Proteomic Identification and Clinicopathologic Characterization of Splenic Amyloidosis

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Abstract: The spleen is a commonly encountered specimen in surgical pathology. However, little is known about the incidence, morphologic pattern, and clinical features of spleens involved by amyloidosis. We retrospectively identified 69 spleen amyloid cases typed using a proteomics-based method between 2008 and 2020. The frequency of amyloid types, clinicopathologic features, and distribution of amyloid deposits were assessed. Four amyloid types were detected: immunoglobulin light chain (AL) (N = 30; 43.5%); leukocyte chemotactic factor 2 amyloidosis (ALECT2) (N = 30; 43.5%); amyloid A (AA) (N = 8; 11.6%); and fibrinogen alpha (AFib) (N = 1; 1.4%). The splenic amyloid showed 5 distinct distribution patterns: (1) diffuse pattern, exhibited by most AL cases; (2) red pulp pattern, exhibited by most ALECT2 cases; (3) multinodular pattern, seen in subsets of AA and AL-kappa cases; (4) mass-forming pattern, seen in the AFib case; and (5) vascular only, seen in a subset of AA cases. Atraumatic splenic rupture was the most common reason for splenectomy in AL cases, while most ALECT2 spleens were removed incidentally during an unrelated abdominal surgery. Splenomegaly was significantly more common in AA spleens than in AL or ALECT2 spleens and was often the reason for splenectomy in this group. In conclusion, splenic amyloid may be underrecognized as it is often an incidental finding. Although, as expected, many of the spleens were involved by AL amyloidosis, ALECT2 emerged as another common spleen amyloid type. Although the spleen amyloid types exhibited characteristic distribution patterns, proteomics-based typing is warranted as some morphologic overlap still exists. Awareness of ALECT2 as a major spleen amyloid type is important for appropriate diagnostic workup and patient management.

Key Words: spleen, amyloidosis, ALECT2, proteomics, typing

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Amyloidosis refers to a group of clinical syndromes characterized by the extracellular deposition of misfolded proteins, leading to end organ dysfunction.[1–5] To date at least 36 proteins are recognized to have amyloidogenic potential.[6] In general, patients with symptoms clinically suspicious for amyloidosis undergo a biopsy of an involved site to demonstrate the presence of Congo red (CR)-positive amorphous deposits with characteristic green birefringence when viewed under cross-polarized light.[3] In other instances, detection of amyloidogenic deposits represents an incidental finding in specimens obtained from patients without an antecedent clinical suspicion for amyloidosis. In both situations, accurate amyloid typing is indicated because amyloid protein type determines both clinical manifestations and patient management

Since 2008, we have performed mass spectrometry-based proteomics to type the amyloid in clinical specimens submitted to our reference laboratory.[7] The 5 most commonly analyzed specimen types, in decreasing order of frequency, are heart, kidney, gastrointestinal tract, bone marrow, and fat aspirate/biopsy. Across all anatomic sites, the most prevalent amyloid types detected were AL (immunoglobulin light chain; 59%) and ATTR (transthyretin; 28%), which together comprise the vast majority (> 85%) of the cases. However, the relative frequency with which the types are detected differs across the anatomic sites. For example, while ATTR is more common than AL in the heart, it is extremely rare in breast and renal specimens.

The spleen is a commonly encountered specimen in surgical pathology. There are many possible reasons leading to the clinical decision to perform splenectomy, which can be divided into several broad categories: (1) therapeutic (eg, hypersplenism due to immune thrombocytopenic purpura [ITP], hemolytic anemia, involvement by known hematologic malignancies, trauma); (2) diagnostic (identifying a splenic mass visualized in imaging studies, determining the cause of unexplained splenomegaly, classifying hematolymphoid

neoplasms); (3) incidental to another procedure (eg, pancreatectomy/Whipple procedure).[8]

While splenic involvement by systemic amyloidosis leading to functional hyposplenism is well-recognized,[9] the frequency of splenic involvement by different amyloid types has not been previously characterized. Herein, we report our experience with 69 cases of mass spectrometry-typed spleen amyloid specimens. The goal of this study was to determine the frequency of the different amyloid types and their distribution in spleen specimens involved by amyloidosis, and to correlate the findings with clinicopathologic parameters.

METHODS

Study Design

The study was approved by the Mayo Clinic Institutional Review Board and conducted in accordance with the Declaration of Helsinki. The cohort consists of all spleen amyloid cases (N = 69; 66 resection cases and 3 biopsies) sent from extramural institutions to the Mayo Clinic Amyloid Proteomic Typing Laboratory between January 1, 2008, and December 31, 2020. Three cases were from patients seeking further care at our institution and requiring secondary review and diagnosis confirmation, while the rest were sent to our laboratory specifically for amyloid typing. All available clinical data extracted from the submitted pathology reports, clinic notes, and/or electronic medical records (for intramural patients) are summarized in Table 1.

Amyloid Typing

CR-positive amyloid deposits present in each case were typed using a previously published method.[7,10] In brief, 10-mM-thick formalin-fixed paraffin-embedded tissue sections were obtained from each case and mounted on a Director slide. Sections were stained with CR, and amyloid deposits were visualized under the fluorescent light of a laser microdissection apparatus. Amyloid material corresponding to a total area of 60,000 mm^2 was collected by dissection, and 2 independent dissections were performed for each case. Proteins present in the collected material were denatured via heat and sonication, and cysteine disulfide bridges were reduced using dithiothreitol. Following overnight trypsin digestion, the peptide material from each dissection was independently analyzed using a QExactive (Thermo-Fisher Scientific, Waltham, MA) mass spectrometer connected to a nanospray ionization liquid chromatography system. Tandem mass spectra (MS/MS) were acquired using the traditional data-dependent acquisition mode. Resulting MS/MS data were processed using a previously described bioinformatics pipeline to detect the peptides and proteins present in each patient's sample.[11–13] Proteins that were detected with an identification probability of at least 90% were considered to be present in the patient's amyloid deposit. A pathologist scrutinized the protein identification profile and determined the amyloid type by correlating the patient's clinical phenotype (whenever available) with the most abundant amyloidogenic protein that was consistently detected across all dissections.

Morphologic Assessment

Morphologic assessment was performed on the resected spleen specimens. The biopsies were excluded from morphologic assessment because the specimens were too small to accurately determine splenic architecture. Hematoxylin and eosin–stained and CR-stained histologic sections from 62 available splenectomy cases were retrieved from the Mayo Clinic Tissue Registry and evaluated for the predominant pattern of amyloid deposition with respect to the spleen compartments: red pulp (cords and sinuses), white pulp (follicles and periarteriolar lymphoid sheath [PALS]), and arteriolar vessels.

Statistics

Ages of patients and spleen weights with different amyloid types were treated as continuous variables, and they were compared using an analysis of variance test with post hoc contrasts. Each morphologic feature assessed was treated as a discrete variable, and its homogeneity was tested using a χ^2 test with Yates's correction. All statistics were carried out using BlueSky statistics software (version 7.40). Statistical tests with a *P* value < 0.05 were highlighted wherever appropriate.

RESULTS

Figure 1 shows protein identification profiles from examples of the spleen amyloid types identified in this study (AL-lambda, AL-kappa, leukocyte chemotactic factor 2 amyloidosis [ALECT2], AA, and AFib). The assay detects amyloid signature proteins (apolipoprotein E, serum amyloid P component, and apolipoprotein A-IV),[10] biochemically verifying the presence of amyloid in the analyzed proteomes. Type-determining amyloid precursor proteins are also detected, thereby enabling unbiased typing of the amyloid deposit.

Table 1 summarizes the patient demographics and frequency of amyloid subtypes detected in spleen specimens, while Table 2 summarizes the indications/clinical setting for splenectomy. While many of the cases (N = 30; 43.5%) were of AL-type, including AL-kappa (N = 6) and AL-lambda (N = 24), an equal number of ALECT2 cases was detected (N = 30; 43.5%). AA comprised most of the remaining cases (N = 8; 11.6%),

TABLE 1. Demographics and Frequency of Amyloid Types Detected in Spleen Specimens by Proteomic Analysis

Type	# of Cases, n (%)	Age (Mean ± SD)	Sex (F/M/U)
AL	30 (43.5)	63.8 ± 11.8	9/19/2
AL-kappa	6 (8.7)	54.3 ± 16.1	0/5/1
AL-lambda	24 (34.8)	63.8 ± 11.9	9/14/1
ALECT2	30 (43.5)	67.6 ± 8.3	20/10/0
AA	8 (11.6)	59.5 ± 12.4	4/4/0
AFib	1 (1.4)	79	1/0/0

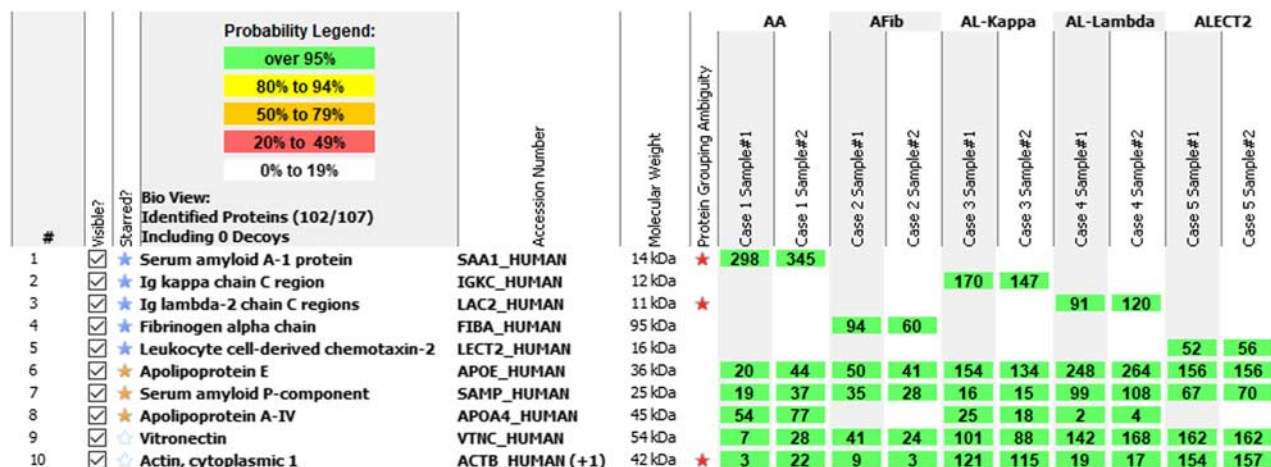F indicates female; M, male; U, unknown.

FIGURE 1. Proteome profiles of the 4 amyloid types identified in this study. Representative cases from formalin-fixed paraffin-embedded spleen specimens are presented on the x-axis, and evaluated proteins are presented on the y-axis. Kappa and lambda light chain amyloidoses are presented separately here as AL-kappa and AL-lambda, respectively. The blue stars highlight the amyloidogenic proteins, and the yellow stars highlight the amyloid signature proteins (apolipoprotein E, serum amyloid P component, and apolipoprotein A-IV).[14] Numbers in the green boxes indicate total number of tandem mass spectrometry spectra matching a protein in a sample, which is a surrogate measure of its abundance.[15]

while a single case of AFib with a mutation in the fibrinogen alpha chain gene (p.E545V variant) was identified. AL spleen cases showed male predominance (male:female = 2:1), while there was a female predominance in ALECT2 spleen cases (male:female = 1:2). There was no gender bias in AA cases (male:female = 1:1). There were no significant differences in the mean ages (in years) of AL (64.0, range: 27 to 90), ALECT2 (67.6, range: 50 to 84), and AA (59.5, range: 48 to 84) spleen cases. A concomitant or remote history of malignancy was reported in 16 cases, including 12 cases (all ALECT2) of nonhematologic solid tumors (pancreatic carcinoma/neuroendocrine tumor, N = 7; renal cell/urothelial carcinoma, N = 3; and colonic/appendiceal tumor, N = 2). Splenectomy was performed as part of a surgical procedure for these intra-abdominal malignancies in N = 11 cases. The remaining 4 cases included 2 cases (both AL-lambda) of low-grade B-cell lymphoma; 1 case (AL-lambda) of plasma cell myeloma, and 1 case (serum amyloid A [SAA]) of myelodysplastic syndrome. Complete clinical information was unavailable for the remaining 53 cases.

The indication for the splenectomy was known in 54 cases (excluding biopsies), which is summarized in Table 2. Of these, 43 patients had a splenectomy with an indication other than autopsy (AL, N = 16; ALECT2, N = 22; SAA, N = 4; AFib, N = 1). The indication for 75% of the AL patients was related to underlying splenic pathology (N = 12; 75%), including atraumatic splenic rupture (N = 10), splenomegaly (N = 1), and splenic cyst (N = 1). Splenectomy performed for other reasons, including unrelated abdominal surgery (N = 2) and trauma (N = 2), were less common. Conversely, the indication for splenectomy for ALECT2 patients was usually unrelated to underlying primary splenic pathology (N = 19; 86.3%) and included indications such as unrelated abdominal surgery (N = 12), trauma (N = 4), and ITP (N = 3). Splenectomy performed due to underlying splenic pathology was uncommon in ALECT2 patients and included atraumatic splenic rupture (N = 1) and splenic cyst/mass (N = 2). The indication for splenectomy for most SAA cases (N = 3; 75%) was splenomegaly and, in the remaining case, was an atraumatic splenic rupture. Overall, splenectomy was performed due to primary splenic pathology in less than half of the cases (N = 20; 46.5%). Finally, the spleen was resected in the single AFib case due to atraumatic splenic rupture.

Of the 11 spleens from autopsy cases, only 2 patients had an antecedent history related to their amyloid diagnosis. Both had splenic AL-lambda, and an antecedent lambda light chain restricted plasma cell neoplasm.

Gross examination findings were available in 44 spleen resection cases. The mean weight of the resected spleens was 413.1 g for AL, 280.5 g for ALECT2, and 2001.8 g for AA cases. Compared with AL and ALECT, splenomegaly was significantly more common in AA spleens ($P < 0.005$ and < 0.001, respectively), although not

TABLE 2. Indication for Splenectomy by Amyloid Type in Spleen Amyloid Specimens (N = 66)

Indication for Splenectomy	AL	ALECT2	SAA	AFib	Total
Incidental to another surgery	2	12	0	0	14
Atraumatic splenic rupture	10	1	1	1	13
Autopsy	9	1	1	0	11
Trauma	2	4	0	0	6
Splenomegaly	1	0	3	0	4
Splenic cyst/mass	1	2	0	0	3
ITP	0	3	0	0	3
Not provided	4	5	3	0	12

TABLE 3. Spleen Weight and Morphologic Analysis of Spleen Amyloid Types

| Type | Weight* (Mean ± SD) (g) | Predominant Pattern†, n (%) | | | White Pulp (Preserved/ Effaced)‡ | Reactive Follicles Present§, n (%) |
		Red Pulp	Diffuse	Multinodular		
AL (N = 27)	413.1 ± 267.6	0	24 (88.9)	3 (11.1)	0/27	1 (3.7)
ALECT2 (N = 27)	280.5 ± 450.9	25 (92.6)	2 (7.4)	0	25/2	20 (74.1)
AA (N = 8)	2001.8 ± 1807.1	1 (12.5)	1 (12.5)	4 (50.0)	2/6	2 (25.0)
AFib (N = 1)	194	0	0	0	1/0	0

*There was a statistically significant difference between spleen weights of AL, ALECT2, and AA types (analysis of variance $P < 0.001$).

†There was a statistically significant difference ($P < 0.00001$) between the frequency of observed predominant patterns (ie, red pulp, diffuse, and multinodular) across amyloid types as tested using a χ^2 test with Yates's correction.

‡The presence of preserved versus effaced white pulp architecture differed significantly ($P < 0.00001$) between the amyloid types.

§The presence/absence of reactive follicles also differed significantly ($P < 0.00001$) between the amyloid types ($P < 0.00001$).

all AA spleens were enlarged. Aside from unrelated parenchymal lesions such as hematoma, cysts, and subcapsular infarct; the cut sections of the spleen specimens were generally described as either being unremarkable or having firm and/or waxy consistency. A miliary pattern of white pulp nodularity or discrete masses were not observed.

The distribution of amyloid deposits in resected spleen specimens with available slides (N = 63) is summarized in Table 3. Based on morphologic review, the amyloid deposits showed one of the following 5 distinct patterns: (1) predominantly diffuse (Figs. 2A–C), where the amyloid extensively involved the red pulp and effaced the white pulp architecture, obliterating the follicles and/ or PALS; (2) predominantly red pulp, where the amyloid was largely confined to the red pulp (Figs. 2D, E) with cordal or perisinusoidal distribution, with relative preservation of the white pulp architecture; (3) multinodular (Fig. 2F), where the amyloid deposits formed multiple distinct macronodules, typically centered around the arterioles/PALS, throughout the spleen parenchyma; (4) mass-forming (Fig. 2G), in which the amyloid formed a dominant large mass focally distorting the spleen architecture; and (5) vascular only (Fig. 2H), where the amyloid deposits were found only within the walls of small and large arteriolar vessels without the involvement of other spleen compartments. Patterns 1 to 4 also showed variable degrees of arteriolar wall involvement. The morphologic pattern in most of the spleen amyloid cases was predominantly diffuse or predominantly red pulp.

The diffuse pattern was seen in 27 cases, including 24/27 (88.9%) AL (3/6 AL-kappa and 21/21 AL-lambda), 2/27 (7.4%) ALECT2, and 1/8 SAA (12.5%) cases. The red pulp pattern was observed in 25/27 (92.6%) ALECT2 and 1/8 (12.5%) AA cases but was not seen in any of the AL cases. The multinodular pattern was seen in 3/27 (11.1%) AL cases, all of which were AL-kappa (3/3, 100%); and 4/8 (50.0%) SAA cases. The vascular-only pattern was seen in only 2 cases, both of which were SAA. Finally, amyloid deposition with mass formation was seen in only the single AFib case. Most of the AL cases showed diffuse involvement, with effacement of the white pulp and absence of reactive follicles, while most

of the ALECT2 cases showed red pulp pattern with preservation of the white pulp and associated reactive follicles. The frequency of the common architectural patterns (diffuse, red pulp, and multinodular) differed significantly ($P < 0.00001$) across amyloid types. Similarly, there were statistically significant differences in preservation/effacement of white pulp architecture ($P < 0.00001$) and presence/absence of reactive follicles ($P < 0.00001$) among the amyloid types.

DISCUSSION

Amyloidosis represents a complex group of clinical syndromes caused by deposition of misfolded proteins with beta-pleated sheet conformation. Depending on the amyloid type, the tissue deposits may become progressive and widespread, leading to end organ damage. Splenic involvement by amyloidosis has been estimated to occur in ~5% to 10% of primary systemic (AL) amyloidosis patients.[16] However, functional hyposplenism (defined as the presence of Howell-Jolly bodies in the peripheral blood due to impaired splenic function) is more common, detected in ~24% of patients with primary systemic amyloidosis; suggesting that there may be some degree of splenic involvement in these patients.[9] It may not be possible to determine the incidence of spleen involvement in amyloidosis with certainty, as the spleen is rarely biopsied or resected for the purpose of diagnosing amyloidosis since there are other more easily accessible diagnostic sites such as bone marrow and fat pad aspirate. Furthermore, as seen in our cohort, the spleen is frequently normal in size or only mildly enlarged, especially in ALECT2 and AL cases, and therefore may not prompt clinical decision to perform a splenectomy. In our study, the splenectomy was usually performed incidentally rather than due to underlying primary splenic pathology (53.5%), and only 20% of the autopsy patients had an antecedent amyloid-related history. Furthermore, the frequency of splenectomy due to underlying amyloid-related splenic pathology varied widely depending on amyloid type: atraumatic splenic rupture was common in AL, splenomegaly was common in AA, while in most ALECT2 patients, the amyloid diagnosis was an incidental finding.
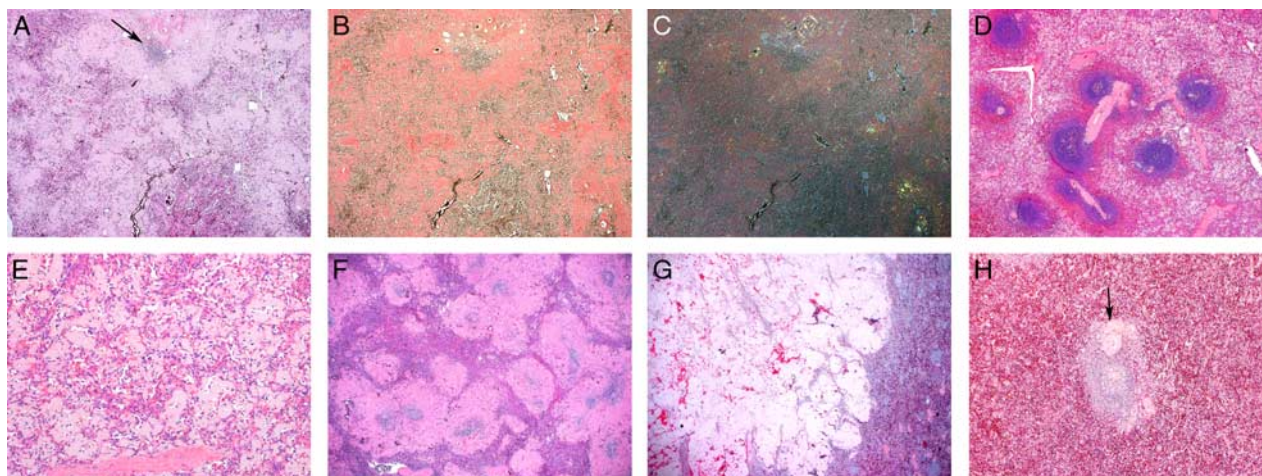
FIGURE 2. Amyloid distribution patterns in spleen amyloidosis specimens. A, Spleen specimen in a case of AL-lambda amyloidosis showing diffuse parenchymal replacement by eosinophilic amorphous deposits; only rare residual follicles are seen (arrow). The deposits are positive for CR (B) which show apple-green birefringence under polarized light (C). D, A spleen specimen involved by ALECT2 showing amyloid deposition confined to the red pulp. The white pulp follicles are normal in number and show reactive changes. On higher magnification (E), the amyloid deposits are seen surrounding the sinusoids and in the red pulp cords. F, Multinodular pattern in a spleen involved by AL-kappa. There are multiple distinct large nodules throughout the spleen parenchyma, which are typically centered around the arterioles/PALS. G, The single case of AFib spleen amyloid showed the formation of a dominant large mass by the amyloid distorting the spleen architecture. H, A spleen involved by SAA showing multiple small vessels with thickened walls due to amyloid deposition (arrow).

These findings suggest that splenic amyloid may be more common than is currently recognized, and the frequency of subclinical/asymptomatic splenic amyloidosis likely varies according to amyloid type.

AL amyloidosis is the most common amyloid type detected in patients with amyloidosis and accounts for 59% of cases in our laboratory practice.[7] It is usually caused by an underlying clonal plasma cell disorder but may also be associated with other monoclonal protein-secreting disorders such as lymphoplasmacytic lymphoma or MALT lymphoma.[17] AL amyloidosis has a propensity for multiorgan involvement such as heart, kidney, liver, and among many others; leading to the rapid functional deterioration of involved organs. Therefore, a diagnosis of AL-amyloid warrants prompt treatment aimed at elimination of the underlying plasma cell clone.

Due to its propensity for multiorgan involvement, AL-amyloid, as expected, comprised a major subset (43.5%) of the spleen amyloid cases. Approximately one-third of AL-amyloid spleen specimens were resected due to spontaneous rupture (ie, not associated with trauma). Atraumatic rupture of the spleen involved by amyloidosis is rare and has been reported in the literature mostly as single cases.[18–20] Most of the previously published cases were reported as AL-type (87%), while a small number of AA cases (13%) were also described; with the caveat that these were presumed types based on patient history and/or histochemical/immunohistochemical staining. While atraumatic splenic rupture in our study did indeed occur mostly in patients with AL-amyloid, it also occurred rarely in ALECT2, AA, and AFib amyloid types. The exact mechanism of splenic rupture is unclear. It has been

suggested that since the spleen is a relatively friable vascular organ, it is susceptible to rupture when provoked by adequate trauma or in disease states where the architecture is compromised by infections (eg, infectious mononucleosis) inflammatory disorders, metabolic disorders, or hematological diseases/neoplasms.[19] Our observations that most AL-amyloid cases (88.9%), particularly AL-lambda (100%), showed the diffuse pattern of amyloid of deposition with extensive architectural effacement of splenic compartments, and were more frequently associated with splenic rupture compared with other types, appeared to support this hypothesis.

While ATTR is the second most commonly detected amyloid type in our laboratory practice (28%), it was not detected in any of the spleen specimens. However, ALECT2, which accounts for only 3% of cases in our laboratory practice overall, was detected as often as AL in the spleen. First reported in 2008, ALECT2 amyloidosis has emerged as an important type of amyloidosis with a predilection for renal and liver involvement, where it is the second most commonly detected amyloid type (after AL) at these sites.[7,21] Leukocyte chemotactic factor 2 (LECT2) is synthesized by the liver and secreted into the circulation, and its physiological function appears to be involved in cell cycle, immunomodulation, and bone growth.[22] ALECT2 amyloidosis usually presents in older adults, with a median age in the 60s at diagnosis, and has a strong ethnic bias affecting mainly Hispanics, Native Americans, Egyptians, and Punjabis.[23–26] The pathogenesis of ALECT2 amyloidosis remains to be elucidated, although it has been suggested that factors leading to increased LECT2 expression or defective LECT2 metabolism may

result in increased tissue LECT2 concentration and initiation of amyloid fibril formation. All ALECT2 cases were found to be homozygous for valine to isoleucine polymorphism resulting from single-nucleotide polymorphism in codon 58, position 40 of the protein, although this polymorphism is also common in the general population, suggesting this polymorphism by itself is not sufficient for the pathogenesis.[24,25,27]

Clinically, ALECT2 manifests primarily as proteinuria and/or progressive renal insufficiency and is now recognized as an important cause of end-stage renal disease in afflicted patients.[24] Hepatic ALECT2 amyloidosis is also common, accounting for 25% of hepatic amyloidosis cases.[27,28] The diagnosis is often incidental during evaluation for unrelated conditions such as biopsy of a liver cyst, elevated liver enzymes, chronic viral hepatitis, and steatohepatitis.[27] Interestingly, patients with hepatic ALECT2 usually have no evidence of renal disease at the time of diagnosis. The abnormal liver function tests can usually be explained by underlying nonamyloid pathologic findings. Therefore, the significance of isolated hepatic ALECT2 amyloidosis is unclear. Due to the limited clinical data provided, the incidence of kidney and/or liver involvement in the spleen ALECT2 amyloidosis cases of our cohort is uncertain, although the single autopsy case did not show evidence of involvement at these sites.

Similar to hepatic ALECT2 amyloidosis, in the spleen ALECT2 amyloidosis was often diagnosed in specimens removed for unrelated reasons; these include organ resection (eg, pancreatomy, nephrectomy, colectomy) for tumor, trauma, laceration, and ITP. Also similar to hepatic ALECT2 amyloidosis, the clinical significance of splenic ALECT2 amyloidosis is unclear. Unlike AL and ATTR, cardiac involvement by ALECT2 amyloidosis is exceedingly rare.[29] Therefore, despite the lack of specific treatment, the prognosis of ALECT2 amyloidosis is generally excellent. In contrast to AL amyloidosis, atraumatic splenic rupture is a rare event in ALECT2; perhaps this could be attributed to the relatively intact architecture in involved spleens. The presence of reactive lymphoid hyperplasia in many of these cases suggests the abnormal protein in ALECT2 may by induced by or elicit an immune response. It is of interest to note that all 3 ITP cases in our cohort had concurrent ALECT2 amyloidosis.

AA represents the remaining major spleen amyloid type in our cohort, seen in 12% of the cases. AA, like ALECT2, accounts for ~3% of amyloid cases across all organ types in our laboratory practice.[7] Half of AA spleen cases showed amyloid deposits in a multinodular pattern, while the remaining cases showed perivascular, red pulp, and diffuse patterns. AA amyloidosis, also known as secondary amyloidosis, is a systemic syndrome triggered by conditions associated with long-standing inflammatory activation including chronic infections (eg, tuberculosis), autoimmune disorders (eg, rheumatoid arthritis), hereditary conditions (eg, familial Mediterranean fever), and various hematological and solid tumors.[30,31] This leads to tissue deposition of SAA protein, an acute phase reactant

with amyloidogenic potential. Similar to ALECT2, the kidney is the most common site of involvement, followed by the liver. Sporadic cases of splenic amyloidosis characterized as AA based on clinical and histochemical/immunohistochemical grounds have been described, again as single case reports in patients presenting with splenic rupture.[32–34] In contrast to AL and ALECT2, where the spleen is usually mildly to moderately enlarged, splenomegaly is significantly more prominent, and splenomegaly is the most common reason for resection in this small group. Splenic rupture was the indication for splenectomy in only 1 AA case. Treatment of AA amyloidosis is usually directed to the underlying disease to subdue the inflammatory process the triggered amyloid formation.[31]

Last, we detected a single case of fibrinogen alpha (AFib) in a patient with atraumatic splenic rupture. This was the only case where the amyloid formed a large focal mass in the spleen. Similar to ALECT2 and AA, the kidney is the most commonly involved organ in AFib (93%) followed by liver (3%).[7] AFib is the most common type of hereditary renal amyloidosis, which usually manifests later in life (mean age: 62 y) and is caused by an unstable mutant fibrogen alpha chain produced by the liver.[7,35,36] There is a high prevalence of atherosclerotic cardiovascular disease in AFib amyloidosis patients. Although uncommon, splenic involvement by AFib amyloidosis has been previously described.[37]

In conclusion, in our study of spleen amyloidosis, the largest to date, we have demonstrated that although AL is far more common than ALECT2 overall, ALECT2 is detected at the same frequency as AL in the spleen. AA is less common, and AFib is rare. However, the incidence of splenic amyloidosis and the frequency of different amyloid types cannot be determined with certainty as subclinical/asymptomatic splenic amyloidosis may be more common than is currently recognized, and the likelihood varies according to amyloid type. We also demonstrated that spleen amyloid types exhibit characteristic amyloid distribution patterns, although there are some degrees of overlap requiring confirmation of amyloid type. Preferably this would be done by a robust and objective proteomic-based method such as MS/MS, as typing by antibody-based methods such as immunohistochemistry or immunofluorescence may suffer from technical limitations,[38,39] while presumptive type determination (eg, AL) based on clinical history (eg, monoclonal gammopathy or plasma cell myeloma) may be even more unreliable as we have recently demonstrated in a large study of bone marrow amyloidosis.[40] In spleen specimens showing characteristic red pulp pattern of amyloid deposition, especially those associated with certain ethnic groups, immunohistochemical staining for LECT2 may also be attempted as the initial step to detect ALECT2 amyloid. Although ALECT2 amyloid in the spleen likely represents an incidental finding with undetermined significance, awareness of this major spleen amyloid type is nevertheless crucial to avoid unnecessary additional workup and/or treatment, and possibly prompt further investigation for renal involvement.

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