81.0% male): 59 wild-type (ATTRwt) and 15 hereditary (ATTRv) mutation (13 Val122Ile, 2 Thr60Ala). Absolute changes from baseline were measured and stratified into terciles. Major Adverse Cardiovascular Event (MACE) was defined as unplanned cardiovascular hospitalization or death.

Results: Serum TTR increased by 9.01 ± 6.56 mg/dL (p<0.001), 49.0% ± 46.1% (p<0.001) and was associated with baseline levels (r= -0.48, p=0.004). TTR levels increased in females more than males (12.67 vs. 8.15 mg/dL, p=0.028); in African Americans more than Whites (12.71 vs. 8.06 mg/dL, p=0.034), and in ATTRv more than ATTRwt (12.18 vs. 8.20 mg/dL, p=0.063). Median follow-up on tafamidis was 396 days (range 110-465 days). 13 subjects (17.6%) experienced a MACE: 4 for decompensated heart failure, 3 for complete heart block, 3 for Atrial Fibrillation w/rapid ventricular response, 2 for acute cerebrovascular event and 1 with a fatal out of hospital cardiac arrest. TTR increased by 6.17 mg/dL in subjects with MACE vs. 9.61 mg/dL without MACE (p=0.093). Those with the greatest increase in TTR levels had lower event rates than those in lower terciles (see table).

Conclusion: Increases in serum TTR on tafamidis are associated with baseline TTR levels, gender and race. Preliminary analyses suggest that absolute increases in TTR may be associated with a lower risk for MACE, suggesting its potential utility as a biomarker.
Table 1: Baseline Clinical Characteristics

<table>
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<th>2nd (n=24)</th>
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<td><strong>Demographics</strong></td>
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<tr>
<td>Age (years)</td>
<td>79.4 [77-82]</td>
<td>78.7 [75-82]</td>
<td>75.4 [71-80]</td>
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<td>Male (%)</td>
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<td>87.5</td>
<td>68.0</td>
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<td>Race</td>
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<tr>
<td>White</td>
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<td>21 (88)</td>
<td>16 (64)</td>
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<td>3 (12)</td>
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<td>TTR Genotype (n, %)</td>
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<td>10 (40)</td>
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<td>11 (46)</td>
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<td>3 (12)</td>
<td>4 (17)</td>
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<td>Gilmore Stage (n, %)</td>
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<td><strong>Laboratory Data</strong></td>
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<tr>
<td>Serum TTR (mg/dL)</td>
<td>25.0 [20.5-28.3]</td>
<td>21.7 [17.6-25.1]</td>
<td>19.6 [14.5-22.2]</td>
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</tr>
<tr>
<td>NT-proBNP (pg/mL)</td>
<td>2426.4 [864.5-2941]</td>
<td>2936.6 [1816.5-3955]</td>
<td>2628.4 [887.3-3563.8]</td>
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<tr>
<td>Troponin T (ng/mL)</td>
<td>0.04 [0.02-0.04]</td>
<td>0.04 [0.02-0.05]</td>
<td>0.04 [0.03-0.04]</td>
<td>0.783</td>
</tr>
<tr>
<td>eGFR (mL/min/1.73 m²)</td>
<td>59.28 [50.73-66.91]</td>
<td>64.80 [43.00-83.62]</td>
<td>53.54 [39.75-62.05]</td>
<td>0.095</td>
</tr>
<tr>
<td>Absolute serum TTR change (mg/dL)</td>
<td>2.13 [0.70-4.50]</td>
<td>8.94 [7.08-10.28]</td>
<td>15.95 [12.20-18.90]</td>
<td></td>
</tr>
<tr>
<td>Percent change in serum TTR (%)</td>
<td>10.50 [3.48-19.52]</td>
<td>46.50 [34.49-51.09]</td>
<td>90.88 [58.69-111.17]</td>
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</tr>
<tr>
<td><strong>Outcomes</strong></td>
<td></td>
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<tr>
<td>Number of MACE Events</td>
<td>6</td>
<td>6</td>
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</table>

Values are reported as Mean [IQR]
TOLCAPONE LEVELS AND TTR STABILIZATION IN CEREBROSPINAL FLUID OF PATIENTS WITH LEPTOMENINGEAL AMYLOID TTR MUTATIONS

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Background: Familial transthyretin amyloidosis (hATTR) results from misaggregation of variant transthyretin (vTTR) produced by the liver, predominantly affecting the heart, peripheral and autonomic nerves. Approximately 12 TTR mutations induce leptomeningeal amyloidosis (LMA) derived from choroid plexus TTR. None of the TTR stabilizers or gene silencers appear to cross the blood brain barrier to potentially treat LMA. Tolcapone (CRX-1008), a Parkinson’s Disease treatment designed to cross the blood brain barrier, stabilizes tetrameric TTR in the sera of patients with hATTR. If capable of stabilizing brain derived TTR, CRX-1008 could represent a first treatment for LMA.

Objectives: We administered CRX-1008 for 28 days to 10 patients with vTTR conferring LMA to determine the degree of cerebrospinal fluid (CSF) drug penetration and to compare TTR stabilization in the sera and CSF.

Methods: 10 patients with vTTR conferring LMA received 2 weeks of CRX-1008 100 mg TID followed by 2 weeks of 200 mg TID. Patients returned 2 weeks after completing drug dosing (day 42) for a safety assessment. Liver function tests (LFT), serum creatinine, and thyroid functions were measured at screening, day 0, 14, 28, and 42. Neurologic impairment scores (NIS) were assigned day 0 and 28. Neuropsychological testing was conducted before and after 28 days CRX-1008 administration. Sera and CSF for drug and metabolite levels, as well as TTR tetramer stability testing before and after urea denaturation, were collected at days 0 and 28.

Results: Patients averaged 39.2 years (30-59, range), were 70% male, with a NIS 5.6 points (0-21, range), PND <1 (0-2, range), NYHA class I (0-2); 40% had congophilic biopsies. TTR genotypes included Y114C (4), F64S (2), Y89H (2), T49P (1), A18G (1). All but one patient had >93% drug compliance by pill counts. Alkaline phosphatase, AST, and ALT declined an average 3.0, 4.4, and 7.2 U/L (-49 to 14, range), respectively. eGFR diminished a mean 5.8% (-23.4 to 13.3%); TSH fluctuated a median 1.0%. Two SAEs (seizure, acute hydrocephalus) occurred unrelated to study drug. A median 2.7 AE/patient were reported (0-7, range) potentially related to study drug in 2 patients. Serum and CSF drug levels and TTR tetramer stability data are under analysis.

Conclusions: CRX-1008 up to 200 mg three times daily for 28 days was well tolerated by clinical and biochemical measures in patients with LMA-related vTTR. Results of serum and CSF CRX-1008 drug level determinations, and TTR tetramer stabilization assays before and after 100 mg and 200 mg TID dosing, will be presented.
FEASIBILITY OF THE COMBINATION OF TAFAMIDIS AND MRNA INTERFERING THERAPY IN PATIENTS WITH TRANSTHYRETIN (ATTR) AMYLOIDOSIS

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Introduction: ATTR amyloidosis is usually characterized by peripheral and autonomic neuropathy as well as restrictive cardiomyopathy. TTR-derived amyloid may result from a mutation, of which >80 have been described. In addition, amyloid cardiomyopathy may occur in the elderly in the absence of a mutation, termed wild-type TTR. Both hereditary and wild-type TTR cardiac amyloidosis are characterized by progressive restrictive cardiomyopathy. New treatment paradigms have emerged with the use of mRNA interfering (mRNAi) therapies as well as the TTR protein stabilizer, tafamidis.

Objectives: To describe real world use of tafamidis with concurrent mRNAi therapy in patients with ATTR amyloidosis.

Methods: We performed a retrospective chart review of patients with ATTR amyloidosis with cardiac involvement who were evaluated at the Ohio State University. Patients’ gender, date of birth, date of diagnosis, use of tafamidis, concurrent use of mRNAi therapy and comorbid conditions were evaluated.

Results: We identified 30 patients with ATTR amyloidosis. Median age was 76 yrs. (57-95). 27 pts (90%) were male and 13 (43%) were African American. 20% of pts had concurrent neuropathic symptoms. 5 pts had concurrent MGUS. On review of medical history, 17 pts (56.7%) were found to have carpal tunnel in the past. 13 pts (43.3%) were found to have mutations in the TTR gene with Val142Ile being the most common (84.6%). All 30 pts were prescribed tafamidis and median dose was 80 mg. 10 pts (33%) were also prescribed mRNAi therapy; with 90% of those getting patisaran. On a median follow up of 2 months, all 10 pts were able to tolerate both therapies. 7 out 10 pts developed fatigue. 5 developed shortness of breath but no change in diuretic regimen was required. 1 (10%) had to decrease dose of tafamidis (dose reduction from 40 mg to 20 mg daily). None of the pts had to stop therapy due to side effects.

Conclusions: ATTR amyloidosis is an underdiagnosed cause of progressive cardiomyopathy with both hereditary and wild type phenotypes. Clinical trials did not allow for pts to be on both mRNAi therapies and protein stabilizers. In our experience, the combination of mRNAi therapies and tafamidis is tolerable, feasible and safe to use.
LONG–TERM EXPERIENCE WITH INOTERSEN IN PATIENTS WITH TRANSTHYRETIN AMYLOID CARDIOMYOPATHY

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Introduction: Transthyretin amyloid (ATTR) cardiomyopathy is a fatal disease which results in congestive heart failure and cardiac arrhythmias. Stabilizer therapy with Tafamidis (Vyndaqel™) was recently shown to decrease all-cause mortality and cardiovascular hospitalizations and slow the decline in walking distance and quality of life compared to placebo. Gene silencers have demonstrated effectiveness in patients with hereditary ATTR amyloidosis with polyneuropathy to reduce and/or improve neuropathy and while many hereditary patients had concomitant cardiomyopathy, cardiac parameters were not a primary endpoint. A compassionate use trial was started in August of 2014 at Indiana University to test the safety and tolerability of the antisense oligonucleotide, inotersen in patients with hereditary or wild-type transthyretin cardiomyopathy without significant neuropathy. We have previously reported our findings for patients on inotersen for up to 3 years.1,2 We now report our long term experience with inotersen for patients on treatment for up to 5 years.

Methods: 6 minute walk (6MWT) and echocardiograms with LV longitudinal systolic strain are performed every 6 months. MRIs are performed every year if not contraindicated. Safety monitoring includes weekly platelet counts and periodic assessment of serum creatinine and urine protein.

Results: As of August 2019, 39 people have enrolled in the study. Of the 28 patients currently actively enrolled, 14 active patients have been on therapy with inotersen for greater than 3 years and 14 less than 3 years. 11 patients have voluntarily withdrawn from the study and one patient suffered a non-drug related death while taking inotersen. Despite many of the patients requiring antiplatelet or anticoagulant therapy, no serious adverse events have been noted due to thrombocytopenia with routine monitoring. There have been no cases of glomerulonephritis identified. Analysis for patients who have completed 2 or 3 years showed a decline in the left ventricular mass on MRI, decrease in interventricular septal thickness on echocardiogram and decrease in BNP as well as an improvement in 6 minute walk and global longitudinal systolic strain. The hereditary patients, who on average were 10 years younger than patients with ATTRWT, appeared to show the greatest benefit from therapy with inotersen. No new safety signals have emerged and patients appear to show continued stabilization or improvement with long term therapy.

Conclusion: Inotersen is a safe and effective therapy for Transthyretin amyloid cardiomyopathy. This noncontrolled study suggests that gene silencer therapy with inotersen has the potential to reverse amyloid cardiomyopathy and improve long term outcomes. The anticipated phase 3 trials with gene silencer therapy in amyloid cardiomyopathy will hopefully verify our findings and have the potential to significantly alter the natural history of transthyretin amyloid cardiomyopathy.

References:
**PW023**

**Efficacy and Safety of Midodrine in Patients with Amyloidosis and Orthostatic Hypotension: A Case Series.**

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**Background:** Orthostatic hypotension is a frequent manifestation of autonomic dysfunction in patients with amyloidosis. Midodrine is an alpha-1 agonist that increases vascular tone and elevates blood pressure. The efficacy and safety of midodrine in patients with amyloidosis is not well described.

**Objectives:** Describe the efficacy and side effects of midodrine in patients with orthostatic hypotension and amyloidosis.

**Methods:** This is a retrospective analysis of a series of patients with a diagnosis of light chains amyloidosis (AL) or transthyretin amyloidosis (ATTR) treated with midodrine for symptomatic orthostatic hypotension. Orthostatic hypotension was defined as a >15 mmHg decrease in the systolic blood pressure from supine to standing. The delta (standing - supine) systolic blood pressure (ΔSBP), delta diastolic blood pressure (ΔDBP) and delta heart rate (ΔDHR) were calculated for every patient pre initiation of midodrine and then during follow-up on midodrine. These delta measurements were compared respectively and then between AL and ATTR. Side effects such as hypertension, urinary retention, paresthesia and pruritus were documented.

**Results:** Five patients with AL and five with ATTR were included in the analysis. In patients with AL amyloidosis, the mean ΔSBP from supine to standing pre midodrine was -26 mmHg and it improved to -5.4 mmHg on midodrine (20.6 mmHg difference). In patients with ATTR amyloidosis the mean ΔSBP from supine to standing pre midodrine was -20.4 mmHg and it improved to -7.8 mmHg on midodrine (12.6 mmHg difference). As for side effects, one patient reported pruritus, another constipation and none developed supine hypertension. All patients demonstrated symptomatic improvement on midodrine.

**Conclusions:** Midodrine improves symptoms and orthostatic vital signs in in patients with orthostatic hypotension and amyloidosis without significant side effects.

**Table 1.** Mean ΔSBP, ΔHR and ΔDBP in patients with amyloidosis and orthostatic hypotension treated with midodrine.

<table>
<thead>
<tr>
<th></th>
<th>AL Amyloidosis N = 5</th>
<th>ATTR Amyloidosis N = 5</th>
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<tbody>
<tr>
<td></td>
<td>Pre-midodrine</td>
<td>On-midodrine</td>
</tr>
<tr>
<td>Mean Δ SBP, mmHg</td>
<td>-26.0 ± 7.41</td>
<td>-5.40 ± 6.91</td>
</tr>
<tr>
<td>Mean Δ HR, bpm</td>
<td>12.40 ± 17.31</td>
<td>12.40 ± 10.94</td>
</tr>
<tr>
<td>Mean Δ DBP, mmHg</td>
<td>-11.40 ± 8.62</td>
<td>2.4 ± 14.60</td>
</tr>
</tbody>
</table>

AL= Light Chains Amyloidosis, ATTR= Transthyretin Amyloidosis, Δ SBP= Delta Systolic Blood Pressure, Δ HR= Delta Heart Rate, Δ DBP= Delta Diastolic Blood Pressure

**Key Words:** amyloidosis, midodrine, orthostatic hypotension
AG10 IN PATIENTS WITH TRANSTHYRETIN AMYLOID CARDIOMYOPATHY (ATTR-CM): FURTHER ANALYSES OF 15-MONTH FOLLOw-UP OF PHASE 2 OPEN LABEL EXTENSION


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Background: AG10 is a small molecule TTR stabilizer under investigation for the treatment of ATTR. AG10 was well tolerated, demonstrated near-complete stabilization of TTR, and increased serum TTR levels to normal in all treated subjects with ATTR cardiomyopathy in a randomized, double-blind, 28-day Phase 2 study (Judge, DP, et al., 2019; JACC 74(3):285-295). The current study is an open label extension (OLE) of that trial.

Methods: This multi-center OLE study enrolled eligible subjects who completed the randomized Phase 2 study. All subjects participating in the OLE (n=47) were administered 800 mg AG10 orally twice daily. The primary endpoint is an assessment of long-term safety and tolerability of AG10. The key secondary endpoints include AG10 pharmacokinetics, pharmacodynamic assessments of TTR stabilization, and serum TTR (prealbumin) concentration. Cardiac biomarkers and echocardiographic parameters were measured in exploratory analyses.

Results: As of 8/31/2019, after a median of 65 weeks since initiation of the Phase 2 study, 41 participants continued on open label AG10. AG10 was generally well tolerated with a pattern of adverse events consistent with underlying disease severity, concurrent illnesses, and age of participants. Rates of all-cause mortality (including either death or cardiac transplantation) and cardiovascular hospitalization were 8.5% and 25.5%, respectively. Near-complete TTR stabilization was observed using established ex vivo assays. Mean serum TTR levels were elevated by 39% and 56% in wild-type and TTR mutation-carriers, respectively, after 180 days of the OLE study. Cardiac biomarkers and echocardiographic parameters were stable throughout trial duration.

Conclusions: This interim analysis of the OLE study data supports continued evaluation of AG10 in an ongoing Phase 3 study (NCT03860935).
DIFFERENTIAL EX VIVO STABILIZATION OF TRANSTHYRETIN BY AG10 AND TAFAMIDIS IN SAMPLES FROM PATIENTS WITH MODERATE OR SEVERELY DESTABILIZING MUTATIONS

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1 Eidos Therapeutics, Inc.

Introduction: Transthyretin (TTR) amyloidosis is a progressive, fatal disease caused by destabilizing TTR mutations and age-related factors. Dissociation of tetrameric TTR initiates protein misfolding, the mechanism of disease. Destabilizing TTR mutations accelerate the process. The more destabilizing the mutation, the more severe the clinical phenotype. TTR stabilizers including diflunisal and tafamidis have demonstrated clinical benefit by stabilizing tetrameric TTR and slowing disease progression. AG10 is a novel stabilizer under development for the treatment of TTR amyloidosis.

Hypothesis: Pathogenic TTR variants with varied intrinsic instability display differential stabilization by AG10 or tafamidis.

Methods: Two in vitro assays were used to assess TTR stabilization: fluorescent probe exclusion (FPE) to measure binding site occupancy in patient serum, and immune blot quantitation of tetrameric TTR following accelerated dissociation at pH 3.8 for 72h. Individual patient samples representing a spectrum of intrinsic instability and clinical phenotypes (V122I, T60A, A97S) were assayed following addition of AG10 or tafamidis at their respective therapeutic plasma concentrations. Commercially available tafamidis was used in this study.

Results: At therapeutic target trough concentration, AG10 bound serum TTR to a greater extent than either peak or trough levels of tafamidis (representative FPE assays, Figure A). Immune blot assays showed that adding AG10 resulted in greater and more durable TTR tetramer stabilization than adding tafamidis in all individual patient plasma samples tested (Figure B).

Conclusions: At therapeutic concentrations, AG10 more completely stabilizes variant TTR samples representing a range of destabilizing mutations and clinical phenotypes than does tafamidis. AG10 has the potential to be clinically efficacious in patients with a variety of genotypes associated with both TTR cardiomyopathy and polyneuropathy.

Figure A: TTR binding site occupancy by AG10 of tafamidis (serum FPE assay)

Figure B: TTR stabilization by AG10 or tafamidis added in vitro (Plasma immune blot)
ATTRibute-CM: A RANDOMIZED, DOUBLE-BLIND, PLACEBO-CONTROLLED, MULTI-CENTER, GLOBAL PHASE 3 STUDY OF AG10 IN PATIENTS WITH TRANSTHYRETIN AMYLOID CARDIOMYOPATHY (ATTR-CM)


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11 Medical University of South Carolina.

Background: Transthyretin (TTR) amyloidosis (ATTR) is an under-diagnosed cause of heart failure driven by TTR destabilization due to pathogenic mutations and/or aging. AG10 is a small molecule TTR stabilizer under development for the treatment of ATTR. In a randomized, double-blind Phase 2 study in patients with symptomatic ATTR cardiomyopathy (ATTR-CM), AG10 was well tolerated, demonstrated near-complete stabilization of TTR, and increased serum TTR levels to normal in all treated subjects (Judge, DP, et al., 2019; JACC 74(3):285-295). Serum TTR levels have been reported to be predictive of survival in ATTR-CM, and treatment with TTR stabilizers increases serum TTR levels to an extent that is correlated with their ability to increase stability of tetrameric TTR ex vivo.

Methods: ATTRibute-CM is a Phase 3 study designed to enroll approximately 510 patients with symptomatic ATTR-CM, including those with either wild-type or mutant TTR, with New York Heart Association Class I-III symptoms (NCT03860935). Participants will be randomized 2:1 to AG10 800 mg or placebo twice daily and followed for 30 months. After 12 months, change in six-minute walk distance (6MWD), the primary endpoint, will be compared between treatment and placebo groups using mixed model repeated measures as an early assessment of functional clinical benefit. Change in Kansas City Cardiomyopathy Questionnaire (KCCQ) quality-of-life scale will be a key secondary endpoint at this stage. At 30 months, the primary analysis will be the hierarchical combination of all-cause mortality and frequency of cardiovascular-related hospitalizations compared between AG10 and placebo using the Finkelstein-Schoenfeld method.

Conclusions: ATTRibute-CM is a prospective, randomized, double-blind, placebo-controlled, global Phase 3 clinical trial in patients with ATTR-CM designed to evaluate AG10’s ability to slow or halt progression of ATTR-CM as measured by function (6MWT), quality of life (KCCQ), and a hierarchical combination of all-cause mortality and frequency of cardiovascular-related hospitalizations.
PW027

DIFFERENTIAL TRANSTHYRETIN BINDING, KINETIC STABILITY AND ADDITIVE EX VIVO STABILIZATION BY AG10 COMPARED TO TAFAMIDIS

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1 Eidos Therapeutics, Inc.

Introduction: Transthyretin (TTR) amyloidosis is a progressive, fatal disease. Dissociation of tetrameric TTR is the triggering event in the pathogenic mechanism; destabilizing TTR mutations accelerate the process. TTR stabilizers including diflunisal and tafamidis have demonstrated clinical benefit by slowing disease progression. AG10, a novel TTR stabilizer which mimics the putative mechanism of the disease-protective variant T119M, is under development for the treatment of TTR amyloidosis.

Hypothesis: By mimicking the stabilizing characteristics of T119M, AG10 displays differential TTR binding, kinetic stability, and a higher degree of stabilization compared to other TTR stabilizers.

Methods: We characterized the relative TTR binding affinity of AG10 and tafamidis. Commercially available tafamidis was used in this study. Thermodynamic stability (Kd) of TTR interaction was determined by microscale thermophoresis (MST). Kinetic stability was assessed by surface plasmon resonance (SPR). The ability of each stabilizer to prevent accelerated tetramer dissociation alone or in combination was measured by immune blots.

Results: The affinity of AG10 for purified TTR, as measured by MST, is greater than that of tafamidis (TABLE). Kinetic stability by SPR reveals over 4X longer residence time for AG10 bound to TTR as compared to tafamidis. When tested at therapeutically attained plasma concentrations, tafamidis does not completely stabilize tetrameric TTR. Addition of AG10 at its therapeutic target plasma concentration to plasma samples containing therapeutic concentrations of tafamidis results in complete stabilization of TTR (FIGURE).

Conclusions: The extended residence time of AG10 compared to tafamidis results in improved TTR binding site occupancy and stabilization. AG10 is a potent stabilizer of TTR as assessed by TTR binding affinity (MST) and kinetic stability (SPR) and demonstrated complete stabilization of TTR in plasma samples with or without therapeutic concentrations of tafamidis.

Table: TTR Binding Characteristics

<table>
<thead>
<tr>
<th>Parameter</th>
<th>AG10</th>
<th>Tafamidis</th>
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<tbody>
<tr>
<td>Thermophoresis (Kd (nM))</td>
<td>32 ± 11</td>
<td>110 ± 20</td>
</tr>
<tr>
<td>Residence Time (s)</td>
<td>50 ± 4</td>
<td>12 ± 3</td>
</tr>
<tr>
<td>Kd (nM)</td>
<td>16 ± 2</td>
<td>120 ± 30</td>
</tr>
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</table>

Figure: Dose responsive effect of TTR stabilization by tafamidis in human plasma: Additive of AG10
RESULTS FROM THE TAFAMIDIS ENHANCED SURVEILLANCE FOR PREGNANCY OUTCOMES (TESPO) PROGRAM

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Introduction: Since 2011, the Tafamidis Enhanced Surveillance for Pregnancy Outcomes (TESPO) program has followed pregnancy outcomes in women exposed to tafamidis. Given the information from animal studies which show that in rabbits dosed with tafamidis during organogenesis, adverse effects on fetal development (mortality, reduced body weight and malformations) were seen at exposures 9-times the human exposure at maximum recommended human dose (MRHD), with fetal skeleton variations seen at exposures equivalent to exposures at the MRHD. In offspring of pregnant rats administered ~2-times the MRHD during gestation and lactation, postnatal mortality, delayed male sexual maturation and impaired memory and learning were seen. The tafamidis prescribing information advises of the potential fetal risk of drug exposure; to date, the limited available data from pregnant women receiving tafamidis 20 mg per day for the treatment of transthyretin (ATTR) amyloidosis have not identified any drug-associated risks of major birth defects, miscarriage, or adverse maternal/fetal outcomes.

Objective: The aim of TESPO is to evaluate outcomes of pregnancy (including 1-year follow up of live births), in patients with ATTR amyloidosis exposed to tafamidis during, or within 1 month prior to, pregnancy.

Methods: Pregnancy exposure information is collected through multiple sources including the Transthyretin-Associated Amyloidosis Outcomes Survey (THAOS) registry, Pfizer-sponsored tafamidis interventional trials, reports from patients and health care professionals (HCP) via the Medical Information Division, sales representatives, and employees. Information on how to contact the sponsor to report pregnancy exposure is provided through safety training of clinical trial sites and in country-specific product labeling.

Results: Of the 20 reported pregnancies (22 fetuses, 2 twin pregnancies), 17 pregnancies resulted in 18 live births (1 set of twins), 1 twin pregnancy led to abortion due to intensive bleeding, 1 was a voluntary termination and 1 provided no information. Of the 18 newborns born to mothers who were directly (7), or indirectly through their partner (11, includes the set of twins) exposed to tafamidis, 15 were full term birth and 3 were preterm (includes the set of twins). Of the 18 live births, 8 had 12-month post-natal follow-ups. All 8 infants survived the first year of life and met age-appropriate development milestones. No infant had a congenital malformation.

Conclusions: Understanding and following the outcome of pregnancies in women exposed to tafamidis will provide safety data for HCPs to use in treating and counseling patients with ATTR amyloidosis who are (or wish to become) pregnant. As of September 2019, the clinical data gathered provide no clear evidence of a safety risk associated with tafamidis exposure during pregnancy, via paternal or maternal exposure. The TESPO program will continue to collect additional safety data and survey pregnancy outcome and newborns to further build upon current safety information.
DEVELOPMENT OF NTLA-2001, A CRISPR/CAS9 GENOME EDITING THERAPEUTIC FOR THE TREATMENT OF ATTR

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1 Intellia Therapeutics.

Introduction: Transthyretin amyloidosis (ATTR) is a progressive disease caused by accumulation of amyloid deposits of misfolded transthyretin (TTR) protein in multiple tissues including the heart, nerves and gastrointestinal tract. Reduction of TTR monomer via stabilization of circulating tetramer and silencing of TTR gene expression in hepatocytes of ATTR patients have emerged as successful therapeutic strategies for chronically-administered medicines. As such, specific disruption (or knockout) of the TTR gene in hepatocytes using the CRISPR/Cas9 gene editing system is a potentially attractive next-generation treatment for ATTR, which may durably reduce the expression of TTR without the need for chronic therapy.

Objectives: To develop NTLA-2001, a lipid nanoparticle (LNP) formulated CRISPR/Cas9 genome editing therapeutic targeting the human TTR gene for the treatment for ATTR.

Methods: We examined the ability of CRISPR/Cas9-LNP to durably reduce the expression of serum TTR in multiple rodent and non-human primate (NHP) preclinical model systems using NTLA-2001 components and/or species-specific surrogates.

Results: A single dose of LNP containing CRISPR/Cas9 and TTR-specific guide (TTR LNP) in mice resulted in >97% reduction in circulating serum TTR protein that was sustained for at least 12 months. Additionally, in a humanized mouse model of hATTR expressing the V30M mutant form of the human TTR protein, rescue of TTR deposition in multiple tissues after a single dose of TTR LNP was demonstrated. NHPs receiving a single dose of TTR LNP achieved a therapeutically meaningful serum TTR reduction (>95%) that correlated with robust whole liver editing. Different effective doses were well tolerated. In NHP, components of TTR LNP were cleared from plasma and liver with half-lives of 23 hours and 17 hours, respectively.

Conclusions: One-time gene disruption of the TTR gene by CRISPR/Cas9-LNP resulted in durable and robust decrease in serum TTR protein levels in vivo. Favorable tolerability of the delivery system was aided by transient exposure to the CRISPR/Cas9 and LNP components. These findings show pharmacologic activity of TTR LNP and support further development of NTLA-2001 for the treatment for patients with ATTR polyneuropathy and/or cardiomyopathy.

Keywords: CRISPR/Cas9, Genome editing
LONG-TERM EFFECTS OF TAFAMIDIS IN ATTR KIDNEY DISEASE: MORE THAN THE INFLUENCE ON ALBUMINURIA

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2 Unidade Corino de Andrade, Centro Hospitalar Universitário do Porto, Porto, Portugal.
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5 Department of Neurophysiology, Centro Hospitalar Universitário do Porto, Porto, Portugal.

Introduction: Hereditary transthyretin (ATTRm) amyloidosis due to Val30Met mutation in the TTR gene is associated with nephropathy, which is characterized by a wide range level of albuminuria and progressive chronic kidney disease (CKD). Higher grades of albuminuria incrementally augment the loss of renal function. Although tafamidis has been associated to significant decrease of albuminuria in ATTRm, the prediction of renal function long-term outcomes remains unclear.

Objective: The aim of this retrospective, one center, study was to investigate the long-term influence of tafamidis on estimated glomerular filtration rate (eGFR) in a real-life setting.

Methods: We evaluate, consecutively, ATTRm Val30Met patients from July 2012 to March 2019. Our cohort was comprised by anti-amyloid treatment-naïve patients that accomplished at least 60 months of tafamidis therapy. All were treated in a post-marketing practice, 20 mg QD without interruption. Eligibility: neuropathy stage I, 18 to 85 years of age, eGFR ≥ 45 mL/min/1.73 m2 (based on cystatin C) and any grade of albuminuria. Exclusion: malignancies, diabetes and concurrent etiologies for nephropathy. Outcomes included eGFR during the 5-year treatment, the annual change in eGFR and changes in kidney function considering the level of albuminuria. Urinary albumin-to-creatinine ratio (ACR) >30 mg/g was considered pathological.

Results: There were 366 patients, biopsy-confirmed ATTR amyloidosis, treated with tafamidis. Our 60 months cohort: 122 patients (56 males, 66 females), mean age of 41 ± 13 years, BMI of 24.5 ± 3.7 kg/m2 at baseline; 28 had pathological ACR; ACR > 300 mg/g was found only in females. At the end of follow-up, when pathological ACR was present at baseline, we verified a significant decrease in the median ACR level: 321.7 (34.3 – 4761.4) compared to 24.4 (6.6 – 1066) mg/g. The eGFR (mean, SD) significantly decreased from the baseline to the end of follow-up, irrespectively the non-pathological (131.1 ± 26.4 vs 105.9 ± 23.9 mL/min, P<0.01) or pathological (98.5 ± 32.7 vs 76.5 ± 25.5 mL/min, P<0.01) grade of ACR; the mean annual decline of eGFR was 5.51 vs 4.51 ml/min/year, P=NS. Table I classifies the patients according to KDIGO stages before and after 5 years of therapy. No significant changes in the annual eGFR were detected considering gender and age at baseline and time from the onset of neuropathy and initiation of treatment.

Conclusions: This study demonstrated a long-term benefit provided by tafamidis, concerning reduction of albuminuria and retardation of CKD. Overall, the five-year treatment analysis shows a decline of renal function, irrespectively the degree of albuminuria at baseline. This effect should be taken into account if a switch to a second line treatment is considered. Potential cofactors for amyloid fibrils toxic effect, present in glomerular basement membrane or in mesangium, may be halted by tafamidis therapy.
Table 1 - KDIGO stages according estimated Glomerular Filtration Rate (eGFR) by Larsson and albuminuria level in patients with urinary albumin-to-creatin ratio in normal (Albuminuric) and pathological range (Non-albuminuric).

<table>
<thead>
<tr>
<th>N (%)</th>
<th>Albuminuric patients</th>
<th>Non-albuminuric patients</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patients</td>
<td>28 (23%)</td>
<td>94 (77%)</td>
</tr>
<tr>
<td>Female</td>
<td>19 (68%)</td>
<td>47 (50%)</td>
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<td>3b</td>
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The authors thank Inês Cardoso for the technical assistance.

Keywords: ATTR, chronic kidney disease, tafamidis
Novel therapies in ATTR amyloidosis
A LEAD CANDIDATE FOR TACKLING CENTRAL NERVOUS SYSTEM SYMPTOMS OF TRANSTHYRETIN AMYLOIDOSIS: IN VITRO CHARACTERIZATION OF PD AND PK FEATURES

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Introduction: Patients living with hereditary ATTR with polyneuropathy (hATTR-PN) are increasingly confronted with cerebral complications due to amyloid deposition in the central nervous system (CNS). A mounting number of reports suggests that at least 30% of patients develop CNS symptoms, on average 14.6 years after disease onset [¹]. Stabilization of transthyretin (TTR) variants responsible for amyloid deposition in the CNS represents opportunity, not only from the scientific perspective of treating a protein misfolding disease in the brain, but also from the viewpoint of bringing clinical benefit to hATTR-PN patients. We recently reported results on a series of TTR stabilizers displaying potent in vitro stabilization of three TTR variants, and capable of dose-dependently stabilize the protein in human cerebrospinal fluid (CSF) ex vivo [²]. Here, we subjected one of our advanced TTR stabilizers, BSIMpc02, to a set of in vitro assays with the goal of further characterizing its pharmacodynamics (PD) and pharmacokinetics (PK) features.

Methods: To assess the extent of TTR engagement by BSIMpc02, isothermal titration calorimetry (ITC) experiments were performed on a Malvern high precision VP-ITC titration calorimetry system. Direct titrations were performed using the classic setup, with ligand solutions in the syringe and protein in the calorimetric cell. The permeability of BSIMpc02 was assessed using a Caco-2 cell monolayer, measured in both directions (A-B and B-A). The assays were run for 1h, in duplicate, and the amount of compound present in each compartment was quantified by LC-MS/MS. The plasma protein binding (PPB) profile for BSIMpc02 was studied by equilibrium dialysis, using a semi-permeable membrane that separates two compartments containing protein and buffer. The compound was mixed with human plasma and quantified in each compartment by LC-MS/MS.

Results: The association constants of BSIMpc02 to the two TTR binding sites were obtained via non-linear regression of the ITC experimental data. The dissociation constants were then calculated: $K_{d1} = 0.86$ nM and $K_{d2} = 25.10$ nM – implying high ligand affinity and modest negative cooperativity. The Caco-2 A-B and B-A permeabilities of BSIMpc02 have been determined in $17.1 \times 10^{-6}$ and $3.2 \times 10^{-6}$ cm/s, respectively. Based on the BA-AB ratio, we infer that interactions with efflux transporters, namely with P-glycoprotein 1 (P-gp), are unlikely to occur. Finally, a mean PPB of 84% obtained for BSIMpc02, which suggests a moderate free fraction available for distribution to the CSF.

Conclusions: BSIMpc02 displays PD and PK features that distinguish it from current hATTR-PN therapies in the market, representing a promising drug candidate for the treatment of hATTR-PN patient groups – be they untreated, treated or liver transplanted cases, who currently lack an therapeutic solution capable of stalling disease progression both in the peripheral nervous system and the central nervous system.


Keywords: Transthyretin amyloidosis, CNS co-morbidities, Novel therapies.
A PHASE 1/2 STUDY TO ASSESS SAFETY AND DOSE OF Ixzomib IN COMBINATION WITH CYCLOPHOSPHAMIDE AND DEXAMETHASONE IN NEWLY DIAGNOSED PATIENTS WITH LIGHT CHAIN (AL) AMYLOIDOSIS


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Introduction: AL amyloidosis is an incurable clonal plasma cell disorder characterized by tissue deposits of immunoglobulin light chain fragments leading to organ dysfunction and death. Standard treatment for newly diagnosed patients (pts) has traditionally included oral melphalan + dexamethasone as well as high-dose melphalan + ASCT. Although the combination of bortezomib, cyclophosphamide and dexamethasone (CyBorD) has activity, better tolerated treatment approaches are needed. Here we report preliminary results of a Phase 1/2, open-label, multi-institution study of ixazomib (I) in combination with cyclophosphamide (Cy) and dexamethasone (D) in newly diagnosed AL amyloidosis.

Methods: Eligible pts are ≥18 years with newly diagnosed, untreated biopsy-proven AL amyloidosis according to standard criteria. A total of up to 30 pts will be enrolled, with up to 18 in the dose escalation arm (phase 1) and 12 in the maximum tolerated dose (MTD) expansion arm (phase 2) according to a classical 3+3 design. Four dose levels were evaluated in phase 1. I and Cy are given orally (PO) on days 1, 8, 15, and D 20mg PO on days 1, 8, 15, 22 of each 28-day cycle. Treatment continues for a total of 6 cycles or until disease progression, significant toxicity or withdrawal. The primary study objective in phase 1 is to establish the MTD and in phase 2 is to determine hematologic/organ response rate.

Results: As of September 2019, 19 pts have been enrolled; 16 in phase 1 and 3 in phase 2. The MTD was established at dose level 3 (I 4mg and Cy 500mg). Median age is 66 years (range 46-79), 12 (63%) are male. Light chain isotype is lambda in 15 (79%). Seven pts (37%) have cardiac, 5 (26%) renal, 5 (26%) gastrointestinal, 1 (5%) hepatic, 2 (10%) soft tissue involvement, with 21% having multi-organ involvement. Five pts (26%) completed 6 cycles of therapy and 4 (21%) remain on study with a median of 3 cycles completed. Ten pts (53%) have been taken off study prior to completing 6 cycles due to no response in 7 (37%) after a median of 3.5 cycles (2-5), grade 4 hyperbilirubinemia unrelated to study drug in 1 (5%), cardiac decompensation in 1 (5%), and 1 death attributed to advanced disease. Most common drug-related adverse event (AE) (any grade) were edema (19%), fatigue (19%), dizziness/lightheadedness (13%) and lymphopenia (13%). Grade 3/4 AEs were rare with grade 3 lymphopenia, anemia, and hyponatremia occurring in 13%, 6%, and 6% of pts, respectively. Of 19 evaluable pts, 7 (37%) achieved ≥VGPR with the median time to best response 2 cycles (1-5).

Conclusion: The combination of ICyD for pts with newly diagnosed AL amyloidosis is safe and well tolerated. Phase 1 is completed and the recommended phase 2 dose has been established. Deep hematologic responses (≥VGPR) have occurred and time to response appears similar to standard of care induction regimens, ie CyBorD. Phase 2 response data will be updated at the meeting.

Keywords: Ixzomib, Cyclophosphamide, AL Amyloidosis.
PW033

A REAL-LIFE COHORT STUDY OF IMMUNOGLOBULIN LIGHT-CHAIN (AL) AMYLOIDOSIS PATIENTS WITH SEVERE CARDIAC INVOLVEMENT OR ADVANCED DISEASE

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² Amyloidosis Center of Expertise.

Objective: To study the outcome of patients with AL amyloidosis who were ineligible for high dose melphalan (HDM) and autologous stem cell transplantation (ASCT).

Methods: A real-life retrospective observational cohort study of Dutch patients with AL amyloidosis ineligible for HDM and ASCT was performed at the University Medical Center Groningen from January 2001 until April 2017. Primary outcome measure was overall survival (OS). Secondary outcome measures were haematological response (HR), organ responses, and treatment toxicity.

Results: Eighty-four patients were included. Ineligibility was due to NYHA class III/IV (n=58), otherwise advanced disease (n=11), advanced age (n=14), or treatment refusal (n=1). Early death (<3 months) rate was high (44%). Median OS improved from 4 months in period 2001-2009 (n=36) to 8 months in period 2009-2017 (n=48, p=0.02). HR was seen in 29%, and 42% of the patients, respectively. Median OS was 36 months after induction treatment with bortezomib (n=32) and 18 months with IMID (n=16), both higher than median OS (7 months) with other regimens (n=27). Incidence of toxicity was high (51%).

Conclusion: OS improved in this high-risk group over the years after introduction of new treatment modalities. However, early death rate remains high, illustrating the need of more effective treatment.
Background: Matrix metalloproteinases (MMPs) play a role as inflammatory components in amyloid diseases. Elevated tissue MMPs have been found in the heart and kidneys in light chain (AL) amyloidosis. Doxycycline has anti-amyloid properties that have been linked to MMP inhibition but the mode of action in amyloid is unknown. It is unclear how serum MMPs change over the course of AL amyloid therapy and if changes correlate with treatments and organ response.

Objectives: Determine whether there are changes in serum MMP activity during the first year of treatment in AL amyloidosis with doxycycline.

Methods: Twenty-five patients with systemic AL amyloidosis were enrolled on a prospective clinical trial of doxycycline in AL amyloidosis (DUAL trial, NCT01392573). Patients were given 100 mg twice daily of oral doxycycline monohydrate for 1 year in addition to chemotherapy. Organ response was measured using validated criteria. MMP-1, -3, -8, -9 and -10 activity and expression were measured at enrollment, 6 months and end of study. Activity was measured fluorometrically using commercial kits. A statistical mixed model was developed to study changes in activity and expression over time adjusting for disease stage and transplant status.

Results: Median age was 61.3 years with 64% males and 68% AL (lambda) subtype. 2012 stage was 1 in 12%, 2 in 36%, 3 in 24%, and 4 in 28% with 60% cardiac and 72% renal involvement. All patients received induction chemotherapy with cyclophosphamide, bortezomib and dexamethasone (CyBorD). At 1-year, mortality was 20%, organ response was 36% response, 32% stable disease, and 36% progression/death; 60% of patients underwent autologous transplant with 0 day-100 transplant-related mortality. Significant elevations in MMP activity at 12 months compared to baseline were seen with MMP-1: estimate ratio 1.14 (95% CI, 1.05, 1.24), p 0.001, MMP-3: estimate ratio 1.12 (95% CI 1.04, 1.21), p 0.003, and MMP-8: estimate ratio 1.2 (95% CI 1.06, 1.36), p 0.004; these changes were more pronounced with stage 3/4 compared to stage 1/2. Transplant led to reduction in MMP activity across all MMP subtypes but was statistically significant only for MMP-10 activity: estimate ratio 0.67 (95% CI 0.45, 1.00), p 0.05. Patients with organ progression had higher MMP-3 activity at 12 months compared to patients with organ response/stable disease.

Conclusions: Compared to contemporaneous data, this study showed low early mortality and high transplant utilization in newly diagnosed AL amyloid patients treated with doxycycline. No reduction in MMP activity was observed with doxycycline over time, though we did not have a control arm. Serum MMP activity may be useful as a biomarker of organ progression in AL amyloidosis. Though our numbers were small in this exploratory analysis, MMP activity over time varied by stage, organ response, and reduced with transplantation. Serum MMPs should be further explored as biomarkers of organ AL response in larger cohorts.
THE OUTCOME OF AL AMYLOIDOSIS PATIENTS UNDERGOING AUTOLOGOUS HEMATOPOIETIC STEM CELL TRANSPLANTATION

S. E. Yoon¹, S. J. Kim¹, K. K. Kim¹, J. H. Min¹, D. Kim¹, J. Choi¹, M. Park¹, E. Kim¹, J. Y. Choi¹, J. Kim¹, J. E. Lee¹

¹ Samsung Medical Center.

Introduction: The present incidence of amyloidosis in Korea has increased approximately 50 times compared with the 1990s. The development of a therapy to eliminated monoclonality have improved survival outcome in AL amyloidosis. Alkylating agents and steroids are still a potent option. If transplantation is not possible, the proteasome inhibitors (PIs) and immunomodulatory drugs (IMiDs) also contributes to the survival outcome. However, in transplantable patients, Autologous hematopoietic stem cell transplantation (ASCT) is the best option to destroy the amylogenic clone. Thus, based on our registry data, we would report the results of the ASCT outcome in AL amyloidosis.

Methods: We have conducted a registry study for amyloidosis patients since 1990 at Samsung Medical Center. A total of 302 patients were enrolled, with 96 patients enrolled in the 1995-2011 retrospective study of amyloid, and 206 patients enrolled in the prospective study in 2012-2018. Among the patients treated in our Center, 302 patients who were able to evaluate the efficacy of treatment included in this study. We excluded AA amyloidosis and hereditary amyloidosis. And, we also excluded patients without enough data or short follow up.

Results: Of these, 192 (63.6%) of those were aged younger than 65 years, and there were 231 cases of amyloidosis involvement in 2 or more organs. The stage III defined by Mayo 2004 was 131 patients (67.9%), and stage III or IV defined by Mayo 2012 was 141 patients (74.2%). The median follow up duration was 19 months (range 0-154). The median overall survival of total patients was 41 months (95% CI 28.6-53.4). The treatment applied to amyloid patients changed slightly over time and the number of patients who received ASCT gradually increased. None of the 17 patients in 1995-2003 received ASCT. The number of patients enrolled in 2004-2008 was 39, of which 15.4% (n=6) of patients received ASCT after induction chemotherapy. Of 91 patients who participated in 2009-2013, 25 patients (27.5%) of all patients received ASCT. The number of patients enrolled in 2014-2019 was 155, most of whom (43.2%) received bortezomib combination chemotherapy, and 23.2% (n=23) patients receive ASCT. The median OS of 302 AL amyloidosis patients who received ASCT was better than those who received chemotherapy alone (NR vs. 33, P-value <0.00). However, there was no difference in the comparison of survival rated every four-years (P-value = 0.16). Of the 165 patients who experienced death during the follow-up period, 87 (28.8%) patients died early within six months. The early death occurrence of patients who received ASCT was lower (4.5%) than systemic chemotherapies alone or the observation group (p-value < 0.00).

Conclusion: The outcome of AL amyloidosis patients compared to 10 years ago is improving with the introduction of new myeloma drugs and collaborative approaches. In the study, ASCT has identified as the treatment for reducing premature death regardless of induction chemotherapy. However, early death is still a big unmet need in patients with amyloidosis. Once the high-risk AL amyloidosis doubted, we suggest that clinicians need to actively consider ASCT or new clinical trials for declining early death.
MANAGEMENT AND OUTCOMES IN AL AMYLOIDOSIS: A RETROSPECTIVE ANALYSIS OF AN INSTITUTIONAL REGISTRY OF ARGENTINA

M. L. Posadas-Martinez ¹, M. A. Aguirre ¹, S. Saez ¹, C. Belziti ¹, R. Luxardo ¹, J. Arbelbide ¹, D. Fantl ¹, G. Machnicki ¹, M. Fernandez ¹, E. Nucifora ¹

¹Hospital Italiano de Buenos Aires.

Background: AL amyloidosis is a monoclonal plasma cell proliferative disorder characterized by tissue deposits leading to organ dysfunction. There is no standard of care approved in Argentina. Disease extension, number of sites, comorbidities and treatment availability may impact treatment outcomes.

Aim: Describe treatment patterns, overall and progression free survival (OS/PFS) of AL amyloidosis in one institution from Argentina.

Methods: Retrospective cohort (2010-2019) of adult AL amyloidosis patients from Italian Hospital of Buenos Aires - Institutional Registry of Amyloidosis (IRA - ClinicalTrials.gov#: NCT01347047). First line treatment was defined as first regimen received, regardless of subsequent changes. Treatment was categorized as ASCT upfront or following induction, CyBorD, Thalidomide-Dexamethasone/Cyclophosphamide, Rituximab+ Cyclophosphamide + Vincristine + Prednisone(R-CVP), Lenalidomide/Dexamethasone, or other. All patients were followed to death for all causes or follow-up loss. Hematological best response was defined as complete remission (CR), very good partial remission (VGPR), partial remission (PR) or no response. PFS was evaluated and survival rates are expressed as the % surviving by Kaplan Meier method.

Results: 91 AL amyloidosis patients were included. Organ compromise at diagnosis (median of 2 – CI:1-3) cardiac 72% (65), renal 71% (62), gastrointestinal 37% (34) and neurological 36% (33). Median ECOG of 2 (1-2) and Charlson score 2 (CI 1-3). The median follow-up of this cohort was 37 months (CI 12-114). Sixty-nine (76%) patients received treatment, 9 (10%) required cardiac transplant and 1 kidney, while 13 (14%) underwent ASCT. From those who received chemotherapy, regimens used were 76% (51) CyBorD, 11% (7) Thalidomide-Dexamethasone/Cyclophosphamide, 2% (1) rituximab, 2% (1) Lenalidomide/Dexamethasone, 2% (1) R-CVP and 9% (6) others; with mean 5 cycles (DS 5). Twenty-six patients received second line with median 4 cycles (CI 2-6), using Lenalidomide/Dexamethasone in 42% (11) and Daratumumab in 19% (5), and 3 patients received 3rd line with median 6 cycles (CI 1-6). Hematologic best response (intention-to-treat) was: CR 48% (32), VGPR 7% (5), PR 5% (3), no response 24% (16). Response was not evaluable in 16% (11) patients - 2 deaths, 1 discontinued treatment by patient decision, 2 insufficient follow-up time and 6 were referred to other institutions. Overall mortality was 42% (n=38, CI 32-53), median survival was 126 months (CI 23-170), and OS at one year was 86% (77-90), 5 years 65% (53-74) and 10 years 53% (39-65). 43% (29) presented deep responses. The relapse rate was 21% (CI 11-38, n 9). The median PFS rate was 91% (78-97) at 5 and 79% (63-89) at 10 years.

Conclusion: Bortezomib based treatment was the most used for first line and 48% of patients achieved CR. Only 38% of patients received second line. OS and PFS are similar to other published cohorts.
PW037

BORTEZOMIB NEUROPATHY: CLINICAL AND ELECTROPHYSIOLOGICAL FEATURES AND ITS PREDICTIVE FACTOR

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Background:
Bortezomib is one of the proteasome inhibitors which are now considered as a major treatment option for systemic light chain (AL) amyloidosis. However, its neurotoxic side effect has emerged as a problem because some bortezomib-treated patients develop subsequent peripheral polyneuropathy.

Objectives:
The aim of this study is to clarify the clinical and electrophysiological features of bortezomib-induced neuropathy and to investigate the possible predictive factor of this side effect.

Methods:
All AL patients, except for those with amyloid neuropathy, who were treated with bortezomib-dexamethasone (BD) between Oct. 2012 and Apr. 2018 were enrolled. Clinical and electrophysiological findings of the patients with bortezomib-neuropathy were retrospectively investigated.

Results:
Among 40 enrolled patients, ten patients (25.0%) were found to develop bortezomib-neuropathy. Mean cumulative dose of bortezomib at neuropathy onset was 15.8 mg/m². Clinical presentation was length-dependent symmetrical sensory neuropathy without any motor-nerve symptom. Electrophysiological examination revealed sensory-nerve axonal involvement. Although this abnormal electrophysiological findings recovered within 6 to 12 months after treatment, clinical symptom remained in all patients (the number of patients with complete recover: 0, partial improvement: 7, no improvement: 3). Many clinical parameters were compared between patients who developed bortezomib-neuropathy and who did not, but no significant predictive factor was detected. Neuropathy development did not affect hematological response rate or overall survival.

Conclusion:
25% of bortezomib-treated AL patients developed sensory-nerve axonal neuropathy with mean cumulative dose of bortezomib 15.8 mg/m². There was no significant relationship between neuropathy development and treatment outcomes. It is important to pay attention to cumulative dose of bortezomib because neuropathy onset was unpredictable but clinical symptom was likely to remain.

Keywords: AL amyloidosis, bortezomib, neuropathy
CLINICAL IMPACT OF AN EARLY RESPONSE AND OF EARLY INITIATION OF SALVAGE THERAPY IN PATIENTS WITH SYSTEMIC LIGHT CHAIN (AL) AMYLOIDOSIS

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Background: A rapid and deep hematologic response is fundamental in order to improve outcome of patients with AL amyloidosis while in patients not optimally responding, early change in therapy may salvage some of them, but there is limited data.

Objectives: to evaluate the clinical impact of an early deep response and of early introduction of salvage therapy, in previously untreated AL patients.

Methods: The analysis included 227 consecutive, previously untreated patients with AL amyloidosis, who received bortezomib-based primary therapy, in a single center (Department of Clinical Therapeutics, Athens) and had evaluable clonal disease and available assessments at 1 and at 3 months from start of therapy.

Results: Median age was 64 years (range 40-84), 57% were males. Heart was involved in 64%, kidneys in 72%, peripheral/autonomic nerve in 18% and liver in 21%. Per Mayo stage 23% were stage 1, 52% stage 2, 18% were 3A and 7% were 3B. After 1 month of therapy, 40.5% had a VGPR, 23% a PR and 36% had not achieved a response (NR). Among patients with <VGPR after 1 month (n=135), only 21 (15%) reached a VGPR by 3 months, so that at 3 months response were ≥VGPR: 54%, PR: 24%, NR: 22%. Median OS of patients with VGPR, PR and NR at 1 month was 9.5 vs 3.9 vs 1.8 years, p<0.001 while according to response at 3 months, median OS for patients with VGPR, PR and NR was 9.3 vs 3.6 vs 1.1 years respectively, p<0.001. Patients that at 3 months improved their response to VGPR (from PR or NR) had an OS of 7 years vs 2 years for those that remained in PR or improved their response from NR to PR while it was 1.6 years for those with NR (p<0.001); however, the OS among those that achieved VGPR at 3 months was shorter than of those that already achieved a VGPR at 1 month (9.5 vs 7, p=0.041).

We then analyzed the outcome of patients that after 3 months had not achieved VGPR. Of those with NR, 32% started a new therapy and 68% continued same therapy, while among those in PR, 80% continued the same therapy. At 6 months, 37% (10% VGPR, 27% PR) of patients who had NR and remained on the same therapy achieved a response vs 44% (20% VGPR, 24% PR) of those that switched therapy. Among those in PR at 3 months, only 12.5% of those continuing with the same therapy improved their response to ≥VGPR. Patients with <VGPR at 3 months that continued their initial regimen had an OS of 41 vs 6 months for those that received salvage therapy (p=0.009), probably because these were sicker (50% vs 12% were stage 3). In multivariate analysis, ≥VGPR after 1 month (p=0.001) or at 3 months (p=0.001) and Mayo stage 1 or 2 vs 3 (p<0.001) were the stronger prognostic factors for survival.

Conclusions: in patients with AL amyloidosis, a rapid (at 1 month) and deep response is associated with better survival. Early introduction of salvage therapy may improve response and survival for some patients, but starting with the most effective therapy, especially in patients at higher risk, offers the most benefit.
REAL-WORLD TREATMENT PATTERNS OF AL AMYLOIDOSIS IN US ADULTS, 2010-2018

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Introduction: Light chain (AL) amyloidosis is a rare plasma cell dyscrasia characterized by organ deposits of amyloid derived from misfolded immunoglobulin light chains. Currently, there are no approved treatments for AL, but guidelines recommend off-label use of multiple myeloma (MM) regimens. Limited information is available on how these treatments are given in real-world clinical settings.

Objectives: To explore the timing, type, and order of treatments received for AL after diagnosis among US adults.

Methods: We explored treatment patterns in the Optum De-Identified Clinformatics® Datamart, which includes commercial and Medicare supplemental health insurance claims. Adults with AL were defined as having ≥1 inpatient or ≥2 outpatient codes (ICD-9-CM 277.30, ICD-10-CM E85.81, E85.89, E85.9) and ≥1 MM treatment any time after diagnosis (01Jan10-31Dec18). Median time to treatment after AL diagnosis was summarized with Kaplan-Meier estimates. Oral drug periods were defined as the fill date plus days supply until a 90-day gap. To reflect 3-4 week treatment cycles, infusion/injection periods were defined as the treatment date plus 7-14 days depending on the drug until a 90-day gap. Regimens were defined as ≥14 day overlap between periods. Drugs classified in ≥2 treatment lines were grouped with the initial line except for dexamethasone.

Results: Among 703 AL patients with guideline-directed AL treatment after 2010, the median time to first treatment was 43 days (IQR:15-117). There were 179 combinations of treatment received over four treatment lines. About 65% of patients only received first-line treatment. Forty-six percent of patients received first-line bortezomib-based regimens, and 3.6% received bortezomib in subsequent lines (Table 1). Cyclophosphamide combinations were received by 24% of patients, with half of the regimens including bortezomib. Besides dexamethasone, the main drugs used were lenalidomide (7.6% first-line; 11.7% other lines), melphalan (5.3% first-line; 3.1% other lines), pomalidomide (1.7% first-line; 1.5% other lines), carfilzomib (1.4% first-line; 2.2% other lines) and daratumumab (1.2% first-line; 2.7% other lines). About 7.6% received first-line autologous stem cell transplant (ASCT); 18% received ASCT in any line. Patients receiving ASCT were more likely to be younger (median age=61 vs 70 years), male (61 vs 57%) and have prior MM than AL patients without ASCT. ASCT patients had fewer comorbidities than AL patients without ASCT, such as heart failure (18 vs. 42%), COPD (5 vs. 22%), cerebrovascular disease (9 vs 27%), and pleural effusion (15 vs. 29%).

Conclusion: In contemporary AL treatment, bortezomib-based regimens represent about 50% of frontline regimens, which is lower than expected. A minority of patients undergo frontline ASCT and are usually healthier and younger, suggesting more use of induction therapy in AL, especially for sicker patients. Future studies will explore outcomes after treatment combinations.
Table 1. Percentage of 703 patients diagnosed with AL amyloidosis after 2010 who received each treatment by treatment line. Patients could receive multiple ingredients or procedures, so the values below are not mutually exclusive.

<table>
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Suggested Keywords: treatment, AL amyloidosis
Category: Treatment of newly diagnosed AL amyloidosis eligible for transplant
SIMILAR SURVIVAL WITH DEFERRED VERSUS SALVAGE AUTOLOGOUS STEM CELL TRANSPLANT IN LIGHT CHAIN AMYLOIDOSIS

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¹ Weill Cornell Medicine.

Background: Light chain (AL) amyloidosis is characterized by deposition of misfolded monoclonal immunoglobulin light chains leading to organ dysfunction. AL amyloidosis has traditionally been treated with multiple myeloma therapies and autologous stem cell transplantation (ASCT). A retrospective study comparing patients (pts) with AL amyloidosis undergoing upfront ASCT to those undergoing ASCT following induction (deferred ASCT) revealed that deferred ASCT was associated with prolonged overall survival (OS) (Afrough et al, Biol Blood Marrow Transplant, 2018). Given the number of effective new therapies for AL amyloidosis, the potential to delay ASCT after one or more lines of therapy (salvage ASCT) is feasible. To our knowledge, transplant outcomes of AL amyloidosis pts undergoing deferred vs salvage ASCT following at least one relapse have not been reported. A retrospective chart review was conducted to compare AL amyloidosis pts receiving deferred vs salvage ASCT for progression-free survival (PFS) and OS.

Methods: 24 pts with AL amyloidosis who underwent deferred or salvage ASCT at Weill Cornell Medical College between 2000-2018 were included in the analysis. PFS was calculated from date of ASCT to first relapse. OS was calculated from date of diagnosis to death. PFS and OS of patients who underwent deferred vs salvage ASCT were compared. Log-rank tests were used to evaluate Kaplan-Meier PFS/OS curves. Cox proportional hazards models were used to derive hazard ratios (HR) using deferred ASCT as the reference.

Results: Among 24 pts, 13 underwent deferred and 11 salvage ASCT with the latter group receiving a median of 3 prior lines (Table 1). Ten pts had cardiac amyloidosis, 15 had renal amyloidosis, and 6 had multi-organ involvement. Neither PFS nor OS was significantly different between patients receiving deferred vs salvage ASCT (Figure 1). After median follow-up of 2.9 years, median PFS was 6.5 years in patients receiving deferred ASCT and was not reached in those receiving salvage ASCT (P=0.47), with a HR of 1.74 (95% CI, 0.38-7.85). Median OS was not reached in either group after a median follow-up of 5.0 years (P=0.79), with a HR of 0.77 (95% CI, 0.10-5.73).

Conclusions: No significant differences in PFS or OS were seen between pts undergoing deferred vs salvage ASCT in this single center study. Unlike the superior OS seen in deferred vs upfront ASCT, our findings show that deferred vs salvage ASCT may be associated with comparable outcomes. Limitations include small sample size and lack of daratumumab-based therapy, which may have improved outcomes in either or both groups. Nevertheless, the lack of difference seen in both PFS and OS between the groups suggests that eventual hematologic relapse and progression with death occur at similar times regardless of receiving ASCT in the deferred vs salvage setting. Achievement of deep responses to prevent organ progression and death regardless of timing of ASCT remains an important treatment goal.
AL AMYLOIDOSIS: MAINTENANCE THERAPY FOLLOWING AUTOLOGOUS STEM CELL TRANSPLANT (ASCT)

M. Ozga 1, Q. Zhao 1, D. Benson 1, Jr., MD 1, P. Elder 1, N. Williams 1, N. Bumma 1, A. Rosko 1, M. Chaudhry 1, A. Khan 1, S. Devarakonda 1, R. Kahwash 1, A. Vallakati 1, C. Campbell 1, S. Parikh 1, S. Almaani 1, J. Prosek 1, J. Bittengle 1, K. Pfund 1, S. LoRusso 1, M. Freimer 1, E. Redder 1, CLT-Lana 1, Y. Efebera 1, N. Sharma 1.

1 The Ohio State University Wexner Medical Center.

Introduction: Autologous stem cell transplantation (ASCT) remains an effective treatment option for many patients with systemic AL amyloidosis (AL). However, the decision to utilize maintenance therapy following ASCT remains controversial and largely unexplored. Furthermore, the growing relevance of cytogenetics in AL may help dictate this maintenance decision as well.

Objectives: The present study aims to a) determine the prognostic significance of utilizing maintenance therapy following ASCT, and b) assess the potential impact of cytogenetics on this decision.

Methods: A retrospective chart review was performed on 50 consecutive AL patients who underwent ASCT. Patients received high dose Melphalan (140 or 200 mg/m²) prior to ASCT. Patients were divided into subgroups based on FISH, i.e. abnormal (presence of any abnormality) versus normal, and whether or not they had received maintenance therapy following transplant. Primary endpoints were progression free survival (PFS) and overall survival (OS), both estimated via the Kaplan-Meier survival function. The log-rank test was used to test the equality of survivor functions between different groups of patients.

Results: Twenty-eight patients (56%) received maintenance and 22 (44%) did not. There was no difference in age, dose of melphalan used, disease stage and number of organs involved between the two groups. There was no statistically significant difference in OS (p=0.32) and PFS (p=0.66) between patients who received maintenance versus those patients who did not receive maintenance (Figure 1A). Specifically, among patients with abnormal FISH, there was no difference in survival between the two groups in OS (p=0.65) and PFS (p=0.15) (Figure 1B). Upon further stratification by degree of organ involvement, i.e. patients with 2 or more organs involved, the decision to utilize maintenance vs no maintenance showed no difference in OS (p=0.80) and PFS (p=0.34) (Figure 1C).

Conclusion: In this small retrospective analysis, maintenance therapy post ASCT did not impact PFS or OS in patients with AL, even among patients with cytogenetic abnormalities or ≥2 organs involved. A prospective study is needed to assess the benefit of maintenance therapy post ASCT in AL amyloid patients.

Figure 1
Introduction: Renal involvement is one of the most common presentations of AL Amyloidosis. Newer therapeutic regimens directed at the plasma cell clone have become available, including high-dose melphalan with autologous stem cell transplantation (ASCT), bortezomib, and monoclonal antibody-based therapies. Chemotherapy in AL amyloidosis is associated with improved survival, but its effect on renal outcome has not been well established.

Objectives: Patients who achieve complete response (CR) have similar outcomes to those achieving very good partial response (VGPR). We seek to describe renal outcomes in patients with AL amyloidosis according to their response (CR or VGPR) after ASCT.

Methods: We performed a retrospective chart review of 50 AL Amyloidosis patients treated at The Ohio State University who underwent ASCT. We then identified patients with renal involvement who achieved VGPR/CR as best hematological response and assessed their renal response. Hematological and renal responses were assessed according to consensus guidelines. Primary endpoints were progression free survival (PFS) and overall survival (OS). PFS was defined as the time from date of transplant to day of progression or death from any cause. OS was defined as the time from date of transplant to death from any cause, with censoring those who were still alive at the last follow up. Kaplan-Meier survival function was used to estimate the PFS and OS. The log-rank test was used to test the equality of survivor functions between different groups of patients.

Results: Out of those undergoing ASCT, 16 patients (32%) had achieved VGPR or CR as their best hematological response. 100% had renal involvement. Median age in that group was 60 years (40-70); 50% were female. 68.8% were lambda restricted. Median number of involved organs was 1.5 and 25% had concurrent cardiac involvement. There was no difference in OS and PFS in patients who achieved CR or VGPR as best hematological response. 6 patients achieved partial response (PR) as best renal response and 10 had stable disease (SD). No patient required hemodialysis. PFS was similar in those achieving renal PR with median PFS 4.1 years compared to those with stable disease (p=0.89). There was no difference in OS with median follow-up of 5.6 years v/s 11.9 years respectively, (p= 0.44).

Conclusions: Hematological response is important in AL amyloidosis with VGPR or CR being the goal for therapy. In patients with renal involvement undergoing ASCT, outcomes are similar in those achieving PR or SD as best renal response.
FAVORABLE OUTCOMES WITH AUTOLOGOUS STEM CELL TRANSPLANT IN AL AMYLOIDOSIS PATIENTS WITH EXTENSIVE ORGAN INVOLVEMENT.

N. Bumma1, M. Ozga1, Q. Zhao1, N. Sharma1, D. Benson 1, A. Rosko1, M. Chaudhry1, A. Khan1, S. Devarakonda1, S. Parikh1, S. Almaani1, J. Prosek1, J. Bittengle1, K. Pfund1, M. Speiser1, M. Freimer1, E. Redder1, S. LoRusso1, A. Vallakati1, R. Kahwash1, Y. Efebera1

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Conclusions: Hematological response is important in AL amyloidosis with VGPR or CR being the goal for therapy. In patients with renal involvement undergoing ASCT, outcomes are similar in those achieving PR or SD as best renal response.
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$^1$Median ± standard error
$^2$Missing 4 observations
$^3$Defined by presence of del17p, del17, t(4;14), t(14;16), t(14;20), and/or chr 1 abnormalities
$^4$Missing 1 observation in the deferred group and 2 in the salvage group
AUTOLOGOUS STEM CELL TRANSPLANTATION MAY HALT PERIPHERAL NEUROPATHY PROGRESSION AND IMPROVE SURVIVAL IN AL AMYLOID NEUROPATHY

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1 Department of Neurology, Mayo Clinic, Rochester, MN, USA.
2 Department of Medicine, Division of Pulmonary and Critical Care Medicine, Mayo Clinic, Rochester, MN, USA.
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Introduction: Immunoglobulin light chain (AL) amyloid neuropathy is a progressive sensory, motor and autonomic neuropathy with a median survival of 25 to 45 months. Although autologous stem cell transplantation (ASCT) has shown improved survival in AL amyloidosis, little is known about its effects on peripheral neuropathy.

Objective: To study the clinical characteristics of peripheral neuropathy and outcomes in a large cohort of patients with AL amyloid neuropathy who underwent ASCT.

Methods: We included patients with AL amyloid neuropathy who underwent ASCT at our institution from 01/01/1998 to 03/31/2018. Neuropathy was defined by neuropathic symptoms plus signs (per a neurologist) or neuropathic symptoms plus neurophysiologic evidence of neuropathy by nerve conduction studies/electromyography (NCS/EMG) or autonomic reflex screen (ARS). Clinical, neurophysiological and survival data were extracted.

Results: 70 patients with AL amyloid neuropathy were identified. Median age was 60 (range:35-72) years old. At baseline (before ASCT), 45% of patients had muscle weakness, 70% sensory loss, 73% prickling, 69% autonomic symptoms and 43% neuropathic pain requiring medications. Baseline median neuropathy impairment score (NIS) was mild at 9 (0-82), median clinical autonomic severity score (CASS) was moderate at 5 (0-10) and median modified Rankin scale (mRankin) was moderate at 2 (1-3). Baseline median sural sensory nerve action potential (SNAP) amplitude was 4 (0-17) µV and median ulnar motor compound motor action potential (CMAP) amplitude was 8.1 (3-12.9) mV. After transplant the median survival was 78 months. 37 (53%) patients died. The probability of survival at 1-, 5- and 10- years after ASCT was, respectively 87, 62.3 and 31.7%. In univariate cox regression model, mortality was associated with heart involvement (HR: 2.0; CI 1.02-3.88; p=0.0417), number of organs involved (HR: 1.82; CI 1.18-3.16; p=0.005) and severity of autonomic neuropathy by CASS (HR: 1.4; CI 1.09-1.85; p=0.007). Prolonged neurological follow-up was available in 23 patients (32.8 %) who had at least one neurology visit, NCS/EMG or ARS after ASCT. Of those, median NIS (n=15; pre-ASCT 38.5; last f/u 38.25), median mRankin (n=15; pre-ASCT 2; last f/u 2), median Ulnar CMAP amplitude (n=16; baseline 7.95; last f/u 6.95) and median CASS (n=4; baseline 5; last f/u 5.75) remained stable at last follow-up.

Conclusion: AL amyloid neuropathy patients that undergo ASCT have a mild sensorimotor peripheral neuropathy but have more severe (moderate) autonomic impairment. Compared to historical AL amyloid neuropathy studies, our study suggests that ASCT prolongs survival. Mortality risk factors include heart involvement, number of organs affected, and autonomic neuropathy (elevated CASS). ASCT seems to halt progression of AL amyloid neuropathy.
A PHASE II STUDY OF PROPYLENE GLYCOL-FREE MELPHALAN IN PATIENTS WITH AL AMYLOIDOSIS UNDERGOING AUTOLOGOUS STEM CELL TRANSPLANT: PRELIMINARY RESULTS

S. Sarosiek¹, J. M. Sloan¹, K. Quillen¹, D. Brauneis¹, R. Cruz¹, A. Schulman¹, S. Fennessey¹, A. Shelton¹, V. Sanchorawala¹

¹Boston University Amyloidosis Center.

Introduction: High dose melphalan and autologous stem cell transplantation (HDM/SCT) is an effective treatment for select patients with light-chain (AL) amyloidosis. Despite rigorous patient selection, ≈21% of patients with AL amyloidosis develop acute renal injury and ≈22% develop cardiac arrhythmias in the peri-transplant period. This is potentially exacerbated by the co-solvent propylene glycol in melphalan hydrochloride. This clinical trial investigates the effect of propylene glycol-free melphalan on organ toxicity (NCT02994784).

Objectives: Primary objectives include evaluation for renal dysfunction (creatinine increase ≥1 mg/dL or creatinine doubling to ≥1.5 mg/dL for ≥2 days), cardiac dysfunction (new arrhythmia), and autonomic dysfunction (drop in SBP ≥20 mmHg). Secondary objectives are time to engraftment, treatment related mortality, overall hematologic response, organ response, and number of hospitalizations.

Methods: Enrollment began in April 2018. Eligibility criteria include: histologic diagnosis of systemic AL amyloidosis, involvement of ≥1 vital organ, eGFR ≥30 mL/min, DLCO ≥50%, LVEF ≥40%, ECOG <3, NYHA class <3, and no prior HDM/SCT. Subjects underwent HDM/SCT per institutional guidelines with 140 or 200 mg/m² propylene glycol-free melphalan IV on Days -3 and -2. Stem cells were infused on Day 0.

Results: At data cut-off (10/21/2019), 19 patients were enrolled. Median age was 60 years (range, 44-76) with 13 males. Median number of organs involved was 1 (range, 1-3). 15 (79%) and 7 (37%) patients had renal and cardiac involvement, respectively. Median NT-proBNP was 396 pg/mL (range, 63-6220), urine total protein was 4 g/24 h (range, 0-22.9), serum creatinine was 0.9 mg/L (range, 0.7-2.1), and dFLC was 45 mg/L (range, 3-700). Subjects received 140 mg/m² (n=3) or 200 mg/m² (n=16) of propylene glycol-free melphalan. There were 0 deaths in the peri-transplant period. Per protocol criteria, 2 patients (11%) had acute renal dysfunction, 3 (16%) developed orthostatic hypotension, and 3 (16%) had a new arrhythmia (atrial fibrillation or ventricular tachycardia). Median time to neutrophil engraftment was 10 days and time to platelet engraftment was 17 days. Peri-transplant hospitalization occurred in 15 subjects (79%). Most common non-hematologic adverse events were nausea/vomiting (84%), fatigue (84%), and diarrhea (89%). Of the 13 evaluable subjects the hematologic response rates are: 6 CRs (46%), 3 VGPRs (23%), 2 PRs (15%) and 2 stable disease (15%).

Conclusion: In this ongoing trial the cardiac toxicities appear comparable to historical data (16% developing a cardiac arrhythmia) and renal toxicity is potentially improved (11% with renal dysfunction), although additional data are required to confirm these findings. Time to engraftment and hematologic responses are comparable to historical controls. Ongoing patient enrollment will determine the safety of propylene glycol-free melphalan in patients with AL amyloidosis.

Keywords: HDM/SCT, melphalan, AL amyloidosis
Primary systemic amyloidosis (AL) is an acquired disease caused by extracellular fibrils deposition that results in organs dysfunction. The median age at diagnosis is 67 years. Prognosis depends on the severity of cardiac involvement and hematologic responses.

The use of new class of drugs, such as proteasome inhibitors and immunomodulatory drugs has led to rapid and profound hematologic responses and huge improvement in survival in the whole population of patients with AL amyloidosis. Is it true for older patients with their lot of comorbidities? The correlation between hematologic data and geriatric status provides insight into elderly treatment choice and outcomes in clinical practice.

We performed a retrospective study from September 2013 to April 2019. Inclusion criteria were: All patients more than 70 years old, diagnosis and organ involvement according to consensus criteria, repeated geriatric assessment (CGA) according to the SIOG recommendations, available clinical and biological data, requiring a treatment. All patients baseline and clinical characteristics were collected.

Patients outcomes were monitored during hematological and geriatric follow up (monthly chemotherapy, quarterly consultations, case management, increasing supportive care and modification of the home care plan).

Thirty-eight patients were included. Eighteen were male (47%) with a median age of 77 [70-91]. Main organ involvements were heart (68%) and kidney (53%). MAYO clinic score >=2 for 85% of patients. Twenty Patients were assigned to the unfit group and 18 to the fit group according to the CGA. Excepting NTproBNP and eGFR in multivariate analysis there were no significant difference in the haematological presentation between the two groups at first CGA. Significative geriatrics difference were found at diagnosis between fit and unfit group. Treatment Choices and chemotherapy duration were homogeneous. Survival of the whole cohort was estimated by Kaplan-Meier was 67% at 3 years after diagnostic and no significant difference was observed between the fit and the unfit group.

In this cohort we show that geriatric status did not results in significant difference in overall survival with a median follow up of 59 month. On multivariate analysis, NTproBNP, eGFR and response to treatment are independent predictor of survival. There is a correlation between Geriatric status, heart and kidney severity.

Those results show no significant difference in treatment efficiency and survival between the fit and unfit patients. The patients initial screening helps practitioners to manage frailty and to choice of the best strategy in multidisciplinary team. We think that the proactive program management in our department erases geriatrics frailty. It’s justifying that age and comorbidty is no longer a reason not to treat the patients with AL amyloidosis. We will try to compare their results with those of centers that do not have this geriatric program.
**Introduction:** Cyclophosphamide, Bortezomib and Dexamethasone (CyBorD) is an effective regimen for the treatment of patients with AL amyloidosis. CyBorD produces rapid and deep hematological responses (HR). However, it remains inadequate to enhance outcomes in patients with advanced cardiac disease. Recently, it has been proposed that the substitution of dexamethasone by methylprednisolone could potentially induce rapid and tolerable responses for patients with stage IIIA/B disease. Methylprednisolone has a biological half-life of 24-36 hrs compared to 36-54 hrs for dexamethasone which might account for a rapid onset of action.

**Objectives and Methods:** All consecutive newly diagnosed AL amyloidosis treated with CyBorD and CyBorMe from 01/2012 to 11/2019 were evaluated. CyBorD was given as reported elsewhere and CyBorMe in contrast, used Methylprednisolone at 500 mg IV q 7 days for 3-4 weeks instead of dexamethasone. Hematological and Organ Response was assessed as per standard guidelines. Patients with at least 1 cycle of treatment were included.

**Results:** 43 patients were treated with CyBorD and 7 with CyBorMe. Clinical characteristics are seen in Table 1. After a median of 4 cycles in the CyBorD and 2 cycles in the CyBorMe group, HR was seen in 90.6% and 86% of cases, including CR in 28.5% (both CyBorD and CyBorMe), VGPR in 33.3% and 28.5% and PR in 30.9% and 28.5% for CyBorD and CyBorMe, respectively. Time to first response was slightly faster in the CyBorMe group (4 vs 6 weeks, p=0.3). Further, dFLC at 1 month was similar (31 vs 39, p=0.2) and cardiac response was of 40% in the CyBorMe group even with a shorter number of cycles and follow-up. Median OS has not been reached for both groups but not initial differences have been observed. At the time of analysis, 58% of patients on the CyBorD group and 85% in the CyBorMe group are alive.

**In Conclusion,** CyBorMe appeared to be efficacious and well tolerated in patients with advanced stage disease. Prospective studies with CyBorMe in the stage III/IV group are warranted aiming to minimize toxicity.

**Keywords:** CyBorMe, CyBorD and AL amyloidosis

**Category:** Treatment of newly diagnosed AL amyloidosis non eligible for transplant
Table 1. Clinical Characteristics of patients with AL amyloidosis treated with CyBorMe and CyBorD at our Institution

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>N=43 (CyBorD)</th>
<th>N=7 (CyBorMe)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (median)</td>
<td>64</td>
<td>69</td>
<td>0.02</td>
</tr>
<tr>
<td>Gender</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>24 (56%)</td>
<td>4 (57%)</td>
<td>0.09</td>
</tr>
<tr>
<td>Female</td>
<td>19 (44%)</td>
<td>3 (43%)</td>
<td></td>
</tr>
<tr>
<td>Hb (g/L)</td>
<td>126</td>
<td>104</td>
<td>0.1</td>
</tr>
<tr>
<td>Creatinine (µmol/L)</td>
<td>92</td>
<td>127</td>
<td>0.3</td>
</tr>
<tr>
<td>B2microglobulin (µmol/L)</td>
<td>2.81</td>
<td>2.9</td>
<td>0.3</td>
</tr>
<tr>
<td>Albumin (g/L)</td>
<td>29</td>
<td>33</td>
<td>0.8</td>
</tr>
<tr>
<td>Stage I</td>
<td>6 (13.9%)</td>
<td>1 (14.2%)</td>
<td></td>
</tr>
<tr>
<td>Stage II</td>
<td>10 (23.2%)</td>
<td>1 (14.2%)</td>
<td></td>
</tr>
<tr>
<td>Stage III</td>
<td>10 (23.2%)</td>
<td>1 (14.2%)</td>
<td>0.7</td>
</tr>
<tr>
<td>Stage IV</td>
<td>15 (34.8%)</td>
<td>4 (57.1%)</td>
<td></td>
</tr>
<tr>
<td>Unknown</td>
<td>2 (4.6%)</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>LDH (IU/L)</td>
<td>208</td>
<td>308</td>
<td>0.2</td>
</tr>
<tr>
<td>BMPC (%)</td>
<td>7.5%</td>
<td>15%</td>
<td>0.1</td>
</tr>
<tr>
<td>NTproBNP ng/L</td>
<td>1810</td>
<td>2278</td>
<td>0.5</td>
</tr>
<tr>
<td>Light chain:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Kappa</td>
<td>6 (14%)</td>
<td>2 (28%)</td>
<td>0.5</td>
</tr>
<tr>
<td>Lambda</td>
<td>36 (86%)</td>
<td>5 (72%)</td>
<td></td>
</tr>
<tr>
<td>Organ Involvement</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cardiac involvement</td>
<td>72%</td>
<td>71%</td>
<td>0.9</td>
</tr>
<tr>
<td>Kidney involvement</td>
<td>72%</td>
<td>85.7%</td>
<td>0.5</td>
</tr>
<tr>
<td>Liver involvement</td>
<td>9.3%</td>
<td>28%</td>
<td>0.1</td>
</tr>
<tr>
<td>Nerve involvement</td>
<td>25.5%</td>
<td>14.2%</td>
<td>0.5</td>
</tr>
<tr>
<td>GI involvement</td>
<td>25%</td>
<td>0%</td>
<td>0.1</td>
</tr>
<tr>
<td>Lung involvement</td>
<td>6.9%</td>
<td>0%</td>
<td>0.4</td>
</tr>
<tr>
<td>Hematological Response</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ORR</td>
<td>90.6%</td>
<td>86%</td>
<td>0.5</td>
</tr>
<tr>
<td>CR</td>
<td>28.5%</td>
<td>28.5%</td>
<td></td>
</tr>
<tr>
<td>VGPR</td>
<td>33.3%</td>
<td>28.5%</td>
<td></td>
</tr>
<tr>
<td>PR</td>
<td>30.9%</td>
<td>28.5%</td>
<td></td>
</tr>
<tr>
<td>Time to first response (weeks, median)</td>
<td>6</td>
<td>4</td>
<td>0.3</td>
</tr>
<tr>
<td>dFLC at 1 month (median)</td>
<td>31</td>
<td>39</td>
<td>0.2</td>
</tr>
<tr>
<td>Time to Best Response (months)</td>
<td>12 weeks</td>
<td>4 weeks</td>
<td>0.06</td>
</tr>
<tr>
<td>Median number of cycles</td>
<td>4</td>
<td>2</td>
<td>0.5</td>
</tr>
<tr>
<td>Discontinuation</td>
<td>25%</td>
<td>14.2%</td>
<td>0.1</td>
</tr>
<tr>
<td>Cardiac Response</td>
<td>34%</td>
<td>40%</td>
<td>0.6</td>
</tr>
</tbody>
</table>

BMPC: Bone marrow plasma cells; FLC: Free-light chains only.
RESULTS OF DEFERRED AUTOLOGOUS STEM CELL TRANSPLANTATION IN PATIENTS WITH ADVANCED STAGES OF SYSTEMIC AL AMYLOIDOSIS

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Introduction: Systemic immunoglobulin light chain (AL) amyloidosis is the form of plasma cell disorders, characterized by various organ dysfunction due to amyloid light chains depositions. Results of autologous stem cell transplantation (ASCT) showed improvements in the outcome compared with standard chemotherapy, but only 20% of patients are eligible for upfront ASCT due to amyloid-induced organ damage.

Patients and methods: Among 151 patients who were treated in First St. Petersburg State I.P. Pavlov Medical University during the past 15 years 27 patients received deferred ASCT. The median age was 55 years (39-66). All patients had 3 or more organs involved at the time of diagnosis with advanced heart or renal stages. The number of patients with Standard Mayo cardiac stage I, II, III was the following: 15% (n=4), 26% (n=7), and 59% (n=16) respectively. Median level of NT-proBNP was 2203 ng/L (338-34772). The number of patients with I, II, III renal stage were: 18% (n=5), 51% (n=14), 22% (n=6). Induction chemotherapy included bortezomib-based regimes. The median number of cycles was 4 (2–9) and median time to ASCT was 299 days (158-2013). 74% of patients received reduced doses of melphalan as conditioning regimen (140-180 mg/m2).

Results: After induction chemotherapy hematologic response (HR) was achieved in 55% patients: complete response (CR)-21% (n=8), very good partial response (VGPR)-14% (n=4), partial response (PR)-12% (n=3). Organ response was achieved in 17 patients with high frequency VGPR (70%). After ASCT 5 patients improved their response to CR, 8 maintained CR, 1 achieved VGPR, 2-PR. 6 patients with NR and 2 with disease progression on day+ 100 continued chemotherapy and achieved HR later. There was only 1 case of transplant-associated mortality. Overall survival (OS) was 81.5%, progression-free survival - 66.7%. 5 years OS in compared with non-ASCT patients was also significantly higher 78% vs 57% (p=0.008).

Conclusions: Deferred ASCT is an effective and relatively safe treatment option in patients with advanced stages of systemic AL amyloidosis who achieved organ response after induction chemotherapy.

Keywords: Systemic light chain (AL) amyloidosis, ASCT
AUTOLOGOUS STEM CELL TRANSPLANTATION IN AL AMYLOIDOSIS: SURVIVAL RESULTS FROM A SINGLE CENTER

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Background: AL amyloidosis is a progressive and life-threatening disease with multiorgan involvement, caused by neoplastic plasma cell clones. High dose melphalan with autologous stem cell transplantation (ASCT) remains a backbone of treatment in fit patients.

Objectives: The aim of the study was to describe hematologic and organ responses, as well as survival of patients with AL amyloidosis treated with ASCT in our center.

Methods: This retrospective analysis included all consecutive pts who underwent ASCT at our center due to AL amyloidosis in years 2001 - 2019. The diagnosis was confirmed by tissue biopsy, amyloid typing was done by immunohistochemistry. The ASCT eligibility criteria were ejection fraction >55%, NT-pro-BNP <5000 pg/mL, troponin T <0.01 ng/mL and systolic blood pressure ≤90 mmHg. Kaplan-Meier curves were used to estimate survival.

Results: The study included 37 pts (mean age 54.8 years, range 37-69). Mayo 2012 stage was 1-2 in 22 (59%) and 3-4 in 15 (41%). Mean number of involved organs was 2 (range 1-3), cardiac involvement was present in 15 (40%), renal involvement in 29 (78%). Median difference between involved and uninvolved free light chain (dFLC) was 101 mg/l (range 5.2-11384). Mean bone marrow plasma cell percentage was 18% (range 0-75%). Four pts (10.8%) had symptomatic multiple myeloma. Five pts (13.5%) required hemodialysis prior to ASCT. Tandem ASCT was performed in 5 pts. Induction treatment was instituted in 20 pts (54%), and was mainly based on bortezomib. Four pts (10.8%) died before day +100 after ASCT due to cardiovascular events. Twenty three out of 27 evaluable pts (85%) achieved response to treatment, including 17 pts (63%) with CR and 5 pts (18.5%) with PR, whereas 4 patients (15%) had stable disease. Organ response was achieved by 13 pts (48%). After a mean observation time of 4.2 years median OS was not reached, whereas median PFS was 4.06 years. Twelve out of 29 evaluable patients (41.4%) had disease progression after a median observation time of 2.74 years (range 0.9 – 5.7), and half of them (6 pts) died due to progressive disease after a median of 2.3 years post ASCT (range 1.1 – 6.4). Longer PFS was observed in patients achieving hematologic CR (HR = 0.15; P = 0.007), hematologic PR (HR = 0.1356; P = 0.001), as well as organ response (HR = 0.3; P = 0.046). Higher risk of progression was seen in patients with heart involvement (HR = 3.77; P = 0.006). Factors adversely correlated with OS were heart involvement (HR = 10.7; P = 0.002), Mayo Stage > 2 (HR = 7.2; P = 0.003) and symptomatic involvement of more than two organs (HR = 8.8; P = 0.04).

Conclusions: Our data confirm that ASCT is feasible and efficacious in the treatment of AL amyloidosis. Heart involvement remains the main predictor of death. Prospective studies are warranted to better delineate the role of induction and melphalan dose.
COMPLETE HEMATOLOGIC RESPONSE AT 6 MONTHS AFTER HIGH DOSE MELPHALAN AND STEM CELL TRANSPLANTATION IS ASSOCIATED WITH PROLONGED OVERALL SURVIVAL AND HIGHER RATES OF BIOMARKER-BASED ORGAN RESPONSE IN AL AMYLOIDOSIS

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Introduction: Organ involvement and dysfunction in AL amyloidosis predicts for survival and morbidity, in addition to impacting quality of life. Current therapies are directed towards the underlying plasma cell dyscrasia and organ response following such treatments are not clearly reported. Moreover, organ response criteria have evolved since 2012 (Palladini et al, J Clin Oncol. 2012, Palladini et al, Blood. 2014). Here, we sought to evaluate the impact of hematologic response at 6 months after initial therapy with high dose melphalan and autologous stem cell transplantation (HDM/SCT) on overall survival and more importantly organ response per the contemporary validated criteria.

Methods: We retrospectively analyzed patients with AL amyloidosis treated with HDM/SCT between 2002 and 2015, and evaluated hematologic response according to the 2012 classification criteria defining complete response (CR), very good partial response (VGPR), partial response (PR) and no response (NR). Organ responses were defined by >30% reduction in 24 hours proteinuria in the absence of a decrease in estimated glomerular filtration rate ≥25% (Palladini et al, Blood. 2014), >30% reduction of BNP (Lilleness et al, Br J Haematol. 2019) and >50% decrease in alkaline phosphatase for renal, cardiac and hepatic involvement, respectively. Patients with previous lines of treatment before HDM/SCT, patients deceased or lost to follow-up in the first 6 months following SCT, patients requiring hemodialysis, and patients treated with organ transplantation were excluded.

Results: 184 out of 384 patients treated with HDM/SCT between 2002 and 2015 met the inclusion criteria of this study. 75 patients were women (41%) and the median age was 57 years (range, 32-77). AL amyloidosis isotype was lambda in 82.5%. Of the 184 patients, 117 patients (64%) had renal involvement with 24 hour proteinuria >500 mg, 70 (38%) had cardiac involvement with BNP >150 pg/mL and 26 (14%) had liver involvement with Alk Phos >1.5 ULN. At 6 months after HDM/SCT, 29.5%, 45.4%, 12.5% and 12.5% of patients achieved hematologic CR, VGPR, PR or NR, respectively. Patients in hemCR had significantly prolonged overall survival (median 8.92 years) in comparison to patients achieving VGPR, PR or NR, respectively. Patients in hemCR had significantly prolonged overall survival (median 8.92 years) in comparison to patients achieving VGPR (median 5.93 years), PR (median 3.21 years) or NR (median 3.52 years) (p<0.0001). Organ responses were more in those achieving hemCR compared to those not achieving hemCR. Renal response was achieved by 80% vs 50% for those who achieved a hemCR vs non-CR (p=0.016). Cardiac response, similarly, was achieved by 90% vs 53% for those who achieved a hemCR vs non-CR (p=0.006). Liver response was experienced by 63% vs 25% for those achieving hemCR vs non-CR (p=0.09).

Conclusion: Complete hematologic response at 6 months after frontline HDM/SCT confers prolonged overall survival and is associated with higher rates of renal, cardiac and liver response according to the most recent biomarker based organ response criteria. Our data suggest that complete hematologic response at 6 months should be considered a therapeutic goal to obtain higher rates of organ response and prolonged overall survival. Prospective studies are needed for validation.
PRIMARY TREATMENT OF LIGHT CHAIN (AL) AMYLOIDOSIS WITH BORTEZOMIB, LENALIDOMIDE AND DEXAMETHASONE (VRD): UPDATED RESULTS

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4 1st Department of Pathology, National and Kapodistrian University of Athens.

Background: Bortezomib/dexamethasone with either cyclophosphamide (CyBorD) or melphalan (BMDex) are the most used primary regimens for AL amyloidosis patients. Combinations of bortezomib with an IMiD [thalidomide (VTD) or lenalidomide (VRD)] are widely used in newly diagnosed myeloma patients, with higher response rates than bortezomib combinations with alkylators. However, the use of VRD in patients with AL amyloidosis is far less common with limited published data. Herein, we present an update or our previous study with VRD with a longer follow up.

Aim: to evaluate efficacy and toxicity of VRD as primary therapy in patients with AL amyloidosis.

Methods: VRD (28-day cycles, for 8 cycles) included bortezomib SC (1.3 mg/m² on days 1, 8 & 15), with lenalidomide (5-15 mg) on days 1-21 but starting at 5 mg in patients with any of the following: eGFR<50 ml/min, proteinuria ≥5 gr/d and/or serum albumin <2.5 gr/dl, age >75 years, Mayo stage 2 or 3 with NTproBNP>4000 pg/ml; dexamethasone 20 mg was given weekly.

Results: 34 patients were treated with VRD; median age was 66.5 years (range 46-84), 79% had cardiac involvement, median NTproBNP was 3649 pg/ml (81->30000) and per Mayo stage 14%, 54%, 14% and 18% were rated as stage 1, 2, 3A and 3B respectively; 54% had renal involvement, median eGFR was 59 ml/min/1.73 m² (range 10-133) and renal stage distribution was 13%, 53% and 33% for stages 1, 2 & 3. Twenty-two patients completed the planned 8 cycles; 9 patients died prior to completion of therapy. After the first cycle of VRD, 24/34 (70.5%) patients had achieved a hematologic response (42% ≥VGPR and 27% a PR). On intent to treat, hematologic response rate was 88% (CR in 32%, VGPR in 50%, PR in 7%); among CR patients 5/11 were tested as MRD negative at 10-5 and among those in VGPR, 9/17 had dFLC <10 mg/L. All patients with t(11;14) (N=7) achieved ≥VGPR (CR: 3, VGPR: 4). Median follow up of living patients is 25 months; renal response was documented in 47% and heart response in 48%. Only 3 patients have relapsed and in one patient second line therapy started for inadequate response; 12-month and 24-month survival is 71% (100%, 85%, 71% and 20% for stage 1, 2, 3A & 3B patients respectively). Hematologic toxicity of VRD was mild; non hematologic toxicities were more common and included rash (Gr≥3:15%), infections (≥Gr3:9%), constipation (≥Gr3:9%), neuropathy (Gr2:20%). When compared to VCD/CyBorD treated patients matched for Mayo stage and baseline dFLC, there was a trend for deeper responses at 3 months with VRD and higher ≥VGPR rate at 6 months (p=0.049). Renal responses were observed in 31% and cardiac responses in 38% of CyBorD/VCD treated patients.

Conclusions: VRD with weekly bortezomib and low dose lenalidomide is an effective, rapidly acting regimen inducing deep hematologic responses, including MRDneg CRs and organ responses. The toxicity of VRD, however, is higher than CyBorD/VCD and patients need close follow up and appropriate interventions.
AL AMYLOIDOSIS WITH DEEP REMISSION: MASS FIX NEGATIVE AND MARROW MRD NEGATIVE AFTER THERAPY WITH MELPHALAN, DEXAMETHASONE, BORTEZOMIB, AND VENETOCLAX

K. Barton¹, N. Leung², P. Hagen¹, M. Picken¹

¹Loyola University Medical Center.

A 51-year-old female with a history of hypertension presented with proteinuria and underwent renal biopsy in January 2016. The biopsy demonstrated AL-lambda light chain amyloidosis involving glomeruli, blood vessels, and interstitium. Abdominal fat biopsy was also Congo red positive. Liquid chromatography tandem mass spectroscopy detected a peptide profile consistent with AL (lambda) - type amyloid deposition. Serum immunofixation did not show any monoclonal paraprotein. Serum free light chain studies showed kappa 1.14 mg/dL (0.3300 - 1.94 mg/dL), lambda 3.69 mg/dL (0.5700 - 2.63 mg/dL), ratio 0.3089 (0.26-1.65). 24-hour urine showed 4 g proteinuria. Bone marrow examination showed 7% lambda restricted plasma cells. FISH demonstrated monosomy 13 and t (11;14). Cardiac markers showed undetectable troponin T < 0.01 ng/ml, and NT proBNP 232 pg/ml (normal < 150). Echocardiogram reported mild increase in LV wall thickness, but the strain was not suggestive of cardiac amyloid.

The patient began therapy with melphalan and dexamethasone in April 2016. Stem cells were collected after 2 cycles of therapy. Bortezomib was added to melphalan and dexamethasone in November 2016 because of lack of response. The patient’s serum free light chain studies showed mild but persistent elevation of the lambda free light chain. The patient’s 24-hour urine studies continued with about 4 g of proteinuria. Melphalan was held after a pneumonia.

Venetoclax 400 mg daily was added to bortezomib and dexamethasone in August 2017. She tolerated this therapy well. Her serum free light chain studies showed slow steady improvement of both the lambda free light chain and the kappa/lambda ratio. 24-hour urine studies started to show improvement of her proteinuria.

Serum mass fix was negative in September 2018. Her bortezomib was stopped. Venetoclax was continued. Antibiotic prophylaxis was begun. She continued with improved light chain studies and improved 24-hour proteinuria of 988 mg in April 2019.

She underwent bone marrow examination in June 2019. Plasma cells were 2% and kappa and lambda ISH showed a polyclonal pattern. Cytogenetic and FISH studies did not detect any abnormalities. The patient’s bone marrow was MRD (minimal residual disease) negative by flow at 10-5. Venetoclax was stopped.

Venetoclax may have contributed to the depth of this patient’s remission. This patient provides an interesting case for discussion regarding the role of sensitive paraprotein studies such as mass fix and MRD testing in patients with AL amyloidosis. We will plan for some discussion regarding this issue.
AUTOLOGOUS STEM CELL TRANSPLANTATION CANDIDACY IN PATIENTS WITH NEWLY DIAGNOSED SYSTEMIC LIGHT-CHAIN AMYLOIDOSIS

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3 Nephrology and Renal Transplantation Department, Amyloidosis and Myeloma Unit, CSUR en Enfermedad Glomerular Compleja del Sistema Nacional de Salud, Hospital Clinic - Universitat de Barcelona, Spain.  
4 Pathology Department, Amyloidosis and Myeloma Unit, CSUR en Enfermedad Glomerular Compleja del Sistema Nacional de Salud, Hospital Clinic - Universitat de Barcelona, Spain.  
5 Immunology Department, Amyloidosis and Myeloma Unit, Hospital Clinic - Universitat de Barcelona, Spain.  
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7 Amyloidosis and Myeloma Unit, Fundació Clinic, Barcelona, Spain.  
8 Hematology Department, Bone Marrow Transplant Unit, Hospital Clinic, IDIBAPS, Universitat de Barcelona, Spain.

Introduction: High-dose melphalan (HDM) followed by autologous stem cell transplantation (ASCT) has shown its potential to obtain deep and durable hematologic responses, a high organ response rate and prolonged overall survival in systemic light chain (AL) amyloidosis. However, the procedure portends an elevated transplant-related mortality (TRM). Although TRM has decreased over time based on the acquisition of experience in transplanting centers, patient’s selection remains critical and limits the applicability of this effective therapy.

Objective & Methods: The aim of our study was to investigate the real proportion of patients able to receive HDM in our center as well the reasons for being considered a non-transplant candidate. Therefore, we analyzed a series of patients consecutively diagnosed with a systemic AL amyloidosis and evaluated for therapy at our institution from January 2008 to March 2019.

Results: In the study period, 130 patients with a newly diagnosed systemic AL amyloidosis (median age: 65 years; 53% women) were evaluated to receive therapy at our institution. Light-chain isotype was lambda in 76% of them, with a median involved serum free light chain (FLC) of 340 mg/L and bone marrow plasma cell infiltration of 10%. Heart involvement was present in 72% of patients and 38% were classified as revised Mayo stage III-IV. In our series, 35 patients (27%) received an ASCT while 95 (73%) were considered as non-transplant candidates. Among the latest, 41 were not selected for HDM because of their age over 70 years. In the remaining 54 patients, reasons for exclusion from the transplant program were: advanced amyloid cardiac disease in 36 (67%), severe renal failure or gastrointestinal involvement in 6 and 3 respectively, age over 65 and clinical trial availability in 3, patient’s decision in 3, severe macroglossia in 2 and lung involvement in one. These patients were treated with combinations of bortezomib, alkylating agent and corticosteroid in most of the cases (CyBorD regimen in 65% of them) and, in an intention-to-treat analysis, the overall hematologic response rate (partial or better) was 72%. Mortality rate during first year after diagnosis was 24% in this population (13/54), resulting in an event-free survival and overall survival of 15 and 37 months, respectively, as compared to 48 and 116 months, respectively, in the 35 patients who received an ASCT in the same period of time.

Conclusions: In our series, 27% of patients with newly diagnosed AL amyloidosis received an ASCT and had a favorable outcome. Among the remaining patients, 57% were potential transplant candidates based on their age but did not receive HDM, mainly due to an advanced amyloid cardiac disease. Most of these patients received bortezomib-based therapies and had poor survival outcomes due to an elevated early mortality.
ARE BORTEZOMIB-BASED REGIMENS ABLE TO CHANGE TRANSPLANT-CANDIDACY STATUS IN PATIENTS WITH SYSTEMIC LIGHT CHAIN (AL) AMYLOIDOSIS?

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Introduction: Immunoglobulin light chain (AL) amyloidosis is a plasma-cell disorder characterized by organ deposition of monoclonal light chain-derived amyloid fibrils. Based on the well demonstrated efficacy of high-dose melphalan (HDM) followed by autologous stem cell transplantation (ASCT), this procedure has become the standard of care for patients who are considered transplant-candidates based on their age and absence of significant organ involvement, particularly of the heart. For young patients who are considered non-transplant candidates, a bortezomib-based regimen without oral melphalan is preferred in order to avoid stem cell damage. However, there is scarce information on the ability of these regimens to change the transplant candidacy status in this population.

Objective & Methods: The aim of our study was to investigate the real ability of bortezomib-based regimens to change the transplant candidacy status of patients with newly diagnosed systemic AL amyloidosis up to 70 years of age but considered non-transplant candidates. For this purpose, we retrospectively analyzed a series of patients consecutively treated at our institution from January 2008 to March 2019.

Results: In the study period, 130 patients with newly diagnosed systemic AL amyloidosis were evaluated to receive treatment at our institution and 89 of them were younger than 70 years and therefore potential candidates to receive an ASCT. Fifty-nine of these patients (66%) received a bortezomib-based regimen as first-line therapy, being the most commonly used the combination of cyclophosphamide, bortezomib and dexamethasone (CyBorD) (46 patients), followed by bortezomib plus dexamethasone (BorD) (7), BorD plus melphalan (3) and oral melphalan combined with prednisone and bortezomib (MPV) (3). After a median of 6 cycles of therapy (range, 1-17), only 7 out of the 59 patients (11.8%) changed their transplant-candidacy status. Hematologic response was observed in all 7 patients, with 2 complete responses (CR), 4 very good partial responses and 1 partial response. Moreover, organ responses were observed in all of them, with 5 renal, 3 cardiac and 2 liver responses, as well as resolution of severe bleeding in one patient. One of these patients also received a heart transplant after CyBorD and before ASCT. Four of the 7 patients received intermediate dose of melphalan (140 mg/m2) and all achieved a hematologic CR after transplant. Five of them remain alive at the last follow-up, with a median overall survival of 57 months.

Conclusions: A small proportion of patients (11.8%) with AL amyloidosis not initially candidates for ASCT become eligible after first-line therapy with a bortezomib-based regimen, high-dose therapy resulting in great outcomes.
WHEN TO PERFORM AUTOLOGOUS STEM CELL TRANSPLANTATION WITH NO NEED OF INDUCTION THERAPY IN SYSTEMIC LIGHT-CHAIN (AL) AMYLOIDOSIS?

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Introduction: Immunoglobulin light chain (AL) amyloidosis is a plasma-cell disorder characterized by organ deposition of monoclonal light chain-derived amyloid fibrils. Given the high hematologic response rates achieved with bortezomib, CyBorD and other bortezomib-containing regimens are being extensively used as first line therapy before high-dose melphalan (HDM) and autologous stem cell transplantation (ASCT). However, few studies have addressed the role of induction therapy before transplant in this disease or its potential clinical indications according to the disease characteristics.

Objective & Methods: The aim of our study was to analyze the outcome of patients who received ASCT without previous therapy based on a low-burden disease defined by a difference between involved and uninvolved serum free light chain (FLC) ≤ 100 mg/L and bone marrow plasma cell infiltration ≤ 10%, as well as absence of progressive organ damage by amyloid. For this purpose, we retrospectively analyzed a series of patients consecutively treated at our institution from January 2008 to March 2019.

Results: In the study period, 130 patients with newly diagnosed systemic AL amyloidosis were evaluated for treatment at our institution and 35 of them received an ASCT according to the selection criteria at the time of first consultation. Seven (20%) of these 35 patients presented at diagnosis low-burden disease as defined before and did not receive induction therapy before transplant. The median age of these 7 patients was 56 years (range, 49-60). Light-chain isotype was lambda in 6 of the 7 patients, with a median involved serum FLC of 26 mg/L (range, 0-82) and bone marrow plasma cell infiltration of 6% (range, 1-10). Renal involvement was present in 6 patients (86%) and was the primary organ involved in 5 (isolated in 3 of them), while heart and lung were the primary organs involved in the remaining two patients. Cardiac involvement was present in 3 patients (43%). All were classified as revised Mayo Stage I except one patient with stage II. Median time from diagnosis to ASCT was 4.5 months (range, 1.6-11.9), with the longest time being 11.9 months in a patient who received a heart transplant before HDM. All patients achieved a hematologic response after transplant, including 5 complete responses (CR), one very good partial response and one partial response. Organ responses were also obtained in all except 1 patient. After a median follow up of 66 months, 3 patients have relapsed (2 of them were subsequently treated and obtained a second CR) and one patient died during a second ASCT.

Conclusions: A subgroup of selected patients with newly diagnosed systemic AL amyloidosis and low-tumor burden can benefit from an ASCT without induction therapy, avoiding unnecessary induction-related toxicity and preserving bortezomib-based therapies for future phases of the disease.
EFFICACY OF MAINTENANCE IN PATIENTS WITH SYSTEMIC LIGHT CHAIN AMYLOIDOSIS; SINGLE CENTRE RETROSPECTIVE ANALYSIS

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Introduction: Amyloidosis is the generic term that refers to extracellular tissue deposit of misfolded proteins, and light chain amyloidosis (AL) is the most common form of amyloidosis. The main objective of treatment is to target the plasma cells responsible for immunoglobulin light chain production and to achieve the best possible haematological response. Deeper haematological responses are associated with better organ responses and overall survival. Despite the chosen first line of treatment, a group of patients will not achieve a deep response.

Objectives: Investigate the efficacy of maintenance after first line of treatment in patients with AL.

Methods: Records of patients diagnosed with AL between 2000 and 2018 were reviewed. Patients included in the analysis were those who did not achieve at least a very good partial hematological response and those considered candidates for maintenance therapy by treating physician after first line of treatment. For analysis purposes the patients were divided in two groups according if they received maintenance or not. Subjects with more than 30 percent of plasma cell bone marrow infiltration or those with myeloma features were excluded from the analysis.

Results: 38 patients were included in the present analysis with a median age at diagnosis of 55 years (30-85) with female predominance (53 vs. 47 percent). 68% presented with more than 2 organs involved at diagnosis being kidney (45%) followed by heart (37%) the most frequent involved organs. Of the 38 patients, 21 patients received maintenance and 17 were considered candidates for maintenance but did not received it. All age variables such as gender, number of organs involved, mayor organs involved, BNP and free light chain difference were very similar in both groups with non-significant P values for all variables. In the maintenance group, as first line of therapy, 52% of patients received proteasome inhibitor (PI), high dose chemotherapy and autologous stem cell transplant (HDT-ASCT) 38% and 10% received alkylating agents. In the non-maintenance group as first line therapy 29% received PI, 58% HDT-ASCT and 13% alkylators. The difference was non significant between both groups. Regarding the haematological response to first line therapy in both groups in the maintenance arm no patient achieved complete response (CR), 29% achieved a very good partial response (VGPR), 38% partial response (PR) and 33% stable disease (SD). In the non-maintenance group, no patient achieved a CR, 24% VGPR, 65% PR and 12% SD. The difference between groups was non-significant P=0.2. In the maintenance group 45% received PI based maintenance, 45% immunomodulatory drugs (IMiDs) and 10% alkylator based. In terms of outcome in the maintenance group were 11 (52%) events. The median and range of the follow-up times was 30.8 (9.9-111.2) months. The median progression free survival (PFS) was 61.6 with 95% confidence Interval (CI) (9.2,80.7). In the non-maintenance group were 14 (82%) events. The median and range of the follow-up times is 20.2 (1.8-72.5) months. The median PFS time was 30.6 with 95% CI (5.9,30.6). The difference in both groups was of statistical significance with a P value=0.0045 favouring the maintenance group. For the overall survival (OS) the difference was not significant for both groups.

Conclusion: Patients that received maintenance have a significant longer PFS compared to those patients that did not. OS was similar in both groups.
The depth of response prior to autologous stem cell transplant predicts survival in AL amyloidosis

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Introduction: The role of induction therapy prior to autologous stem cell transplant (ASCT) in immunoglobulin light chain (AL) amyloidosis remains controversial. Data on the prognostic impact of response to induction in a transplanted cohort are lacking.

Objectives: The aim of this study was to assess the impact of response to induction therapy on progression-free survival (PFS) and overall survival (OS) in patients undergoing ASCT for AL amyloidosis.

Methods: We conducted a retrospective study of all newly diagnosed AL amyloidosis patients who received induction treatment prior to ASCT between January 2007 and August 2017 at Mayo Clinic, Rochester, Minnesota. Patients receiving only corticosteroids prior to transplant were excluded as were those with an involved light chain of less than 5 mg/dL (not measurable for response). Response assessment was according to consensus criteria. PFS was calculated from day 0 of ASCT to biochemical progression, initiation of a second-line treatment or death from any cause. OS was defined from day 0 of ASCT until death from any cause. Survival curves were calculated by JMP software using Kaplan-Meier statistics.

Results: Of the 376 patients who were transplanted during the study period, 242 were excluded due to no pre transplant induction or treatment with steroids only. 134 patients met inclusion criteria for the study and were included in the analysis. The median age at diagnosis was 60 (range 36-74), 63% were men. Patients’ characteristics are listed in table 1. The most common induction regimen used was proteasome inhibitor-based (n=98, 74 received bortezomib, cyclophosphamide, dexamethasone, 24 received bortezomb and dexamethasone). The overall response rate to induction was 83%, complete response 17%, VGPR 30% and PR 36%. With a median follow up of 56.5 months for the surviving patients, the median PFS and OS for the whole cohort in months was 48.5 (interquartile range: 36.8-63.7) and not reached (interquartile range: 127-not reached), respectively. Response depth to induction therapy was associated with improved PFS and OS. The median PFS was not reached for patients achieving CR/VGPR prior to ASCT and 33.8 for patient achieving PR or less prior to ASCT (P=0.001). The median OS was not reached for patients achieving CR/VGPR prior to ASCT and 128 months for patients achieving PR or less prior to ASCT (P=0.02). Four patients died within 100 days of transplant corresponding to a treatment related mortality (TRM) of 3%. No difference was found between the different induction regimens given prior to ASCT on OS or PFS. On univariable analysis, Mayo stage, age and depth of response prior to ASCT were predictive of PFS while the conditioning melphalan dose, age and depth of response prior to ASCT were predictive of OS. On multivariable analysis, the only predictor of PFS was depth of response prior to transplant (RR 9.7; P=0.002). Predictors of OS include conditioning dose (RR 4.7; P=0.03) and depth of response prior to transplant (RR 5.3; P=0.02).

Conclusions: Hematologic response prior to transplant predicts post transplant outcomes in patients with AL amyloidosis.
Table 1:

<table>
<thead>
<tr>
<th>Variable</th>
<th>Cohort n=134</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, median years (range)</td>
<td>60 (36-73)</td>
</tr>
<tr>
<td>Male, n (%)</td>
<td>85 (63)</td>
</tr>
<tr>
<td>Organ involved, n (%)</td>
<td></td>
</tr>
<tr>
<td>Cardiac</td>
<td>71 (53)</td>
</tr>
<tr>
<td>Renal</td>
<td>67 (50)</td>
</tr>
<tr>
<td>&gt;2</td>
<td>26 (19)</td>
</tr>
<tr>
<td>% Plasma cells at diagnosis, n (%)</td>
<td></td>
</tr>
<tr>
<td>&lt;10%</td>
<td>61 (46)</td>
</tr>
<tr>
<td>≥10%</td>
<td>71 (54)</td>
</tr>
<tr>
<td>Light Chain type</td>
<td></td>
</tr>
<tr>
<td>Lambda</td>
<td>80 (60)</td>
</tr>
<tr>
<td>Kappa</td>
<td>54 (40)</td>
</tr>
<tr>
<td>Mayo 2012 Stage, n (%)</td>
<td></td>
</tr>
<tr>
<td>I</td>
<td>42 (32)</td>
</tr>
<tr>
<td>II</td>
<td>55 (42)</td>
</tr>
<tr>
<td>III</td>
<td>21 (16)</td>
</tr>
<tr>
<td>IV</td>
<td>11 (8)</td>
</tr>
<tr>
<td>Induction Treatment, n (%)</td>
<td></td>
</tr>
<tr>
<td>PI based</td>
<td>98 (73.1)</td>
</tr>
<tr>
<td>IMiD based</td>
<td>15 (11.2)</td>
</tr>
<tr>
<td>PI+IMiD</td>
<td>11 (7.5)</td>
</tr>
<tr>
<td>Melphalan based</td>
<td>7 (5.2)</td>
</tr>
<tr>
<td>Other</td>
<td>3 (3)</td>
</tr>
<tr>
<td>Conditioning regimen and dose, n (%)</td>
<td></td>
</tr>
<tr>
<td>200 mg/m²</td>
<td>94 (70)</td>
</tr>
<tr>
<td>140 mg/m²</td>
<td>37 (28)</td>
</tr>
<tr>
<td>BEAM</td>
<td>3 (2%)</td>
</tr>
<tr>
<td>Response to induction</td>
<td></td>
</tr>
<tr>
<td>CR</td>
<td>23 (17)</td>
</tr>
<tr>
<td>VGPR</td>
<td>40 (30)</td>
</tr>
<tr>
<td>PR</td>
<td>49 (36)</td>
</tr>
<tr>
<td>SD</td>
<td>16 (12)</td>
</tr>
<tr>
<td>PD</td>
<td>6 (5)</td>
</tr>
</tbody>
</table>

n=number; PI=proteasome inhibitors; IMiD=immunomodulatory drugs; CR=complete response; VGPR= Very good partial response; PR= partial response; SD=stable disease; PD=progressive disease.

**Keywords:** AL amyloidosis, Stem cell transplant, response.
LOCALIZED AL AMYLOIDOSIS: CLINICAL ASPECTS, BIOCHEMISTRY AND LONG-TERM FOLLOW-UP

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Introduction: AL amyloidosis may be a systemic disease, significantly associated with plasma cell dyscrasia or lymphoproliferative disease, or occur in a localized form with predilection for specific organ systems, notably throat and tongue, ocular, skin, breast, lung, and genitourinary system. Localized AL has been the subject of two large reviews, with mass spectroscopy revealing distinct patterns of V-region gene usage compared to systemic AL (Mahmood, S et al, 2015; Kourelis, TV et al 2017). In particular, these presentations of AL may be benign, with symptomatology that remains static over long periods of time. Pathogenesis has been linked to plasmacytic infiltrates contiguous to sites of deposition which may share κ/λ specificity. Although in most cases a kappa or lambda predominance is apparent, there have been reported instances in which both forms of light chain may constitute the amyloid.

Materials and Methods: The core of this series includes 30 cases of nasopharyngeal amyloidosis seen, evaluated and followed over the past 25 years, but extends also to localized AL in skin (10), breast (2), GU tract (7), eye (4), lung (11) and joint (1). Amyloid was extracted from fresh or formalin-fixed tissue and identified as kappa or lambda light chains by N-terminal sequencing of tryptic peptides or mass spectroscopy after laser capture microscopy, the latter also to identify the predominance of light chain sequences and co-deposition of P-component and apolipoproteins.

Results: Laryngeal amyloids had an equal gender distribution and median age ~50 at presentation. Supraglottic involvement was most common, with extra-laryngeal subsites at presentation including nasopharynx, oropharynx and trachea. Following diagnosis, and in some cases debulking, patients remained stable for follow-up ranging up to 20 years, though a tendency for long-term recurrence was noted, even with initial complete excision. Two required treatment for associated plasmacytomas and one laryngeal carcinoma; no patient developed systemic AL. An association between localized AL and Primary Sjogrens syndrome (SS) was identified in 6 patients, with cutaneous disease in 3, lung in 2 and breast in one; one patient subsequently developed systemic AL and was treated for multiple myeloma. Among these patients, 5 were λ and one κ by sequence analysis. Overall, κ/λ was 1.3/1, in contradistinction to the 2/1 λ predominance seen in systemic AL; in two instances-one laryngeal, one tracheobronchial-MS yielded both κ and λ predominant light chains.

Discussion: Localized AL may develop in a variety of clinical settings, and has also been described in other animal species; pulmonary amyloidomas were among the first forms of AL to be subjected to light chain sequence analysis. An association with SS has been corroborated in other reports, and may be a consideration in evaluating nodular cutaneous amyloid. Although biochemical analysis confirms clonality is most, though not all, instances, this is not reflected by serum/urine immunofixation or abnormal Ig free light chain (FLC) assays.
SAFETY, TOLERABILITY AND RESPONSE RATES OF DARATUMUMAB IN PATIENTS WITH RELAPSED LIGHT CHAIN (AL) AMYLOIDOSIS: RESULTS OF A PHASE II STUDY

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Background: Daratumumab is a human IgG1k monoclonal antibody targeting the CD38 surface antigen on plasma cells with proven efficacy in multiple myeloma. Infusion-related reactions (IR) of 48% are reported in patients who had daratumumab as monotherapy for relapsed multiple myeloma. Therefore, we designed a clinical trial to study tolerability of daratumumab in those with relapsed AL amyloidosis (NCT02841033).

Objectives: The primary objective was to determine the safety and tolerability of infusion of daratumumab, with respect to IR. The secondary objectives were to assess hematologic response, clinical response and time to next treatment.

Methods: Accrual began in April 2017. Patients with AL amyloidosis after ≥1 prior therapy, and eGFR of >20 mL/min, AST/ALT < 3xULN, NT-proBNP ≤ 8500 pg/mL, LVEF ≥ 30%, FEV1 ≥ 50% in patients with COPD or chronic smokers, and ECOG performance status of ≤ 3, received daratumumab at 16 mg/kg IV infusion weekly for weeks 1-8, followed by every 2 weeks for weeks 9-24 and every 4 weeks thereafter until progression or unacceptable toxicity, for up to 24 months. The first infusion was given in 1000 mL, the second infusion in 500 mL if no grade 1 or greater reactions occurred, and subsequent doses were administered in 500 mL of saline. All patients received acetaminophen, diphenhydramine, loratadine, famotidine, montelukast and methylprednisolone (100 mg for first 2 infusions and 60 mg thereafter) 30-60 mins prior to infusion. Ondansetron was added after development of grade 1 nausea/vomiting in the first 2 patients. Diphenhydramine, famotidine and methylprednisolone (40 mg) were also administered 2 hrs after start of infusion during the first 2 infusions even if there was no reaction. Methylprednisolone 20 mg (or its equivalent) and montelukast were given 24 and 48 hrs after start of infusion for the first 2 infusions and then it was optional. All patients received prophylaxis with acyclovir.

Results: At data cut-off (Aug 1, 2019) 25 patients were screened and 22 enrolled. The median age was 63 (range, 42-84), and the median of prior therapies was 2 (range, 1-7): 16 (68.1%) had received SCT, 9 (40.9%) immunomodulatory agents, and 16 (72.7%) proteasome inhibitors. Eleven (50%) patients were refractory to prior therapy and median time from last therapy was 9 months (range, 1-180). The median time from diagnosis to enrollment was 48 months. The median number of organ systems involved was 2 (range, 1-5): 11 (50%) had involvement of ≥ 2 organ systems, 20 (90.9%) had cardiac biomarker stage II or III disease. Median NT-proBNP level was 1264 pg/mL (range, 32-3962) and urine protein excretion was 0.53 g/24 h (range, 0-10.1), while median dFLC was 80.7 mg/L (range, 2.9-854). Seven patients remain on study and 594 infusions have been completed. The median number of infusions received per patient is 31 (range, 7-34). Nine patients have completed 24 months of protocol directed therapy, 3 were removed from the protocol due to progression of AL amyloidosis, 2 were removed after 8 cycles due to patient choice, and 1 removed due to persistent grade 3 adverse event of muscle weakness. No patient experienced a grade 3-4 IR. Four (19%) patients experienced grade 1 nausea and vomiting during initial infusion, which resolved after an antiemetic. The median time of first infusion was 7 hrs and 2nd infusion was 4 hrs 29 mins. Grade 3/4 adverse events were noted in 20 (91%) patients. Respiratory illnesses were experienced by 13 (59%), 4 (18%) of which were grade 3 events (Influenza A, Rhino/Enteroviral infection, PCP). Nine (40.1%) patients were noted to be iron deficient and required parenteral iron infusion or oral supplementation. Hematologic responses were rapid as shown in figure. Hematologic CR and VGPR were achieved by 85.7% (18/21), 85.7% (18/21) and 94% (16/17) patients at 3 months, 6 months and 12 months respectively. Renal response was achieved by 66.7% (8/12) and cardiac response was achieved by 50% (7/14) of patients.

Conclusions: Daratumumab infusion is well tolerated in patients with AL amyloidosis when administered with appropriate supportive care. A rapid hematologic response after 1 dose of daratumumab is seen in patients with AL amyloidosis.
TREATMENT WITH DARATUMUMAB IN PATIENTS WITH RELAPSED/REFRACTORY AL AMYLOIDOSIS: A MULTICENTRIC RETROSPECTIVE STUDY

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5 Hematology Department, Hospital Clínico Universitario de Santiago-CHUS, Spain.
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10 Hematology Department, Hospital Universitario Vall d’Hebron, Barcelona, Spain.
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Background: Management of patients with relapsed or refractory (R/R) AL amyloidosis is complex. Some initial reports have shown positive results with daratumumab in heavily pre-treated AL amyloidosis patients.

Objective: To further evaluate the efficacy of daratumumab in patients with R/R biopsy proven AL amyloidosis.

Methods: In this multicentric study, we retrospectively analyzed a series of patients with R/R AL amyloidosis treated with daratumumab. Daratumumab was administered according to the approved posology for multiple myeloma.

Results: Thirty-eight patients (mean age 64±9 years) were included in the study. Cardiac and renal involvement was present in 76% and 74% of patients respectively, and 42% had 3 or more organs involved. According to the revised Mayo staging system, 16% of patients were stage I, 24% stage II, 32% stage III and 26% stage IV. Median number of previous lines of therapy was 2 (range 1-8). All patients had previously received bortezomib, 47% IMiDs and 39% had undergone an ASCT with melphalan-based conditioning. 37% of the patients had achieved a very good partial response (VGPR) or better with any of the previous lines of therapy. 63% were refractory to the last administered regimen. Median number of daratumumab infusions per patient was 18 (range, 1-38). 36 patients were evaluable for efficacy. Overall hematological response (HR) was 72%, including 28% complete responses (CR). The median time to first HR was 2 weeks. A high-quality response (≥ VGPR) was obtained in 65% of patients who had never achieved such depth of response previously. Cardiac and renal organ response rates were 37% and 59%, respectively. After a median follow-up of 10 months (range, 1-28) from the start of daratumumab, median progression-free survival (PFS) is 18 months and median overall survival (OS) has not been reached. OS and PFS at 6 months were 72% (95% CI 0.53-0.85) and 70% (95% CI 0.51-0.83). At 12 months, OS and PFS were 59% (95% CI 0.36-0.77) and 52% (95% CI 0.29-0.70). Regarding safety, in 5 patients treatment was stopped due to infusion reaction during the first administration (1 patient), infectious complications (3 patients), and a second neoplasm (breast cancer in a heavily pre-treated patient).

Conclusions: Daratumumab is a safe and effective drug in the treatment of R/R AL amyloidosis and should be considered early in the course of the disease.

Keywords: AL amyloidosis, relapsed/refractory, daratumumab.
FACTORS INFLUENCING RESPONSE TO DARATUMUMAB IN PATIENTS WITH RELAPSED/REFRACTORY AL AMYLOIDOSIS: RESULTS FROM A MULTICENTRIC RETROSPECTIVE STUDY

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4 Hematology Department, Hospital Universitario Donostia, San Sebastián, Spain.
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13 Hematology Department, Hospital Universitario de Canarias, Tenerife, Spain.
14 Hematology Department, Hospital Universitario Galdakao-Usansolo, Vizcay, Spain.
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Background: Management of patients with relapsed or refractory (R/R) AL amyloidosis is complex. Daratumumab is an attractive treatment alternative in this setting, as shown by several case series.

Objective: Our aim was to evaluate possible clinical variables that could influence the efficacy of daratumumab in patients with R/R biopsy proven AL Amyloidosis.

Methods: In this multicentric study, we retrospectively analyzed a series of patients with R/R AL amyloidosis treated with daratumumab. Daratumumab was administered according to the approved posology for multiple myeloma.

Results: Thirty-eight patients were included in the study. Cardiac and renal involvement were present in 76% and 74% of patients, respectively, and 42% of them had 3 or more organs involved. Median number of previous lines of therapy was 2 (range, 1-8). All patients had previously received bortezomib, 47% immunomodulatory drugs (IMiDs) and 39% had undergone an autologous stem cell transplant (ASCT). Median number of daratumumab infusions per patient was 18 (range, 1-38) and 36 patients were evaluable for efficacy. Overall hematological response (HR) rate was 72%, including 28% complete responses (CR) and 36% very good partial responses (VGPR). No significant differences in the overall HR or the rate of CR/VGPR was found depending on age (over or below 65 years), involved light chain isotype (kappa or lambda), type of monoclonal component (Bence-Jones or with a heavy chain), revised Mayo staging (I/II vs III/IV), number of involved organs (≥ or < 3), serum dFLC (> or ≤ 180 mg/L), pro-BNP levels (> or ≤ 5000 pg/mL), bone marrow plasma cells (≥ or < 10%), previous treatment with ASCT or IMiDs, achievement of a VGPR or better with any of the previous lines of therapy, or refractoriness to the last treatment received. Interestingly, HR was more frequent among patients that received daratumumab as second-line therapy compared to subsequent therapies: 12/13 (92.3%) vs. 14/23 (60.9%); p=0.06. HR was also more frequent in patients with glomerular filtration rate ≥45 mL/min: 15/17 (88.2%) vs 11/19 (57.9%); p=0.06.

Conclusions: Daratumumab is an effective drug in the treatment of patients with R/R AL amyloidosis. According to this exploratory analysis it should be considered early in the course of the disease. Further studies are needed to confirm other possible factors that could influence its efficacy.

Keywords: AL amyloidosis, relapsed/refractory, daratumumab.
DARATUMUMAB/BORTEZOMIB/DEXAMETHASONE AS SALVAGE THERAPY IN SIXTY-TWO PATIENTS WITH ADVANCED SYSTEMIC LIGHT-CHAIN AMYLOIDOSIS

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2 Department of Hematology, University Hospital Essen, Germany.
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4 Institute of Human Genetics, University Heidelberg, Germany.
5 Department of Internal Medicine V, Division of Hematology/Oncology, Heidelberg University Hospital, Germany.

Background: Daratumumab/bortezomib/dexamethasone (DVD) is FDA-approved as 2nd line treatment for multiple myeloma since 2016. A randomized phase 3 trial currently investigates the addition of subcutaneous daratumumab to the established 1st line regimen CyBorD in systemic light-chain amyloidosis (AL). Translocation t(11;14) has been reported as adverse for response to bortezomib in AL.

Objective: To evaluate efficacy and safety of DVD as salvage therapy for AL.

Methods: This is a retrospective analysis of 62 consecutive patients treated with DVD for AL starting from June 2017. Hematologic remission, cardiac and renal response were assessed according to consensus criteria. Patients with a dFLC of 20-50mg/l were assessed according to Milani et al. (Blood, 2017). Hematologic progression-free-survival (PFS) and overall survival (OS) were calculated from start of DVD. All times and continuous data are presented as median [range], if not stated otherwise.

Results: DVD was initiated 5.9 [0.2-105] months after start of 1st line therapy and 1.0 [0-72] months after end of last therapy. DVD was mostly utilized as 2nd (n=47) or 3rd (n=8) line regimen. 59 patients had been treated with bortezomib in the past, overall 27 of them were previously refractory to the drug. The median age was 60 [38-79] years and 53% of patients were male. Baseline bone marrow plasma cell percentage was 19.5% [8-81] and amongst 23 of the 43 analyzed patients t(11;14) was detected. Lambda subtype was present in 76%. A median of three [1-5] organs were involved, namely: heart (87%), kidney (65%), soft tissue (45%), liver (24%), GI tract (19%) and neuropathy (5%). Ahead of DVD, the NT-ProBNP level was 5.475 [78-87.654]ng/l, dFLC was 117 [0-1997]mg/l and eGFR in patients not on dialysis was 54 [11-106]ml/min/1.73m². Daratumumab 16 mg/kg body weight was applied intravenously after standard premedication eight times weekly, eight times every other week and every four weeks thereafter combined with weekly subcutaneous bortezomib 1.0 or 1.3mg/m² and dexamethasone. Hematologic remission and organ response rates are presented in Table 1. After a median follow-up time of 16 months median OS and median PFS had not yet been reached. Fourteen patients died from progressive AL and two from infectious complications. Fourteen patients still received DVD on data cut-off date, 7 patients were solely treated with daratumumab while another 23 patients were off therapy without progression. At 12-months 74% of patients were alive and 57% were without a hematologic event. Hematologic remission rates, OS and PFS showed no significant differences between patients with or without t(11;14). The most common complications were infections (29%, 16% grade 3/4), congestive heart failure (CHF, 10% grade 3/4), atrial fibrillation (6%) and infusion-related reactions (5%).

Conclusions: DVD is effective in AL and not adversely affected by t(11;14). Nevertheless, there is a substantial risk of serious infection and CHF.

Table 1:

<table>
<thead>
<tr>
<th>Time</th>
<th>VGPR rate</th>
<th>OR rate</th>
<th>CR rate</th>
<th>Organ response assessment in evaluable patients</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>cardiac</td>
</tr>
<tr>
<td>Best</td>
<td>63% (30/48)</td>
<td>75% (41/55)</td>
<td>24% (13/55)</td>
<td>response</td>
</tr>
<tr>
<td>3 months</td>
<td>54% (26/48)</td>
<td>66% (35/53)</td>
<td>11% (6/53)</td>
<td>39% (14/36)</td>
</tr>
<tr>
<td>6 months</td>
<td>51% (21/41)</td>
<td>61% (28/46)</td>
<td>20% (9/46)</td>
<td>46% (11/24)</td>
</tr>
</tbody>
</table>

CR, complete remission; OR, overall response; VGPR, very good partial response

Keywords: salvage therapy, Daratumumab/bortezomib/dexamethasone, translocation t(11;14)

Category: Treatment of relapsed/refractory AL amyloidosis
Venetoclax Shows High Efficacy in Relapsed/Refractory AL Amyloidosis: A Multicenter, International Retrospective Study

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7 Paris 7 University and APHP, Immuno-Hematology Department, Saint-Louis Hospital, Paris, France.
8 Amyloidosis Center, Boston University School of Medicine, Boston Medical Center, Boston, MA, USA.
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12 Department of Clinical Therapeutics, National and Kaposidrissian University of Athens, School of Medicine, Alexandra General Hospital, Athens, Greece.
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Introduction: Up to 60% of patients with AL amyloidosis (AL) harbor t(11;14), resulting in a higher BCL-2:MCL-1 ratio. Venetoclax (VCL), a small molecule BCL-2 inhibitor FDA-approved for use in CLL and AML, is under investigation in multiple myeloma (MM). Limited data from case reports show high efficacy of VCL alone or with proteasome inhibitors (PI) or daratumumab (dara). Here is the first systematic retrospective multicenter analysis of relapsed/refractory (RR) AL patients treated with VCL.

Objective: Our objective is to provide preliminary efficacy and toxicity data of VCL in RR AL to form a basis for future prospective trials.

Methods: Local IRB approval was obtained and de-identified data regarding efficacy and toxicity (among others) of RR AL patients treated with VCL-containing regimens were collected from 14 Amyloidosis centers in the US and Europe.

Results: We report pooled data from 24 patients (data of 35 patients is expected). Regimens included VCL +/- dex (45.8%), VCL + PI +/- dex (29.2%), VCL + dara +/- PI (12.5%), VCL + elotuzumab (Elo) + PI (4.2%), VCL + Cyclophosphamide (Cy) + PI (4.2%) and VCL + dara + lenalidomide (4.2%). Dosage for VCL ranged from 100-800mg po daily. Patient demographics are shown in Table 1; of note 15 patients (62.5%) harbored t(11;14). Overall response rate (ORR) for all patients was 75% (18/24 patients) with a ≥VGPR of 66.7%. For t(11;14) patients, the ORR was 86.7% (13/15 patients) with 12 patients (75%) achieving ≥VGPR (6 VGPR, 6 CR). Non-t(11;14) patients had an ORR of 55.6% (5/9 patients) with a ≥VGPR of 44.4% (2 VGPR, 2 CR) with an odds ratio for ≥VGPR of 0.2 (p = 0.084). Organ response was seen in 5/20 evaluable patients. Toxicity is noted in Table 2. Of note, one patient died of bacteremia as a consequence of therapy and one patient died of heart failure, not attributed to VCL. Toxicities were generally reversible and manageable.

Conclusion: This first time pooled analysis shows VCL-containing regimens are highly efficacious in RR AL with manageable toxicity, warranting further prospective study in RR AL.
Table 1. Demographics

<table>
<thead>
<tr>
<th>Sex</th>
<th></th>
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<tbody>
<tr>
<td>Male</td>
<td>16 (66.7%)</td>
</tr>
<tr>
<td>Female</td>
<td>8 (33.3%)</td>
</tr>
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</table>

<table>
<thead>
<tr>
<th>Light Chain</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Lambda</td>
<td>16 (66.7%)</td>
</tr>
<tr>
<td>Kappa</td>
<td>8 (33.3%)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Median age in years (range)</th>
<th>68.5 (49-83)</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;65</td>
<td>9 (37.5%)</td>
</tr>
<tr>
<td>65-75</td>
<td>10 (41.7%)</td>
</tr>
<tr>
<td>&gt;75</td>
<td>5 (20.8%)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>t(11;14) status</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>+</td>
<td>15 (62.5%)</td>
</tr>
<tr>
<td>-</td>
<td>9 (37.5%)</td>
</tr>
</tbody>
</table>

| Concurrent symptomatic MM | 4 (16.7%) |

| Median prior lines of therapy (range) | 4 (1-10) |

<table>
<thead>
<tr>
<th>Prior therapies</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Cy</td>
<td>19 (79.2%)</td>
</tr>
<tr>
<td>Melphalan</td>
<td>5 (20.8%)</td>
</tr>
<tr>
<td>Bendamustine</td>
<td>2 (8.3%)</td>
</tr>
<tr>
<td>PI</td>
<td>24 (100%)</td>
</tr>
<tr>
<td>Lenalidomide</td>
<td>17 (70.8%)</td>
</tr>
<tr>
<td>Pomalidomide</td>
<td>9 (37.5%)</td>
</tr>
<tr>
<td>Dara</td>
<td>18 (75%)</td>
</tr>
<tr>
<td>Elo</td>
<td>3 (12.5%)</td>
</tr>
<tr>
<td>Liposomal doxorubicin</td>
<td>1 (4.2%)</td>
</tr>
</tbody>
</table>

| Prior ASCT | 3 (12.5%) |
THE ROLE OF INDUCTION THERAPY BEFORE AUTOLOGOUS STEM CELL TRANSPLANTATION IN AL AMYLOIDOSIS PATIENTS WITH BONE MARROW PLASMA CELLS LESS THAN 10%

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Background: Nowadays, induction therapy before autologous stem cell transplantation (ASCT) is only recommended for immunoglobulin light chain (AL) amyloidosis patients with 10% or greater bone marrow plasma cells (BMPCs). The role of induction therapy before ASCT in patients with BMPC less than 10% still unknown.

Methods: In this study we evaluated the role of induction therapy and its impact on the outcome of ASCT in AL patients with BMPC less than 10%. A total of 227 patients with AL amyloidosis were included in this study. Among 227 patients 124 patients received bortezomib based induction prior to ASCT and defined as group A, 35 patients received other chemotherapeutic induction and defined as group B, and the other 68 patients without induction defined as group C.

Results: The hematologic ORR of the group A, B and C were 90.2% (111/123), 65.7% (23/35) and 74.2% (49/66), respectively. The CR rates of the group A, B and C were 48.8% (60/123), 25.7% (9/35) and 19.7% (13/66), respectively. Both the ORR and CR of group A was significantly higher than that of both group B and group C. The renal response rates of the group A, B and C were 64.2% (79/123), 45.7% (16/35) and 46.9% (31/66), respectively. The cardiac response rates of the group A, B and C were 74.0% (54/73), 45.0% (9/20) and 40.0% (10/25), respectively. The renal and cardiac responses of group A was also significantly higher than the other two groups, but there is no difference between group B and group C. After a median follow-up of 44 months, the median OS has not been reached. The 5-year estimated OS rates of the group A, B and C were 81%, 57% and 67%, respectively. The median PFS was 83 months of all patients. The 5-year estimated PFS rates of the group A, B and C were 61%, 38% and 49%, respectively. Both of the OS and PFS of group A was higher than that of both group B and group C. On multivariate analysis, baseline dFLC > 5mg/dl (P =0.005, Hazard Ratio: 3.26, 95% CI: 1.64 – 6.48) were associated with worse survival, but induction with bortezomib (P =0.001, Hazard Ratio: 0.34, 95% CI: 0.16 - 0.72) were associated with better survival.

Conclusion: Our study demonstrated that AL patients with BMPCs less than 10% who eligible for ASCT still can benefit from bortezomib based induction therapy. The benefits of induction therapy are attributed to an improvement in the rate of hematologic ORR and CR, renal and cardiac responses, PFS and OS.
LONG TERM FOLLOW-UP OF PATIENTS WITH AL AMYLOIDOSIS TREATED ON A PHASE 1 STUDY OF ANTI-AMYLOID MONOCLONAL ANTIBODY CAEL-101

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Background: AL amyloidosis with cardiac involvement continues to carry a very poor prognosis with median survival of less than a year from diagnosis and a median survival of only 3 months in patients with cardiac stage 3b disease (1). CAEL-101 (11-1F4 mAb) is a monoclonal IgG1 antibody that directly binds to a conformational epitope present on both human kappa and lambda light-chain amyloid fibrils. In preclinical studies the antibody was able to localize to the amyloid tissue and induce a humoral immune response which led to rapid decrease in size as well as elimination of the amyloid (2). We have previously reported results of an open-label phase 1a/1b clinical trial of the CAEL-101 has been completed and showed a very promising organ response rate of 63% (3). Here we are reporting long term survival of patients enrolled on the trial.

Objective: To assess the long term survival of patient treated with CAEL-101 in the prospective clinical trial.

Methods: Patients with relapsed/refractory systemic AL amyloidosis were enrolled and treated in a phase 1a (N=8) 1b study (N=19) using the anti-amyloid mAb CAEL-101, five patients enrolled on phase 1a also received treatment on phase 1b. Thus, for survival analysis a total of 22 patients are reported. CAEL-101 was administered weekly for 4 weeks with sequential doses of 0.5, 5, 10, 50, 100, 250 and 500 mg/m2 in a dose-escalation design. Updated disease assessment and survival evaluations were conducted with a data-cut off of 7/30/2019.

Results: A total of 22 patients were enrolled and received at least one dose of CAEL-101. The median age was 66 years (range 34-79), gender (M/F 70/30%) and lambda type disease was present in 56% of patients. The majority of patients had cardiac (54%, 12/22) and renal involvement (45%, 10/22). The median number of prior anti-plasma cell regimen was 2 (range, 1 – 9). Evidence of active hematological disease was present in 12/22 patients at the time of enrollment. In terms of the organ responses cardiac response was seen in 67% (8/12) and renal responses were seen in 40% (4/10) patients. At the last data cut-off on 7/30/2019, long-term survival data is available for 19/22 patients. With a median follow-up of 37 months (range 30-45), the median survival has not been reached and 78% (15/19) of patients are alive and 4 patients have died from progressive AL Amyloidosis. Among the evaluable patients 7/12 have experienced hematological progression and only 2/12 patients have experienced organ progression.

Conclusion: Therapy with CAEL-101 can lead to durable long-term organ responses as well as excellent overall survival in patients with systemic AL Amyloidosis. These results compare favorably with previous studies of patients treated with anti-plasma cell therapy for R/R AL amyloidosis. At this time a phase 3 trial randomized trial of CAEL-101 is being planned for patients with newly diagnosed systemic AL Amyloidosis.

References:


Figure 1: Long term survival of patients
COMBINING ANTI-FIBRIL ANTIBODY NEOD001 WITH ANTI-CD38 ANTIBODY DARATUMUMAB IN SYSTEMIC LIGHT-CHAIN AMYLOIDOSIS

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Introduction: Systemic light-chain amyloidosis (AL) results from deposition of immunoglobulin free light chains (FLC) as amyloid protein, causing progressive organ damage. About 30% of AL patients die within a few months of diagnosis due to cardiac complications. Daratumumab (dara) is a CD38-directed antibody that has shown high efficacy in AL amyloidosis. NEOD001 (D1) is an anti-amyloid antibody that was used in several clinical trials (NCT02632786, NCT02312206). A futility analysis showed that the trials would not meet the primary endpoints and clinical development of D1 was terminated. After the assignments of patients on D1 studies were unblinded, we identified a series of patients who received concurrent treatment with both dara and D1 and now report their outcomes in this retrospective analysis.

Results: From 2016 through 2018, 19 patients with AL amyloidosis received dara as later therapy after disease progression. They were a median of 68 years old (range, 52-75) and 63% were male. Median iFLC level at baseline was 95 mg/L (28-1050 mg/L). Nine of these patients received dara and D1 simultaneously in the second-line setting and 10 received dara alone (Table 1). Infusions of both monoclonals were separated by 2 days and continued for a median of 258 days (47-637 days). In the 9 patients who received dual antibody therapy, hematologic response ≥VGPR was seen in all (3 CR and 6 VGPR), achieved at a median of 33 days of treatment (19-161), and organ responses were seen in 7 of 8 patients with cardiac involvement. Infusions of both monoclonals were well tolerated without unexpected toxicity. The 10 patients who received dara alone had a ≥VGPR rate of 80% and 4 of 6 had cardiac responses. Six patients in the dual antibody group and 7 in the daratumumab group are alive at this time.

Conclusion: In 9 AL patients with cardiac involvement in this series of 19 patients, we combined two monoclonal antibodies safely without unexpected toxicities, encouraging future combinations of monoclonals in this population. High rates of hematologic and organ responses occurred at a rapid pace with the combination of dara and D1, suggesting possible synergistic activity that requires further validation in a prospective randomized study.

Table 1. Baseline characteristics of patients at the time of starting daratumumab

<table>
<thead>
<tr>
<th></th>
<th>Daratumumab + NEOD001 (n= 9)</th>
<th>Daratumumab (n= 10)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age in years (median, range)</td>
<td>68 (52-75)</td>
<td>67 (54-74)</td>
</tr>
<tr>
<td>Gender</td>
<td>Male: 45% Female: 55%</td>
<td>Male: 80% Female: 20%</td>
</tr>
<tr>
<td>Light chain isotype</td>
<td>λ: 89% κ: 11%</td>
<td>λ: 70% κ: 30%</td>
</tr>
<tr>
<td>Cardiac involvement (Mayo Stage 2004)</td>
<td>88%</td>
<td>70%</td>
</tr>
<tr>
<td></td>
<td>Stage 2 33% Stage 3 57%</td>
<td>Stage 2 100%</td>
</tr>
<tr>
<td>Renal involvement</td>
<td>44%</td>
<td>80%</td>
</tr>
<tr>
<td>Number of prior therapies</td>
<td>1 (1-3)</td>
<td>3 (1-5)</td>
</tr>
<tr>
<td>Prior stem-cell transplant</td>
<td>22%</td>
<td>70%</td>
</tr>
<tr>
<td>Prior therapy with PI</td>
<td>100%</td>
<td>100%</td>
</tr>
<tr>
<td>iFLC (mg/L)</td>
<td>91 (30-1050)</td>
<td>102 (37-303)</td>
</tr>
<tr>
<td>NT-proBNP (pg/ml)</td>
<td>3807 (1326-13193)</td>
<td>960 (369-3134)</td>
</tr>
<tr>
<td>Proteinuria (g/24 hr)</td>
<td>3.5 (1.5-21)</td>
<td>3.4 (0.2-11)</td>
</tr>
<tr>
<td>Days from diagnosis</td>
<td>261 (51-2037)</td>
<td>1540 (245-3327)</td>
</tr>
<tr>
<td>Days of dual monoclonal antibody therapy</td>
<td>258 (47-637)</td>
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</tbody>
</table>
Upfront treatment of systemic non-IgM AL amyloidosis is well defined in the recommendations of the French National Reference Center for AL amyloidosis since 2008, with chemotherapy at a standard dose. Indeed, nowadays very few patients still receive high-dose melphalan and autologous stem cell transplantation at first line. Therapy objective is to rapidly obtain a hematological response (HR), modulating treatment according to severity of heart involvement and decrease of serum free light chains (FLC). However, in the case of relapse there are no precise guidelines. Combinations of other drugs which are effective in multiple myeloma may be proposed, but studies are rare and often include transplanted patients, so a consensus remains to be established. Here we report a multicenter retrospective study assessing different treatment at relapse and evaluating its impact on HR, time to next event (TTNE), overall survival (OS), organ responses (OR) and toxicity. We included 136 patients who received non-intensive chemotherapy as a first line treatment, one third of them were classified Mayo Clinic stage III. The HR was assessed at the end of treatment based on FLC. The median follow-up of the cohort was 57.4 months (9-214). Among the 17 different therapies used at first relapse, increased HR were seen with triplet drug regimens and proteasome inhibitor (PI) based regimens resulting in 75.86% and 69.01% of very good partial response (VGPR) or better response respectively compared with doublet regimens and other combinations without PI drugs, p<0.0001 for both. No differences were seen regarding OR or median TTNE and OS except for median TTNE for PI combination with 17.05 months (2.9-111) versus 9.95 months (2-126.8) for other drugs (p=0.0027). No severe toxicities were observed in the triplet or PI group. Furthermore, the VRD (bortezomib, lenalidomide and dexamethasone) combination appears to be the most effective in patients treated with the immunomodulatory drugs (IMIDs) based combination, with 75% of VGPR or better, 66% of organ response and a significant advantage in terms of median TTNE with 30 months (5.9-90.9) compared to the doublet IMIDs subgroup result of 8.9 months (2-126.8), p=0.009. Patients having cardiac progression at the second round of treatment and a depth of HR lower than VGPR after second line therapy had shorter survival time on multivariate analysis. In conclusion, this study highlights the need for clear recommendation of good practices. By comparing regimens and their effectiveness we show that bortezomib remains a key molecule for treatment of relapses. VRD has been used in a few patients but we found no cumulative toxicity with the combination of PI and lenalidomide and very encouraging TTNE, hematologic and OR rates. VRD should be tested more systematically in relapsing or refractory patients. Furthermore, therapy should be started early before cardiac progression and the depth of HR still remain a prognosis factor at relapse.
PW068

DARATUMUMAB THERAPY IN PATIENTS WITH IMMUNOGLOBULIN LIGHT CHAIN AMYLOIDOSIS, A SINGLE CENTER EXPERIENCE

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Introduction: Immunoglobulin light-chain amyloidosis (AL) is an underdiagnosed disease characterized by organ deposition of a misfolded monoclonal light-chain secreted by malignant plasma cells, in virtually any organ, most frequently in the heart and kidneys. In most patients, transplantation is not applicable, bortezomib-based combination is the standard of care. However, almost all patients eventually experience hematologic relapse or progression of organ involvement. Daratumumab, a CD38-directed monoclonal antibody approved for the treatment of multiple myeloma (MM), is highly effective inducing rapid and deep hematologic responses. We reported 7 consecutive cases of AL amyloidosis treated with daratumumab in this setting.

Results: Over the past 2.5 years, 7 patients (4 males, 3 females) with proven AL amyloidosis were treated with daratumumab at our centre. Mean age at diagnosis was 59. Plasma cell disorders concerned MGUS in 3, MM in 3, WM in 1. lg-light chain was involved in all cases. Organ involvement concerned the heart in 5 (4 with a MAYO stage ≥2), kidneys in 4 (mean albuminuria >5g/24h), soft tissues in 1. Three patients had ≥2 organs affected. All patients were previously exposed to bortezomib. Daratumumab was introduced as second line treatment in 3, third line in 2, and fifth line in 1 patient. All but one patient, achieved at least partial hematological response. Partial cardiac response was reported in 3 patients and partial renal response in 1, 3 patients being not yet evaluable or refractory. Interestingly, daratumumab was the first therapy to allow an organ improvement, within a 1-4 months delay. Therapy was well tolerated, even among patients with cardiac involvement. Treatment was discontinued in 3 patients because of progressive haematological/organ disease after a median follow-up of 6 months.

Conclusions: Daratumumab represents an effective treatment after conventional treatment failure in AL amyloidosis. In our experience, daratumumab-based regimens induce organ improvement in heavily pre-treated AL amyloidosis, and should be started promptly in bortezomib-refractory patients.

Keywords: Daratumumab, AL Amyloidosis

<table>
<thead>
<tr>
<th>ID</th>
<th>Age at diagnosis</th>
<th>Haematological disease</th>
<th>Organ involved</th>
<th>Mayo Stage</th>
<th>Proteinuria (mg/24h)</th>
<th>lg chain value at diagnosis (mg/L)</th>
<th>Treatments PRIOR to daratumumab</th>
<th>Daratumumab based treatment and number of cures</th>
<th>Haematological response to daratumumab treatment</th>
<th>Organ response after daratumumab</th>
</tr>
</thead>
<tbody>
<tr>
<td>1, F</td>
<td>60</td>
<td>MM</td>
<td>H</td>
<td>2</td>
<td>120</td>
<td>129,1</td>
<td>VCD/Rd</td>
<td>DRd 8x</td>
<td>CR</td>
<td>Partial cardiac response</td>
</tr>
<tr>
<td>2, F</td>
<td>67</td>
<td>MGUS</td>
<td>H/K</td>
<td>3</td>
<td>3120</td>
<td>431,1</td>
<td>VCD</td>
<td>DRd 7x</td>
<td>No response</td>
<td>Progression †</td>
</tr>
<tr>
<td>3, H</td>
<td>47</td>
<td>MM</td>
<td>H/K/N</td>
<td>3</td>
<td>5128</td>
<td>488,9</td>
<td>VCD</td>
<td>DR 2x</td>
<td>CR</td>
<td>Progression †</td>
</tr>
<tr>
<td>4, F</td>
<td>63</td>
<td>MM</td>
<td>S</td>
<td>1</td>
<td>260</td>
<td>17200</td>
<td>Vd/VCD/Rd/P</td>
<td>Dd 6x</td>
<td>Progression after PR</td>
<td>NA †</td>
</tr>
<tr>
<td>5, H</td>
<td>69</td>
<td>MGUS IgG lambda</td>
<td>K</td>
<td>1</td>
<td>3860</td>
<td>262</td>
<td>VCD</td>
<td>DRd 5x</td>
<td>VGPR</td>
<td>Partial kidney response</td>
</tr>
<tr>
<td>6, H</td>
<td>53</td>
<td>WM, IgM Lambda</td>
<td>H</td>
<td>3</td>
<td>&lt;5</td>
<td>369,4</td>
<td>Rtx-Vd/ Rtx-B</td>
<td>Dd 5x</td>
<td>CR</td>
<td>Partial cardiac response</td>
</tr>
<tr>
<td>7, H</td>
<td>56</td>
<td>MGUS</td>
<td>H/K</td>
<td>1</td>
<td>8750</td>
<td>76,1</td>
<td>VCD</td>
<td>DRd 3x</td>
<td>PR</td>
<td>Partial cardiac response</td>
</tr>
</tbody>
</table>

F= female, M= male, H= heart, K= kidney, N= nerves, S= skin, soft tissues, MM= multiple myeloma, WM= Waldenstrom Macroglobulinemia, MGUS= monoclonal gammopathy of unknown significance, B= Bendamustine, C= Cyclophosphamide, D= daratumumab, d= Dexamethasone, P= Pomalidomide, Rtx= Rituximab, R= Lenalidomide, V= Bortezomib, CR= complete response, PR= Partial Response, †= lethal event
CARFILZOMIB IS A POTENTIALLY EFFECTIVE REGIMEN IN RELAPSED/REFRACTORY SYSTEMIC AL AMYLOIDOSIS: RESULTS OF A PHASE I DOSE ESCALATION STUDY (CATALYST TRIAL)

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Introduction: Systemic AL amyloidosis is a multi-system disorder caused by deposition of misfolded light chains (as insoluble amyloid fibrils) produced by an underlying plasma cell dyscrasia. Bortezomib based therapy, in combination with Dexamethasone, Thalidomide or Cyclophosphamide is the current standard of care. Carfilzomib (Kyprolis™), a second generation irreversible inhibitor of the proteasome, is licenced for treatment of relapsed/refractory myeloma. We present the results of a prospective phase 1 multicentre study (CATALYST Trial), designed to define the maximum tolerated dose (MTD) and recommended dose (RD) Carfilzomib in combination with a fixed dose of thalidomide and dexamethasone (KTD) in patients with relapsed/refractory AL amyloidosis.

Trial protocol: Patients were treated in escalating dose cohorts using a 3+3 dose escalation design at three planned dose levels (Table 1). All patients received carfilzomib 20mg/m² on cycle 1, day 1. The primary endpoint was to define the MTD, RD and assess the safety and tolerability of weekly carfilzomib used in combination with Thalidomide and Dexamethasone. The key secondary endpoint was a preliminary assessment of the efficacy of the regimen in AL amyloidosis. Responses were determined centrally according to amyloidosis consensus criteria 2005.

Results: Eleven patients were assessed and ten eligible patients were recruited (6 male, 4 female); median age was 64 years (30% older than 70 years). The mean time to treatment from diagnosis for Dose levels 0 and 1 was 7.8 years (SD ±3.87 yrs.) and 3.2 years (SD ± 1.80) respectively. All non-refractory participants had received at least two previous lines of therapy, with a maximum of four previous lines of therapy. The median creatinine, NT-proBNP and dFLC was 77.0µmol/L, 53.3pmol/L and 77.0mg/L at start of treatment. Three evaluable participants were recruited at Dose Level 0 with no dose limiting toxicities (DLTs). The next cohort of three patients were recruited at Dose Level 1. One patient had a serious adverse event following cycle 1 day 1 (20 mg/m²) and did not receive any further treatment; hence, deemed not evaluable for DLT. Another patient had a DLT (acute kidney injury). The data monitoring committee decided that it was not in the patients' best interest to proceed to Dose level 2 as there was significant evidence of activity at dose level 1 and concern about potential toxicity. Therefore, the subsequent cohort of patients was recruited at Dose level 1 and there were no further DLTs. The MTD of carfilzomib was not reached. Six patients received the full course of six cycles (60%). Two patients discontinued treatment due to toxicity and two patients received 2 and 5 cycles respectively. The overall hematologic response rate (ORR) was 70%; complete response – 1 (10%); very good partial response - 3 (30%) and partial response 3 (30%). Of the 3 patients at Dose level 1, 1 participant had a VGPR, 1 had PR and 1 had no response (discontinued at the end of cycle 2). A total of three SAE’s were reported (all at dose level 1): one of which was a DLT. One patient developed pyrexia, hypotension and hypoxia requiring intensive care admission following cycle 1 day 1 (20 mg/m²); there was no clear evidence of an infection and this was deemed a SAE due to Carfilzomib (but not a DLT). Another patient developed grade 3 abdominal pain deemed unrelated to the investigational agent and the third patient developed grade 3 acute kidney injury that was deemed to be DLT; it recovered with supportive care. 66 grade 1 or 2 adverse events were also reported from 9 patients. There were no SUSARs or deaths in the study reported to date. None of the patients had worsening cardiac function. Detailed toxicity data will be presented.

Conclusion: Carfilzomib 45 mg/m² administered weekly in combination with fixed dose Thalidomide and Dexamethasone is an effective and reasonably well tolerated regimen in relapsed/refractory systemic AL amyloidosis with a comparable efficacy to other regimens in relapsed AL amyloidosis. Further studies are required to validate these results.

Table 1:

<table>
<thead>
<tr>
<th>Dose level</th>
<th>Carfilzomib IV (mg/m²) (Days 1, 8 &amp; 15)</th>
<th>Thalidomide (mg) (Days 1-28)</th>
<th>Dexamethasone (mg) (Days 1, 8 &amp; 15)</th>
</tr>
</thead>
<tbody>
<tr>
<td>-1</td>
<td>27</td>
<td>50</td>
<td>20</td>
</tr>
<tr>
<td>0</td>
<td>36</td>
<td>50</td>
<td>20</td>
</tr>
<tr>
<td>1</td>
<td>45</td>
<td>50</td>
<td>20</td>
</tr>
<tr>
<td>2</td>
<td>56</td>
<td>50</td>
<td>20</td>
</tr>
</tbody>
</table>
REAL WORLD DATA ON THE EFFICACY AND SAFETY OF DARATUMUMAB FOR TREATMENT OF RELAPSED/REFRACTORY AL AMYLOIDOSIS: A MULTI-CENTER RETROSPECTIVE STUDY

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Background: Response rate and progression free survival (PFS) for relapsed light chain (AL) amyloidosis remained unsatisfactory. Recently, few phase II and retrospective trials reported encouraging results with the anti CD38 monoclonal antibody Daratumumab (DARA). However, real world data are scarce.

Aim: Analyze real-world outcomes of DARA alone and in combinations for the treatment of relapsed AL amyloidosis, among a multi-site cohort.

Methods: A retrospective, multi-institutional study conducted in 9 centers in Israel. Adult (≥18 years) patients with confirmed histopathological evidence of relapsed AL amyloidosis who received at least one treatment cycle of DARA, as monotherapy or in combination with other drugs, between October 2016 and march 2019, were included. Hematologic response was defined according to consensus guidelines, on the basis of dFLC, serum and urine immunofixation. Organ response was defined as 30% improvement in target organ biomarker.

Results: Forty-nine patients were included. Baseline characteristics are described in table 1a. Patients received a median of 1 (range 1-6) prior lines of therapy. DARA was combined with IMids in 29 (60%) patients, proteasome inhibitors in 10 (21%) or Cyclophosphamide (5 patients, 10%), 7 (15%) patients received DARA monotherapy. Hematologic Overall response rate (ORR) was 85% (41/48), with 31% (15), 33% (16) and 21% (10) of the patients achieving CR, VGPR and PR respectively. Median time to hematologic response was 2 (range 0.5-10) months. Factors associated with achieving CR were lower dFLC before DARA treatment and kappa light chain clonality. The latter remained significant in a multivariate model incorporating also age and dFLC (hazard ratio 4.2 [95% confidence interval 1.1-15.7]; p=0.03). Twenty-six patients were evaluable for cardiac response. Fifteen (57%) patients achieved at least partial organ response. Median time to response was 5 (range 1-10) months. NYHA score improved, was unchanged or deteriorated in 12 (46%), 15 (58%) and one patient (4%) respectively. EF improved, unchanged or decreased in 8 (53%), 6 (40%) and 1 (7%) of fifteen evaluable patients. Renal response was documented in 7 (30%) of 22 evaluable patients. Median time to renal response was 6 (range 2-12) months. Twelve months PFS rate was 68.8±7.5%. Twelve months OS rate was 80.1±6.5%. Predictors of hematologic and organ response and for PFS and OS are described in tables 2a and 2b, respectively. Thirty-four out of 47 evaluable patients (72%) remain on treatment at data cutoff. Fifteen patients stopped treatment: 14 due to progression or death, and one due to referral to transplantation. None of the patients stopped DARA treatment due to toxicity.

Conclusion: Our data suggests a favorable safety, tolerability and efficacy profile of DARA, as monotherapy or in combinations, among nonselective relapsed AL amyloidosis patients in a real-world setting, with high rates of hematological and organ responses.
**Table 1a: patient’s characteristics**

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age at initiation of DARA, years median (range)</td>
<td>61 (47-89)</td>
</tr>
<tr>
<td>Males n (%)</td>
<td>30 (61)</td>
</tr>
<tr>
<td>Myeloma defining event n (%)</td>
<td>13 (27)</td>
</tr>
<tr>
<td>FISH t(11;14) n (%)</td>
<td>9 (18)</td>
</tr>
<tr>
<td>BM plasmocytosis at diagnosis, %; median (range)</td>
<td>17 (2-100)</td>
</tr>
<tr>
<td>Light chain type by IHC n (%)</td>
<td></td>
</tr>
<tr>
<td>kappa</td>
<td>19 (40)</td>
</tr>
<tr>
<td>Lambda</td>
<td>28 (60)</td>
</tr>
<tr>
<td>dFLC mg/L before Dara treatment mean (IQR)</td>
<td>134 (10-3730)</td>
</tr>
<tr>
<td>Revised Mayo score≥3; n (%)</td>
<td>31 (91)</td>
</tr>
<tr>
<td>Number of organs involved; n (%)</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>14 (28)</td>
</tr>
<tr>
<td>2-3</td>
<td>24 (50)</td>
</tr>
<tr>
<td>4-7</td>
<td>11 (22)</td>
</tr>
<tr>
<td>Organs involved; n (%):</td>
<td></td>
</tr>
<tr>
<td>Heart</td>
<td>35 (71)</td>
</tr>
<tr>
<td>Kidney</td>
<td>28 (57)</td>
</tr>
<tr>
<td>GI</td>
<td>15 (31)</td>
</tr>
<tr>
<td>PNS</td>
<td>27 (55)</td>
</tr>
<tr>
<td>Soft tissue</td>
<td>11 (22)</td>
</tr>
<tr>
<td>liver</td>
<td>7 (14)</td>
</tr>
<tr>
<td>Other</td>
<td>6 (12)</td>
</tr>
</tbody>
</table>

DARA: daratumumab; FISH: fluorsence in-situ hybridaztion; BM: bone marrow; IHC: immunohistochemistry; GI: gastrointestinal tract; PNS: peripheral nervous system; dFLC: free light chain difference; IQR: intra-quartile range

**Table 2a: predictors of hematologic and organ response:**

<table>
<thead>
<tr>
<th></th>
<th>CR</th>
<th>VGPR or less</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hematologic response (n=48)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lambda on IHC: n(%)</td>
<td>5 (19)</td>
<td>22 (81)</td>
<td>0.04</td>
</tr>
<tr>
<td>Organ involved, median [IQR]</td>
<td>3 [2-4.25]</td>
<td>2 [1-3]</td>
<td>0.03</td>
</tr>
<tr>
<td>Nadir of NT proBNP in prior lines, pg/ml; median [IQR]</td>
<td>700 [493-2500]</td>
<td>3000 [1973-5243]</td>
<td>0.019</td>
</tr>
<tr>
<td>dFLC g/L; median [IQR]</td>
<td>96 [44-164]</td>
<td>183 [90-290]</td>
<td>0.052</td>
</tr>
<tr>
<td>Organ response Organ response No organ response</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cardiac (n=26)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male Sex n(%)</td>
<td>9 (47)</td>
<td>10 (53)</td>
<td>0.02</td>
</tr>
<tr>
<td>Female</td>
<td>7 (100)</td>
<td>0 (0)</td>
<td></td>
</tr>
<tr>
<td>Kappa on IHC n (%)</td>
<td>8 (80)</td>
<td>2 (20)</td>
<td>0.051</td>
</tr>
<tr>
<td>dFLC at diagnosis; g/L; median [IQR]</td>
<td>577 [341-1690]</td>
<td>299 [140-680]</td>
<td>0.04</td>
</tr>
<tr>
<td>EF before DARA, %; mean±SD</td>
<td>54±13</td>
<td>35±17</td>
<td>0.04</td>
</tr>
<tr>
<td>Renal (n=22)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>BM plasmocytosis, %; median [IQR]</td>
<td>10 [5-28]</td>
<td>25 [14-40]</td>
<td>0.056</td>
</tr>
</tbody>
</table>

CR: complete response; VGPR: very good partial response; IHC: immunohistochemistry; IQR: intraquartile range; NT proBNP: N-terminal prohormone of brain natriuretic peptide; dFLC: free light chain difference; EF: ejection fraction; SD: standard deviation; BM: bone marrow.
Table 2b: predictors of PFS and OS

<table>
<thead>
<tr>
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<th>HR [95% CI]</th>
<th>P</th>
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<td><strong>Progression</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Female sex</td>
<td>0.09 [0.01-0.7]</td>
<td>0.02</td>
</tr>
<tr>
<td>BM plasmocytosis (for +10%)</td>
<td>1.19 [0.99-1.42]</td>
<td>0.058</td>
</tr>
<tr>
<td>Myeloma defining event</td>
<td>4.5 [1.5-13.8]</td>
<td>&lt;0.00</td>
</tr>
<tr>
<td>Creatinine at diagnosis (for +1 mg/dl)</td>
<td>1.47 [0.9-2.4]</td>
<td>0.11</td>
</tr>
<tr>
<td><strong>Mortality</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>female sex</td>
<td>0.17 [0.02-1.4]</td>
<td>0.1</td>
</tr>
<tr>
<td>NT proBNP before initiation of DARA (for each +1000 ng/ml)</td>
<td>1.1 [1.0-1.2]</td>
<td>0.04</td>
</tr>
<tr>
<td>EF (for +5%) before DARA</td>
<td>0.64 [0.40-0.95]</td>
<td>0.03</td>
</tr>
<tr>
<td>after DARA initiation</td>
<td>0.73 [0.50-1.03]</td>
<td>0.08</td>
</tr>
</tbody>
</table>

PFS: progression-free survival. OS: overall survival; HR: hazard ratio; CI: confidence interval; BM: bone marrow; NT proBNP: N-terminal prohormone of brain natriuretic peptide; EF: ejection fraction; DARA: daratumumab.
OUTCOME OF RENAL TRANSPLANTATION IN AA AMYLOIDOSIS AND THE ROLE OF RECURRENT DISEASE

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Introduction: AA amyloidosis is the second commonest cause of amyloidotic end stage renal disease. Patients with AA amyloidosis are often relatively young and biological therapies enable improved underlying disease control. Previous studies suggested poorer patient and renal allograft survival with renal transplantation compared to other causes of end stage renal disease although more recent studies suggest that this may be improving.

We assessed the patient and allograft outcomes of forty-eight patients with AA amyloidosis, and identified risk factors for, and the implications of, recurrent disease. We compared these to non-matched diabetic nephropathy and autosomal dominant polycystic kidney disease patients (ADPKD).

Objectives: Assess renal allograft and patient outcomes in patients with AA amyloidosis
Investigate significance of recurrence on graft and patient outcomes
Compare outcomes to those of ADPKD and diabetic nephropathy patients

Methods: Forty-eight patients with end stage renal failure due to AA amyloidosis undergoing their first renal transplant being followed up at the National Amyloidosis Centre were identified. Graft failure was defined as a return to dialysis; recurrence was defined as transplant biopsy demonstrating amyloid, or abnormal uptake in the renal allograft on SAP scintigraphy. Patient and renal allograft survival were calculated from the National Amyloidosis Centre database and from National Health Service Blood and Transplant (NHSBT) data. Patient and graft outcomes for 1473 diabetic nephropathy and 5389 ADPKD patients were obtained from NHSBT for comparison.

Results: The median patient survival for AA patients post transplantation was 12.4 years which was significantly worse than ADPKD (19.9 years; p<0.05) but no different to diabetic nephropathy (10.4 years)(Table 1). Neither mean serum Amyloid A (SAA) >10 mg/L post-transplantation (p=0.88), median SAA > 10 mg/L in the year prior to transplant, or recurrent amyloid (0.84) were significant predictors of mortality. The median uncensored graft survival for AA patients was 10.1 years which was significantly worse than ADPKD (16.8 years; p<0.05) but no different to diabetic nephropathy (8.7 years). Graft losses were as follows: twelve died with a functioning graft, four from recurrent amyloid, two due to post-operative complications, one from acute rejection, and one multifactorial loss including recurrence. Eight patients had amyloid recurrence diagnosed at a median of 3.8 years; four lost their graft at a median of 4.3 years. Recurrence had a hazard ratio of 2.83 (p=0.15) for death censored graft loss. Median SAA concentration post-transplant correlated with recurrence of amyloid in the allograft (19.7mg/L and 9.0mg/L for those with and without recurrence respectively) (p=0.004; Table 2).

Conclusions: In summary, patients with end stage renal disease from AA amyloidosis who are selected to undergo renal transplantation have comparable outcomes to diabetic nephropaths. Patients with ESRD from AA amyloidosis should be considered for transplantation. Recurrence is associated with poorly controlled SAA levels and likely plays a role in graft failure.

Table 1.

<table>
<thead>
<tr>
<th></th>
<th>AA Amyloidosis</th>
<th>Diabetic nephropathy</th>
<th>ADPKD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patient survival (years)</td>
<td>Median (SE)</td>
<td>12.4 (10.4-14.4)</td>
<td>10.4 (9.1-11.6)</td>
</tr>
<tr>
<td>Uncensored graft survival (years)</td>
<td>Median (SE)</td>
<td>10.1 (5.4-14.8)</td>
<td>8.7 (7.9-9.4)</td>
</tr>
</tbody>
</table>
### Table 2.

<table>
<thead>
<tr>
<th>Patient Survival</th>
<th>Hazard Ratio</th>
<th>(95% CI; p value)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Median SAA &gt; 10 in the year prior to transplant</td>
<td>1.2</td>
<td>(0.38-3.82; 0.76)</td>
</tr>
<tr>
<td>Recurrence</td>
<td>1.13</td>
<td>(0.34-3.75; 0.84)</td>
</tr>
<tr>
<td>Large amyloid load at diagnosis</td>
<td>1.56</td>
<td>(0.48-5.04; 0.46)</td>
</tr>
<tr>
<td>SAA &gt; 10 since transplant</td>
<td>1.1</td>
<td>(0.33-3.66; 0.88)</td>
</tr>
<tr>
<td>Death censored graft survival</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Recurrence</td>
<td>2.83</td>
<td>(0.69-11.6; 0.15)</td>
</tr>
<tr>
<td>Median SAA &gt; 10 post-transplant</td>
<td>1.61</td>
<td>(0.27-9.73; 0.60)</td>
</tr>
</tbody>
</table>

**Keywords:** AA amyloidosis; kidney transplantation; recurrent disease; amyloid recurrence; patient survival; allograft survival

**Category:** Organ transplant in systemic amyloidosis
LIVING DONOR KIDNEY TRANSPLANTATION IS AN EFFECTIVE OPTION OF RENAL REPLACEMENT THERAPY IN PATIENTS WITH AL AMYLOIDOSIS

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2 Department of Hematology, Amyloidosis and Myeloma Unit, Hospital Clinic-Universitat de Barcelona, Spain.
3 Department of Pathology, Centro de Referencia en Enfermedad Glomerular Compleja del Sistema Nacional de Salud (CSUR), Hospital Clinic-Universitat de Barcelona, Spain.
4 Department of Hematology, Hospital de la Santa Creu i Sant Pau.

Background: Renal disease is a frequent manifestation in systemic AL amyloidosis and often is the major source of morbidity for individuals with this disorder. Without treatment, amyloidosis-associated kidney involvement usually progresses to end-stage renal disease (ESRD). The introduction of autologous stem cell transplantation and the development of new antiplasma cell agents have markedly improved the survival in this disease. Although the role of kidney transplantation for patients with amyloidosis-associated ESRD remains controversial, it might be a reasonable option, improving patients’ survival and quality of life.

Objective-Method: Herein we describe our experience in three patients with AL amyloidosis who received a living donor renal transplantation. Results: AL organ involvement was limited to the kidney in the 3 patients (2 women, 1 man). Two patients achieved complete hematological response with high-dose melphalan and autologous hematopoietic stem cell transplantation and one patient with bortezomib and dexamethasone before kidney living donor transplantation. Two patients were transplanted from an unrelated donor and one from a HLA identical donor, all three achieved complete recovery of renal function during the first week. Two patients developed allograft dysfunction derived from recurrence of their disease (confirmed by biopsy at 27 and 84 months post transplantation) and received a regimen consisted of 6 cycles of bortezomib, cyclophosphamide and dexamethasone followed by 6 more cycles without bortezomib. Complete hematologic response was achieved without detectable clonal plasma cells in the bone marrow by flow-cytometry, as well as progressive organ response with proteinuria < 1 g/24h and preserved kidney function.

Conclusions: Kidney transplantation in AL amyloidosis might be more than a reasonable option in patients with amyloidosis-associated ESRD who obtained a complete hematologic response after treatment, if there is no significant extrarenal involvement. Outcomes in this group of patients are similar to those observed in others high-risk patient subgroups that frequently, undergo kidney transplant. Nevertheless, the risk of histological recurrence after kidney transplantation exists, even when the transplant is performed in a recipient in hematologic remission. Based on our clinical experience, the treatment should be established at the time of hematological recurrence and/or when the organic renal recurrence is mainly histological. Allograft dysfunction derived from amyloid deposition in the transplanted kidney is an avoidable co-morbidity, especially if hematologic relapse is early diagnosed and the recurrence of renal deposition is detected in an early histological stage. Moreover, living donor transplants may provide remarkable advantages for the recipients with AL amyloidosis such as a better genetic match that lessens the risk of rejection and immediate allograft function that minimize the risk of co-morbidities.

Key words: light chain amyloidosis; amyloidosis-associated kidney disease; living donor kidney transplantation; recurrence; allograft biopsy
INCREASING NUMBER AND IMPROVED SURVIVAL IN SOLID ORGAN TRANSPLANTATION FOR PATIENTS WITH AMYLOIDOSIS. AN ANALYSIS OF THE UNITED NETWORK FOR ORGAN SHARING DATABASE.

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2 Huntsman Cancer Institute.
3 Intermountain Medical Center.

Introduction: Amyloidosis results from accumulation of mis-folded proteins causing organ failure and historically has been associated with a poor prognosis. In the last decade therapeutic options for amyloidosis have increased with improved outcomes. We hypothesized that the trends and outcomes of solid organ transplantation (Tx) have increased and improved respectively.

Objectives: Describe and compare the findings among different solid organs including kidney, heart and liver transplantation.

Methods: We analyzed the United Network for Organ Sharing (UNOS) database and included patients with diagnosis of amyloidosis who received a primary single organ (Kidney, Liver and Heart) between January 1987 and March 2018. We compared post-transplant outcomes in two eras: January 1987 to December 2007 (Previous Era) and January 2008 to March 2018 (Recent Era).

Results: From 1987 to 2018 a total of 1,150 patients with amyloidosis were transplanted. 589(50.4%) kidney, 359(30.8%) heart and 215(18.7%) liver transplantsations were performed. The 1-year survival for kidney tx between the previous and recent eras improved from 88% to 94%; for heart tx from 79% to 89%; and for liver tx from 83% to 90% - Table. The 5-year survival for kidney tx between the previous and recent eras also improved from 67% to 82%; for heart tx from 46% to 79%; and for liver tx remained similar 58% to 59%.

Conclusion: More patients with amyloidosis are considered for solid organ transplantation. Outcomes continue to improve and are now approaching survival rates comparable to non-amyloid patients.

Key words: amyloidosis, organ transplant survival, UNOS
HEART TRANSPLANTATION AND CONCOMITANT CHEMOTHERAPY FOR PATIENTS WITH ADVANCED CARDIAC AL AMYLOIDOSIS: 20 YEARS OF EXPERIENCE AT A SINGLE CENTER

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¹Amyloidosis Center, Heidelberg University Hospital

Introduction: Patients with systemic light-chain (AL) amyloidosis and advanced cardiac involvement have a very poor prognosis with a median overall survival (mOS) of <6 months. One reason is that highly increased therapy-related mortality (TRM) in these patients prohibits the application of an effective chemotherapy regimen. However, due to the systemic nature of the disease and the shortage of organs the justification of a heart transplantation (HTx) as treatment in AL is controversially discussed.

Objective: To evaluate chances and limitations of HTx, combined with chemotherapy and/or high-dose Melphalan and autologous stem cell transplantation (HDM/ASCT) as treatment for high-staged cardiac AL patients.

Patients & Methods: Clinical data of 46 patients (23m, 23f) with advanced cardiac AL who were treated at our center and high-urgency (HU) listed for orthotopic HTx between 2000 and 2019 was retrospectively analyzed. Two eras with different selection criteria and different available chemotherapies were compared: 2000 – 2007 (era 1, n= 18) and 2008 – 2019 (era 2, n= 28). Survival estimates were calculated using the Kaplan-Meier method, starting with date of high urgency listing if not otherwise stated.

Results: Patient characteristics, transplantation and chemotherapy data are stratified by era and reported in Table 1. The median follow-up of era 1 and 2 was 13.1/12.7 years and 7.4/7.5 years from initial diagnosis / HU listing, respectively. Due to stricter selection criteria, only 2 patients of the modern era had significant extracardiac organ involvement (both gastrointestinal tract), compared to 7 patients of era 1 (kidney 4x, gastrointestinal tract 2x, kidney and gastrointestinal tract 1x). Era 2 was further characterized by a longer median waiting time until HTx and more extensive chemotherapeutical pretreatment, including the frequent use of proteasome inhibitors. The most frequent first therapy after HTx in both eras was HDM followed by ASCT, whereby TRM happened in 1/15 patients. Only 1/15 patients showed no hematologic response and 8/15 patients achieved a very good partial response or better. Overall 27 patients died. Causes of death were either progression of AL (n=16), sepsis (n=6), heart transplant rejection (n=3) or other (n=2). Median OS after HU listing of the patients in era 1 and 2 were 2.6 and 6.5 years, respectively (logrank p= 0.22), one-year OS was 61.1% and 67%. Two patients in era 1 and 6 patients in era 2 deceased before receiving HTx with a mOS of 87 and 25 days, respectively (p= 0.27). Patients of era 1 who underwent HTx had a mOS of 3.7 years from HTX, while the median was not reached in era 2 (logrank p= 0.036).

Conclusions: HTx with concomitant chemotherapy is a feasible treatment approach in patients with advanced, but sole cardiac amyloidosis. Novel agents allow for longer bridging and most patients who reach HTx can be treated with HDM + ASCT, which comes with acceptable TRM and very good hematologic response rates.
Table 1:

<table>
<thead>
<tr>
<th>Parameter</th>
<th>2000-2007 (n= 18)</th>
<th>2008-2019 (n= 28)</th>
<th>P values</th>
<th>Missing%</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Patient characteristics and disease activity at date of high urgency listing</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age, years</td>
<td>54 [40, 62]</td>
<td>52 [36, 64]</td>
<td>0.910</td>
<td>0.0</td>
</tr>
<tr>
<td>Sex male</td>
<td>10 (55.6)</td>
<td>13 (46.4)</td>
<td>0.763</td>
<td>0.0</td>
</tr>
<tr>
<td>LC isotype = lambda</td>
<td>16 (88.9)</td>
<td>22 (78.6)</td>
<td>0.615</td>
<td>0.0</td>
</tr>
<tr>
<td>PC count, %</td>
<td>0.16 [0.04, 0.35]</td>
<td>0.12 [0.04, 0.43]</td>
<td>0.289</td>
<td>0.0</td>
</tr>
<tr>
<td>dFLC, mg/L</td>
<td>387 [10, 1648]</td>
<td>132 [9, 1779]</td>
<td>0.408</td>
<td>8.7</td>
</tr>
<tr>
<td>NTproBNP, ng/L</td>
<td>6149 [2247, 32344]</td>
<td>10634.5 [1809, 90754]</td>
<td>0.079</td>
<td>0.0</td>
</tr>
<tr>
<td>cTnT [µg/l]</td>
<td>0.09 [0.03, 0.46]</td>
<td>0.06 [0.03, 0.13]</td>
<td>0.300</td>
<td>52.2*</td>
</tr>
<tr>
<td>hsTnT [pg/ml]</td>
<td>Not available</td>
<td>121 [16, 546]</td>
<td>NA</td>
<td>47.8'</td>
</tr>
<tr>
<td>Significant extracardiac organ involvement</td>
<td>7 (38.9)</td>
<td>2 (7.1)</td>
<td>0.023</td>
<td>0.0</td>
</tr>
<tr>
<td>Proteinuria, mg/d</td>
<td>259 [66, 1798]</td>
<td>136 [95, 1466]</td>
<td>0.052</td>
<td>4.3</td>
</tr>
<tr>
<td>eGFR [ml/min]</td>
<td>82 [23, 126]</td>
<td>58 [30, 111]</td>
<td>0.177</td>
<td>4.3</td>
</tr>
<tr>
<td><strong>Chemotherapy before high urgency listing</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Any therapy before HU</td>
<td>12 (66.7)</td>
<td>23 (82.1)</td>
<td>0.397</td>
<td>0.0</td>
</tr>
<tr>
<td>&gt;1 therapy line</td>
<td>1 (5.6)</td>
<td>6 (21.4)</td>
<td>0.297</td>
<td>0.0</td>
</tr>
<tr>
<td>Bortezomib included</td>
<td>1 (5.6)</td>
<td>21 (75.0)</td>
<td>&lt;0.001</td>
<td>0.0</td>
</tr>
<tr>
<td><strong>Heart transplantation</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No HTx performed</td>
<td>2 (11.1)</td>
<td>6 (21.4)</td>
<td>0.615</td>
<td>0.0</td>
</tr>
<tr>
<td>Days on HU list until HTx</td>
<td>23 [3, 267]</td>
<td>51 [8, 260]</td>
<td>0.011</td>
<td>0.0'</td>
</tr>
<tr>
<td><strong>Chemotherapy after heart transplantation</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HDM + ASCT</td>
<td>6 (33.3)</td>
<td>9 (32.1)</td>
<td>1.000</td>
<td>0.0</td>
</tr>
<tr>
<td>Days HTx until ASCT</td>
<td>180 [136, 341]</td>
<td>244 [129, 903]</td>
<td>0.469</td>
<td>0.0'</td>
</tr>
</tbody>
</table>

Categorical data is shown as count (% of respective total), continuous data is shown as median [range].
* At least one troponin value is available in 44/46 patients.
† Significant organs: heart, kidney, liver, gastrointestinal tract, peripheral nervous system
‡ non-applicable patients not counted as missing
Background: The first liver transplantation as a treatment for hereditary ATTR-amyloidosis was performed in 1990 in Stockholm. Registry data indicate a 55% 20-year survival after liver transplantation. This case presents the development of the symptoms of ATTR amyloidosis over time in a long-term surviving liver transplanted patient.

Patient: In 1989 a 37 y old male patient with a family history of FAP presented with polyneuropathy and gastrointestinal symptoms. Rectal biopsy showed presence of ATTR amyloidosis and genetic testing confirmed a V30M-mutation. There was no significant cardiomyopathy and liver transplantation was performed in 1992. Since transplantation he has been treated with cyclosporine and corticosteroids. Corticosteroids was tapered and later stopped in 2000.

The patient, now 66 y old, has no clinically significant progression of polyneuropathy, however in 2000 EMG showed a mild to moderate peripheral neuropathy of axonal type and neurography a slight progression compared to 1995. The same year he experienced worsened gastrointestinal symptoms and investigations showed steatorrhea, bile acid malabsorption, and impaired ventricular emptying. He has over the years developed bilateral glaucoma requiring both drugs and surgery. Prostate biopsy due to lower urinary tract symptoms and slightly elevated PSA showed no malignancy but presence of amyloid.

In 2000 echocardiography revealed mild hypertrophy. In 2006 he was diagnosed with paroxysmal atrial fibrillation that later became chronic. In 2007 there was moderate hypertrophy with septal thickness 16mm that progressed to 18mm in 2019. In 2009 MRI showed signs consistent with cardiac amyloidosis. Latest MRI was performed in 2019 and showed progression with ECV 50% and involvement of left ventricle, right ventricle as well as intra atrial septum. DPD-scintigraphy in 2019 was almost normal with exception of a very low uptake located only in the atria. Abdominal fat biopsy in 2019 showed ATTR-amyloidosis with only full-length (type-B) fibrils. Patient is still in FAP stage 1, NYHA class 2 and experiences a good quality of life.

Long term survival after liver transplantation is feasible in selected patients with ATTRv amyloidosis. This patient is one of those with the longest survival after transplantation for this indication. Progression of systemic amyloidosis after liver transplantation due to recruitment of wild type TTR is a concern in ATTR type A but obviously sometimes in patients with type B-fibrils. Consistent with previous studies, DPD-scintigraphy was almost normal in this patient with type B fibrils in spite of significant ATTR cardiac amyloidosis. This might provide a clue to understanding DPD-scintigraphy positivity.
OUTCOMES OF RENAL TRANSPLANTATION IN AL AMYLOIDOSIS PATIENTS:
AN INTERNATIONAL COLLABORATION


1 Memorial Sloan Kettering Cancer Center, New York, USA.
2 Boston University Amyloidosis Center, Boston, MA, USA.
3 Mayo Clinic, Rochester, MN, USA.
4 Weill Cornell Medicine/NewYork-Presbyterian Hospital, New York, USA.
5 Amyloidosis Research and Treatment Center, University of Pavia, Italy.

Introduction: Therapies for AL amyloidosis have dramatically improved, leading to longer patient survival. Consequently, more patients with AL amyloidosis are now reaching end stage renal disease (ESRD). Renal transplantation in patients with AL amyloidosis has been controversial due to lack of long term outcome data and appropriate and validated eligibility criteria.

Objectives: The goal of this study is to define eligibility criteria for kidney transplantation in patients with AL amyloidosis.

Methods: We describe 95 patients with AL amyloid renal involvement who underwent renal transplantation at various centers throughout the world over the last 30 years.

Results: Our preliminary data is shown here. Additional patient data is expected at the time of the meeting. Median age at diagnosis and renal transplantation were 53.1 (range 26-73) and 60 (range 30-75) years (yrs) respectively. With the exception of two, all patients were white and 65% were male. Lambda light chains were involved in 63% of patients. One third had isolated renal involvement and 35% had heart involvement. The most common treatment modality was high dose melphalan/stem cell transplantation (HDM/SCT) in 58%. Approximately half of the transplants were from live donors and 13% were preemptive. Median time from diagnosis to ESRD was 2.3 years (range 0-19) and from renal replacement therapy to renal allograft was 2.2 years (0.2-11). During a median follow up of 8.1 years (range 0-22), median overall survival (OS) from diagnosis was 17.2 yrs (range 1-27.2), and from renal transplantation was 11.5 yrs (range 0.2-24). Most of the patients achieved a hematologic complete response (CR) or very good partial response (VGPR) at time of renal transplantation and their renal transplant outcomes were comparable to other non-amyloid patient groups. Amyloid recurrence in the graft was overall low (21%) and successful salvage plasma cell directed therapy for hematologic relapse likely prevented graft loss in this setting. There was a trend towards better OS and graft survival in patients who underwent HDM/SCT any time during their disease course vs patients who were treated only with chemotherapy however the difference did not reach statistical significance.

Conclusions: Overall and renal graft survival in patients with AL amyloidosis were excellent and comparable to outcomes achieved in non-AL amyloidosis patients (based on data from the USRDS Database). Most of the patients in the study achieved a CR and VGPR to plasma cell directed therapy at the time of renal transplantation. These favorable outcomes were independent of the type of treatment that was used for the underlying plasma cell dyscrasia. There was a striking disparity with respect to race in our cohort. With the expanding treatment options for AL amyloidosis, it is critical to recognize the benefit of renal transplantation in this population to ensure access for all AL patients.

Keywords: AL Amyloidosis, Renal Transplantation, Graft Survival
HEART TRANSPLANTATION FOR SYSTEMIC LIGHT CHAIN AMYLOIDOSIS IN THE MODERN ERA: A MULTICENTER COLLABORATION

H. Landau 1, M. Rosenzweig 2, S. Goodman 3, M. Maurer 4, M. Zucker 5, J. Liu 1, R. Comenzo 6

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2 City of Hope Comprehensive Cancer Center.
3 Vanderbilt University Medical Center.
4 Columbia University Medical Center.
5 Newark Beth Israel Medical Center.
6 Tufts University Medical Center.

Background: Patients with advanced heart failure due to light chain (AL) amyloidosis often do not survive long enough to benefit from plasma cell directed (PCD) therapies. The risk of disease progression in other organs and recurrent amyloid deposition in the transplanted heart has limited the use of orthotopic heart transplantation (OHT) in these patients. Earlier studies suggested inferior outcomes of OHT for AL compared with non-amyloid indications. Recently, effective PCD therapies have become available for AL. Yet, the role of OHT remains controversial and many centers consider AL to be a contraindication to OHT.

Objective: To describe the characteristics and outcomes of patients with AL who underwent OHT for AL in the modern era.

Methods: The medical records of patients with AL who underwent OHT from 2008-2018 and were followed at collaborating academic medical centers in the US were reviewed.

Results: 8 patients (63% male, 63% Caucasian) with median age 56.5 years (yrs) (range 38 – 64), Mayo/European cardiac stage II (N= 2), IIIA (N= 5) and IIIB (N= 1) and had New York Heart Association (NYHA) class II (N= 5) and III (N= 3) heart failure at diagnosis were included in this preliminary analysis. Additional patient data are expected at the time of the meeting. Extra-cardiac involvement was present in 75% of patients (renal in 6, GI in 2, neuro in 2). All but 1 received PCD therapy prior to OHT and achieved complete hematologic remission (CR) (N= 5, 63%) or very good partial hematologic remission (VGPR) (N= 2, 25%). Despite clonal disease control, all patients had cardiac progression and qualified for OHT. There were no deaths postoperatively and all patients remain alive. The median overall survival (OS) from diagnosis and OHT is 10.2 yrs (range 2.3 – 14.4) and 4.3 yrs (range 0.8 – 11.4), respectively. Post-OHT, 2 (25%) patients were treated for chronic rejection and 4 (50%) required PCD. Three patients required salvage therapy for hematologic relapse/progression at 2, 3.5 and 6 yrs following OHT; 1 is receiving maintenance Ixazomib due to <CR prior/post OHT. Hematologic disease is currently controlled in all patients (CR 6, VGPR 2). No patient has experienced cardiac progression although one patient developed ESRD at the time of hematologic progression. Three patients requiring PCD therapy have been treated for grade 3 infections (CMV, C Difficile, MAC/nocardia) while receiving salvage daratumumab and one for Aspergillus following ASCT.

Conclusions: Selected patients who received OHT for AL have excellent outcomes with OS that exceeds what is expected for AL patients who have cardiac progression. Adequate hematologic disease control (at least a VGPR) is likely a prerequisite for consideration of OHT although patients with very advanced heart failure may benefit from OHT prior to PCD therapy. Successful treatment for hematologic relapse likely prevented cardiac progression but can lead to infectious complications.
THE PROGRESSION OF LEFT VENTRICULAR THICKNESS IS GREATER IN MEN COMPARED TO WOMEN IN WILD-TYPE TRANSTHYRETIN AMYLOID CARDIOMYOPATHY

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1 University of Pittsburgh Medical Center

Background: Left ventricular (LV) thickness is an important echocardiographic feature of wildtype transthyretin cardiac amyloidosis (ATTR-CA wt). The rate of progression of LV thickness in ATTR-CA wt is unknown.

Objective: We aimed to determine the progression of LV thickness and its association with ECG changes, serum biomarkers and clinical symptoms in untreated ATTR-CA wt.

Methods: Measurements were made on 2 echocardiograms, at least 18 months apart, before and after the diagnostic Tc-99m pyrophosphate (PYP) scan. Echocardiographic measurements were obtained according to the American Society of Echocardiography guidelines. ECG, serum biomarkers and New York Heart Association Functional Class (NYHA FC) were obtained through chart review. The measurements closest to the echocardiograms were analyzed.

Results: (Table 1). In 50 patients, on echocardiograms that were 24 ± 6 months apart, there was progressive increase in LV thickness (mean difference=1.6mm, 95% CI 1.3-2.0, p<0.001). than women (mean difference=1.4mm, n=19, p<0.001). There was concomitant decrease in voltage-to-LV mass index ratio on ECG, worsening diastology, higher NYHA Class (NYHA FC ≥2) and increase in BNP and troponin (p<0.001).

Conclusion: Our data demonstrates greater progression in LV thickness in men compared to women with ATTR-CA wt.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Pre-PYP Measurements</th>
<th>Post-PYP Measurements</th>
<th>Mean Difference (95% Confidence Interval)</th>
<th>p-value (Paired T-test)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Interventricular Septal thickness (IVSd/mm)</td>
<td>17.1±3.0</td>
<td>18.7±3.4</td>
<td>1.6 (1.3-2.0)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>-IVSd in men</td>
<td>18.1±2.9</td>
<td>19.9±3.3</td>
<td>1.8 (1.5-2.4)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>-IVSd in women</td>
<td>14.8±2.0</td>
<td>16.2±2.0</td>
<td>1.4 (1.1-1.5)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Posterior Wall thickness (PWd/mm)</td>
<td>16.0±3.2</td>
<td>17.4±3.5</td>
<td>1.3 (1.1-1.7)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>LV Mass Index (g/m²)</td>
<td>157.4±37.5</td>
<td>180.7±40.7</td>
<td>23.3 (17.9-28.8)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Voltage-to-LV mass indexratio (mm/g/m²)</td>
<td>0.23±0.09</td>
<td>0.16±0.08</td>
<td>-0.07 [-0.08-(-0.06)]</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>B-type Natriuretic Peptide (BNP / pg/mL)</td>
<td>531.5±328.91</td>
<td>821.8±559.5</td>
<td>290.4 (207.5-373.2)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Troponin I (ng/mL)</td>
<td>0.10±0.10</td>
<td>0.20±0.24</td>
<td>0.09 (0.03-0.14)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Diastolic Dysfunction Grade</td>
<td>1.9±0.8</td>
<td>2.8±0.8</td>
<td>0.09 (0.70-0.95)</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>
SYSTEMATIC REVIEW OF THE GENDER DISTRIBUTION OF PATIENTS WITH WILD-TYPE TRANSTHYRETIN AMYLOID CARDIOMYOPATHY

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³ Pfizer B.V., Netherlands.

Background: Transthyretin amyloid cardiomyopathy is a progressive and fatal disease which is underdiagnosed. It may arise from mutations in transthyretin, but the majority of cases are due to the aggregation and deposition of wildtype transthyretin (ATTRwt) amyloid. Previous studies have shown that males are disproportionately affected by ATTRwt cardiomyopathy. However, the gender distribution of patients diagnosed with ATTRwt cardiomyopathy has yet to be formally established in the literature.

Objectives: The primary aim of this study was to provide the first estimate of the gender distribution of patients with ATTRwt cardiomyopathy. The secondary objective was to review available evidence of demographic and clinical differences by gender.

Methods: A systematic review of the literature was conducted using PubMed. Predefined search terms included: “Cardiomyopathy, Amyloidosis”, “Transthyretin”, and “Heart Failure”. Studies published before 1990, those not reporting the number of males and females separately, those reporting overlapping patient cohorts, and those that focused solely on hereditary type or light chain amyloidosis were excluded. Data on the number of patients, gender, age, geographical location, study type, method of diagnosis (clinically diagnosed or ICD codes), ethnicity and various other clinical characteristics were extracted from the selected studies.

Results: A total of 182 studies were identified with 9 additional studies identified through snowballing. After removing studies with potential overlapping patient cohorts, 19 studies were left including 2,494 patients. The majority (79%, n=15) of studies were published after 2015. Most studies focused on the US (42%, n=8) and Europe (32%, n=6), the remaining studies were performed in Japan (11%, n=2), Australia (5%, n=1), or multiple regions (11%, n=2). The median age of the overall population upon diagnosis ranged from 75.1 to 79.4 years. The mean percentage of male patients with ATTRwt cardiomyopathy was 92.5% (2,308 out of 2494). There was, however, a wide range varying from 78% up to 100% males. Only 4 studies specifically reported differences between genders. These studies noted that women were significantly older upon diagnosis, were more likely be diagnosed post-mortem, and more likely to have a non-invasive diagnosis.

Conclusion: This is the first review to estimate the gender distribution of ATTRwt cardiomyopathy patients. Our study showed that 92.5% of patients with ATTRwt cardiomyopathy are males although we did observe a wide range across the included studies. Questions remain about biases in gender distribution between the individual studies observed. Future studies should investigate whether females affected by ATTRwt cardiomyopathy are underdiagnosed or if males are primarily affected.
A CURIOUS FAMILIAL CLUSTER OF WILD-TYPE TRANSTHYRETIN CARDIAC AMYLOIDOSIS

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Background: Wild-type transthyretin amyloid cardiomyopathy (ATTR-CA wt) is thought to be a result of age-related spontaneous misfolding and aggregation of transthyretin (TTR). It is distinguished from the hereditary type (ATTR-CA h) by the absence of known point mutations in the TTR gene. Here, we describe 3 brothers with transthyretin amyloid cardiomyopathy (ATTR-CA) without demonstrable gene mutations.

Case description: Sibling A is an 80-year-old Caucasian man who presented with recurrent pleural effusions and was found to have amyloid deposits on a lung biopsy. Past history revealed bilateral carpal tunnel syndrome and paroxysmal atrial fibrillation. The echocardiogram showed severe left ventricular hypertrophy (LVH), bilateral atrial enlargement and grade 3 diastolic dysfunction, highly suspicious for cardiac amyloidosis. Tc-pyrophosphate (PYP) scintigraphy showed prominent myocardial uptake of tracer and serum studies for AL amyloidosis were negative, thus establishing a diagnosis of ATTR-CA. Comprehensive cardiomyopathy and TTR gene testing identified a variant of unknown significance (VUS) identified in LITAF gene which is associated with Charcot-Marie-Tooth disease Type 1 disease, but no TTR mutation.

Sibling B is 69-year-old and self-referred himself to the amyloid clinic after the diagnosis in his brother. He had a history of persistent atrial fibrillation, bilateral carpal tunnel syndrome and spontaneous quadricep tendon rupture. An echocardiogram showed severe LVH and global longitudinal strain with an apical sparing pattern suggestive of cardiac amyloidosis. He underwent Tc-pyrophosphate (PYP) scanning which showed significant myocardial uptake. Serum studies for AL amyloidosis were negative. Gene testing for TTR mutations was negative.

Sibling C, 87-year-old, also had a history of bilateral carpal tunnel release, severe spinal stenosis with multiple back surgeries and coronary artery disease with 4-vessel coronary artery bypass graft surgery. Prior echocardiograms showed severe LVH. Given high suspicion for cardiac amyloidosis, Tc-99m PYP scan was performed which demonstrated avid tracer uptake suggestive of ATTR-CA along with concomitant negative serum studies for AL amyloidosis. Comprehensive cardiomyopathy and TTR gene testing identified a VUS in the CACNA1C gene, which is associated with long QT syndrome and hypertrophic cardiomyopathy, but no mutations in the TTR gene.

Discussion: This case series reports a curious familial clustering of transthyretin cardiac amyloidosis without a demonstrable mutation in the TTR gene. It raises relevant questions about as yet unrecognized potential genetic or epigenetic mechanisms in the pathogenesis of ATTR-CA wt.
ELECTRODIAGNOSTIC AND CLINICAL FEATURES OF NEUROPATHY IN PATIENTS WITH WTATTR

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Introduction/background: Wild type transthyretin amyloidosis (wtATTR) manifests with cardiomyopathy and affects mostly older men. Some patients with wtATTR may also develop peripheral neuropathy that generally follows a less progressive course than is seen with hereditary transthyretin amyloidosis (hATTR); dysautonomia is also uncommon. Another manifestation is spinal stenosis with radiculopathies associated with amyloid deposits and symptoms including distal numbness, dysesthesias and weakness. The clinical phenotype of neuromuscular complications of wtATTR has not been well described in the literature.

Objectives: To describe electrodiagnostic feature of neuropathy in 7 patients with wtATTR - cardiomyopathy.

Methods: Review of electrodiagnostic and clinical features of neuropathy in patients with wt-ATTR cardiomyopathy who were evaluated in Cardiac Amyloidosis Center at University of Pittsburgh Medical Center in Pittsburgh, PA, USA.

Results: There were 7 subjects with with wtATTR who complained of numbness, weakness and unsteadiness of gait and were suspected to have peripheral neuropathy. The patients were all men with median age of 76 years. They developed neuropathy on the average 7.5 years earlier. None had diabetes, and 1 patient had IgG kappa MGUS. Electrodiagnostic testing showed the common occurrence of bilateral carpal tunnel syndrome (n=7), ulnar neuropathy (n=3; bilateral 1, unilateral 2), sensorimotor axonal length-dependent polyneuropathy (n=5) and chronic lumbar and cervical radiculopathies (n=4). The clinical phenotype included 5 patients with mild neuropathy (PND stage I) and 2 patients with moderate neuropathy (PND stage III). Five patients with PND stage I presented on the average at 5.2 years after onset of neuropathy symptoms, and two patients with PND stage III presented at 16.5 years after onset.

Conclusions: Neuropathy associated with wtATTR typically follows a mild, slowly progressive course contrasting with the natural history and rapidly progressive course seen with hATTR-associated neuropathy. In addition to carpal tunnel syndrome, which was present in all patients, clinical patterns included sensorimotor axonal polyneuropathy (n=5), chronic radiculopathies (n=4) and ulnar neuropathies (n=3). Our findings are concordant with the previously described slowly progressive axonal sensorimotor polyneuropathy in patients with wtATTR and also suggest more frequent occurrence of ulnar entrapments and chronic radiculopathies. However, larger series are needed to define the prevalence and clinical significance of neuromuscular complications associated with wtATTR and the potential role of comorbidities.
THE PROGRESS RATE IN SEPTAL THICKNESS IN PATIENTS WITH WILD TYPE TRANSTHYRETIN AMYLOIDOSIS.

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**Background:** Wild type cardiac amyloid (ATTRwt) amyloidosis is today known to be more common in heart failure than previously recognized. However, the rate of progression of inter-ventricular diastolic septal (IVSD) thickness in ATTRwt amyloidosis is not well characterized. We aimed to investigate the increase in IVSD over time in a retrospective study of patients who had been diagnosed with ATTRwt amyloidosis, and who have had an echocardiographic examination performed at least 3 years before diagnosis.

**Material & methods:** We identified 23 patients (mean age at diagnosis was 84±6 years, 22 males, all diagnosed due to symptoms of heart failure) with a diagnosis of ATTRwt (negative for both TTR mutation and AL amyloidosis). The echocardiograms were available from at least three years (mean of 6±3 years) prior to diagnosis. All patients had undergone echocardiography at the time of diagnosis (between 2013-2019).

**Results:** The echocardiographic data from the two examinations are summarized in table 1. IVSD thickness increased from 14±3 mm to 18±3 mm in the group, p<0.001. The mean increase in IVSD thickness for the group was 0.6 mm/year. Individual differences in IVSD between baseline and follow up are shown in figure 1. During the follow up time, there were also reductions in cardiac output and LV global longitudinal strain, the latter borderline significantly.

**Table 1.**

<table>
<thead>
<tr>
<th></th>
<th>Prior to diagnosis</th>
<th>At diagnosis</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>IVSD, mm</td>
<td>16±4</td>
<td>19±3</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>LVEF, %</td>
<td>49±12</td>
<td>48±8</td>
<td>0.467</td>
</tr>
<tr>
<td>LV GLS, %</td>
<td>14±5</td>
<td>11±3</td>
<td>0.061</td>
</tr>
<tr>
<td>CO ml/min</td>
<td>5.0±1.3</td>
<td>3.9±1.0</td>
<td>0.002</td>
</tr>
</tbody>
</table>

IVSD=interventricular septum thickness, LVEF=left ventricular ejection fraction, GLS=global longitudinal strain, CO=cardiac output.

**Figure 1.**

Septal thickness on Y axis and years of follow up on X-axis

**Conclusion:** In this study in ATTRwt amyloidosis patients the increase in IVSD over time was approximately 0.6 mm/year in the years prior to diagnosis. In addition to increased septal thickness, LV systolic function decreased. However, great variation in progress rate of IVSD was observed. The pathophysiological mechanisms behind this difference in IVSD rate progression between subjects needs to be further studied.
WILD-TYPE TRANSTHYRETIN, MORE THAN WHAT MEETS THE HEART

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Introduction: In today’s aging population, there is growing awareness that amyloid deposition is an important and under recognized cause of cardiomyopathy. Wild-type transthyretin amyloidosis (ATTRwt), caused by misfolded transthyretin protein, is associated with widespread amyloid deposition in post-mortem analysis but historically, is thought to present with a narrow phenotype marked primarily by cardiomyopathy. Recently, novel treatments have been approved for both wild type and hereditary TTR amyloidosis, making it more essential than ever to identify cases expeditiously. Clarifying the phenotype of ATTRwt can aid in early detection. Clinically, there is growing recognition of ATTRwt’s non-cardiac features including carpal tunnel syndrome and lumbar stenosis but, until now, no strong association to polyneuropathy or autonomic neuropathy has been made in the literature.

Objectives: The goal of this case series is to describe the neurologic features seen in patients with ATTRwt who were referred to neurology at a single academic institution.

Methodology: ATTRwt patients evaluated by the Utah Amyloidosis Program between 4/2017-4/2019 were screened for neurologic symptoms and, when appropriate, sent for a neurologic consultation. This case review highlights the neurologic features in six sequential wild type patient evaluations and categorizes the variability and overlap of neurologic symptoms.

Results: In this case series, six (12%) of the 51 patients confirmed with wild type cardiomyopathy at this amyloidosis center were sent for a neurologic evaluation during this time interval. The patients’ average age at evaluation was 77.3 and among these patients four (67%) were found to have a sensory predominant neuropathy, three (50%) had autonomic dysfunction, six (100%) had carpal tunnel syndrome, three (50%) had trigger finger, two (34%) had voice changes marked by dysphonia and two (34%) had lumbar spinal stenosis (image 1). Carpal tunnel syndromes predated the diagnosis of cardiomyopathy by an average of 14 years and was bilateral in all cases.

Conclusion: Despite the limitations of a small case series, these findings suggest that bilateral carpal tunnel syndrome maybe a manifesting symptom of ATTRwt predating cardiomyopathy by over a decade and sensory and autonomic neuropathies is commonly seen in ATTrwt. Neurologic symptomatology in a patient with cardiomyopathy is an important diagnostic clue for cardiologists to consider. Future prospective studies evaluating prevalence of sensory and autonomic neuropathies using histologic confirmation will be helpful to confirm and expand the known phenotype of ATTRwt.

Key Words: wild-type, transthyretin, neuropathy

Image 1.

Neurologic features in patients with wild type ATTR (percentages)

- Sensory neuropathy: 67%
- Autonomic dysfunction: 50%
- Carpal tunnel syndrome: 100%
- Trigger finger: 50%
- Voice changes: 34%
- Lumbar stenosis: 34%
STEep INCREASE IN THE NUMBER OF TRANSTHYRETIN-POSITIVE CARDIAC BIOPSY CASES IN JAPAN: DATA FROM THE NATION-WIDE PATHOLOGY CONSULTATION SYSTEM

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3 Department of Neurology, Kumamoto University.
4 Department of Pathology, Yokohama City University.
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6 Department of Pathology, Keio University.
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Introduction/Background: On March 26, 2019, the Ministry of Health, Labour and Welfare (MHLW) of Japan approved tafamidis for transthyretin (TTR)-related (ATTR) cardiomyopathy (CM). By referring to the inclusion criteria of the TTR amyloidosis CM clinical trial (ATTR-ACT), MHLW also announced the patient criteria to officially cover the cost of tafamidis therapy, which included the pathological confirmation of amyloid deposits on biopsy specimens and, in patients without TTR mutation, the immunohistochemical confirmation of TTR in amyloid deposits. Back in April 2018, the MHLW-funded group for surveys and research of amyloidosis in Japan started the nation-wide pathology consultation of amyloidosis.

Objectives: To confirm that the approval of tafamidis for ATTR-CM has elicited interest in this disease among Japanese cardiologists, which increased the number of cardiac biopsy cases for the definite diagnosis of this disease.

Methods: We added up the number of cardiac biopsy cases, Congo-red-positive and ATTR-positive cases from April 2018 to March 2019, and from April 2019 to October 2019, respectively to assess the increase in the number of cases after the approval of tafamidis. Six institutes attended the consultation and shared the panel of anti-κ chain(116-133), anti-λ chain(118-134) (Hoshii et al. 2001), anti-TTR(115-124) (Gustavsson et al. 1994), anti-AA (Dako) and anti-β2-microglobulin (Dako) antibodies. When the typing diagnosis was unavailable by immunohistochemical analysis, we performed proteomic analysis.

Results: From April 2018 to October 2019, we diagnosed 347 cases of cardiac biopsy, in which the number of ATTR-CM cases was 235. After the approval of tafamidis, cardiac biopsy cases, Congo-red-positive and ATTR-positive cases increased from 15.3, 14.5 and 10.0 cases/month to 23.4, 21.6 and 16.4 cases/month, respectively. Among cardiac biopsy cases, the ratio of ATTR-CM cases increased from 65.6% to 70.1%.

Conclusions: These data indicate that the approval of tafamidis in Japan has increased the number of cardiac biopsy cases for the definite diagnosis of ATTR-CM. In January 2020, we will start to register all patients who are prescribed tafamidis for ATTR-CM, which, together with our consultation system will elucidate the precise clinical picture of ATTR-CM in Japan.

Keywords: transthyretin, cardiac biopsy, pathology consultation
PRESENCE OF TRANSTHYRETIN AMYLOID IN CARPAL TUNNEL BIOPSIES

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Background and Introduction: Transthyretin (TTR) is a normal serum protein produced mainly in the liver which acts as a carrier protein for thyroxine and retinol. Genetic variants of TTR, of which more than 130 are known to be pathogenic, are associated with widespread tissue aggregation and deposition of TTR in an insoluble fibrillary conformation called amyloid, usually causing a predominant neuropathy and cardiomyopathy. Wild-type (non-variant) TTR is deposited as amyloid in association with advancing age, usually in the synovial tissues, tendons and heart. Wild-type ATTR amyloid deposition in certain tissues may be asymptomatic, although extensive cardiac infiltration can lead to a cardiomyopathy which is typically restrictive in nature (wild-type ATTR cardiac amyloidosis). Wild-type ATTR amyloidosis is currently diagnosed in only ~200 individuals in the UK per year despite post-mortem studies showing presence of wild-type ATTR amyloid deposits in the hearts of up to 30% of male individuals over 80 years of age.

This discrepancy in conjunction with the knowledge that the majority of wild-type ATTR amyloidosis patients treated at the UK National Amyloidosis Centre (NAC) have a history of carpal tunnel (CT) decompression surgery, often as long as 12 years prior to presentation with cardiac amyloidosis, prompted this preliminary study in which we sought to identify amyloid in excised tissue from the flexor retinaculum from patients over 50 years of age undergoing carpal tunnel decompression surgery. In cases in which amyloid was identified, the amyloid fibril protein was determined by immunohistochemistry (IHC) using a panel of antibodies. The study is ongoing.

Materials and Methods: Samples from the flexor retinaculum were taken from 31 males and 34 females, with a median age of 61 and 67 years respectively, who underwent routine carpel tunnel decompression surgery. Samples were fixed in 10% formalin and processed into a paraffin blocks (FFPE) for routine histology, Congo red staining and IHC. If amyloid was identified, IHC, using a panel of monospecific antibodies against known amyloid fibril proteins, was performed in order to amyloid type. Interpretation of results was undertaken by two experienced individuals.

Results: Of the 65 biopsies tested 39% (25 cases) were found to contain amyloid deposits. Of those, 72% (18 cases) were from males, with a median age of 61 (range 55-88) years and 28% were from (7 cases) females, with a median age 67, (range 52-92) years. The amyloid type in all but one case was ATTR; the amyloid was too scanty to type by IHC in the remaining case.

Discussion and Conclusions: This preliminary study has shown that approximately 40% of samples taken for routine carpel tunnel decompression contain TTR amyloid, and may be a simple procedure that may identify individuals at risk of developing cardiac ATTR amyloidosis, at an early age.

Key words: Carpel Tunnel, Transthyretin, Immunohistochemistry
DIFFERENT BEHAVIOR IN TTR STABILITY IN PLASMA FROM CARDIO AND NEUROPATHIC MUTANT TTR CARRIERS

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Introduction: Familial ATTR is triggered by mutations in the TTR gene who promote destabilization of the tetrameric structure of TTR. The generation of amyloidogenic monomers who deposits into tissues causes organ injury. Depending on the mutation, the phenotype can be cardiologic, neurologic, or mixed. Neurological manifestations, particularly polyneuropathy is the most common manifestation of the majority of the mutations namely V30M, the most common prevalent amyloidogenic TTR mutation. Among TTR mutations that cause cardiomyopathy such Val122Ile and Ile68Leu, wild-type TTR is the most common cause of cardiomyopathy in the elderly.

Objective: To develop a method that can discriminate the TTR stability in plasma of cardio from neuropathic ATTR carriers.

Methodology: Plasma (n=3 from each group) from V30M asymptomatic, patients and domino transplanted recipients were tested. As for cardiomyopathies V122I, I68L and ATTR WT were included. A semi-denaturing gel system, with minor denaturing conditions was performed, after TTR immunoprecipitation, to assess the dissociation into monomers.

Results: We observed that under these conditions the neuropathic V30M presents monomers in opposition to the cardiomyopathic V122I, I68L and plasma of ageing subjects carrying WT amyloid cardiomyopathy who display only dimeric TTR as control plasma does. As for patients with both cardio and neuropathic symptoms, different ratios of monomer/dimer are found.

Conclusions: This assay can discriminate a distinct behavior in TTR stability in plasma from cardio and neuropathic of ATTR amyloidosis,
NATURAL PROGRESSION OF WILDKYPE TRANSTHYRETIN AMYLOIDOSIS AND EVALUATION OF RISK PREDICTION PARAMETERS DERIVED FROM CLINICAL ROUTINE

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Introduction: In transthyretin (TTR) amyloidosis deposition of fibrillar TTR in tissue leads to progressive organ dysfunction. In contrast to hereditary transthyretin amyloidosis, in wildtype transthyretin amyloidosis (ATTRwt) no mutation in the transthyretin (TTR) gene is detectable. ATTRwt occurs in elderly, predominantly male patients and typically primarily affects the heart. Deposition of amyloid in the myocardium leads to restrictive cardiomyopathy with progressive symptoms of heart failure. Despite increased awareness little is known about the natural progression of the disease.

Objectives: Therefore this study longitudinally analyses clinical findings and patients symptoms over a medium period of twelve months, to gain more detailed information about the clinical course of ATTRwt.

Methods: Clinical characteristics and diagnostic findings of 65 patients suffering from ATTRwt were collected and analyzed over a period of 12 months. Therefore ECG, echocardiographic parameters, laboratory test and clinical functional test (NYHA class, Karnofsky Index and BMI) were analyzed.

Results: In most of the patients (69%) the leading clinical symptom was dyspnea. Some patients (11,6%) presented signs of polyneuropathy, but after non-invasive neurological examination only one patient needed nerve biopsy to exclude ATTR-polyneuropathy. Furthermore a part of the patients (16,9%) were already treated with cardiac pacemakers. Two third of the patients (67,7%) presented with abnormal ECGs. Prolonged PQ and QRS durations as well as low voltage patterns were frequently found. After 12 months intraventricular septum was significantly elevated (19,9±0,5mm) compared to baseline visits (18,9±0,5mm) and ejection fraction (EF) declined significantly from 45,4±2,1% to 38,9±2,6%. During the follow up period of twelve months 16 patients (24,6%) died. Especially patients with low EF, reduced renal function, impaired right ventricular function and need for pacemaker implantation presented an elevated risk for death within the one year follow up period.

Conclusions: In ATTRwt precise evaluation of routine cardiac diagnostic helps to identify patients with elevated risk of mortality. Especially patients with progressive myocardial hypertrophy, right ventricular failure, reduced renal function and need for cardiac pacemakers are at risk for elevated one year mortality.

Keywords: wild type TTR amyloidosis, prognosis, routine care
PW088

A MACHINE LEARNING FRAMEWORK FOR PREDICTING RISK OF WILD-TYPE TRANSTHYRETIN AMYLOID CARDIOMYOPATHY

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¹ Pfizer Inc.

Introduction: Wild-type transthyretin amyloid cardiomyopathy (ATTRwt-CM) is a rare, underdiagnosed and fatal disease that is increasingly recognized as a cause of heart failure. Despite increasing awareness, a majority of patients with ATTRwt-CM continue to remain undiagnosed owing to its similarities in clinical presentation to more common etiologies of heart failure. Here, we adapted our previously published machine learning algorithm that identified patients with ATTRwt-CM using ICD codes from medical claims data transformed into a tool for educating clinicians to estimate risk of ATTRwt-CM in hypothetical patient scenarios.

Methods: US medical claims data sets were sourced from IQVIA (n >300 million patients, 10 years of medical history) and ICD codes were extracted to define patient cohorts as well as provide diagnostic history for patients within each cohort [medical claims cohort 1 ATTRwt-CM: N=1,071; HF: N=9,639]. Random Forest (RF) classification model was setup to differentiate ATTRwt-CM from HF patients (matched for age, gender, and medical history). A set of 15 clinical conditions along with age and biological sex were selected as features from the RF model. Performance metrics and operating characteristics were calculated on a 20% holdout/validation sample (test data). Additional internal validations were conducted on three different EHR and medical claims data sources.

Results: RF model features reflect the cardiac and non-cardiac clinical manifestations of ATTRwt-CM. The model was able to correctly classify ATTRwt-CM and HF patients with AUC of 0.84 (sensitivity 74%, specificity 77%, PPV 27%, NPV 96%, and accuracy 77%). The model was internally tested by classifying ATTRwt-CM or Cardiac Amyloid (CA) patients from HF patients in 3 additional cohorts derived from medical claims and Electronic Health Records (EHR) data: EHR cohort 2 –ATTRwt-CM: N=213, HF: N=1,696, medical claims cohort 3 – CA: N=7,198, HF: N=62,161, EHR cohort 4 – CA: N=1,941, HF: N=15,618.

Conclusion: We developed and internally tested a novel approach that uses physician input for a machine learning model with an aim to educate clinicians to estimate an empirical probability of ATTRwt-CM. This framework may serve as a simple and easily implementable tool for clinician ascertainment of patient risk for ATTRwt-CM. Ongoing external validation work will further inform use in clinical practice.
AMYLOID IN THE LIGAMENTUM FLAVUM OF PATIENTS WITH SPINAL STENOSIS AND ATTR (WILD-TYPE) CARDIAC INVOLVEMENT

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6 Department of Pathology, Tufts Medical Center, Boston, MA, USA.

Background: Amyloidosis is a protein misfolding disorder, wherein the deposition of the fibrillar form of precursor protein causes displacement of tissue architecture. Transthyretin amyloid (ATTR) deposits cause age-related (wt) and mutant (m) amyloidosis and have also been identified in tissues such as carpel tunnel ligament, ligamentum flavum (LF). Several therapies have been recently approved for patients with ATTR cardiac and ATTRm neuropathy, but strategies for early identification of these patients remain elusive. Preliminary data have indicated that TTR may play a role in the pathogenesis of spinal stenosis (SS) and may, like carpal tunnel syndrome (CTS), be a harbinger of systemic ATTR. In a preliminary approach to this question, we investigated patients undergoing spinal surgery for neurogenic reasons and asked whether amyloid was present in the LF and whether there was a risk of cardiac amyloidosis.

Methods: We created an IRB-approved registry of patients who underwent SS surgery, had the surgical specimen (LF) analyzed by pathology with Congo-red staining and if indicated, typed for amyloid by mass spectrometry. We included demographic information, baseline comorbidities, disease presentation, imaging studies and associated conditions such as CTS, non-ischemic cardiomyopathy (NICM) and atrial fibrillation. Patients with positive specimens were offered clinical evaluation as standard of care with fat pad aspirates, serum protein electrophoresis, light-chain assays and serum immunofixation, TTR gene sequencing and 99mTc-PYP scintigraphy.

Results: In 2018 and 2019, a total of 325 patients had surgery for symptomatic SS with Congo-red staining of the LF (Table 1). Sixty-two had amyloid in the LF of whom 61 had typing by mass spectrometry. Forty-four (71%) had ATTR, 17 (27%) had indeterminate type, and 1 (2%) with multiple myeloma had dual deposits with ATTR and AL-kappa type. None had ATTRm by gene sequencing. Amyloid cases were significantly older and had a significantly higher prevalence of CTS (Table 1). Among the amyloid cases, 39 had fat pad biopsies and 48 had PYP scans; 7 had positive fat pads and 7 PYP scans were strongly suggestive of ATTR. Four ATTRwt patients had ATTRwt in other tissues (1 cardiac, 1 CTS, 2 at other spinal levels). Two confirmed ATTRwt cardiac cases have subsequently received treatment (cardiac transplant and tafamidis respectively).

Conclusions: In this series of 325 patients undergoing laminectomy for SS, amyloid was found in the LF of 19% and ATTRwt identified in 14%. Patients with amyloid in the LF were significantly older and had a higher prevalence of CTS, suggesting a systemic form of ATTR involving connective tissue. Fat pad aspirates were positive in 7 of 39 cases (18%) and PYP scans were suggestive in 7 of 48 patients (15%). ATTRwt cardiac disease was confirmed and treated in 2 patients. Further prospective study of patients with spinal stenosis at risk for systemic amyloidosis is warranted.
Table 1. Characteristics of patients undergoing spinal stenosis surgery

<table>
<thead>
<tr>
<th></th>
<th>Congo-red positive (N=62)</th>
<th>Congo-red negative (N=263)</th>
<th>P value (Mann-Whitney/Fisher’s exact test)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Male</strong></td>
<td>58%</td>
<td>56%</td>
<td>NS</td>
</tr>
<tr>
<td><strong>Age, years (range)</strong></td>
<td>73 (46-88)</td>
<td>66 (30-95)</td>
<td>&lt;&lt;0.01</td>
</tr>
<tr>
<td><strong>Carpal tunnel syndrome</strong></td>
<td>27%</td>
<td>13%</td>
<td>0.01</td>
</tr>
<tr>
<td><strong>Race</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>82% Caucasian</td>
<td>84% Caucasians</td>
<td>NS</td>
</tr>
<tr>
<td></td>
<td>8% Asian</td>
<td>6% African American</td>
<td></td>
</tr>
<tr>
<td></td>
<td>5% African American</td>
<td>6% Asian</td>
<td></td>
</tr>
<tr>
<td></td>
<td>3% Hispanic</td>
<td>3% Hispanic</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2% Other</td>
<td>1% Other</td>
<td></td>
</tr>
<tr>
<td><strong>Hypertension</strong></td>
<td>75%</td>
<td>63%</td>
<td>0.07</td>
</tr>
<tr>
<td><strong>Hyperlipidemia</strong></td>
<td>65%</td>
<td>57%</td>
<td>NS</td>
</tr>
<tr>
<td><strong>Diabetes mellitus</strong></td>
<td>18%</td>
<td>21%</td>
<td>NS</td>
</tr>
<tr>
<td><strong>Body mass index (range)</strong></td>
<td>28 (22.2-56.8)</td>
<td>29.1 (17.3-51.4)</td>
<td>NS</td>
</tr>
<tr>
<td><strong>Non-ischemic cardiomyopathy</strong></td>
<td>7%</td>
<td>3%</td>
<td>NS</td>
</tr>
<tr>
<td><strong>Atrial fibrillation</strong></td>
<td>16%</td>
<td>10%</td>
<td>0.18</td>
</tr>
<tr>
<td><strong>Level of spinal surgery</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Lumbar 86%</td>
<td>Lumbar 70%</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Cervical 8%</td>
<td>Cervical 28%</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Thoracic 6%</td>
<td>Thoracic 2%</td>
<td></td>
</tr>
</tbody>
</table>

**Congo-red Positive Patients (N=62)**

<table>
<thead>
<tr>
<th><strong>Fat pad biopsy</strong></th>
<th>Positive: 7/39 (18%)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>PYP scan</strong></td>
<td>Strongly suggestive (grade 2 or H/CL ratio &gt;1.5): 7/48 (15%)</td>
</tr>
<tr>
<td></td>
<td>Equivocal (grade 1 or H/CL ratio 1-1.5): 37/48 (77%)</td>
</tr>
<tr>
<td></td>
<td>Not suggestive (grade 0 or H/CL ratio &lt;1): 4/48 (8%)</td>
</tr>
</tbody>
</table>
FACTORS ASSOCIATED WITH A DELAYED DIAGNOSIS IN WILD TYPE TRANSTHYRETIN CARDIAC AMYLOIDOSIS

F. Domínguez 1, E. González-López 1, A. Hernández 1, A. López Sáinz 1, A. Rivas Pérez 1, L. Escobar López 1, S. Vilches Soria 1, M. Cobo Marcos 1, J. F. Oteo Domínguez 1, C. Salas Antón 1, J. Segovia Cubero 1, L. Galan 2, P. García Pavía 1

1 Hospital Universitario Puerta de Hierro, Madrid, Spain.

Background: Diagnosis of wild type TTR cardiac amyloidosis (ATTRwt) is challenging. ATTRwt clinical spectrum is very heterogeneous and is not restricted to the classical phenotype of elderly males with restrictive cardiomyopathy and low QRS voltages on ECG. New treatments for ATTRwt have shown to be more effective when administered early highlighting that an early recognition of ATTRwt is desirable. We sought to study which factors are associated with a delayed diagnosis in ATTRwt.

Methods: Clinical, electrocardiographic and echocardiographic characteristics were studied in a cohort of 100 patients (77% male, mean age 80.2 ± 8.1 years) with ATTRwt evaluated at Hospital Universitario Puerta de Hierro (Madrid, Spain) from 2008 to 2018. Diagnostic delay was defined as the time elapsed from the initial onset of symptoms until diagnosis. Factors associated with delayed diagnosis were evaluated.

Results: Mean diagnostic delay in ATTRwt patients was 2.9 ± 4.5 years. Patients under 70 years of age at the onset of symptoms were diagnosed later as compared to older ones (6.3 ± 7.4 years vs 1.9 ± 2.6 years; p=0.01). LVH of 15 mm or less was found in 30% of patients and showed a statistical trend towards an increased diagnostic delay (4.7 ± 6.9 vs 2.2 ± 2.7 years; p=0.06). In 28% of patients, diagnostic delay was of 3 years or longer. In this subgroup of patients, there was a trend towards a higher rate of patients with pacemakers implanted (11/28: 35.7%, vs 15/72: 20.8%; p=0.07). However, the type of LVH distribution, the presence of low or high ECG voltages, LVEF systolic dysfunction, gender or history of hypertension did not have a significant impact on time until diagnosis. Interestingly, patients with concomitant coronary artery disease (20%) exhibited also increased diagnostic delay (6.7 ± 7.7 vs 2.0 ± 2.6 years; p=0.01) (Table 1).

Conclusions: Patients with ATTRwt under 70 years of age, with pacemakers or mild LVH show an increased diagnostic delay. The classical picture for ATTRwt phenotype is only present in a minority of patients. Clinicians should be aware of the broad clinical spectrum of ATTRwt to correctly identify an entity for which a number of disease-modifying treatments are already available or under investigation.

<table>
<thead>
<tr>
<th></th>
<th>n (%)</th>
<th>Mean time to diagnosis (years)</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male</td>
<td>77 (77%)</td>
<td>3.1 ± 5.0</td>
<td>2.3 ± 2.6</td>
</tr>
<tr>
<td>Female</td>
<td>23 (23%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age under 70 years</td>
<td>23 (23%)</td>
<td>6.3 ± 7.4</td>
<td>1.9 ± 2.6</td>
</tr>
<tr>
<td>Age ≥ 70 years</td>
<td>77 (77%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>HTN</td>
<td>77 (77%)</td>
<td>2.5 ± 3.7</td>
<td>4.3 ± 6.7</td>
</tr>
<tr>
<td>No HTN</td>
<td>23 (23%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>CAD</td>
<td>20 (20%)</td>
<td>6.7 ± 7.7</td>
<td>2.0 ± 2.6</td>
</tr>
<tr>
<td>No CAD</td>
<td>80 (80%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>No LVH* in ECG</td>
<td>82 (93.2%)</td>
<td>2.3 ± 3.8</td>
<td>3.4 ± 6.3</td>
</tr>
<tr>
<td>LVH in ECG</td>
<td>6 (6.8%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>LVH ≤ 15 mm</td>
<td>30 (30%)</td>
<td>2.2 ± 2.7</td>
<td>4.7 ± 6.9</td>
</tr>
<tr>
<td>LVH &gt; 15 mm</td>
<td>70 (70%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>LVEF &lt; 50 %</td>
<td>27 (27%)</td>
<td>3.3 ± 4.6</td>
<td>2.8 ± 4.5</td>
</tr>
<tr>
<td>LVEF ≥50 %</td>
<td>73 (73%)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table 1. Clinical, ECG and echocardiographic associated with a delayed diagnosis in ATTRwt.
CAD: Coronary artery disease, HTN: Hypertension
*Sokolov criteria for LVH
**CLINICAL FEATURES AND TREATMENT OF WILD-TYPE TRANSTHYRETIN CARDIAC AMYLOIDOSIS – SINGLE CENTER EXPERIENCE**

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3 3rd Department of Internal Medicine, University Hospital Olomouc, Olomouc, Czech Republic.

**Introduction:** Wild-type transthyretin amyloidosis (wtATTR) is caused by the deposition of molecules of native transthyretin. Wild-type transthyretin amyloidosis is manifesting mainly as heart disease characterized by restrictive/hypertrophic cardiomyopathy with clinical symptoms of heart failure. The prevalence estimate represents approximately 25% of the autopic findings in male population > 80 years. Diagnosis and differential diagnosis depend on the use of imaging methods and on exclusion of light-chain and hereditary types of amyloidosis, caused by the deposition of mutated transthyretin.

**Material and Methods:** The analysed group consisted of 13 patients (11 males, 2 females) with wtATTR amyloidosis and cardiac impairment (NYHA II-IV). The aim of our study was to analyze the clinical features and treatment outcome of our patients.

**Results:** In 9/13 patients, supraventricular arrhythmias, such as atrial fibrillation, were detected; all patients had clinical symptoms of carpal tunnel. Echocardiographic examination confirmed in all patients restrictive/hypertrophic cardiomyopathy with depression of LVEF in 8/13 patients. Cardiac magnetic resonance was performed in 11/13 patients, with positive results. In patients was initiated usual symptomatic therapy and all patients started a specific treatment combination of TUDCA (tauroursodeoxycholic acid) with doxycycline (duration of treatment 10 – 72 months). In 12/13 patients, clinical status stabilized and we also registered decrease in NT-proBNP levels and LVEF improvement in 8/13 patients.

**Discussion and Conclusion:** Wild-type transthyretin amyloidosis is a relatively common, but rarely diagnosed disease with dominant cardiac impairment. Although treatment has been so far predominantly symptomatic, the combination TUDCA and doxycycline appears to be effective and well tolerated.

Supported by grant from the Ministry of Health of the Czech Republic (16-31156A).
PW092

DIAGNOSIS AND CLINICAL CHARACTERISTICS OF PATIENTS WITH WTATTR CARDIOMYOPATHY: A SYSTEMIC DISEASE BEYOND THE HEART

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1 Department of Clinical Therapeutics, National and Kapodistrian University of Athens.

Background: Cardiomyopathy due to wild type ATTR is now recognized as a common cause of restrictive cardiomyopathy, especially in older males. Despite recent advances and the use of scintigraphy the diagnosis is often missed, and the disease is still underdiagnosed. The introduction of TTR stabilizing drugs may improve the outcomes of these patients, and new therapies targeting TTR production are explored in this setting too. A detailed description of the patients’ characteristics and the natural history of this disease, outside clinical trials, is important in order to define diagnostic algorithms, efficacy benchmarks and evaluate cost effectives of new therapies.

Aim: To describe the diagnostic approach, clinical characteristics and outcomes of patients with ATTR CM in a single center from Greece (Department of Clinical Therapeutics, Athens).

Methods: The report includes data from 50 patients with a diagnosis of ATTR CM, which were diagnosed after 1/1/2015.

Results: The rate of diagnosis of ATTR CM was doubling each year. Median age was 81 years (range 53-89), 85% were males. All had positive PYP-Tc99m scans; median H/CL ratio was 1.7 (range 1.6-2.3). A fat aspirate was performed in 60% and was Congo red positive in 35% of biopsy samples; an endomyocardial biopsy was performed in 8% and was positive in all cases; a monoclonal protein was found in 9% of patients and typing (by IHC, IEM or MS) was performed in 12% of all patients. Genetic testing was performed in all patients and in 2 (6%) patients (1 was a female) a TTR mutation was found; none of the two had neuropathy. Median serum creatinine was 1.12 mg/dl and median eGFR 65 ml/min/1.73 m2. All patients had symptoms of restrictive CM; median NYHA stage was 2 and 20% were NYHA stage ≥3. At echo studies, median IVS was 15.5 mm, PW was 15 mm, median EF was 50% and median GLS -9.6%. Median NTproBNP was 4541 pg/ml (range 843-35000), 58% had atrial fibrillation, 23% a pacemaker, 25% a history of CAD, 98% were receiving diuretics (furosemide in all, spironolactone/eprelenone in 10%), 45% an ACE-I/ARB and 72% a b-blocker. Other findings and symptoms at the time of diagnosis included carpal tunnel syndrome in 36%, purpura in 10%, weight loss in 36%, tendon rupture in 14%, history of spinal stenosis in 4%, peripheral edema in 38%, symptoms of peripheral neuropathy in 34% and of autonomic neuropathy in 16%. In 42% of patients, doxycycline was given. The median follow-up is 24 months and the overall survival rate is 92%.

Conclusions: symptoms of severe heart failure dominate the clinical presentation of patients with TTR CM, however, symptoms from other organ systems indicate that ATTR is a systemic disease. Only few patients carry TTR mutations, but genetic testing should be considered in all patients with TTR CM. In this elderly population, with several co-morbidities, the management of the disease becomes challenging and current and future therapies must have a very favorable toxicity profile.
INTERACTION BETWEEN ATTR FIBRILS AND ELASTIN IN LIGAMENTUM FLAVUM

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1 Uppsala University.
2 Stockholm Spine Center.
3 Karoliska institutet.
4 Umeå University.

Background: ATTR amyloid deposits are common in ligaments and are associated with aging. ATTR amyloidosis occurs in two main forms, hereditary and sporadic. While hereditary forms depend on mutations in the TTR gene sporadic ATTR amyloidosis involves the wild-type (wt) protein. It has been suggested that deposition of ATTR amyloid in ligaments is an early sign of systemic disease. There is also evidence that the local deposits in ligaments may induce clinical consequences and association with lumbar spinal stenosis and osteoarthritis have been proposed. In a prospective study of patients undergoing surgery for lumbar spinal stenosis we found ATTR amyloid in 92 out of 250 individuals. The elastin-rich ligamentum flavum (LF) is a site often harboring ATTR amyloid. Some other amyloid forms, particularly those derived from gelsolin (1) and medin (2) are specifically associated with elastin. A relationship between ATTR deposits and elastin in LF is unknown.

Objective: To study a possible morphological relationship between ATTR deposits and elastin ultrastructurally.

Methods: About 1 cubic mm pieces of LF from two patients with ATTR deposits were processed for electron microscopy with routine methods including post-fixation in osmium tetroxide. Ultrathin sections on formvar-coated nickel grids were immunolabelled with rabbit antiserum 1898 against ATTR50-127, contrasted with uranyl acetate and lead citrate and examined in an electron microscope.

Results: Amyloid fibrils appeared as short, haphazardly oriented fibrils, sometimes in large areas. A constant feature was the close topographical association of amyloid fibrils with elastic fibers. ATTR labelling sometimes appeared as a thin cover of elastic tissue and seemed to be intermingled with outer part of elastin fibers, which often were broken into smaller fragments.

Conclusions: ATTR fibrils in LF occur in close connection with elastin in a manner that reminds of AMed fibrils in aorta. Elastin fibers have hydrophobic surfaces that may interact with amyloidogenic proteins and be of importance in generation of amyloid fibrils. Such interaction seems to be associated with degradation of elastic tissue and may thereby be involved in the pathogenesis of diseases with loss of elastin.

References:
**Background:** Autopsy studies have shown that ATTR amyloidosis (presumably wild-type) is very common in aging populations and reaching 25% over the age of 80 years. A number of reports have shown that deposits of ATTR amyloid are surprisingly common in many ligament tissues including those of the carpal tunnel and ligamentum flavum of the spinal canal. There is also increasing evidence that carpal tunnel syndrome and lumbar spinal stenosis (LSS) in some cases can be early signs of a systemic disease later manifested as progressive amyloid cardiomyopathy. In a study of materials resected at surgery for LSS in 250 patients we found ATTR amyloid in ligament specimens from 92 (Eldhagen et al., manuscript in preparation). Deposits were graded 1-4+ and 41 (16%) of the patients had extensive deposits (3-4+).

**Objectives:** To investigate whether materials taken at surgery for LSS show histologic signs of systemic disease including deposits in adipose tissue and vessel walls in proximity to the ligament tissue.

**Methods:** Ligament materials from all patients with amyloid deposits of grade 3-4+ were studied after staining with mab 7X against ATTR and with Congo red. Some of the materials were double stained with the endothelial cell marker CD31 for visualization of blood vessels and then with Congo red. Congo red stained sections were analysed in a polarization microscope.

**Results:** The material was initially collected for finding amyloid in ligament tissue. However, adipose tissue was identified in the material outside ligaments from 14 patients, and 7 of them contained ATTR amyloid encircling fat cells similar to what is seen in fat tissue of subcutis or pericardium. Furthermore, ligament is almost completely non-vascularized but ATTR amyloid was seen in small vessel walls outside ligament in material from 3 patients.

**Conclusion:** Although not a definite proof, these findings indicate that ATTR amyloid deposits in lumbar spinal ligaments are part of systemic amyloidosis albeit not (yet) giving clinical symptoms from other organs.
TIME COURSE OF TRANSTHYRETIN REDUCTION AFTER INOTERSEN THERAPY GIVEN WITHOUT A LOADING DOSE FOR TRANSTHYRETIN AMYLOID CARDIOMYOPATHY

R. Falk ¹, E. Wong ¹, C. Walker ¹, S. Cuddy ¹

¹Brigham and Women’s Hospital, Harvard Medical School.

Introduction: Inotersen significantly slows the progression of TTR familial amyloid polyneuropathy (FAP). In the pivotal trial leading to its approval in N. America and Europe, inotersen was administered as 300 mg sc weekly, after an initial loading dose of 300 mg x 3 in week 1. Mean TTR change from baseline was approximately -72%, with almost all this reduction achieved by 6 weeks. Unlike FAP, most patients with clinically significant amyloid cardiomyopathy have wild-type (ATTRwt), and it is unknown whether their TTR response to inotersen will differ from ATTRm. We here present data evaluating this.

Methods: The INOCARD study is an ongoing open label study of sc inotersen, given without a loading dose, to determine the effect of inotersen on multiple functional, echo, MRI and biomarker parameters in patients with predominant ATTR cardiomyopathy over a 2-year period. 300 mg sc inotersen is self-administered weekly, with platelet and renal function monitored every 2 weeks. TTR is measured at baseline, weeks 4, 6 and 8 and then monthly. No loading dose is given.

Results: 20 patients (1 woman) age 63-82 (mean 72 +/- 7) were studied. 17 had ATTRwt and 2 had ATTRm (2 due to the V122Ile mutation). All had symptomatic heart failure. Mean baseline TTR level was 23.5 mg/dl, with a 39.7% reduction at 6 wk. and near-maximal reduction by 20 weeks. (table). The 2 patients with V122Ile mutation had the most rapid decline in TTR, and the lowest nadir, with a reduction by 6 weeks of 85.8% and 72.2% respectively.

| PERCENTAGE REDUCTION IN TTR LEVELS FROM MEAN BASELINE TTR OF 23.5 MG/DL |
|---------------------------|----------------|----------------|----------------|----------------|----------------|
| Week 4 (20*)             | Week 6 (20)   | Week 8 (19)   | Week 12 (18)  | Week 16 (14)  | Week 20 (9)   |
| 39.7%                    | 48.3%         | 62.2%         | 63.7%         | 70.8%         | 74.7%         |

*Numbers in parentheses represent number of patients with TTR levels.

Conclusions: Inotersen 300 mg administered subcutaneously without a loading dose in patients with ATTR cardiomyopathy produces a similar suppression of TTR levels as seen in ATTRm FAP, but over a longer time frame (16-20 weeks versus 6 weeks). Whether the longer time to nadir represents the lack of a loading dose or the predominance of ATTRwt patients is unclear, but the observation that the 2 ATTRm patients had a more rapid fall may indicate at least a component of differential response between ATTRm and ATTRwt TTR suppression.
EFFECT OF TAFAMIDIS ON TTR LEVELS: A POST-MARKETING STUDY IN “REAL-WORLD” PATIENTS

S. Cuddy 1, M. Coyle 1, S. Dorbala 1, R. H. Falk 1

1 Brigham and Women’s Hospital, Harvard Medical School.

Introduction: Transthyretin (TTR) stabilization results in an increase in serum TTR and levels of increase may act as a surrogate for the potency of the drug, or drug dose. The ATTR-ACT trial for TTR amyloid cardiomyopathy found that both 20 mg and 80 mg tafamidis meglumine were superior than placebo on clinical outcome, but no data on TTR levels were published. Subsequently the US Food and Drug Administration has approved tafamidis meglumine 80 mg, or the bioequivalent tafamidis 61 mg, for the treatment of TTR cardiomyopathy. The ATTR-ACT results implied drug-induced slowing of cardiac disease but not cessation and it has been suggested that more potent TTR stabilizers may have had a greater clinical effect. We therefore determined the effect of approved doses of tafamidis 80mg and 61mg on TTR levels in a group of patients prescribed the drug following FDA approval and release in the USA, and compared the change in TTR levels seen to those published in the literature in clinical trials of tafamidis 20mg, diflunisal and AG10. Pre-amyloid TTR oligomers have been shown to be toxic in vitro. We therefore also sought to determine whether stabilization of TTR, which presumably decreases toxic pre-amyloid oligomers, would have any effect of markers of cardiac dysfunction, specifically NTproBNP and troponin levels (similar to the decrease seen in AL amyloidosis when toxic light chains are lowered).

Methods: Serum TTR was measured at baseline and after 1-3 months therapy in 24 pt. prescribed tafamidis 80 or 61 mg for TTR amyloid cardiomyopathy. NtproBNP and high sensitivity troponin levels were also measured.

Results: 21 pt had ATTRwt, and 3 had ATTRm. Mean age was 80.0±5 years, with 5 women. Nine had a baseline TTR level below the normal range of 20-40 mg/dl. Following tafamidis, all TTR levels were normal.

<table>
<thead>
<tr>
<th>TTR LEVEL *</th>
<th>PRE-TAFAMIDIS</th>
<th>POST-TAFAMIDIS</th>
<th>P VALUE</th>
</tr>
</thead>
<tbody>
<tr>
<td>NTproBNP*</td>
<td>4380 (652) pg/ml</td>
<td>4424 (647) pg/ml</td>
<td>NS</td>
</tr>
<tr>
<td>HS-troponin*</td>
<td>64 (7.3) ng/l</td>
<td>72 (10.1) ng/l</td>
<td>NS</td>
</tr>
</tbody>
</table>

*(all values +/-SEM)

Discussion and Conclusions: 1. In this “real world” study of tafamidis therapy in an elderly patient population with ATTR cardiomyopathy, tafamidis therapy successfully increased serum TTR levels measured after approximately 3 months of therapy. The 34.3% increase in TTR levels compares favorably with previously published increase in TTR levels with 20 mg tafamidis (17%), diflunisal 250 mg bid (25%) and AG10 400 mg bid (29%) and is identical to that reported with AG10 800 mg bid (34%); 2. The failure of tafamidis to lower NTproBNP or high-sensitivity troponin despite it stabilizing effect on TTR may suggest that pre-amyloid oligomer cellular toxicity does not play a major role in human amyloid cardiomyopathy.
CONCURRENCE OF AUTOINFLAMMATORY DISEASE AND AA AMYLOIDOSIS: THE MOUNT SINAI EXPERIENCE (1945-2020)

P. Gorevic 1

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Introduction: The first full clinical description of Familial Mediterranean Fever (FMF) was reported in 1945 from Mount Sinai Hospital in New York by Dr. Sheppard Siegal as “Benign Paroxysmal Peritonitis”. One of the 5 patients in this first publication was in fact the author, who was profoundly affected by the disease through his death in 1988. By 1964, Dr. Siegal was able to describe 50 patients, reflecting the melting pot of nationalities referred to his practice, including a sephardic jew who developed renal amyloid. The full sequence of AA protein reported in 1972 included splenic amyloid from a patient with FMF, and positional cloning led to description of the MEFV (pyrin) gene in 1997 by two international groups studying patients from the countries surrounding “marenostrin”.

Objectives: We aimed to update the Siegal clinical and ethnic experience at our institution with a review of 90 carriers of MEFV variants and 62 cases of AA amyloidosis evaluated since 1997

Materials and Methods: This series includes 43 cases of AA and 51 with MEFV mutations previously published (Bunker and Gorevic, 2012; Bunker, Matza and Gorevic, 2015), with special attention to ethnicity and concurrence of the two entities. MEFV variants were determined by full genetic sequencing of pyrin, with reference to the infevers data base, common founder mutations, hetero-/compound hetero- and homozygosity. The prevalence of specific sequences was compared to published population surveys, a control panel used to assess reproducibility before MEFV testing was made part of our institutional carrier screen, and data retrieved from our Biobank. AA amyloidosis was defined by immunohistology using monospecific antibodies, or by direct extraction and characterization of subunit protein biochemically.

Results: Among AA amyloidosis patients, 13/62 (21%) had Inflammatory bowel disease (IBD; Crohns>>Ulcerative colitis); idiopathic AA accounted for 20/62 (31.7%) cases; in 3 instances, IBD associations were through Hidradenitis suppurativa or Turner’s syndrome. Among patients with defined MEFV mutations, 60% were heterozygous, 19% homozygous and 21% compound heterozygotes; phenotypic associations included classic criteria for FMF (Gottorno M. et al, 2019), variant serositis syndromes, persistent perplexing pyrexia, Still’s disease, and recurrent pleurisy or pericarditis. 32 (35%) were Ashkenazi and 23 (25%) Sephardic, geographic origins of the latter ranging from Morocco to Uzbekistan; significant associations included Behcets syndrome, “colitis”, and coexisting Yao (NOD 2 polymorphism) syndrome. 8 patients were from Turkey/Armenia, including one compound heterozygote presenting as MEFV phenotype 2. Variants were also identified for individual patients with idiopathic AA, AA associated with inflammatory arthropathy or crohns disease, localized AL and Lect2 amyloid.

Discussion: Carrier rates for MEFV mutations have been reported in up to 1:5 among moroccan jews and 1:3 among armenians, the latter recently by the Genetics Institute in Yerevan. Concomitance with SAA polymorphisms has been linked to an excess incidence of AA amyloidosis. These associations were likely reflected in Dr. Siegal’s original experience with what we now recognize as FMF, and the identification of patients with AA amyloidosis or MEFV carriers among the ethnically diverse populations being referred to our hospital.
SERUM AMYLOID A: STRUCTURAL BASIS FOR A VITAL FUNCTION AND MISFOLDING

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2 Boston University, Chemistry.
3 Northeastern University, Chemistry.
4 Int. of Protein Biochemistry, Ulm University.

Introduction: SAA (12 kDa), a protein precursor of secondary AA amyloidosis, is a biomarker of inflammation that has been highly evolutionally conserved for over 500 mln years [1]. SAA plasma levels increase swiftly and dramatically in inflammation, infection and injury, suggesting that it must have played a vital yet poorly understood role. This role involves lipid transport [2-5], as most circulating SAA is bound to high-density lipoproteins. In vitro SAA can solubilize diverse lipids and their degradation products to form lipoprotein nanoparticles [5]. Binding to lipoproteins protects SAA from forming amyloid [5-7]. Our goals are: i) to determine the vital beneficial role of SAA in lipid transport and obtain its structural underpinnings, and ii) to dissect the protein misfolding pathway.

Approach: During inflammation SAA increases simultaneously with another lipophilic acutephase reactant, secretory phospholipase A2 (sPLA2). This compelled us to explore potential synergy between these proteins by using model and plasma lipoproteins analyzed by and array of biochemical techniques. To determine the lipid binding mechanism, we used hydrogen-deuterium exchange mass spectrometry, molecular dynamics simulations and other biophysical approaches to explore SAA in lipid-free form and on model lipoproteins. Recombinant murine SAA1 was used in all experiments.

Results and Discussion: Our biochemical studies showed that SAA enhances lipolysis by sPLA2 via a dual effect: it forms lipoprotein nanoparticles that provide substrates for sPLA2, and it solubilize its water-insoluble reaction products [4]. We also established the synergy of SAA and other lipolytic enzymes that are upregulated in inflammation. This synergy, combined with the protein’s ability to solubilize diverse lipids and their degradation products [5], suggests that SAA acts as a lipid scavenger that transports lipids and their potentially toxic bioactive derivatives from the sites of injury, which is prerequisite for tissue healing. We postulate that the ability to safely remove cell membrane debris from the sites of injury constitutes the evolutionally conserved function of SAA. To obtain structural underpinnings for this function, we used experimental and computational approaches. The results showed that solution conformations of lipid-bound and lipid-free murine SAA1 at pH7 agreed in remarkable details with the crystal structures [2,3] but also revealed important differences. Amphipathic α-helices h1 and h3 comprise the lipid-binding site that is partially preformed in solution and is stabilized on lipoproteins, including induced folding of h3. This site sequesters apolar ligands in SAA oligomers via a concave hydrophobic face. The largely disordered C-terminal region mediates promiscuous binding of other functional ligands, including cell receptors. Unexpectedly, the charge-rich h1-h2 linker region forms a β-hairpin that likely represents a previously unknown early amyloidogenic intermediate in the mSAA1 monomer. Specific interactions stabilizing this β-hairpin and its relevance to the fibril structure of human and murine SAA1 [8] are revealed. The results establish structural underpinnings for understanding SAA interactions with lipids and other functional ligands, its evolutional conservation, and amyloid formation.

CAUSES OF AA AMYLOIDOSIS: A SYSTEMATIC REVIEW

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From a clinical perspective there is a need for a reliable and comprehensive list of diseases causing AA amyloidosis. This list could guide clinicians in the evaluation of patients with AA amyloidosis in whom an obvious cause is lacking. In this systematic review a Pubmed, Embase, and Web of Science literature search was performed on causes of AA amyloidosis published in the last four decades. Initially 4066 unique titles were identified, but only 795 full-text articles and letters were finally selected for analysis. Titles were excluded because of non-AA type of amyloidosis, language, no full-text publication or irrelevant. Hundred and fifty diseases were initially reported to be associated with the development of AA amyloidosis. The presence of AA amyloid was proven in 208 articles (26% of all) of which 140 (67%) showed a strong association with an underlying disease process. Disease associations were categorized and 48 were listed as strong, 19 as weak, 23 as unclear, and 60 as unlikely. Most newly described diseases are not really unexpected because they often cause longstanding inflammation. Based on the spectrum of identified causes, a pragmatic diagnostic approach is proposed for the AA amyloidosis patient in whom an obvious underlying disease is lacking.
Introduction: Systemic AA amyloidosis is a worldwide occurring protein misfolding disease of humans and animals. It arises from the formation of amyloid fibrils from the acute phase protein serum amyloid A. AA amyloid fibrils are characterized by a linear morphology and a cross-β structure. A central aspect of the pathology of systemic amyloidosis is that they form massively sized deposits that physically impair and distort the affected tissues. Amyloid fibrils also underlie the prion-like characteristics of systemic AA amyloidosis in mice and several other animal species. Injection of purified amyloid fibrils, fibril fragments, oligomers or spleen extracts from amyloidotic donors into inflamed mice transmits the disease between animals. We are using electron cryo-microscopy (cryo-EM) to determine the structures of AA amyloid fibrils from a patient and from a diseased mouse.

Objectives: The goal is to reconstruct high resolution structures of ex vivo AA amyloid fibril from mouse and human to create a molecular model. This allows to gather detailed structural information on AA amyloid fibrils and to help develop a fundamental molecular understanding of the mechanism of disease by comparing the two structures.

Methods: AA amyloid fibrils were extracted from the kidney of an AA amyloidotic patient and from the spleen of a mouse diseased with systemic AA amyloidosis based on a pre-existing protocol. Samples were plunge frozen. Collected data was reconstructed using single particle based helical reconstruction with RELION.

Results: The obtained resolutions are 3.0 Å and 2.7 Å for the murine and human fibril, respectively. The two fibrils differ in fundamental properties, such as presence of right-hand or left-hand twisted cross-β sheets and overall fold of the fibril proteins. Especially the more C-terminal segments differ substantially in conformation between the human and the murine fibril. Yet, both proteins adopt highly similar β-arch conformations within the N-terminal ~21 residues.

Conclusions: Our data identify the N-terminal ~21 residues of the fibril proteins, the most hydrophobic and amyloidogenic segment of the protein sequence and as crucial for structuring disease-associated AA amyloid fibrils. A single amino acid changes within this region can make the protein incompatible with the observed fibril architecture. For example, hSAA1.1 contains an additional N-terminal Arg residue compared to mSAA1.1. This residue is missing in human fibril protein and incompatible with the packing of the observed fibril structure. The reconstruction shows that the N-terminus of the human protein, which lacks the N-terminal Arg, is located within the tightly packed fibril core. This data demonstrate the importance of the fibril protein N-terminus for the stability of the analyzed amyloid fibril morphologies.

Keywords: AA amyloidosis, amyloid fibril structure, cryo-EM.
Introduction: Secondary amyloidosis associated with chronic inflammatory diseases such as rheumatoid arthritis (RA) is derived from serum amyloid A (SAA). There is known to be an association between SAA gene polymorphisms and amyloidogenesis. The SAA gene consists of subtypes SAA1, SAA2, SAA3 and SAA4. SAA1 is mainly related to the production of both SAA and amyloid precursor protein. There are three major allelic variants (SAA1.1, SAA1.3, and SAA1.5) of the SAA1.1 gene, each being equally represented in the Japanese population whereas SAA1.3 is rare in Caucasians. There is a strong correlation between SAA1.3/1.3 homocarriers and AA amyloidosis, and the SAA1.3 allele is a risk factor for the latter. However, the SAA protein isoforms deposited in various organs and their distribution have been insufficiently studied.

Materials & methods: A 50-year-old woman had been diagnosed RA at the age of 22 years, and renal biopsy had revealed AA amyloidosis at the age of 35 years. She died due to pancreatitis at the age of 50. Autopsy revealed AA amyloidosis and amyloid deposits in all organs including the kidneys. Renal autopsy specimens were stained with Congo red for histopathological diagnosis. Immunohistochemical analysis confirmed these deposits to be AA-type amyloid. Amyloid fibril proteins were investigated biochemically by predicted reaction monitoring with LC-MS/MS. Imaging MS was also used to confirm the location of SAA polymorphisms.

Results: In renal autopsy specimens, Congo red staining demonstrated dense deposits of amyloid protein in the renal medulla and scattered deposits in the cortex. Immunohistochemistry confirmed these deposits to be AA-type amyloid. LC-MS/MS revealed SAA isoforms 1.1, 1.3, 1.4, and 1.5. SAA 1.1 and 1.3 showed strong signals in both the cortex and medulla; SAA 1.4 and 1.5 were also present in both the cortex and medulla, but the signal was weaker in the former than in the latter. Merged images of the SAA 1.1 and 1.3 isoforms showed a similar distribution in the cortex, but SAA1.1 was more strongly deposited than SAA1.3 around interlobar arteries in the medulla. Merged images of the SAA 1.1 and 1.4 isoforms also showed a different distribution.

Conclusions: LC-MS/MS detected SAA subtypes 1.1, 1.3, 1.4, and 1.5 in the renal tissue of this patient with AA amyloidosis associated with RA. Among the SAA isoforms, SAA1.1, 1.3, 1.4 and 1.5 were deposited in both the medulla and cortex of the kidney, but each of these samples showed different signal strengths and distributions. Further investigation of the SAA gene subtypes in this patient will be necessary.

ANALYSIS OF AA PROTEIN SPECIES IN SERIAL BIOPSY SAMPLES FROM AA AMYLOIDOSIS PATIENTS BY MALDI-TOF MS

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Introduction: In AA amyloidosis, AA proteins truncated and modified from serum amyloid A (SAA) are fibrillized and deposited in tissues. Polymorphism of the major isotype SAA1 is known to affect disease susceptibility. This study assessed changes in the relative amount or structure of AA protein species in serial biopsy samples from AA amyloidosis patients by MALDI-TOF-MS.

Materials & methods: Samples: Amyloid-laden tissue samples were obtained by endoscopic biopsy from gastric mucosa of AA amyloidosis patients every year for 2 years. A total of 27 samples from 9 AA amyloidosis patients were analysed. The SAA1 polymorphism was 1.1 / 1.3 for 1, 1.1 / 1.5 for 1 patient, 1.3 / 1.3 for 1, 1.3 / 1.5 for 5, and 1.5 / 1.5 for 1 patient, respectively.

Preparation of AA proteins: One mg of the biopsy sample was treated with 4 M guanidine and the AA proteins were purified by immunoprecipitation using anti-SAA monoclonal antibody-conjugated magnetic beads.

MALDI-tof ms: Mass spectrometry was performed on a matrix-assisted laser desorption time-of-flight mass spectrometer (MALDI-TOF MS; ultrafleXtreme, Bruker Japan). The prepared samples were desalted and concentrated by ZipTip C-18 (Merck Millipore), and then measured in linear positive mode. Spectra were analysed by flex analysis (Bruker Japan).

Results: As AA protein species in the samples, 1-76, 2-76, 5-76 amino residue fragments originating from SAA1 (1.1, 1.3, or 1.5) and SAA2.1 were detected. Analysis of the heterozygote samples containing the SAA1.3 allele revealed that the relative amount of 1.1 or 1.5 derivates slightly increased or decreased among the overall deposited amount of AA proteins, whereas that of 1.3 derivates were often constant. Regarding amino terminal degradation, as for 1.1 or 1.5 derivates, when overall deposition was increased, the relative amount of 1-76 fragments slightly increased, and when overall deposition was decreased, the relative amount of 2-76 or 5-76 fragments slightly increased. On the other hand, as for 1.3 derivates, 2-76 fragments remained relatively stably as the major species regardless of the amount of overall deposition.

Discussion & conclusions: This study confirmed that AA proteins in biopsy samples existed as different amino terminally truncated forms. The presented results suggest the stability of SAA1.3-derived AA proteins in the tissues. This may reflect the disease severity observed in patients with the corresponding SAA1 polymorphism.
A RARE CASE OF SIMULTANEOUS SYSTEMIC AA AND AL AMYLOIDOSIS IN A PATIENT WITH JUVENILE ARTHRITIS

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Introduction: Systemic amyloidosis originates from amyloidogenic precursor proteins misfolding in a beta-sheet structure. Deposition results in severe organ dysfunction [1]. The identification of underlying pathways is crucial to decide on therapy differing widely among subtypes. It is often challenging to deduce the right diagnosis since symptoms overlap. A nephrotic syndrome is often described in AL amyloidosis patients and is also the principal characteristic of AA amyloidosis [2]. Histological work up with e. g. immunohistochemically subtyping of the amyloid is required for discrimination.

Case: Here, we describe the case of a 38 year old female in whom two types of amyloidosis were discovered consecutively. She presented to our outpatient clinic for further consultation due to newly diagnosed AL-amyloid deposits in transplant kidney samples. In her medical history, juvenile arthritis was diagnosed at age of 3 years and typical treatment was commenced immediately. AA amyloidosis was first detected at age of 11 in skin samples. Further biopsies revealed organ involvement of the liver and kidneys at age 13 and 18 respectively. At age 29, allogeneic kidney transplantation was performed due to terminal kidney failure. Monoclonal gammapathy type IgA lambda was detected two years later. In early 2019 the patient presented with reduced physical performance and urine analyses demonstrated a deterioration of proteinuria. Diagnostic renal biopsies exposed AL amyloid fibrils type lambda. Bone marrow biopsies revealed a lambda plasma cell infiltration of less than 10 percent. Cardiac biomarker levels were normal and echocardiography ruled a septal hypertrophy. To prevent additional organ damage chemotherapy with bortezomib, cyclophosphamide and dexamethasone was initiated. So far, two cycles have been administered without further complications and a complete hematologic remission was achieved. Autologous stem cell transplantation can be evaluated, however, may only be conducted in relapsing disease regarding the reduced general condition.

Conclusion: There is only limited research on the coexistence of two types of amyloid deposition in the same patient so far. Studies mainly refer to a simultaneous occurrence of AL and TTR wild type amyloidosis, which will likely be encountered more frequently due to an ageing population with constant improvement of myeloma therapy [3-5]. Nevertheless, monoclonal plasma cell neoplasms are rather often described in rheumatoid diseases, although there is not enough evidence supporting the pathogenic mechanisms linking rheumatoid diseases to MGUS [6]. Chronic inflammation may accelerate the formation of aberrant clonal plasma or B cells. Hence, a screening of AA patients for MGUS and possibly a subsequent re-subtyping of amyloid deposits in the course of disease seems reasonable. A correct subtyping of amyloid fibrils is, nonetheless, essential to initiate adequate therapy and prevent end organ failure.

Keywords: AL amyloidosis, AA amyloidosis, clinical features, therapy.
NEW IMMUNOASSAYS FOR THE SELECTIVE MEASUREMENT OF SERUM AMYLOID A ISOFORMS 1 AND 2 IN HUMAN BLOOD

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Human serum amyloid A (SAA) is a soluble plasma precursor of amyloid A, which is a major component of amyloid fibrils deposited during secondary amyloidosis. The SAA isoforms SAA1 and SAA2 belong to the group of acute phase proteins. SAA is a sensitive marker of inflammation. Under inflammatory conditions total concentration of SAA1 and SAA2 in blood increases by up to 1,000-fold. It has been reported that SAA1 and SAA2 expression levels vary in response to different inflammation stimuli (Thorn et al., 2003).

Consequently, it has been suggested that relative SAA1 and SAA2 content in blood could vary in different inflammatory diseases and that the differential measurement of SAA isoforms might have additional clinical value. SAA1 and SAA2 share 92% identity in the primary structures. Existing immunochemical methods for SAA measurement detect both SAA isoforms. Recently, Xu et al. developed immunoassays that enable the measurement of SAA1 and total SAA (Xu et al., 2006). The SAA2 concentration in samples was determined indirectly by the subtraction of the SAA1 concentration from the total SAA concentration. However, the development of immunochemical methods for the direct measurements of both SAA1 and SAA2 concentrations has not yet been reported.

In this study, we developed three sandwich immunoassays for the detection of SAA1, SAA2, and total SAA. The monoclonal antibody SAA80, which equally recognizes both SAA isoforms, was used as a capture antibody in all of the assays. Meanwhile, the monoclonal antibodies SAA100 (which is specific to SAA1), 2SAA65 (which is specific to SAA2) and VSA31 (which equally recognizes both SAA1 and SAA2) were used as detection antibodies. Recombinant SAA1 was used as a calibrator for assays that were specific for SAA1 and total SAA, while recombinant SAA2 was used as a calibrator for the SAA2 assay.

Developed assays were used in order to evaluate the content of SAA isoforms in the plasma of patients with inflammation caused by tissue injury. SAA1, SAA2 and total SAA concentrations were determined in EDTA plasma samples that were collected from 28 patients who underwent surgery within 7 - 62 hours following the surgery. The total SAA concentration in plasma samples ranged from 2.6 to 293.0 mg/L (mean 52.1 mg/L). SAA1 was the predominant isoform in all tested samples. The SAA1/total SAA ratio was 80.4±5.5% (mean±SD), while the SAA2/total SAA ratio was 15.9±7.1% (mean±SD). The sum of SAA1 and SAA2 concentrations that were determined by isoform-specific assays comprised 96.3±5.8% (mean±SD) of the total SAA concentration for each patient. This confirms the reliability of the proposed method of detection.

To conclude, in order to allow for the selective measurement of SAA isoforms 1 and 2 in human blood, we have developed immunoassays that have not been described before. In the small group of post-operation patients, we were able to demonstrate the accurate measurement of SAA1 and SAA2 by these assays.
Background: Therapeutics of AA are directed against the underlying cause and in the idiopathic ones to the blockade of the inflammation. Organic involvement, evolution time, comorbidities, treatment availability and underlying disease are likely to have an impact on treatment options.

Aim: Describe treatments in AA amyloidosis. Estimate global survival

Methods: Retrospective cohort of patients with amyloidosis AA in the Institutional Registry of Amyloidosis (NCT01347047) at Hospital Italiano de Buenos Aires from 2010-2019. All patients were followed to death for all causes. Survival rates are expressed as the percentage surviving calculated using Kaplan Meier method.

Results: During the period, 160 patients with amyloidosis were included, 14% (23) had diagnosis of AA. 48% were female (11), with a median age of 52 years (RII 49-64). The diseases that caused the deposition of SAA were, in order of frequency, idiopathic, autoimmune and infections. The kidney was the most frequently affected organ (86%), followed by the heart (43%) and the digestive tract (32%).

During follow-up, 4 patients died, the overall mortality rate was 17% (n = 4, CI 6-40%). The cause of death in three of the 4 patients was related to amyloidosis, one patient died of heart failure, one patient died of end-stage renal failure, a patient of refractory septic shock, and one patient died of accidental traumatic cause, not related to underlying disease. Thirteen patients (56%) received treatment, and one died of sepsis. At the beginning of treatment 60% had a PS of 2 and a Charlson score of 4. Biologic agents were administered to 54% of patients with AA (7). Tocilizumab (TCZ) was the biologic agent most frequently indicated (7) as first or second line of therapy, two patients received etanercept and canakinumab as first and second line previously to TCZ, and two patients received cyclophosphamide and mycophenolate. All patients showed good responses to TCZ. Antimicrobial agents were prescribed in 6 patients. All treated patients but one, stabilized their condition, improved inflammation biomarkers and achieved a normal nutritional status.

Conclusion: Half of the patients received treatment, directed to the underlying disease or in idiopathic cases directed to inflammation. Treatment is critic in stabilizing the disease progression Overall prognosis and survival are similar to other series.

Key words: AA amyloidosis, treatment, prognosis
SYMPTOMATIC AA-TYPE AMYLOIDOSIS IN A PATIENT WITH LATE-ONSET CRYOPYRIN-ASSOCIATED PERIODIC SYNDROME DUE TO SOMATIC NLRP3 MOSAICISM

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Introduction: AA amyloidosis is a form of systemic amyloid deposition that complicates the clinical course of different inflammatory or infectious chronic diseases and that are typically associated with long-lasting increased plasma levels of acute phase reactants (APR). Some of these chronic diseases are genetic in nature, being the incidence of AA amyloidosis moderate-to-high in TNF receptor-associated periodic syndrome and cryopyrin-associated periodic syndromes (CAPS).

Objectives: To elucidate the cause of AA amyloid deposition in a patient who suffered since adulthood from recurrent inflammatory episodes and to describe the outcome of anti-IL-1 treatment.

Methods: Clinical data and results of laboratory tests were collected from patient’s medical charts. Genetic tests for monogenic autoinflammatory diseases (AID) were performed by next-generation sequencing using DNA extracted from peripheral blood.

Results: We present the case of a 61-year-old woman, who was born from a Spanish non-consanguineous couple and with no familial history of inflammatory, autoimmune or primary immunodeficiency. Since the age of 40 years she suffered from recurrent episodes of urticaria-like rash, fever, conjunctivitis, oligoarthritis (knees, wrists, elbows, and ankles), and increased APR. At the age of 59 she developed fecal incontinence as well as proteinuria (12 gr/24h), with biopsies revealing AA amyloid deposits. Despite the late age at disease onset, a CAPS diagnosis was suspected. Genetic tests for monogenic AID exclusively revealed the likely pathogenic p.Gln306His NLRP3 variant, with a mutant allele frequency of 5.1%, supporting the diagnosis of late-onset CAPS due to somatic NLRP3 mosaicism. Anti-IL-1 treatment with daily anakinra was started, which resulted in disappearance of inflammatory episodes, decrease of APR and improvement of renal function with progressive reduction of proteinuria.

Conclusions: We identified an adult patient carrying a somatic NLRP3 mosaicism as the cause of a form of late-onset CAPS complicated with AA-type amyloidosis. Despite patients with late onset CAPS are rare and are not usually complicated with amyloidosis (only one patient reported to date), the patient here described highlights the need to consider this disease in the differential diagnosis of AA-type amyloidosis, especially for the consequences that it may have from a therapeutic point of view.

Key Words: Cryopyrin-associated periodic syndromes, NLRP3 mosaicism, AA-type
SERUM AMYLOID A (SAA)-INDUCED INFLAMMASOME ACTIVATION IN INNATE IMMUNE CELLS

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Background: Serum amyloid A (SAA) is an acute phase reactant with significant immunological activities, including cytokine synthesis and neutrophil chemotaxis. IL-1β is a key proinflammatory cytokine and its secretion is controlled by inflammasome.

Objective: We investigated the proinflammatory effects of SAA on NLRP3 inflammasome in innate immune cells.

Methods: Human neutrophils isolated from healthy subjects were stimulated with serum amyloid A (SAA). Cellular supernatants were analysed for IL-1β or caspase-1 by ELISA. IL-1β mRNA expressions and Nod-like receptor family, pyrin domain containing 3 (NLRP3) protein expressions were analyzed by real-time PCR and immunoblot.

Results: SAA stimulation induced pro-IL-1β mRNA expression in neutrophils. Furthermore, SAA engaged inflammasome activation, resulting in the production of cleaved caspase-1 (p20) and IL-1β. SAA induced pro-IL-1β mRNA and NLRP3 protein expression. Analysis of intracellular signaling revealed that SAA stimulation activated NF-κB. Hydroxychloroquine (HCQ) pretreatment significantly inhibited the SAA-induced IL-1β production in human neutrophils, but did not affect the SAA-induced NF-κB activation or NLRP3 protein expression.

Conclusion: Two-step activation process are required for inflammasome activation: an initial signal promoting expression of pro-IL-1β and a second signal involving activation of the NLRP3 inflammasome and caspase-1, allowing processing of pro-IL-1β and secretion of mature IL-1β. SAA stimulation resulted in NLRP3 inflammasome activation and IL-1β secretion in neutrophils without the need for a priming signal. HCQ affects this SAA-induced NLRP3 activation process, resulting in the impaired IL-1β production in human neutrophils, as representative innate immune cells.
FIRST RECORD OF SAA BLOOD LEVELS IN EUROPEAN BROWN HARES (LEPUS EUROPÆUS)

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Introduction: In AA amyloidosis, the protein deposits are derived from the acute phase reactant, serum amyloid-A (SAA). An increase of this acute phase reactant in the blood is therefore linked to the entity. In veterinary medicine, amyloidogenic SAA-isotypes have been recognized in some species (e.g. mouse, mink, horse). However, in wildlife medicine, not much is known about the entity itself, and there are little or no reports in the literature about the blood levels of SAA in wild animal species. In the course of an experiment to induce AA amyloidosis in European brown hares, 4 blood samples were taken from each animal to get a first insight into the serum levels of SAA in this species.

Material and Method: Hares were divided into 3 experimental groups: control (A; n=10); extracted hare amyloid fibrils (HAF) with drinking water (B; n=10); HAF with drinking water + 3 subcutaneous injections of silver nitrate (C; n=10) (ethic forms available on request). Other than this treatment, all hares were kept under the same conditions and received the same foodstuff. Blood samples of each animal were taken 4 times during the experiment and serum samples were frozen at -80°C for further analyses. To determine the concentrations of SAA, a multispecies sandwich ELISA was performed. The ELISA kit used was the PHASE™ range from Tridelta Development Ltd. and all steps were carried out according to the provided manual.

Results: Statistical analyses were carried out using the program R. No statistically significant differences were found between the three experimental groups in terms of SAA concentrations when controlled for sex, age, body weight and day of sampling. Although not statistically significant, the SAA levels in group C are considerably higher. The range of SAA concentrations in the serum of each experimental group are as following: group A: 0,08µg/ml - 36,9µg/ml; group B: 0,15µg/ml -35,4µg/ml; and group C: 0,16 µg /ml - 70,8µg/ml (Tab.1).

Discussion and Conclusion: Although no statistically significant differences were found, all hares in group C showed higher values than hares from the other two groups. Highest values were found at timepoint 2 (T2) - this was 1 day after the first treatment with silver nitrate (group C). Interestingly, no increased SAA values could be determined in the blood samples of hares that showed severe chronic infections at necropsy (i.e. pyometra, abscesses near ovary). However, these results give a first insight into the SAA levels in the blood serum of this species, if these could be used in diagnosis, and how SAA levels are affected when animals are treated with HAF and silver nitrate.

Tab.1 Shows the serum SAA values of each individual animal at 4 different timepoints.
HUMAN AA AMYLOID SUBUNIT PROTEINS - REVISITED BY MASS SPECTROMETRY

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Introduction: While sustained inflammation accompanied by elevated levels of serum amyloid A (SAA) is recognized as the most significant causal factor in AA amyloidosis, the specific triggers involved in conversion of SAA to AA amyloid remain unresolved. To advance understanding of this process we are examining human AA amyloid subunit proteins by mass spectrometry. This approach is providing data regarding the SAA isoform composition of AA fibrils, the C-terminal extent of AA proteins, and the identity and location of post-translational modifications.

Methods: AA fibrils were extracted from human tissues by repeated homogenization and centrifugation in sodium citrate/ sodium chloride followed by homogenization in water. Proteins in the floating fraction were separated by Sepharose CL-6B chromatography and visualized by SDS-PAGE and western analysis. Fractions containing a single low molecular weight species (~8 kDa) immunoreactive with anti-SAA antiserum were pooled, dialyzed into ammonium bicarbonate and analyzed by mass spectrometry (LC-MS/MS).

Results: Mass spectrometry data have been collected on AA proteins from 5 patients; samples from 3 additional patients are ready for analysis. With regard to isoform composition, 2 of the 5 samples contain peptides of both SAA1 and SAA2, while the other 3 are derived solely from SAA1. Samples 1 – 4 contain peptides covering the entire 104-amino acid sequence of SAA; the most C-terminal peptide of sample 5 ends at Arg87 in SAA1-derived AA and Lys84 in SAA2-derived AA. All 5 samples have N-terminal peptides beginning with Arg1 as well as peptides beginning with Ser2. Carbamylated Arg1 was identified in 4 of the 5 samples (not present in sample 5). Other post-translational modifications detected include carbamylated residues Lys34, Lys46, Arg47, Arg62 and Arg71; acetylated Arg1; and oxidized Met17 and Met24.

Discussion: In accordance with previous reports, SAA1 was found to be the predominant precursor of AA amyloid protein. While SDS-PAGE results showed a mass of ~8 kDa for all of the analyzed AA subunit proteins, peptides spanning the entire 104-amino acid sequence of 11.6 kDa SAA were detected. The presence of C-terminal peptides could indicate that cleavage of SAA to AA occurs post fibril formation and that the cleaved C-terminal peptides, while they do not comprise fibrils, are co-extracted. Alternatively, the C-terminal peptides could represent SAA contaminants detected via highly sensitive LC-MS/MS. Sample 5 not only yielded outlier LC-MS/MS data relative to the other 4 samples, but also was derived from a patient whose clinical course differed significantly from the other 4. Taken together, these data may provide insight into the temporal relationship of post-translational modification of SAA, cleavage of SAA to AA, and AA fibril formation.
AA (secondary, reactive) amyloidosis is most often a complication of chronic inflammation. AA amyloid deposition in the kidneys has serious clinical consequences, causing nephrotic syndrome and eventually death. Currently, no specific therapies exist to treat this disease. To facilitate development and testing of AA amyloid specific agents, we have developed a human serum amyloid A (SAA) transgenic (hSAATg/mSAAKO) mouse model. At the previous symposium we presented a transgenic human SAA1 mouse line carrying a 12 kilobase segment of the human genome which includes the SAA1 promoter and all regulatory elements (hSAATg). Deposition of human SAA-derived AA as well as mouse SAA-derived AA amyloid was demonstrated in the hSAATg mice. These mice have now been bred with mice that lack production of the three mice acute phase SAA genes (mSAAKO). The hSAATg/mSAAKO mice generated through this breeding provide an exclusively human model with respect to AA amyloidosis, i.e., co-deposition of murine AA is not a confounding factor. As shown by ELISA and western analysis, hSAATg/mSAAKO mice produce elevated levels of human SAA (1 mg/ml) as part of the acute inflammatory response. The mice are currently undergoing classic amyloid induction via repeated injections of azocasein to establish a chronic inflammatory state. Once the development of human SAA-derived AA amyloidosis has been characterized, the mice will be used to evaluate the ability of human-specific SAA antisense oligonucleotides to suppress production of SAA and assess if reduction of SAA levels in serum slows or halts progression of AA amyloidosis. The mice will also serve as a model for investigating the role that post-translational modification of SAA may play in AA amyloid fibril formation and propagation. The latter study is based on our in vitro finding that SAA modified by amino-terminal carbamylation is particularly susceptible to amyloid formation. Additionally, carbamylated residues have been identified on AA subunit proteins extracted from human amyloid deposits. To begin to probe the significance of these modified residues, newly formed amyloid will be extracted from hSAATg/mSAAKO mice and examined by mass spectrometry for the presence of carbamylated SAA and/or AA.
CURRENT TRENDS IN THE ETIOLOGY AND COURSE OF REACTIVE AMYLOIDOSIS

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Background: The key problem in secondary AA-amyloidosis (AA) is correct diagnosis and treatment of amyloidogenic inflammatory disease approaches to which have changed today. Our clinical study was carried out to study today’s changes in the etiology and course of systemic AA.

Material and methods: Among 140 AA patients hospitalized between 1995 and 2017, a group of 110 patients was followed for a long time. The diagnosis in all patients was confirmed morphologically. Standard clinical observation and laboratory tests was performed.

Results: With time series method it was proposed the exponential predictive model of AA development dependent on the time elapsed since the onset of inflammatory disease ($S_0=4.7$, $T_0=0.98$, $a=0.03$, $\gamma=0.1$, $\delta=0.1$). The greatest risk was observed during the first 20 years with the rate 4-5 out of every 100 patients. An 83% of the AA cases during the first 9 years had high activity of inflammatory disease ($p=0.02$). In the next 20 years, the rate decreased to 1-2 patients out of 100 and after 40 years of inflammation it was reached an absolute minimum. AA was induced by chronic infections in only 8.96%. A 39.6% of patients had AA caused by rheumatoid arthritis (RA). In the patients observed after 2002, when TNF-\(\alpha\) inhibitors were entered into wide clinical practice, the share of RA decreased from 48% (before 2002) to 34%. At the same time, the frequency of autoinflammatory diseases (AID) increased from 46% to 59%. The number of patients with familial periodic fevers (FPF) decreased from 35% to 15% ($p=0.015$) and the proportion of patients with polygenic AID (pAID) increased from 11 to 43% ($p=0.0004$). With the cases of Castleman’s disease (n=3) and sarcoidosis (n=2) the cumulative share of AID was 53.7%. Of the 22 AA patients who died, 14 with FPF and pAID had uremia. Only 1 patient with RA developed uremia and died (OR=4.2, conf.int. – 0.6-26.3). In 70% of RA patients uremia was not developed. By 14 years from the beginning of AA only 16.6% of RA patients had uremia but it was developed in 53% of patients with AID by this time. By 20-25 years – respectively 31.7% and 71.9%. The low renal survival curve in patients with pAID differed from curves of patients with RA, FPF ($p=0.035$). Only 38.7% patients with pAID had cytostatic or anticytokine therapy differing from 55% of RA patients received basis treatment. The median overall survival of AA patients in exponential distribution model ($\chi^2=46.9$, df=15, $p=0.000038$) was 18 years. The frequency of deaths from the AA beginning reached a peak by 16 years coincided with the peak of uremia in 49% of patients with pAID. After the second increase in mortality in 28 years from the AA beginning 18% of patients remained alive.

Conclusion: The most common causes of AA now are AID. The lack of adequate baseline therapy is the leading trigger of AA. The maximum risk of AA is noted in the first 20 years of inflammatory disease, which approves the need for regular screening biopsies during this period.
WHAT IS THE AMYLOID PROTEIN IN CANINE MAMMARY TUMOR-ASSOCIATED AMYLOIDOSIS?

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¹Tokyo University of Agriculture and Technology.

Introduction: Mammary tumor-associated amyloidosis (MTAA) in dogs is characterized by severe amyloid deposition in the stroma of mammary adenoma or carcinoma. Although the casein family is suspected as an amyloid protein of MTAA, the exact amyloid protein has not been identified. In this study, we attempted to identify an amyloid protein of canine MTAA.

Methods: Formalin-fixed paraffin-embedded specimens from 5 cases of canine MTAA were used for the following analyses. Amyloid deposits were collected from Congo red-stained sections using laser microdissection, and digested with trypsin or chymotrypsin, followed by mass spectrometry. Using the total RNA extracted from the paraffin sections, the nucleotide sequence was determined from the gene fragment amplified by RT-PCR. Amyloid fibrillization was evaluated using synthetic partial peptides (Glu64-Arg96, Phe108-Pro135) and recombinant protein (Asn56-Trp158) of canine α-S1-casein.

Results: As a result of mass spectrometry of the trypsin digested samples, α-S1-casein was detected at a high level from all cases, considered as a prime candidate of amyloid protein. Mass spectrometry using chymotrypsin suggested that Asn56-Trp158 fragment forms amyloid. Amino acid mutations were denied by mass spectrometry and genetic analyses. The synthetic partial peptides and recombinant protein of α-S1-casein formed amyloid fibrils in vitro.

Conclusion: In this study, α-S1-casein was strongly suspected as an amyloid protein of canine MTAA. α-S1-casein is a novel amyloidogenic protein that has not been reported so far. The present data suggest that amyloidogenic factors are due to protein fragmentation rather than mutation.

Supplement: Currently, antiserum is being prepared by immunizing mice with recombinant protein (Asn56-Trp158), and immunological analyses (immunohistochemistry, immunoelectron microscopy, Western blotting) using antiserum are planned.

Keywords: Dog, mammary tumor, mass spectrometry.
DEVELOPMENT OF DIAGNOSTIC ANTIBODIES AGAINST THE HEAVY CHAIN VARIABLE REGION FOR AH AMYLOIDOSIS

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3 Tokyo Metropolitan Institute of Medical Science, Tokyo, Japan.

Background: AH amyloidosis is a rare systemic form of amyloidosis that is associated with the deposition of immunoglobulin heavy chains. Since AH amyloid proteins are mainly composed of variable region fragments without the constant region in some patients, AH amyloid heavy chains often go undetected in immunohistochemistry (IHC) using commercially available antibodies. Therefore, detailed investigation of amyloid protein deposits is usually required for AH amyloidosis diagnosis. Although anti-variable region antibodies for IHC-based screening are needed in clinical practice, their development for AH amyloidosis has been elusive owing to the high variability of the variable region.

Objectives: This study aimed to develop easier diagnostic tools for AH amyloidosis, especially antibodies applicable for IHC and western blot analysis.

Methods: Based on the homology of the amyloid protein amino acid sequences in 4 earlier described AH amyloidosis patients, 3 types of rabbit polyclonal anti-heavy chain variable region antibodies were generated. Using them, IHC was performed on 11 AH amyloidosis patients diagnosed by proteomics analysis at our institute and 36 patients with other amyloidosis types (26 AL, 2 AA, 4 ATTR, 4 others) confirmed by IHC. Additionally, western blot analysis was performed on extracted amyloid protein and plasma samples in some AH amyloidosis patients.

Results: Using 2 out of the 3 antibodies in IHC, a positive outcome was obtained in 10 AH patients. Positive results were not seen in patients with other types of amyloidosis except for 2 patients with AL amyloidosis. In western blot analysis, an approximately 11 kDa amyloid protein band was clearly detected, and protein bands of same molecular weight as the amyloid protein were observed in patient plasma samples.

Conclusions: Two of the antibodies generated in this study may represent a useful diagnostic tool for AH amyloidosis associated with variable region fragments. Moreover, our data indicate the presence of free N-terminal variable region fragments in patient plasma, and their detection has potential as a diagnostic and therapeutic marker in AH amyloidosis.

Keywords: AH amyloidosis; variable region; diagnostic antibody
THE HIGH RELAPSE RATE OF LOCALIZED AL-AMYLOIDOSIS AFTER SURGICAL TREATMENT

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**Background and aim.** Localized AL-amyloidosis (AL\textsubscript{L}) is characterized by the local deposition of light chain amyloid fibrils. The optimal type of therapy for these patients is not well established. The aim of our study was to analyze treatment outcomes and relapse rate of AL\textsubscript{L}.

**Material and methods.** We recruited 343 patients (admitted to the Clinic from 1995 to 2017) with different types of amyloidosis. AL\textsubscript{L} was confirmed by isolated single organ involvement proved by Congo red positive and dichroic in polarized microscopy biopsy specimen and by the absence of monoclonal gammapathy by means of immunofixation and the serum free light chain assay (Freelite\textsuperscript{8}). Immunohistochemistry was performed to find immunoglobulin light chains deposition in amyloid and clonal plasma cells in most of the cases, especially in disputable localization (e.g. brain). Recurrence of the amyloid deposits at the site of origin or another site was defined as relapse. Progression-free survival was estimated by the Kaplan-Meier method.

**Results.** Thirty two (9.3\%) out of the 343 patients had AL\textsubscript{L}. The majority of patients were females (24/32). The most frequent types of AL\textsubscript{L} were laryngeal and tracheobronchial (n=13) and conjunctival (n=8). The other sites of AL\textsubscript{L} included pulmonary nodular (n=4), cutaneous (n=3), urine bladder (n=2), brain (n=1) and soft tissue (n=1). Amyloid deposits were tumor-like (n=19) or diffuse infiltrative (n=13). The median time to diagnosis was 23 months (range 8-55 months). Surgical treatment was performed in 26/32 (81\%) patients. Other treatments included dimethylsulfoxide instillation for urine bladder (n=2) and skin (n=2), topical corticosteroids for skin (n=1) with neither symptomatic, nor substantial effect on the local amyloid deposits. The relapse after surgical treatment was registered in 10/26 (38.5\%) cases. The median time to relapse was 62 months (Table). One patient with brain AL\textsubscript{L} recurred 3 times after each surgical excision. One patient with relapse of tracheobronchial AL\textsubscript{L} died of recurrent pneumonia and respiratory failure. One- and 5-year progression-free survival in total group was 95.0\% and 66.5\%, respectively.

**Conclusion.** The first-line treatment in our cohort of AL\textsubscript{L} was surgical excision of the amyloid deposits. We observed the high relapse rate (38.5\%). Thus more efficient treatment strategies are needed (e.g. radiotherapy for local plasma cell clone elimination).

**Key words.** localized AL-amyloidosis, surgical treatment, relapse rate

**Table.** Characteristics of patients with relapse of AL\textsubscript{L}.

<table>
<thead>
<tr>
<th>Patient</th>
<th>Age, years</th>
<th>Sex</th>
<th>Site of origin</th>
<th>Site of relapse</th>
<th>Time to relapse, months</th>
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<tbody>
<tr>
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<td>31</td>
<td>M</td>
<td>larynx</td>
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<td>18</td>
</tr>
<tr>
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<td>50</td>
<td>F</td>
<td>larynx</td>
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<td>F</td>
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NODULAR PULMONARY AMYLOIDOSIS: TO CUT OR NOT TO CUT, THAT IS THE QUESTION – A CASE REPORT

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Background: Localized pulmonary amyloidosis may present in 3 forms – diffuse alveolo-septal, tracheobronchial and focal nodular amyloidosis. The most prevalent of these forms is focal nodular amyloidosis, a disorder with good prognosis, usually treated with surgical resection or watch and wait strategy. Finding of one or more pulmonary nodules is usually incidental, most commonly it is revealed on various means of chest examination performed for different reasons, as patients are usually asymptomatic. The aim of this report is to present 3 cases of focal nodular amyloidosis.

Methods and results: All three patients were men, aged 68, 76 and 79 at the time of diagnosis, without history of significant respiratory diseases. Two of them were smokers presenting with long-term cough, one patient had significant cardiovascular history including symptomatic aortic valve stenosis and therefore presented with dyspnea on exertion. In all cases pulmonary nodules were incidental findings, either on chest X-ray or CT-angiography. As a next step each patient underwent PET/CT examinations which showed unilateral nodules of sizes 25x19x18mm (SUVmax 5.2), 19x20x17mm (SUVmax 3.3) and 18x12x9mm (SUVmax 1.2). Due to a high suspicion of malignant process, resections of affected lobes were carried out in all patients. Histological examination showed presence of amyloid deposits in all three cases and polyclonal lymphoplasmocytic infiltrate was described in one nodule. Systemic form of amyloidosis was excluded in all patients. Immunohistochemistry and mass spectrometry were used for amyloid typing and they revealed that there were two cases of kappa light chain and one case of lambda light chain which supports the usual finding of kappa light chain predominance in localized amyloidosis compared to lambda light chain build-up in systemic amyloidosis. An increased number of macrophages were described in all cases and we assume that they were responsible for the substantially heightened accumulation of fluorodeoxyglucose during PET/CT examination. Follow-up PET/CT scans to assess post-operative state showed no residual disease.

Conclusion: When assessing pathological lesions on chest examination, it is necessary to keep in mind the possibility of lung tumors, infectious granulomas, hamartomas but also localized amyloidosis. Although the patients in our center were treated with lung resections, it is also possible, after histological verification of the lesions, to periodically evaluate their metabolic activity with PET/CT scans, which tends to cease over time, and therefore only observe the patients.

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FOCAL ORBITAL AMYLOIDOSIS TREATED WITH BORTEZOMIB AND DEXAMETHASONE – A CASE REPORT

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Background: Localized amyloidosis is a rare entity among the amyloidosis disorders able to affect various organs and tissues such as urinary bladder, lungs, prostate, skin and gallbladder. The most significant difference from systemic forms of amyloidosis lays within the treatment – unlike the chemotherapy which is the preferred modality for systemic forms, localized forms are treated with resection of the affected tissue, radiotherapy or by watch and wait strategy. The aim of this report is to present a case of a patient with localized orbital amyloidosis, successfully treated with the combination of bortezomib with dexamethasone.

Methods and results: 38-year-old patient was admitted to our hospital with exophtalmos, increased lacrimation and impaired vision. No signs of diplopia nor movement restriction of the ocular muscles were present. Magnetic resonance imaging (MRI) examination showed tumorous finding of left orbit, possibly a neurofibromatous lesion. Patient underwent surgical resection and the histological examination showed amyloid deposits with the presence of monoclonal plasma-cell population secreting kappa light chains. Systemic amyloidosis was excluded. Follow-up MRI showed residual infiltration of orbit, without indication for another surgical resection due to high risk of subsequent nerve damage. Radiotherapy was also contraindicated with the reason being a risk of visual loss. Systemic therapy with 3 cycles of bortezomib and dexamethasone was therefore initiated followed by immediate alleviation of symptoms. Gradual regression of exophtalmos was detected on MRI 18 months after therapy, regular MRI follow-ups were carried out every 6 months until 42 months since the last cycle of bortezomib and dexamethasone. Currently, during his latest assessment 5.5 years after the therapy, the patient is free of clinical symptoms with stable finding on MRI.

Conclusion: Bortezomib-based therapy is in widespread clinical use in the treatment of systemic AL amyloidosis with good response rate. Until now only one case report with poor outcome was described using bortezomib in the treatment of localized hepatic amyloidosis. Our case presents a successful use of biological therapy in a patient with focal amyloidosis of orbit when other treatment options were not possible.

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EGF-CONTAINING FIBULIN-LIKE EXTRACELLULAR MATRIX PROTEIN 1: A NOVEL AGE-RELATED AMYLOID PRECURSOR PROTEIN

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Introduction: To date, 36 human amyloid precursor proteins have been reported [1]. However, unidentified amyloid precursor proteins still exist. In this study, of them, we focused on an amyloid of unknown origin in venous walls and attempted to identify an amyloid precursor protein. In addition, we investigated the occurrence and biochemical features of this amyloid.

Materials and Methods: 1) To identify an amyloid precursor protein, we performed mass spectrometric analysis with amyloid-laden tissues isolated from intestinal portal veins by means of laser microdissection. 2) Autopsied tissue specimens obtained from 98 consecutive patients who had died at the age of 60 years or older were examined by immunohistochemistry with anti epidermal growth factor-containing fibulin-like extracellular matrix protein 1 (EFEMP1) antibody. 3) Extracted amyloid fibrils were analyzed by Western blotting with several anti EFEMP1 antibodies and mass spectrometry. 4) EFEMP1 mRNA expression levels in senescent human umbilical vein endothelial cells and aged mice were examined by reverse transcription-quantitative-PCR.

Results: 1) In amyloid deposits in the intestinal portal veins, EFEMP1 was most abundant in proteins detected by mass spectrometry. ApoE, clusterin and vitronectin were also detected. A monoclonal antibody to this protein reacted specifically with the amyloid deposits in intestinal venous walls by immunohistochemistry. 2) Our postmortem analyses showed that EFEMP1 amyloid deposits frequently developed in systemic venous walls of elderly people. 3) Biochemical analyses indicated that C-terminal portions of EFEMP1 were predominantly in intestinal venous amyloid deposits. 4) EFEMP1 mRNA expression levels in intestinal veins were higher than those in intestinal arteries. In addition, in vitro and in vivo studies showed aging enhanced EFEMP1 mRNA expression.

Conclusions: EFEMP1 is a novel amyloid precursor protein, which causes an age-related venous amyloidosis.

EXPLORING THE IMPACT OF AORTIC MEDIAL AMYLOID ON AORTIC WALL INTEGRITY IN ANEURYSM AND DISSECTION

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Introduction: Aortic medial amyloid (AMA) is the most common localised amyloid, estimated to occur in 97% of Caucasians above the age of 50. The main constituent of AMA is a 50 amino acid polypeptide called medin and is thought to be derived from the proteolysis of milk fat globule-EGF factor 8 protein (MFGE8). The pathological impact of aortic medial amyloid is unknown, but it is believed that extracellular amyloid accumulation contributes to age-related diminished elasticity of the vessels underlying the pathogenesis of thoracic aortic aneurysm.

Objective: To explore the relationship of aortic medial amyloid with biochemical and micromechanical properties of the aortic wall in sub-types of aneurysm and dissection patients.

Methods: Human aortic tissues removed during aneurysm surgery from tricuspid (idiopathic degenerative aneurysm, DA) patients, bicuspid valve (BAV) patients and patients that had an acute type A dissection were subjected to oscillatory nanoindentation experiments to determine localised mechanical properties of the tissue. Collagen, elastin, and glycosaminoglycans concentrations were determined, along with relative levels of aortic medial amyloid-related factors (medin, milk fat globule-EGF factor 8, oligomers and fibrils). FFPE sections were stained with Van Gieson Elastin and the elastin content was quantified using an in-house segmentation script. Measurements were combined with clinical data and statistical analyses performed.

Results: The DA cohort can be divided based on their phenotype. One group shared similar characteristics with BAV patients, termed bicuspid like phenotype-tricuspid valve. The second group had high amyloid oligomer species present with a significantly lower elastic modulus (p=0.01), indicative of reduced elastic response of the tissue, termed amyloid-rich. The amyloid-rich patients had increased levels of elastin fragmentation compared to the other two aneurysm groups. Dissected tissue had higher levels of amyloid factors than aneurysm patients.

Conclusions: We identified a group of DA patients with high amyloid oligomers and altered micromechanical and structural properties of the vessel wall. We propose these findings as a cause for aneurysm formation in these patients. Tissue from dissected patients had even higher levels of amyloid factors potentially indicating a more advanced pathology. Amyloid is not found in BAV patients, suggesting a distinct mechanism for pathogenesis in BAV that does not involve amyloid formation.

Keywords: aortic medial amyloid, dissection, aneurysm
MONOCLONAL GAMMOPATHY OF CLINICAL SIGNIFICANCE: ABOUT A CASE OF NODULAR PULMONARY AMYLOIDOSIS

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Background: Amyloid proteins can infiltrate virtually all organs. Diagnosis is usually difficult owing to its diverse clinical presentation. Involvement of the lung is relatively common, but rarely symptomatic, and most commonly observed in localized (primary) forms of amyloidosis. We report here a case of nodular pulmonary amyloidosis that evolved over a 10-year period.

Case-report: A 61 year-old woman was referred for a suspicion of pulmonary amyloidosis. She had been diagnosed at the age of 30 with rheumatoid arthritis (RA) that required for years, hydroxychloroquine. In 2008, a chest x-ray described lung and pleural calcifications that were attributed to RA. A small IgG lambda monoclonal component was noted in 2013. The patient remained substantially asymptomatic until December 2014, when she developed tachycardia with paroxystic atrial fibrillation. Echocardiography was normal. The association of cardiopulmonary symptoms along with a clinical history of autoimmune disease and the presence of a monoclonal gammopathy raised the suspicion of amyloidosis.

Progressively, she presented a worsening of dyspnea (NYHA grade 2). The cardiac work-up failed to identify any abnormality pointing out cardiac amyloidosis with cardiac biomarkers remaining in the normal range. Respiratory function test highlighted a mixed defect along with signs of impaired CO diffusion. The light chain lambda M-component was measured at 40.8 mg/l (N <26.3 mg/l), with a normal kappa/lambda ratio. Multiple myeloma was ruled out. FDG-18 pet-scan confirmed the presence of multiple sub-pleural and parenchymatous hypermetabolic nodules. Biopsies of a sub-pleural nodule and peri-umbilical fat aspiration were not contributive. Transthoracic CT-guided biopsy identified foci of amorphous eosinophilic material, positive for TTF1 and cytokeratine-7 by immunostaining, consistent with the diagnosis of pulmonary nodular amyloidosis, AL lambda monotypie. So far, the patient did not receive any specific treatment, as suggested in the literature, and her medical condition remains stable. Standard bortezomib-based chemotherapy will be proposed in case of worsening of her respiratory situation.

Conclusions: Nodular pulmonary amyloidosis is an extremely rare condition that can be asymptomatic and misdiagnosed for years. It is usually diagnosed incidentally on chest x-rays. In most cases, it is localized, and association to systemic amyloidosis is uncommon. It is defined as one or more nodular amyloid deposits involving the lung. Management depends on the severity of symptoms. Parenchymal amyloid nodules generally grow slowly and remain asymptomatic with an excellent long-term prognosis. Treatment is usually not required. So far, there is no consensus of the opportunity of systemic chemotherapy in progressive pulmonary localized amyloidosis.

Keywords: amyloidosis, lung, gammopathy
AMYLOIDOSIS APPOINTMENT COMPANION: A NOVEL TOOL TO IMPROVE PATIENT CARE AND COMMUNICATION WITH HEALTH CARE PROVIDERS

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2 University of Utah, Salt Lake City, UT, USA.
3 Weill Cornell Medical Center, New York, NY, USA.
4 Northern California Support Groups, San Francisco, CA, USA.
5 Boston University School of Medicine, Boston, MA, USA.
6 Akcea Therapeutics, Boston, MA, USA.
7 Patient Discovery, Boston, MA, USA.
8 University of North Carolina, Chapel Hill, NC, USA.
9 Oregon Health and Sciences University, Portland, OR, USA.

Background: Amyloidosis is a severe, progressive and life threatening disease. Patients often feel overwhelmed by the complexities of symptoms, treatments, and care management. The Amyloidosis Appointment Companion (AAC) was created to help patients identify their goals of care, changes in their condition, and challenges they are facing and to share these with their health care providers (HCPs).

Objectives: This abstract reports findings and feasibility data from the first pilot study of the AAC with 25 patients across five amyloidosis centers in the US.

Methods: The Amyloidosis Research Consortium in collaboration with Patient Discovery, a healthcare technology platform, created the AAC as an online tool that guides patients through an interactive survey to capture quality of life (using the EQ-5D), concerns, and goals of care. Answers are compiled and prioritized in a one-page ‘Discussion Guide’ to be shared with their health care team prior to their appointment. Patients and HCPs will have access to longitudinal data as utilization of the tool progresses. A steering group of amyloidosis patients, HCPs and industry representatives was assembled to evaluate aggregated de-identified data, discuss opportunities to improve care and oversee data collection. A pilot feasibility study of the AAC was conducted in five amyloidosis centers in the United States.

Results: A total of 25 patients successfully used the AAC to create their Discussion Guide [68% AL, 12% hereditary ATTR, 8% wild-type ATTR, 12% untyped]. Selected discussion topics varied widely between types. Two main types of reported goals of care were identified: 1) short term, isolated goals (such as attending a specific life event) and 2) long term, lifestyle goals (such as regaining exercise tolerance or living independently). While responses to quality of life questions varied, a majority of patients reported moderate levels of anxiety or depression (64%, N=16). Of typed patients, 27% (N=6) reported being satisfied with their treatment while 32% (N=7) reported being neutral or dissatisfied.

Conclusions: Outcomes from this tool are useful to both patients and HCPs for improving individual patient care through identification of priorities, concerns, and misconceptions. Feedback from HCPs and the steering group highlight the particular importance of questions relating to patients’ goals of care as well as treatment satisfaction, which provide novel information that are crucial for managing individual patient expectations, treatment decisions, and appointment schedules. Furthermore, review of aggregated data with industry and physicians shows promise for improving patient care. Insights from this data provide powerful information that may help improve development of assistance programs, clinical trials, and research.
MODULATION OF ENDOTHELIAL CELL HYPOXIC INJURY BY AMYLOIDOGENIC MEDIN: IMPLICATIONS FOR AGING-RELATED CEREBROVASCULAR DISEASE

S. Truran 1, N. Karamanova 1, H. Davies 2, M. Hansen 1, V. Weissig 3, J. Madine 2, R. Migrino 1

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2 University of Liverpool.
3 Midwestern University.

Introduction: Medin is one of the most common human amyloid proteins that accumulate in the vasculature with aging. We recently showed that medin is found in elderly human cerebral arteries with greater amount in vascular dementia versus cognitively normal subjects. We also showed that medin caused endothelial cell dysfunction and immune activation, suggesting its potential role in vascular aging and cerebrovascular disease. Age is the most important risk factor for stroke, characterized by hypoxic/ischemic injury from cerebrovascular disease leading to neurodegeneration and neuroinflammation. The effect of medin on vascular function in setting of ischemic insult is not known.

Objective: We aim to compare the effects of hypoxia, medin and combined hypoxia and medin on human brain microvascular endothelial cell (EC) viability. We also aim to determine whether GM1 ganglioside-containing liposomes (GM1L), found in prior studies to protect against amyloid vasculopathy, could protect ECs against hypoxic and medin-induced injury.

Methods: Primary ECs were exposed for 20 hours to 4 treatments: 1) vehicle control, 2) medin 5µM, 3) medin 5µM+GM1L 300 µg/mL, or 4) GM1L 300 µg/mL under two ambient conditions: room air (RA) or hypoxia (chamber with 1% oxygen). Cell viability was assessed using calcein-AM fluorescence by flow cytometry. Gene expression of antioxidant enzymes heme-oxygenase 1 (HO1), NADPH quinone dehydrogenase (NQO1) and superoxide dismutase 1 (SOD1) were measured separately by qPCR in ECs on RA.

Results: see table.

<table>
<thead>
<tr>
<th></th>
<th>Room Air (RA)</th>
<th>Hypoxia (1% oxygen)</th>
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<tbody>
<tr>
<td></td>
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<td>Medin</td>
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<tr>
<td>Calcein-AM fluorescence relative to control (%) N=4</td>
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<td>59±2*</td>
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</tbody>
</table>

p<0.05 versus veh-RA (*), versus medin-RA (+), versus medin-hypoxia ($) 

Conclusions: Hypoxia and medin reduced EC viability and, combined, had additive adverse effects. Co-treatment with GM1L fully restored EC viability against these insults, likely through upregulation of antioxidant defenses. Because medin vascular accumulation increases with aging around the same period of enhanced risk of ischemic injury, medin may be a promising novel treatment target to reverse the pathophysiology of cerebrovascular disease and vascular dementia.

Keywords: medin, cerebrovascular disease, aging

Category: Other forms of amyloidosis
10 YEAR FOLLOW-UP IN A PATIENT WITH A CEREBRAL AL AMYLOIDOMA

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Introduction: Cerebral amyloidomas are rare tumor-like lesions characterized by dense deposits of amyloid, often lambda light chains, in the brain parenchyma without evidence of systemic amyloidosis. The mass effect of the amyloid deposits can create neurologic symptoms. In most of the reported cases, the amyloid deposits are found supratentorially in the white matter, most commonly in the frontal lobe. The extracellular amyloid deposits may also often lead to cognitive decline.

Objective: To provide 10 year clinical follow-up in a patient with a cerebral AL amyloidoma.

Methods: A 65 year old Caucasian female presented with confusion, cognitive decline and right homonymous hemianopsia in September 2009. MRI demonstrated 2 enhancing lesions in her brain. The larger mass was in the left occipitotemporal region and the smaller lesion in the left frontal region. A stereotactic needle biopsy demonstrated a cerebral amyloidoma that on fibril isolation and amino acid sequencing analysis proved to be from immunoglobulin lambda light chain. She underwent a negative work-up for systemic light chain amyloidosis.

Results: Due to anticipated stability of the mass the patient has undergone observation with serial MRI imaging and clinical follow-up. After 10 years of follow-up the cerebral amyloidomas, they have increased slightly in size and are associated with more surrounding edema. The patient has a right hemianopsia, seizure disorder, and exhibits progressive disinhibited behavior and cognitive decline.

Conclusion: Cerebral amyloidomas are rare. The amyloid deposits can produce neurologic symptoms by mass effect by the tumors and cognitive decline due to extracellular amyloid deposition. In general cerebral amyloidomas are associated with a stable clinical course.


Key Words: Cerebral, Amyloidoma, AL
BONE MARROW SPECIMEN POSITIVE FOR AMYLOID NOT ALWAYS AL – CAN BE AA AND ATTR

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Introduction: Bone marrow biopsy is the method of choice for follow up/diagnosis of patients with most hematologic disorders including plasma cell dyscrasia/multiple myeloma, patients with abnormal serum/urine paraproteins and lymphomas. Meanwhile, detection of amyloid in the bone marrow can be the first evidence of concurrent amyloidosis. Majority of the bone marrow reports merely describe the presence or absence of amyloid and do not specify their spatial distribution.

Objectives: We sought to determine whether location of amyloid deposits in the bone marrow specimens may have clinical relevance. We reviewed Congo red stained bone marrow specimens and subcategorized the location of the amyloid deposits and its relation to type of amyloidosis and associated conditions.

Methods: 809 consecutive bone marrow biopsies were reviewed and 85 cases had amyloid deposits by Congo red stain (10.5%). However, slides were not available for review in 19 cases and hence further studies were limited to 66 cases. The biopsies were subcategorized according to the location and type of amyloid. Clinical/pathological correlation was performed.

Results: There were 57/66 cases (86%) of AL amyloidosis, 2 cases of AA (3.0%), 2 cases (3.0%) of ATTR amyloidosis and 5 patients had insufficient data, including one highly suspicious for wild type ATTR (aka senile amyloidosis). The M:F was 1.3:1 and the mean was 65 y/o. The % of the plasma cells in the marrow ranged 1-80% with the mean of 13.46%, median 8%. In 44/66 [66.6%] specimens plasma cells were <10%, eight of which were post-therapy bone BM specimens. Twenty two of the 66 cases showed ≥10% plasma cells, among which 17 cases met the WHO criteria for plasma cell myeloma. Plasma cells were polyclonal in 8 cases. In this study, 4 cases of AL amyloidosis were associated with near normal counts (<5% plasma cells) and polyclonal plasma cells prior to receiving treatment. The 2 cases with AA amyloidosis had 6% and 11% polyclonal plasma cells and the 2 cases with ATTR amyloidosis had 1% and 5% polyclonal plasma cells, respectively.

The presence of marrow stromal amyloid deposits was associated only with AL- amyloidosis (deposits limited to stroma or in association with non-stromal involvement) while no marrow stromal involvement was seen in the non-AL amyloidoses. The latter were associated with amyloid deposits in the vessel wall or periosteal soft tissue.

In 54/66 (81%) of patient biopsy proven amyloid deposits were detected in at least 1 other organ and 16 patients had abdominal fat biopsy positive for amyloid.

Conclusions: Although AL amyloid is the most common type of amyloid in the bone marrow specimen, the pathologist should have high index of suspicion for non-AL amyloidosis especially if the deposits are limited to the vessel wall or periosteal soft tissue. Whether bone marrow stromal deposits are AL type specific, requires further study for confirmation.
AUTOLOGOUS STEM CELL TRANSPLANTATION FOR THE TREATMENT OF MONOCLONAL GAMMOPATHY OF RENAL SIGNIFICANCE (MGRS): EXPERIENCE FROM A SINGLE INSTITUTION

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Introduction: Monoclonal gammopathy of renal significance (MGRS) is a recently described entity that defines a spectrum of renal diseases resulting from the nephrotoxic effects of paraproteinemia from non-malignant clonal B cells or plasma cells, and by definition does not meet criteria for multiple myeloma, chronic lymphocytic leukemia, or malignant lymphoma. MGRS can result from deposition of monoclonal proteins, dysregulation of complements, and activation of humoral factors.

Objectives and methods: We describe here, two cases of MGRS that have undergone an autologous stem cell transplant as part of the management of the disease.

Results: Case 1. A 50-year-old woman with hypertension and asthma presented to the hospital with history of fatigue and lower limbs edema. Work-up investigations revealed the presence of 2.33 g/day of proteinuria, free kappa of 205 mg/L, lambda 20.09 mg/L and ratio of 10.21. Hemoglobin was 93 g/L, albumin 22 g/L and creatinine of 132 umol/L. A kidney biopsy was performed and the report was consistent with Heavy Chain Deposition Disease (HCDD). Further, bone marrow aspirate showed the presence of a plasma cell clone of 4-5% of the marrow cellularity with kappa light chain restriction. Patient had received 3 cycles of Cyclophosphamide, Bortezomib and Dexamethasone (CyBorD) with no response and ultimately progressed with creatinine of 425 umol/L, free kappa of 457.2 mg/L and 9.95 g/day of proteinuria. Patient proceeded with stem cell collection followed by high-dose melphalan and bortezomib conditioning and stem cell transplantation. At day-100, response assessment was consistent with MRD negative complete response (CR), and decrease of proteinuria to 1.67 g/day (Renal Response).

Case 2. A 54 year-old man with history of cranial sixth palsy, presented with proteinuria and IgG kappa and kappa light chains (LC) MGUS (Free kappa 55.5 mg/L, ratio 10.04 and IgG kappa of 9.2 g/L). Bone marrow and fat pad aspirate were negative for MM or AL amyloidosis at diagnosis. Two years from the initial presentation, patient went to progress with proteinuria of >1g/day and increase of free kappa. Kidney biopsy was performed and findings were consistent with crystalloid podocytopathy. Bone marrow was repeated and was consistent with 10% of kappa-restricted PC’s. PET/CT and MRI of the whole spine did not show evidence of active MM. Patient received 6 cycles of CyBorD with PR only and due to progressive proteinuria and PN proceeded with auto-SCT with high-dose melphalan conditioning. At day-100, response assessment was consistent with MRD positive CR with secondary MGUS and with a decrease of the proteinuria to 0.27g/day.

IN CONCLUSION, auto-SCT is a feasible and efficacious alternative for the treatment of selected patients with MGRS. Further data is needed to assess the impact of auto-SCT in the treatment of the evolving MGRS.

Key Words: MGRS, CyBorD, auto-SCT
RESULTS OF AUTOLOGOUS STEM CELL TRANSPLANTATION IN PATIENTS WITH MONOCLONAL IMMUNOGLOBULIN DEPOSITION DISEASE

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Introduction: Conventional chemotherapy, previously used in patients with multiple myeloma (MM), has shown suboptimal results in the setting of light chain deposition disease (LCDD). In recent years, treatment of younger patients with a bortezomib-based regimen, followed by high dose intravenous melphalan and autologous stem cell transplantation (HDM/ASCT) has been increasingly used. Also, if complete response is achieved and patients are on dialysis, kidney transplantation should be considered.

Objectives: To update and extend our experience in the treatment approach with bortezomib-based regimes in patients with newly diagnosed LCDD and candidates to HDM/ASCT.

Methods: We reviewed the medical records of all patients with a biopsy proved diagnosis of LCDD who had undergone HDM/ASCT between January 2007 and June 2019. Clinical and laboratory test results were analyzed to study the characteristics at diagnosis, during treatment, and at relapse, including their outcomes and subsequent lines of treatment received overtime.

Results: Fourteen patients were diagnosed with LCDD through the biopsy of an affected organ (12 kidney, 2 heart). Median age of the series was 54 years old (9 men and 5 women). The organ more frequently involved was kidney (85.7%). Median values of involved FLC was 4695mg/L (range 37200-299) and median value of bone marrow plasma cell infiltration was 36.4% (range 60-5). All of them were initially treated with bortezomib based regimes (10 VD, 2 CyborD, 2 VTD) followed by HDM/ASCT, with no major complications. At least a partial response (PR) was obtained after induction therapy in 11 patients (78.5 %), including complete response (CR) in 3 patients (21.4 %). All hematologic responses improved after the HDM/ASCT (10 CR, 1 VGPR, 1PR). In six cases, given the sustained complete hematologic response but persistence of renal impairment requiring dialysis, kidney transplantation was successfully performed.

Conclusion: Our results confirm and expand on the use of bortezomib based regimes followed by ASCT as a safe and well tolerated treatment for patients with LCDD. In consequence, induction treatment with bortezomib, followed by ASCT, should be considered the treatment of choice in younger patients with LCDD. Additionally, renal transplant should be taken into consideration in those patients still requiring dialysis and who have achieved a sustained hematologic CR.
HEREDITARY GELSOLIN AMYLOIDOSIS ASSOCIATED WITH A NOVEL IN-FRAME DELETION IN EXON 13 OF THE GSN GENE.

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Introduction: Gelsolin (AGel) amyloidosis, also known as familial amyloidosis Finnish type, is an extremely rare form of hereditary amyloidosis. To date, four pathogenic variants in the GSN gene have been identified. By far the commonest are the two substitutions p.D214N/Y associated with cranial neuropathy, corneal lattice dystrophy, distal sensorimotor neuropathy and skin changes. Two other rare variants: p.G194R and p.N211K cause nephrotic syndrome.

Objectives: To describe novel GSN mutation detected in a patient presenting with corneal lattice dystrophy and peripheral and autonomic neuropathy.

Methods: Evaluation of patients included clinical review, 123I-SAP scintigraphy, Tc-DPD scintigraphy, echocardiography, DNA analysis (using classical Sanger sequencing method and the next generation sequencing), histology and mass spectrometry.

Results: A 51 yr old woman was referred to our centre after the ophthalmology investigations showed corneal lattice dystrophy. She also complained of weakness, numbness and pins and needles affecting her legs, and bowel disturbance suggestive of autonomic neuropathy. There was clinical evidence of cranial neuropathy with facial weakness. SAP scintigraphy showed no evidence of visceral amyloid deposits, echocardiography and Tc-DPD scintigraphy showed no evidence of cardiac amyloidosis. Her rectal and colonic biopsies were Congo-red positive. Genetic testing using Sanger method was used to screen TTR, GSN (exon 4 only) and APOA1 genes were all WT. Next generation sequencing (NGS) of all genes known to cause hereditary amyloidosis (now routinely used in our Centre), revealed a novel variant in exon 13 of the GSN gene, resulting in the deletion of tyrosine residue at position 603 (p.Tyr603Del). Proteomic analysis of the amyloid in her bowel biopsy showed AGel amyloid.

Conclusions: The four previously reported amyloidogenic GSN variants result from single nucleotide substitutions and are all located in exon 4. This is the first report of a GSN deletion outside exon 4 to cause hereditary gelsolin amyloidosis, characterised by a classical disease phenotype including corneal lattice dystrophy and cranial neuropathy. This report highlights the success of combining widened genetic analysis using NGS technology with proteomic analysis of amyloid to determine the final diagnosis in rare cases of amyloidosis.

Key Words: Gelsolin amyloidosis, corneal lattice dystrophy, peripheral neuropathy.
AMYLOIDOSIS IMPACT ON VARIOUS ASPECTS OF A PATIENT'S LIFE: A TRANSVERSAL STUDY FROM THE FRENCH PATIENTS ASSOCIATION


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Introduction: Despite the significant clinical complications of amyloidosis, few studies report information about the patient’s quality of life. The principal aim of this study was to gain insight on the patient’s experience before and after diagnosis of amyloidosis and on changes in patients’ daily life with amyloidosis.

Methods: For 190 days in 2019, in the 5 French expert centers for amyloidosis, every patient who consulted for any type of amyloidosis was offered to answer a unique anonymous survey, some patients treated in other centers and members of the association also responded. Quality of life was measured with EVA score of EQ5-D.

Results: The survey was completed by 603 patients, median age was 67 years; 65% were male. Amyloidosis subtypes were as follow: 36% hereditary transthyretin (TTR), 28% AL, 18% wild type TTR, 4% inflammatory amyloidosis (AA), and 4% other hereditary forms, 10% not known by the patient. There were no social disparities between amyloidosis subtypes except for patients with AA who have more financial difficulties (28%). The first main symptoms reported were breathlessness (49%, 79% for WT TTR and 18% for FAP), tingling (33%), pain (27%), walking difficulties (27%). Patients had been seen by a median of 4 physicians before diagnosis with a median of 15 months since first symptoms with some difference between subtypes, from 10 months in AA to 23 months for FAP; 10.4 months in WT TTR and 13.5 months in AL. Correct diagnosis was most often made by hospital practitioners (84%), 32% of whom were in an expert center. The subtype of amyloidosis barely influences patients’ questions about their disease. Questions mainly concern research progress, amyloidosis outcome and underlying causes. Patients with hereditary forms tend to wonder about the transmission to their descendants (64%) more than those affected by another form (24% for AL). Internet was the main source of additional information mentioned by patients (65%), despite the fact that 85% were satisfied with the announcement and the explanations given, and 71% felt involved in therapeutic choices. Patients were usually satisfied with their follow-up, 50% were seen at least twice a year by their doctor but 20% were asking for more frequent consultations. Only 10% of patients had received a therapeutic education which is particularly appreciated. Quality of life was affected in most patients as 66% of patients considered their illness as a handicap in their daily life. Median EVA score, a quality of life score in oncology, was quite low: 60 (±21). For the 30% of respondents working, 57% were on sick leave or disability for an average time of 162 days (30-244) by year.

Conclusion: Most results were similar regardless of amyloidosis type. Diagnosis and therapeutic delays in amyloidosis cause avoidable complications and worsen deterioration in the patient quality of life. Therapeutic education seems an interesting approach to overcome difficulties in patients’ comprehension.
LEUKOCYTE CELL-DERIVED CHEMOTAXIN 2 PROTEIN STABILITY AND AGGREGATION IN VITRO

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Background: ALECT2 amyloidosis is a rare but increasingly recognized form of systemic amyloidosis caused by the deposition of the leukocyte cell-derived chemotaxin 2 (LECT2) protein. Amyloid deposits occur in patients' kidneys and liver. There are no disease-specific treatments. Two alleles of LECT2 are common in the global population, encoding valine or isoleucine at position 40 of the secreted protein. Many patients investigated to date are homozygous for the valine variant, but heterozygous individuals with amyloid deposition have also been identified. However, valine homozygosity is common in the general population and so the penetrance of this variant for ALECT2 amyloidosis is low. Amyloidosis is often associated with decreased precursor protein stability. We therefore hypothesized that the V40 variant of the protein is less stable than the I40 variant and that this decreased stability contributes to the risk of amyloidosis in combination with other factors.

Objectives: We asked whether the V40 and I40 variants of recombinant LECT2 have different biochemical properties in vitro.

Methods: We expressed both variants of LECT2 in E. coli without the signal sequence or affinity tags and purified them using ion exchange and size exclusion chromatography. We characterized the proteins and measured their stability using fluorescence, CD and NMR spectroscopy. We used thioflavin T fluorescence to measure the aggregation rates of both LECT2 variants.

Results: Both variants of recombinant LECT2 folded to structures with native-like NMR spectra. Denaturant titrations and limited proteolysis indicate that the variants have similar stability. Removal of the structural Zn2+ ion by EDTA chelation disrupted the tertiary structure and decreased the stability of both variants. Both variants readily aggregated under non-reducing conditions at pH 7.4 and this aggregation was accelerated in the presence of EDTA.

Conclusions: The experiments conducted to date do not show a difference in stability between the V40 and I40 variants of LECT2. The Zn2+ ion stabilizes the protein against unfolding and aggregation and removal of the Zn2+ disrupts the folded structure of the protein. Therefore, zinc homeostasis may play a role in pathogenesis. Aggregation of LECT2 in vitro occurs more rapidly in destabilizing conditions, consistent with a model wherein amyloid formation is driven by self-association of non-native, partially unfolded proteins. Therefore, strategies to stabilize the native state of LECT2 may reduce amyloid deposition and benefit patients.
AMYLOID ARTHROPATHY AS INITIAL MANIFESTATION OF MULTIPLE MYELOMA-ASSOCIATED AL AMYLOIDOSIS

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Background: Amyloidosis is an extracellular tissue deposition of fibrils composed of variety of proteins. Synovium is an uncommon site of extramedullary involvement in AL amyloidosis. So far, only isolated biopsy proven cases have been reported. We report a case where synovial amyloidosis was the initial presentation of multiple myeloma-associated AL amyloidosis.

Case presentation: A 73-year old male was referred for a swelling of the left knee associated with pain and stiffness increasing over the past 12 months. MRI showed a large tumoral mass of 64x40 mm, with a diffuse synovial thickening of the knee and several osteolytic lesions, raising the suspicion of amyloidosis. PET scan identified an increased uptake in both knees, hips, shoulders and the left ankle. A CT-guided biopsy recognized characteristic amyloid deposits of lambda light chains. Further investigations demonstrated an elevated IgG lambda (850 mg/dL) and lambda free light chain M-protein (149 mg/l) with an abnormal kappa/lambda ratio and a 36% abnormal plasma cell bone marrow infiltration, confirming the diagnosis of SD stage IA, ISS 2 multiple myeloma. Organ involvement concerned the kidneys (albuminuria, 640 mg/24h) and the heart (NTproBNP, 1920 pg/mL; troponin T, 72 ng/L), in addition to a bilateral carpal tunnel syndrome. A combination of bortezomib-cyclophosphamide-dexamethasone was initiated. Within the first cycle of therapy, we noted a significant improvement in arthritis-related symptoms, in parallel with a significant decrease in the lambda light chain level (partial hematological response).

Conclusion: Amyloid arthropathy as initial manifestation of multiple myeloma is an uncommon presentation of the disease. Amyloid infiltration of synovial tissues may occasionally cause synovial thickening, subcutaneous nodules and erosive bone lesions, responsible for joint stiffness and arthralgia, mimicking rheumatoid arthritis. Therefore, the complete differential diagnosis of chronic polyarthritis should include systemic AL amyloidosis.

Keywords: Amyloid arthropathy, synovial amyloidosis, AL amyloidosis.
AMYLOIDOSIS DERIVED FROM EPIDERMAL GROWTH FACTOR-CONTAINING FIBULIN-LIKE EXTRACELLULAR MATRIX PROTEIN 1 (FIBULIN-3) IS A PROPOSED NOVEL AMYLOID TYPE

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Introduction: Amyloidosis is characterized by extracellular deposits of misfolded protein fibrils aligned in cross beta-sheets. At present, 36 human amyloid proteins have been recognized. Amyloid derived from epidermal growth factor-containing fibulin-like extracellular matrix protein 1 (EFEMP1), also known as fibulin-3 (FBLN3), has recently been proposed as a novel amyloid type (Tasaki M, et al. Journal of Pathology 2018 DOI: 10.1002/path.5203).

Objectives: To characterize the clinical, histologic, immunophenotypic, and proteomic features of EFEMP1-associated amyloidosis.

Methods: We reviewed the protein identification reports generated on all tissue specimens from our liquid chromatography-tandem mass spectrometry (LC-MS/MS) amyloid typing database from 2010 to 2019 (20,612 specimens) in which there were ≥10 spectral counts for EFEMP1. EFEMP1 amyloid cases were defined as having elevated spectral counts for EFEMP1 as well as for proteins commonly deposited with amyloids of all types but without proteins indicative of a separate canonical amyloid type. Clinical data were available on all cases. Histologic examination of hematoxylin and eosin (H&E)- and Congo red (CR)-stained slides was performed on 25 cases. Immunohistochemistry (IHC) for EFEMP1 (clone sc-33722, Santa Cruz Biotechnology, Santa Cruz, CA) was performed on 12 cases. The CR and EFEMP1 stains were evaluated for intensity of staining. As controls, we also evaluated H&E, CR and EFEMP1 stains from 12 colon specimens that were involved by AL (n=10) and ATTR (n=2) amyloidosis per previous LC-MS/MS analysis.

Results: We identified 33 specimens involved by EFEMP1 amyloid from 32 patients. There were 3 males and 29 females with a mean age at diagnosis of 75 years (range 49-92 years). Involved sites included colon (n=26), small bowel (n=3), peritoneum (n=3) and stomach (n=1). One patient had colon and small bowel involvement. The gastrointestinal amyloid primarily involved the submucosa and the walls of submucosal veins, with less frequent deposition in deep mucosa, muscularis mucosa and muscularis propria. Nearly all amyloid deposits were very weakly CR-positive, with subtle birefringence under polarized light. EFEMP1 IHC showed high sensitivity for the amyloid deposits but also showed nonspecific weak staining predominantly in the lamina propria and muscularis mucosa in a subset of the control cases.

Conclusions: EFEMP1-associated amyloidosis, which has recently been proposed as a novel amyloid type, appears preferentially to affect the lower gastrointestinal tract of elderly females. The amyloid deposits tend to show very weak Congo red positivity and are highlighted with high sensitivity but lower specificity by EFEMP1 IHC. LC-MS/MS is the gold standard for detection of EFEMP1-associated amyloidosis. Due to its subtle morphologic features and weak Congo red positivity, this form of amyloid may be under-recognized.
Introduction: The genetic variant of β₂-microglobulin (D76N β₂-m) is associated with a familial form of systemic amyloidosis (Valleix et al, N Engl J Med 2012;366:2276-83). The lack of suitable animal model for β₂-m associated diseases has prompted us to focus our attention on the nematode C. elegans which has been successfully used for other protein aggregation diseases. Previous transgenic C. elegans strains were generated to express wild type β₂-m and other amyloidogenic isoforms demonstrating that a correlation exists between the amyloidogenicity of each variant and the worms biological abnormalities (Diomede et al, PLoS One. 2012; 7:e52314). We have now generated and characterized transgenic C. elegans worms expressing D76N β₂-m in the extracellular space using a temperature-dependent transcriptional system to control the transgene expression.

Objectives: Establishment of an in vivo model where the key biophysical/biochemical steps of D76N β₂-m amyloidogenesis can be replicated, dissected and correlated with pathological features in patients. Development of a rapid, highly informative and automatized system to study the pathological phenotype and screen putative therapeutic molecules at the early stages of drug development.

Methods: Stable chromosomally-integrated lines of transgenic C. elegans were generated to express D76N β₂-m under the temperature inducible control of the body-wall muscle-specific myo-3 promoter. Aggregation of D76N β₂-m in the nematodes was assessed by classical biochemical methods such as size exclusion chromatography and western blot. Characterization of the pathological phenotype was carried out by using the INVertebrate Automated Phenotyping Platform (INVAPP) and the Paragon (Partridge et al, Int J Parasitol Drugs Drug Resist 2018, 8: 8-21) designed for high-throughput, plate-based chemical screening. This system was also used to test the efficacy of fibrillogenesis interactors.

Results: The pathological phenotype in D76N β₂-m expressing strain, analyzed by INVAPP/Paragon system, was strictly correlated to the expression and accumulation of both monomeric and soluble oligomeric species of the protein. The defective phenotype was substantially rescued by targeting the D76N β₂-m gene with RNAi. The INVAPP/Paragon system showed that doxycycline was able to rescue the pathological phenotype confirming the efficacy of the drug as inhibitor of β₂-m fibrillogenesis both in vitro and in vivo. The effect observed with doxycycline was associated with the reduction of the D76N β₂-m oligomeric species in the worms.

Conclusions: C. elegans is a suitable model for D76N β₂-m related amyloidosis. The automated INVAPP/Paragon platform, that has been invaluable to study the proteotoxicity of this variant, will be ideal for high-throughput drug screening as no treatment for D76N β₂-microglobulin related amyloidosis is yet available.
STUDIES ON ALPHA SYNUCLEIN IN BETA CELLS

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Introduction: It has been reported that patients with type 2 diabetes (T2D) have an increased risk to develop Parkinson’s disease (PD) with a more aggressive phenotype. Both T2D and PD are protein-misfolding diseases. In T2D, islet amyloid polypeptide (IAPP) forms amyloid deposited in islets of Langerhans; while in PD, alpha synuclein (aSyn) forms insoluble Lewy bodies and Lewy neurites in neurons.

Objectives: To analyze the subcellular localization of aSyn in pancreatic beta cells and to monitor possible interaction between aSyn and IAPP.

Methods: Quantitative PCR, Western blot, and immunofluorescence staining were used to identify the presence of aSyn in human pancreas and human beta cell line EndoCβH1. Proximity ligation assay (PLA) was used to study the co-localization of IAPP and aSyn. Fibril formation was monitored using the ThT assay. aSyn was down regulated with siRNA and glucose stimulated insulin secretion (GSIS) was monitored using alphalisa. Cell viability was studied by flow cytometry.

Results: aSyn was detected in beta cells, PP cells and a subgroup of alpha cells in human islets as well as in the EndoCβH1 cells. PLA positive signal for IAPP and aSyn was also detected in human pancreatic islets but not in amyloid-laden areas. Addition of preformed aSyn seeds to monomeric IAPP accelerated fibril formation in vitro while preformed IAPP fibrils did not induce fibril formation of aSyn. Knock down of aSyn in EndoC cells did not affect GSIS. Neither knockdown nor overexpression of aSyn in cells affected cell viability.

Conclusion: Despite the close proximity of aSyn and IAPP in islets and the detected capacity of preformed aSyn fibrils to seed IAPP, it is still an open question if this occurs in real life.
INHIBITION OF IAPP AMYLOID FORMATION BY BRICHOS IN CULTURED ISLETS

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**Background:** Formation of IAPP amyloid is one major reason for loss of beta cells in type 2 diabetes. IAPP amyloid deposits are almost constantly present in this disease but is found also in type 1 diabetes (1) and in transplanted human islets of Langerhans. Formation of islet amyloid is linked to beta cell stress. Bri2, also known as Integral Membrane Protein 2B (ITM2B) is a multi domain protein containing the BRICHOS region. BRICHOS is 100 residues long and has been ascribed a chaperone function. Bri2 is expressed in beta cells and IAPP and Bri2 co-deposit in islet amyloid. Bri2 reduction with si-RNA make beta cells more sensitive to lipo-toxicity while a subsequent over-expression of BRICHOS protects the cells (1).

**Objectives:** To investigate if transduction of human islets with the BRICHOS domain can prevent the formation of IAPP amyloid during culture in high glucose.

**Methods:** Adenovirus for expression of BRICHOS (Ad-Bri2 BRICHOS) was produced in HEK293 cells. Isolated human islets supplied from the Nordic Network for Human Islet Transplantation were digested with trypsin prior to transduction with Ad-Bri2 BRICHOS.

**Results:** We have found that islet amyloid is deposited in human islets cultured in 22 mM glucose for 18 days. Semi-digested human islets transduced with Ad-BRICHOS on day 1 reform into larger cell aggregates during culture. Sections from cultured islets were subjected to immunolabeling with an ITM2B antibody which confirmed an increased expression of BRICHOS. Amyloid load determined after Congo red staining was found to be higher in non-transduced islets.

**Conclusions:** This result supports that BRICHOS can retain IAPP in monomeric form and prevent aggregation and amyloid formation.

AMYLOID IN PATIENTS WITH MYELOPROLIFERATIVE NEOPLASMS

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Introduction: Amyloidosis is a protein misfolding disease characterized by deposits of amyloid fibrils in various organs. Patients with myeloproliferative neoplasm (MPN) have rarely been reported to have amyloid deposits. We have observed MPN patients who presented with symptomatic amyloidosis and had incidental amyloid deposits identified on biopsies.

Objectives: This study was conducted to better understand the characteristics of these patients.

Methods: Patients were identified from our electronic database from 1990 to 2019. Demographic data were obtained from chart review. Amyloid was typed by immunohistochemistry and after 2009, by liquid chromatography mass spectrometry (LC-MS/MS).

Results: From 1990 to 2019, 12 patients with MPN were found to have amyloid deposits or amyloidosis. Median age was 78 years and 50% were male. The MPNs were chronic myelogenous leukemia (n = 2), polycythemia vera (PV, n = 3), primary myelofibrosis (n = 4), and unclassified myeloproliferative neoplasm (n = 3). JAK2 mutation was positive in 4, negative in 2, and not tested in 6. BCR/ABL was tested in 3 and was all negative. Heart failure was documented in 4 patients, 2 had cardiac amyloidosis. The amyloid was identified in the heart (17%), bone marrow (BM, 58%), and gut, leptomeninges, and larynx in 8% each. One patient had an autopsy in which amyloid deposits were found in multiple organs. Fat aspirate was performed in 9 patients and was positive in only 2. Amyloid was typed as immunoglobulin light chain (AL) in 3 (2 BM and 1 omentum), serum amyloid A (AA) in 1 BM by LC-MS/MS and transthyretin (assumed ATTR with negative immunohistochemistry stain for AL and AA) in 1 heart. A plasma cell disorder was found in 50% of the patients with 1 patient progressing to multiple myeloma. All 3 patients with verified AL also had a plasma cell disorder. The cause of the AA amyloidosis in the 1 patient is unknown. Only 1 patient with AL had documented heart failure. This patient had progressive kidney failure and 0.3 g/d of proteinuria but no kidney biopsy. Another patient with AL had extensive multiorgan amyloid involvement documented at autopsy but no clinical information was available prior to death. The amyloid in the rest of the patients did not undergo typing.

Conclusions: Amyloid deposits and amyloidosis can occur in patients with MPNs. This most commonly occurred as AL in the setting of a plasma cell clonal disorder. One patient was found to have AA amyloidosis without any identifiable infection or inflammatory condition. ATTR was found in another patient which is most likely due to wild type ATTR. Unfortunately, the type of amyloid deposit was not determined in the other 7 patients. The rate of amyloid deposits could be more common as many of these patients were asymptomatic and the amyloid was found incidentally or on autopsy.

Keywords: Amyloid, Myeloproliferative Neoplasm
Upper Respiratory Tract Amyloidosis: About 3 Consecutive Cases

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Background: Amyloidosis is an extracellular deposition of fibrils composed of a variety of proteins that occurs in virtually any organ, most frequently the heart and the kidneys. Involvement of the upper respiratory tract (URT) is usually a localized phenomenon rarely associated with a systemic disease. Here we report 3 cases of kappa laryngo-tracheal amyloidosis occurring in three different situations.

Cases presentation – Over the past 2 years, 3 patients with a positive history of smoking, were referred for long-lasting hoarseness. Complete work-up for systemic amyloidosis was negative in the 2 first of them. The first patient, a 55-year-old woman, presented a mucoid cyst of the right ventricular band that was surgically resected with no later recurrence. The second patient, a 73-year-old man, presented an infracentimetric nodule of the posterior wall of the larynx, with a significant infra-glottic extension for which no resection was proposed, in the absence of adverse evolution. In the third patient, a 74-year-old man, repeated investigations of the URT remained non conclusive till he developed swallowing problems related to macroglossia. He was finally referred for progressive sensitive polyneuropathy in the context of an IgM kappa MGUS, that raised the suspicion of systemic amyloidosis. Biopsies confirmed a MYD-88 Waldenström macroglobulinemia associated to a kappa AL amyloidosis with soft tissues and peripheral nerves involvement. He failed to respond to rituximab-cyclophosphamide-dexamethasone, achieved a partial hematological response after 6 cycles of rituximab-bendamustine, and finally obtained an organ improvement under rituximab-bortezomib-dexamethasone.

Conclusion: Larynx is the most common site for isolated amyloidosis to occur in the URT. Localized lesions can be removed by endoscopic surgery and require a regular follow-up for early diagnosis of recurrence. An initial complete work-up is however mandatory to rule out a systemic disease that will dictate a more specific management.

Keywords: Isolated upper respiratory tract amyloidosis, AL systemic amyloidosis, IgM Waldenström macroglobulinemia
LARYNGO-TRACHEOBRONCHIAL AL/AH PRIMARY AMYLOIDOSIS WITH AIRWAY COMPROMISE TREATED WITH AUTOLOGOUS STEM CELL TRANSPLANTATION

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**Background:** Amyloidosis of the lower respiratory tract is rare and its management in life-threatening presentations is not established.

**Clinical case:** A previously healthy 44-year-old woman consulted for a 2-year progressive dyspnea on exertion that had worsened abruptly in previous days with stridor. Physical examination was otherwise normal. A fibroscopy (FB) revealed submucosal millimetric nodules in rhinopharynx, larynx and trachea and a circular stenosis 2 cm below the glottis. A fibrobronchoscopy (FBC) revealed involvement down to subsegmental bronchi. Computed tomography imaging was not suggestive of parenchymal lung involvement and respiratory function tests were normal. Laryngo-tracheobronchial (LTB) biopsies were performed, patched tissue infiltration by positive Congo red material was observed that proved to be positive for gamma heavy and lambda light chain. The workup for systemic disease resulted negative. There was no evidence of measurable monoclonal component in serum and urine and the free light chain ratio was normal. Bone marrow plasma cells were normal. No abnormalities were observed in renal or cardiac function. The findings were consistent with the diagnosis of light/heavy chain amyloidosis (AL/AH) with exclusive involvement of the LTB airway. Laryngo-tracheobronchial (LTB) biopsies were performed, patched tissue infiltration by positive Congo red material was observed that proved to be positive for gamma heavy and lambda light chain. The workup for systemic disease resulted negative. There was no evidence of measurable monoclonal component in serum and urine and the free light chain ratio was normal. Bone marrow plasma cells were normal. No abnormalities were observed in renal or cardiac function. The findings were consistent with the diagnosis of light/heavy chain amyloidosis (AL/AH) with exclusive involvement of the LTB airway. Rigid FBC and photoresection with Nd:Yag laser of the subglottic stenosis was performed. Five days later, the patient required urgent attention again due to laryngeal stridor and severe obstructive respiratory failure. A rigid FBC was performed to resect pseudomembranes and dilate the tracheal stenosis, a residual stenosis of 20% remained in a control FBC with extensive nodules affecting most of the airway down to sub-bronquial level. Persistent symptomatology prompted the instauration of systemic therapy with high dose-melphalan (200mg/m2 in 2 days) and autologous stem cell transplantation (ASCT) five months after diagnosis. The patient did not experience complications during or after the procedure and presented clinical improvement, remaining symptom-free except for an isolated episode of stridor requiring local intervention 9 months after diagnosis. Nodules have not disappeared.

**Discussion:** LBT localized amyloidosis is a rare, slowly progressive entity, with few cases reported. Within exclusive respiratory tract clinical presentations, LBT involvement is the most frequent pattern and is associated with a better vital prognosis than interstitial or nodular forms. Standard treatments include laser debridement, dilation or prosthetic devices. Re-interventions are usually needed (a median of 2-5 times per patient). External beam radiation therapy as well as chemotherapy are therapeutic alternatives, although reports are scarce. Our case shows that high-dose melphalan is feasible and does not present specific risks. Long term efficacy remains a matter of concern, although the strategy may allow control of life-threatening episodes of obstruction.

**Key notes:** transplant, AL/AH, Laryngo-tracheobronchial
WORKING TOWARDS AN ALGORITHM TO AVOID UNNECESSARY CARDIAC BIOPSY IN AMYLOIDOSIS

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1 Mayo Clinic.

Introduction: By consensus, a diagnosis of ATTRwt can be made without a biopsy as long as there are negative monoclonal protein studies and a positive PYP. Monoclonal proteins can be found in as many as 25-30% of ATTR patients. Because fat aspirate is only positive in approximately 15% of patients with ATTR, more invasive biopsies may be required, including endomyocardial biopsy. FLC ratios can rise in renal insufficiency and a ratio of 0.37-3.1 has been proposed to account for the altered catabolism in renal failure. AL with a dFLC under 5 is a well described phenomenon, accounting for 15% of AL patients. ATTRwt and AL are different diseases that require different management, making it imperative that diagnostic algorithms are sensitive and specific.

Objectives: Using a cohort of amyloidosis patients, to develop a decision making algorithm to avoid unnecessary heart biopsies.

Methods: We retrieved patients with AL and ATTRwt seen from 1/2012 to 7/2018 within 90 days of their diagnosis. Patients were excluded if they did not have FLCs or serum immunofixation (SIFE) within 90 days of diagnosis. dFLC was defined as the absolute value of κ−λ for all patients. Patients were divided into 3 groups: 1, presumed ATTR defined as a normal SIFE and FLC ratio; 2, indeterminate, not meeting criteria for group 1 or 3; and group 3, presumed AL defined as an abnormal FLC ratio and a dFLC > 5 mg/dL.

Results: A total of 998 patients were included and their characteristics are shown in Table 1. Group 1, the presumed ATTR group, comprised 20% of the population, with only 24 ALs included. It is notable in this cohort that the AL patients were younger, more likely to be female, to have proteinuria, thinner IVS, and positive UIFE. At the other end of the spectrum, group 3, the presumed AL group comprised 60% of the population. Group 2 comprised 20% of the population. AL patients were more likely to be female, have a positive UIFE, thinner IVS, and they had lower NT-proBNP. Of the 21 ‘presumed AL’ pts who were diagnosed with ATTR, 5 had PYP scans that were positive. Biopsy clarified diagnoses in all of the ‘miscalculated’ patients.

Conclusion: This dataset sets the stage for development of an algorithm to interpret and manage abnormal monoclonal protein studies in the context of presumed ATTR. The final system will be presented at the meeting.

<table>
<thead>
<tr>
<th>Actual diagnosis</th>
<th>Group 1 ‘Presumed ATTR’</th>
<th>Group 2 ‘Uncertain’</th>
<th>Group 3 Presumed AL</th>
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<tr>
<td></td>
<td>AL</td>
<td>ATTR</td>
<td>AL</td>
</tr>
<tr>
<td>N</td>
<td>24</td>
<td>175</td>
<td>116</td>
</tr>
<tr>
<td>% study population</td>
<td>2</td>
<td>18</td>
<td>12</td>
</tr>
<tr>
<td>Female, n (%)</td>
<td>11 (46) *</td>
<td>8 (5)</td>
<td>38 (33) *</td>
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<tr>
<td>Age, years</td>
<td>67 *</td>
<td>76</td>
<td>66 *</td>
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<tr>
<td>Albumin, g/dL</td>
<td>2.5 *</td>
<td>3.5</td>
<td>2.8 *</td>
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<td>dFLC, mg/dL</td>
<td>2.7 *</td>
<td>0.47</td>
<td>2.01</td>
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<td>Abn FLC renal ratio, n (%)</td>
<td>7 (29) *</td>
<td>0</td>
<td>47 (40) *</td>
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<td>Urine g/24 h</td>
<td>2.874 *</td>
<td>0.142</td>
<td>1.362 *</td>
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<tr>
<td>UIFE pos, n</td>
<td>12 *</td>
<td>2</td>
<td>37 *</td>
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<td>Creatinine, mg/dL</td>
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<td>1.2</td>
<td>1.2</td>
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<tr>
<td>IVS, mm</td>
<td>14 *</td>
<td>17</td>
<td>12 *</td>
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<td>Troponin T, ng/ml</td>
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<td>NT-proBNP, pg/ml</td>
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<tr>
<td>Endomyocard</td>
<td>1</td>
<td>81</td>
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*p<0.001 for comparisons between AL and ATTR
PW138

WILD-TYPE TRANSTHYRETIN AMYLOID CARDIOMYOPATHY PREDICTS THROMBOEMBOLIC RISK IN ATRIAL FIBRILLATION

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Background: The impact of wild type transthyretin amyloid cardiomyopathy (ATTR-CA wt) on thromboembolic risk in patients with atrial fibrillation (AF) is unknown.

Objective: Our study was aimed to investigate the prevalence of AF as well the incidence of thromboembolism in the patient population with ATTR-CA wt.

Methods: We studied patients who underwent Tc-99m pyrophosphate scintigraphy (PYP) between 6/2015 and 6/2019. Those with positive PYP, negative serum for AL amyloidosis and no identifiable gene mutation were diagnosed with ATTR-CA wt. We compared the prevalence of AF in patients with and without ATTR-CA wt and the incidence of thromboembolism (stroke, TIA or systemic embolism) in AF patients with ATTR-CA wt (AF-ATTR) and without (AF-controls).

Results: Of 277 patients referred for PYP, 77 (28%) had ATTR-CA wt. The prevalence of AF was markedly higher in patients with ATTR-CA wt (n=68, 88%) compared to patients with negative PYP (n=77, 39%, p< 0.01). Compared to AF-controls, AF-ATTR patients had similar age (79±7 vs. 79±8 years, p=0.9), lower CHA2DS2-VASc (4.7±1.4 vs. 5.4±1.2, p=0.001) and lower left atrial indexed volume (LAVI) (46±35 ml/m2, p=0.003). However, the incidence of thromboembolic events was higher in AF-ATTR compared to AF-controls (37.3% vs. 19.5%, p=0.02). AF-ATTR was associated with increased risk of ischemic stroke, TIA or systemic embolism (OR 2.46, 95% CI 1.16-5.21, p=0.02) compared to AF-controls. On multivariable logistic regression analysis adjusting for CHA2DS2-VASc, oral anticoagulation use and LAVI, ATTR-CA wt was an independent predictor of thromboembolism (OR 6.5, 95% CI 2.33-18.08, p=0.001). The incidence of hemorrhagic stroke (p=0.3), intracranial hemorrhage (p=0.9), major bleeding (p=0.7), and all-cause death (p=0.4) did not differ between the 2 groups.

Conclusion: ATTR-CA wt cardiomyopathy is a strong predictor of thromboembolism in patients with AF, independent of CHA2DS2-VASc score or left atrial size.
FUNCTIONAL LIMITATIONS OF PATIENTS WITH TTR AMYLOIDOSIS

E. Redder 1, N. Bumma 1, R. Kahwash 1, A. Vallakati 1, C. Campbell 1, S. LoRusso 1, M. Freimer 1, S. Parikh 1, J. Prosek 1, S. Almaani 1, N. Sharma 1, Y. Bumma 1

1 The Ohio State University Medical Center and James Cancer Hospital.

Introduction/Background: The Comprehensive Amyloidosis Clinic (CAC) at The Ohio State University (OSU) is a multidisciplinary clinic that allows patients to see neurology, nephrology, cardiology, hematology, and physical therapy in one 3 hour visit. Ultimately, this saves the patient, insurance, and providers time and money. Amyloidosis affects different organ systems, Transthyretin amyloidosis (TTR) mainly affects the heart and nerves, causing physical and functional impairments. Currently, specific guidelines for the rehabilitation of amyloidosis patients have yet to be established. The Outpatient Oncology Rehabilitation department at OSU has structured specific guidelines to assess functional impairments at diagnosis and throughout treatment to improve quality of life, functional mobility and reduce the risk of decline. This study aims to assess functional limitations and rehab needs of patients with TTR amyloidosis.

Methods: Examination information was gathered by chart review, observation and patient report from the physical therapy evaluation completed during the clinic visit of hereditary and wild-type TTR patients. Physical therapy evaluation included subjective reporting from the patient of their functional limitations, home set up, goals for rehab, range of motion and manual muscle testing. Objective outcome assessments included the Timed Up and Go (TUG) and the 30 second sit to stand. These scores were compared to age and gender established normative values. The physical function section of the Short Form 36 was chosen to provide a qualitative patient reported value. Therapists address impairments by prescribing exercise, home modifications, adaptive equipment, compression garments or further physical therapy treatment.

Results: The outcome tool scores of TTR amyloidosis patients were examined and compared to the same age and gender established normative data. Patients diagnosed with hereditary and wild-type TTR assessed in the CAC scored below same age and gender normative data on the TUG and 30 second sit to stand test indicating the need for physical therapy intervention to improve mobility, quality of life and reduce the risk of further decline. See Table 1.

Conclusion: Patients with TTR amyloidosis present with physical impairments including reduced activity tolerance, strength and impaired balance as a result of disease side effects when compared to same age and gender normative data increasing their risk for falls and further decline. Physical therapy intervention is beneficial to improve physical functioning, quality of life and reduce the risk of further decline. Future development of the physical therapy assessment within the CAC will include specific cardiovascular and balance outcome tools.

Key words: TTR amyloidosis, physical therapy, functional mobility
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<tr>
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<th>Hereditary TTR</th>
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<tr>
<td>Patients (33 total)</td>
<td>9</td>
<td>24</td>
</tr>
<tr>
<td>Age (30-95 y.o.)</td>
<td>77.2 y.o. (65-95 y.o.)</td>
<td>62.3 y.o. (30-80 y.o.)</td>
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<td>14</td>
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<tr>
<td>Female</td>
<td>0</td>
<td>10</td>
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Organ involvement

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<tr>
<td>heart</td>
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<td>2</td>
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<tr>
<td>neuropathy</td>
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<td>6</td>
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<tr>
<td>≥2 systems involved</td>
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<td>14</td>
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<tr>
<td>other</td>
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<td>1</td>
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<tr>
<td>no symptoms</td>
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TUG average male (normative value)

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<tr>
<th></th>
<th>&lt;60 (NA)</th>
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<td>6.6 sec</td>
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TUG average female (normative value)

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30 sec sit to stand male (normative value)

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<th>70-74 (15)</th>
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<td>16</td>
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<td>7</td>
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30 sec sit to stand female (normative value)

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Note: (NA) – normative data not available or no subjects in this category
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Objectives:

- Review the mechanisms and patterns of cardiac deposition in amyloidosis
- Educate on the importance of red flags to recognize and diagnose ATTR cardiomyopathy
- Discuss the management of ATTR cardiomyopathy

Faculty:

- Pablo Garcia-Pavia, MD, PhD (Chair)
  Inherited Cardiac Diseases Unit
  Hospital Puerta de Hierro
  Madrid, Spain

- Yukio Ando, MD, PhD
  Kumamoto University
  Kumamoto, Japan

- Claudio Rapezzi, MD
  University of Ferrara
  Ferrara, Italy

Time Topic
17:00 - 17:10 Welcome and Introductions
  Pablo Garcia-Pavia, MD, PhD

17:10 - 17:30 Mechanisms and Patterns of Cardiac Deposition in Amyloidosis
  Yukio Ando, MD, PhD

17:30 - 17:50 Recognition and Diagnosis of ATTR Cardiomyopathy
  Claudio Rapezzi, MD

17:50 - 18:15 Management of ATTR Cardiomyopathy
  Pablo Garcia-Pavia, MD, PhD

18:15 - 18:30 Panel Discussion and Q&A
  Moderator: Pablo Garcia-Pavia, MD, PhD
  Panel: Yukio Ando, MD, PhD; Claudio Rapezzi, MD

This event is not Continuing Medical Education (CME) Accredited.
VYNDAQEL® is the first and only treatment that can provide both:

- A reduction in all-cause mortality and CV-related hospitalization in patients with wild-type or hereditary ATTR-CM¹
- A delay in neurologic impairment for adult patients with stage 1 ATTR-PN²

LEARN ABOUT THIS TREATMENT OPTION FOR STAGE 1 ATTR-PN AND ATTR-CM AT OUR VIRTUAL BOOTH
XVII International Symposium on Amyloidosis

Hereditary Transthyretin Amyloidosis

Thursday, 17 September, 2020
16:30 - 18:00 (CEST)

AGENDA

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<td>Potential predictors of progression and response to treatment of hereditary TTR amyloidosis</td>
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<td>Carlos Casasnovas</td>
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<td>Ole Suhr</td>
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AKC-277-001

Symposium organized and funded by Akcea Therapeutics.
Alnylam Symposium at the International Society of Amyloidosis (ISA)
XVII International Symposium on Amyloidosis

**ATTR Amyloidosis**

**UNLOCKING**

The Potential of RNAi Therapeutics

**Online Event | Friday, 18 September 2020 | 16:40–18:10 CET**

**PLEASE JOIN OUR EXPERT FACULTY PANEL**

to learn how organ damage from transthyretin-mediated (ATTR) amyloidosis can affect patients’ lives, discuss how RNA interference (RNAi) can be used to control ATTR amyloidosis, and evaluate the potential of RNAi as a treatment option for patients with hereditary ATTR amyloidosis

**Symposium Chair: Dr Mathew Maurer, MD**
New York-Presbyterian Hospital
Columbia University Medical Center
New York, USA

**Dr Laura Obici, MD**
Amyloidosis Research and Treatment Center
IRCCS Fondazione Policlinico San Matteo
Pavia, Italy

**Professor Julian Gillmore, MD, PhD**
National Amyloidosis Centre
University College London
London, UK

**Professor David Adams, MD, PhD**
APHP, French Reference Center for FAP
INSERM U1195, University Paris Saclay
Bicêtre Hospital
Paris, France

**AGENDA**

**Dr Mathew Maurer**
Welcome and introduction

**Prof Julian Gillmore**
Mechanisms of organ damage in ATTR amyloidosis

**Dr Laura Obici**
Controlling gene expression with RNAi in ATTR amyloidosis

**Prof David Adams**
Interfering with hereditary ATTR amyloidosis using RNAi

**Dr Mathew Maurer**
Closing and Q&A
WILL YOU ACCEPT THE CHALLENGE?

Tuesday 6 October - Friday 9 October 2020
Virtual event, “live” from the Karolinska Institute, Stockholm, Sweden
(2 hour evening session each day)

HEREDITARY ATTRamyloidosis:
exploring possibilities,
transforming outcomes

“To raise new questions, new possibilities, to regard old problems from a new angle requires creative imagination and marks real advance in science.”
— Albert Einstein

Join us for an interactive, educational event focused on translating the latest scientific innovations into improved outcomes for patients with hereditary ATTR amyloidosis.

This multidisciplinary forum brings together an international faculty to advance best practice in the management of this complex disease.

The meeting will be chaired by four international experts:

Prof. David Adams
France

Prof. Teresa Coelho
Portugal

Prof. Arnt Kristen
Germany

Prof. Ole Suhr
Sweden

CLICK HERE TO REGISTER YOUR INTEREST IN JOINING THE MEETING
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For healthcare professionals only. This meeting is organized and funded by Alnylam® Pharmaceuticals

Please note that some of the meeting sessions will include data specific to products from Alnylam®.

ATTR, transthyretin amyloidosis.

TTR02-CEMEA-00359 08.2020

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Turnhoutseweg 30
2340 Beerse, Belgium

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Date of Preparation: August 2020
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2340 Beerse, Belgium
EM-25964
Date of Preparation: August 2020
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• Improving access with ingenuity
• Healing hopelessness with heart
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TTR=transthyretin.