Chapter

5

Amyloidosis: Why We Need to Think About It Sooner?

SHAJI KUMAR

Amyloidosis constitutes a spectrum of diseases, unified by the morphological appearance of the amyloid protein deposits, the cardinal feature of this disorder. The term "amyloid" was first coined in 1838 to describe plant starch and was adopted by Rudolph Virchow in 1854 to describe amyloid deposits in tissues, because of its similarity to cellulose stained with iodine. Amyloidosis results from the extracellular deposition of insoluble fibrillar amyloid protein composed of low molecular weight subunits and is a generic term that includes all forms of systemic amyloidosis and can arise from over 25 different human proteins including immunoglobulin light chains, amyloid A protein, transthyretin, apolipoprotein, lysozyme, or fibrinogen (Table 1). When stained with Congo red amyloid deposits have a green birefringence on polarized microscopy and when stained with thioflavine T, they have intense yellow-green fluorescence[1]. Electron microscopic examination of amyloid deposits demonstrates straight and unbranching fibrils 8 to 10 nm in width, laid down in a β-pleated sheet fashion. Amyloid deposits are frequently associated with other non-fibrillar substances, including serum amyloid P-component, glycosaminoglycans (particularly heparan sulfate), and certain apolipoproteins. The mechanisms driving the conversion of the soluble precursor protein subunits into the insoluble amyloid deposits are not clear and are likely multifactorial[2].

Classification of Systemic Amyloidosis

Broadly the amyloid disorders can be classified into two groups, the hereditary types of amyloidosis and the acquired types of amyloidosis (Table 1). The clinical manifestations of the different types of amyloidosis have much in common and are primarily dictated by the degree and pattern of organ involvement. In the hereditary types of amyloidosis, the disorders result from missense mutations of the precursor proteins and are mostly dominantly inherited heterozygous disorders. In addition, hereditary disorders like Familial Mediterranean fever and TNF receptor associated periodic syndrome can also lead to secondary amyloidosis. In senile systemic amyloidosis (SSA), there is deposition of normal transthyretin in the heart and less commonly other tissues. Light chain amyloidosis (AL), or primary amyloidosis is the most prevalent type of amyloidosis in the developed countries, where as in the developing countries AA amyloidosis (secondary amyloidosis) related to chronic infection, is encountered more often.

Primary Systemic Amyloidosis (AL amyloidosis)

AL amyloidosis belongs to the spectrum of plasma cell disorders including monoclonal gammopathy of undetermined significance and multiple myeloma[3,4]. These patients present with a monoclonal plasma cell population in the bone marrow and presence of a monoclonal protein in the serum or urine and tissue deposition of amyloid derived from the monoclonal kappa or lambda light chains.

Clinical Presentation

The symptoms of this disorder are often nonspecific and vague including fatigue, edema, and weight resulting in considerable delay before a form diagnosis is made. In our experience, patients have symptoms for a median of 2 years before the correct diagnosis is established. The symptoms and physical findings in

Table 1: Types of amyloidosis based on precursor proteins

Clinical syndrome	*Precursor protein*
Hereditary Types	
Hereditary amyloid nephropathy	Apolipoprotein A-II Fibrinogen alpha chain Lysozyme Transthyretin
Hereditary neuropathic and/or cardiac amyloidosis	Transthyretin Apolipoprotein A-I
Senile systemic amyloidosis	Transthyretin
Hereditary amyloid neuropathy associated with corneal lattice dystrophy and cutis laxa (Meretoja syndrome)	Gelsolin
Hereditary and sporadic Alzheimer's disease; congophilic cerebral angiopathy	Amyloid precursor protein
Hereditary and sporadic spongiform encephalopathies	Prion protein
Hereditary dementias (British and Danish types)	BRI gene product
Hereditary cerebrovascular hemorrhage with amyloidosis (Icelandic type)	Cystatin C
Familial corneal dystrophies	TGFB1 Keratoepithelin
Acquired Types	
Primary and myeloma-associated amyloidoses (AL)	Kappa/Lambda light chain
Amyloidosis complicating chronic infections or inflammatory diseases	(Apo) serum AA
Dialysis related amyloidosis	Beta-2 microglobulin
Age-related amyloid occurring in the aortic intima	Apolipoprotein A-I
Amyloid complicating C-cell thyroid tumors	(Pro)Calcitonin
Islet cell amyloid (Insulinomas, type II diabetes mellitus)	Islet amyloid
Isolated atrial amyloidosis of aging	Atrial natriuretic peptide
Prolactinomas	Prolactin
Local amyloid complicating use of the insulin pump	Insulin
Cutaneous amyloid	Keratin

these patients primarily depend on the spectrum of organ involvement (Table 2)[3]. Although virtually all patients have multiorgan amyloid deposition at diagnosis, individuals may present with evidence of only one organ being affected. It is only rarely that a patient is diagnosed prior to onset of symptoms, and usually happens in the context of work of a monoclonal gammopathy or detection of amyloid deposit in tissue biopsies performed for other purposes. Given the non-specific features at presentation, amyloidosis should be considered in the differential diagnosis of any patient with nephrotic-range proteinuria, unexplained cardiomyopathy, peripheral neuropathy, unexplained hepatosplenomegaly, nonspecific gastrointestinal symptoms, unexplained weight loss and fatigue[5].

Screening and Diagnosis

Once amyloidosis is considered in the differential diagnosis, the initial step is the demonstration of a monoclonal protein in the serum or the urine and a monoclonal plasma cell population in the bone marrow. Monoclonal plasma cells are almost always detected in the bone marrow if sufficiently sensitive techniques are employed[6]. Screening serum protein electrophoresis and immunofixation may be normal in as many as 25% of patients, since they may only have a light chain (Bence Jones proteinemia), which can be picked up by urine immunofixation. Immunofixation when performed on serum and the urine will pick up the abnormal protein in over 90% of the patients. In the majority of the remaining patients, a free light chain assay will be able to detect presence of the abnormal free light chain[7]. Radioscintigraphy with radiolabeled amyloid P component is a noninvasive way to detect amyloid deposits, but this technique is not widely available[8]. The next step is the histological demonstration of amyloid deposition and the initial sites to be screened are the bone marrow biopsy (positive in 50 to 60%) and a subcutaneous fat aspirate (70 to 80%). Additional non-visceral sites can be biopsied for detection of amyloid and include minor salivary glands, gingiva, rectum, and skin and if nondiagnostic biopsies can be directed at the

Table 2: Typical presentations of amyloidosis

Renal	Asymptomatic proteinuria Clinically apparent nephrotic syndrome Renal failure with little or no proteinuria
Cardiac	Systolic or diastolic dysfunction Congestive heart failure Syncope due to arrhythmia or heart block Angina or infarction due to accumulation of amyloid in the coronary arteries
Gastrointestinal	Hepatomegaly and or splenomegaly GI bleeding (due to vascular fragility) Gastroparesis and early satiety Constipation Chronic diarrhea (Bacterial overgrowth, Malabsorption) Intestinal pseudo-obstruction (Dysmotility)
Neurological	Mixed sensory and motor peripheral neuropathy Autonomic neuropathy Compression of peripheral nerves (Carpal Tunnel syndrome) Bowel or bladder dysfunction Orthostatic hypotension (due to autonomic nervous system damage) Central nervous system involvement is unusual
Musculoskeletal	Muscle enlargement (pseudohypertrophy) Macroglossia Arthropathy (amyloid deposition in joints and surrounding structures). Scapulohumeral periarthritis Spondyloarthropathy Bone disease
Hematologic	Bleeding diathesis (due to factor X deficiency, advanced liver disease, amyloid infiltration of blood vessels)
Pulmonary	Tracheobronchial infiltration (hoarseness, stridor, airway obstruction, and dysphagia) Persistent pleural effusions (amyloid-induced cardiomyopathy, pleural infiltration with amyloid deposits) Parenchymal nodules (amyloidomas) Pulmonary hypertension
Dermal	Waxy thickening Easy bruising (ecchymoses) Purpura Subcutaneous nodules or plaques

involved organs. Once amyloid is identified by Congo red stain, the type of amyloid should be confirmed by immunohistochemistry and staining for K and λ immunoglobulin light chains, amyloid A, and transthyretin will detect most forms of amyloid. Given the prevalence of monoclonal gammopathies in the general population, subtyping of the amyloid is critical. In one study, 10% of patients thought to have AL amyloid had amyloid of other types, including 5% with fibrinogen amyloid and 4% with unrecognized mutations of transthyretin[9]. When unclear, more sophisticated analysis of the tissue using mass spectrometry to identify the involved protein as well as screening of the serum for common transthyretin mutation to rule out some forms of hereditary amyloidosis can be useful.

Prognosis

The primary determinant of outcome in patients with AL is the extent of organ involvement, most importantly cardiac involvement with the most common cause of death being progressive heart failure or sudden death due to arrhythmias[10]. Presence of overt heart failure is associated with a median survival of only 6 months and is the most important predictor of survival. Echocardiography with doppler evaluation allows measurement of ejection fraction and interventricular septal thickness, detection of diastolic dysfunction as well as presence of abnormal myocardial strain pattern. Newer modalities that are being investigated include magnetic resonance imaging including use of MR elastography. Cardiac biomarkers provide another sensitive estimate of cardiac involvement and include serum troponin T, brain

natriuretic peptide (BNP) and N-terminal fragment of pro-brain natriuretic peptide (NT-pro-BNP)[11,12]. Troponin has been shown to be a powerful predictor of survival in AL allows stratification of patients into groups with markedly different survivals. Levels of troponin T and NT-pro-BNP have been shown to predict survival after the diagnosis of amyloid and have been used to develop a staging system[13]. Presence of a normal NT-pro-BNP virtually excludes cardiac amyloidosis. Involvement of other organ systems can be detected by presence of other laboratory abnormalities such as elevated alkaline phosphatase with hepatic involvement and proteinuria with renal involvement (Table 2).

Nearly 10% of the patients presenting with an amyloid syndrome will have bone marrow plasmacytosis consistent with diagnosis of myeloma, but few have a clinical presentation characteristic of myeloma and have a shorter survival. In a series of 1596 patients with primary amyloidosis seen at the Mayo Clinic between 1960 and 1994, only six (0.4%) showed delayed progression (at 10 to 81 months) to overt myeloma; likely a reflection of short median survival in these patients[14]. Other prognostic factors that have been identified in AL include high β2 microglobulin, presence of circulating plasma cells, and a high marrow plasma cell labeling index.

Occasionally, patients present with amyloid deposits localized to a single site, often dermal or submucosal. Localized amyloid can affect different organs including the ureter, bladder, urethra, prostate, tracheobronchial tree, and pulmonary parenchyma as a solitary nodule. Soft tissue deposits of amyloid may or may not be associated with systemic disease and this need to be ruled out in all these patients. Patients with localized amyloid and no evidence of systemic plasma cell dyscrasia have an excellent prognosis.

Therapy

Therapeutic approaches to AL have been aimed at eliminating the plasma cell clone and limiting the availability of light chains for amyloid formation. While ideal, therapies aimed at dissolution of the amyloid deposits are still in experimental stages. Given this approach, it is not surprising that most of the current treatments for AL are similar to that used for management of multiple myeloma[15].

Melphalan/Prednisone: Melphalan with or without prednisone has been the standard therapy for AL for several decades, in those not undergoing stem cell transplantation[16,17]. In randomized trials, the combination has been shown to have a response rate of nearly 20% with a median survival of around 18 months. Survival was best for those patients with at least a 50 percent reduction in the paraprotein and all long term survivors had a complete response to therapy[18]. However, long-term therapy with melphalan / prednisone regimen increases the risk of myelodysplasia and acute myelogenous leukemia.

Melphalan/Dexamethasone: Dexamethasone alone or in combination has been used for treatment of AL. High dose dexamethasone with interferon followed by interferon maintenance has been shown to improve amyloidosis-related organ dysfunction in nearly half of the patients[19]. The VAD combination, commonly used for myeloma, has been studied in AL with organ responses seen in nearly 40% of the patients. VAD has also been used as initial therapy prior to stem cell transplant. Dexamethasone alone in a pulsed dose fashion results in hematologic complete remissions in a quarter of patients with improvement in amyloid-related organ dysfunction in 45%. Patients with cardiomyopathy had a low response rate, as did patients with peripheral neuropathy.

The median time to a hematologic response was 103 days. Palladini et al combined melphalan with dexamethasone with a hematologic response of 67% with 33% complete responses and 48% organ responses[20]. This regimen is probably considered the current standard therapy in patients not undergoing stem cell transplant.

Thalidomide and Lenalidomide: Thalidomide has been studied in small studies and patients with AL appear to tolerate the drug poorly[21]. In combination with dexamethasone, complete and overall responses of 19 and 48 percent has been noted in patients with disease that was refractory to, or had relapsed after, first-line therapy, complete and overall responses were noted in, respectively. A combination of cyclophosphamide, thalidomide and dexamethasone was recently studied in patient with advanced AL with hematological response rate of 74% and median estimated overall survival of 41 months from start of therapy. Lenalidomide alone or with dexamethasone has been studied in a small phase II study and results in hematological and organ function improvement in a small number of patients, especially in combination with dexamethasone. .

Stem Cell transplantation: High dose therapy and stem cell rescue is the standard of care for patients with myeloma and is being used increasingly to treat patients with AL[22-24]. This modality is limited by the fact that majority of the patients will not be eligible for this approach and the high mortality rate. The presence of significant organ dysfunction puts these patients at high

risk for complications, including sudden cardiac death and profound GI tract bleeding[25]. Complete hematologic responses (CR) are seen in nearly 40% of the patients with majority of these patients being alive beyond 5 years. Improvement in the function of at least one organ system in two thirds of these patients, compared to a third of those not achieving a CR. Dose reductions in melphalan have resulted in decrease response rates. We have generally limited SCT to patients less than 70 years with symptomatic amyloid, adequate performance status, less than three organs involved, ejection fraction >55 percent, creatinine < 2.0 mg / dL, and direct bilirubin < 2.0 mg / dL. Comparison of transplanted patients to historical controls suggest better organ response rates as well as improved overall survival with transplant[26]. However, the strict patient selection criteria used for transplant also results in a group of patients, who have better prognosis[27].

However, preliminary results of a French randomized trial suggest that overall survival may not be superior with HCT compared to melphalan plus high-dose dexamethasone[28]. In a prospective randomized trial comparing SCT (melphalan 140 or 200 mg / m2) to oral melphalan and dexamethasone, no statistically significant difference between the 2 arms was found for hematologic responses. The Kaplan-Meier estimated median survival was 56.9 months for the M-Dex arm and 48.6 months for the ASCT arm. The follow up remains short for this study and continuing follow up of these patients as well as other ongoing studies will help us answer this question.

Secondary Amyloidosis (AA Amyloidosis)

Systemic AA amyloid results from the deposition in tissue of serum amyloid A (SAA) protein, which is a major acute phase reactant, and is usually associated with inflammatory disorders and chronic infections. It can affect multiple organs, most importantly the kidneys and the heart, which usually contributes to death in these patients. The median survival is 4 to 5 years for patients with secondary amyloidosis, with those having cardiac involvement at diagnosis having a particularly poor prognosis. An elevated level of SAA has been associated with poor outcome in these patients. The most important aspect of management of these patients remains adequate control of the underlying inflammatory condition and this can lead to stabilization or improvement in the organ function and regression of amyloid deposits have been observed in some situations using SAP scintigraphy.

Colchicine remains the standard therapy for patients with established secondary amyloidosis, and has been particularly effective in AA associated with Familial Mediterranean fever (FMF). Colchicine at a dose of 1 to 1.5 mg / day markedly reduces the frequency of acute episodes of FMF, decreases the incidence of renal disease, and can stabilize the renal function in patients with mild proteinuria. Colchicine needs to be continued indefinitely in patients unless the underlying condition can be corrected. Newer agents such as Fibrillex®, an oral glycosaminoglycan mimetic is currently undergoing randomized, phase III trial for the treatment of AA amyloid and early results appear promising.

SUMMARY

In conclusion, amyloidosis is a systemic disorder, which is not very well recognized because of the relative rarity as well as the general lack of awareness of the condition. The diagnosis should be entertained in any patient presenting with unexplained cardiomyopathy or renal dysfunction as well as in other situations with unexplained multiorgan dysfunction. An early diagnosis will greatly improve the outcome of patients with primary as well as secondary amyloidosis. Once diagnosed, several treatment options are available currently for these patients and participation in clinical trials evaluating new treatment modalities should be considered for all these patients.

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