Utility of Doppler Myocardial Imaging, Cardiac Biomarkers, and Clonal Immunoglobulin Genes to Assess Left Ventricular Performance and Stratify Risk Following Peripheral Blood Stem Cell Transplantation in Patients with Systemic Light Chain Amyloidosis (AL)

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Background: Cardiac dysfunction is a well-recognized complication of light chain amyloidosis (AL). Autologous stem cell transplant (auto-SCT) has emerged as a successful treatment modality for AL patients. In this study, we examined the effect of clonal immunoglobulin light chain genes (VL), which encodes the immunoglobulin light chain protein that ultimately forms amyloid, on cardiac function, in the context of auto-SCT and its impact on overall survival.

Methods: Longitudinal Doppler myocardial imaging parameters along with cardiac biomarkers were used to assess for cardiac function pre and post auto-SCT.

Results: VL gene analysis revealed that Vl genes, in particular VlVI, were associated with worse cardiac function parameters than Vk genes. Clonal VL genes appeared to have an impact on left ventricular (LV) function post-transplant and also influenced mortality, with specific VL gene families associated with lower survival. Another key predictor of mortality in this report was change in tricuspid regurgitant flow velocity following auto-SCT. Correlations were also observed between systolic strain rate, systolic strain and VL genes associated with amyloid formation.

Conclusions: Clonal VL gene usage influences global cardiac function in AL, with patients having VlVI and VIII-III-associated amyloid more severely affected than those having Vk or VII amyloid. Pulsed wave tissue Doppler imaging along with immunoglobulin gene analysis offers novel insights into prediction of mortality and cardiac dysfunction in AL after auto-SCT. (J Am Soc Echocardiogr 2011;24:444-54.)

Keywords: Doppler myocardial imaging, AL amyloidosis, Immunoglobulin genes, LV function, Cardiac biomarkers

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Immunoglobulin light chain amyloid (AL) amyloidosis is the most common systemic amyloidosis, with an age-adjusted incidence of 5 to 13 per million patients per year in the United States. The traditional low-dose oral melphalan treatment regimen is generally associated with a poor prognosis (median survival, 13 months).[1] The recent introduction of autologous peripheral blood stem cell transplantation (auto-SCT) has dramatically improved median survival in a select group of patients with AL amyloidosis.[1,2] Both hematologic response at day 100 and cardiac function are key determinants of survival after auto-SCT.[1-4]

In earlier studies, we reported the high sensitivity of longitudinal Doppler myocardial imaging (DMI) myocardial velocity and strain and strain rate imaging in detecting left ventricular (LV) impairment in both early and advanced AL amyloidosis.[5,6] Because an association among particular clonal VL genes, organ tropism of amyloid deposition, and outcomes has been observed,[7-10] this study had a twofold aim: (1) to evaluate whether clonal immunoglobulin light chain variable gene (VL) gene usage influences changes in LV function

Abbreviations

AL = Immunoglobulin light chain amyloid

auto-SCT = Autologous peripheral blood stem cell transplantation

BNP = Brain natriuretic peptide

CI = Confidence interval

cTnT = Cardiac troponin T

DMI = Doppler myocardial imaging

FLC = Free light chain

HR = Hazard ratio

Ig = Immunoglobulin

LV = Left ventricular

sS = Systolic strain

sSR = Systolic strain rate

VL = Immunoglobulin light chain variable gene

after SCT, and its impact on long-term mortality, and (2) to test whether serial measurement of cardiac biomarkers and longitudinal DMI measures are useful for monitoring cardiac function and for risk stratification.

METHODS

This study was approved by the institutional review board of the Mayo Clinic (Rochester, MN). Fifty-three patients with AL systemic amyloidosis, consecutively selected from patients undergoing evaluation in the Division of Cardiovascular Diseases and the Division of Hematology at the Mayo Clinic from January 1 2004, to April 30, 2007 and referred to auto-SCT were enrolled in a prospective manner. Patients received either a standard-dose conditioning regimen (melphalan) at 200 mg/m^2 or a reduced-dose regimen (<200 mg/m^2), depending on age, renal function, the presence of cardiomyopathy, or triple-organ involvement. Of the 53 patients who underwent immunoglobulin (Ig) gene analysis, 28 received the standard dose and 25 received the lower dose (see Supplementary Table 1). Follow-up ended June 30, 2009. The diagnosis of AL amyloidosis was made by either subcutaneous fat biopsy or an involved organ biopsy study that demonstrated typical Congo red birefringence under polarized light. Endomyocardial biopsy was performed in four patients, for clinical purposes, and was positive for amyloid infiltration in all the cases. AL amyloidosis was further confirmed by the presence of a monoclonal protein in the serum or urine specimen and/or a monoclonal population of plasma cells in the bone marrow. Clinical, biomarker, and echocardiographic examinations were accomplished at two different time points (in a subset of 39 patients): the first assessment was performed <1 month before auto-SCT and the second assessment on day 100 after SCT.

Exclusion criteria were familial, secondary, or senile amyloidosis ($n = 2$); all other causes of cardiomyopathy (including other forms of restrictive cardiomyopathy, hypertrophic cardiomyopathy, or any cause of LV hypertrophy other than AL amyloidosis); diabetes mellitus ($n = 0$); a history of moderate or greater systemic or pulmonary hypertension ($n = 1$); more than mild valvular heart disease ($n = 0$); and coronary artery disease or previous myocardial infarction ($n = 0$). Atrial fibrillation ($n = 3$) was not an exclusion criterion.

Clinical Classification of Patients

Patients with AL amyloidosis were evaluated for the extent of amyloid-related organ involvement and for dominant organ involvement, integrating standard criteria described elsewhere[11] with information provided by the assessment of cardiac biomarkers and diastolic function. Patients were categorized according to clinical presentation as having renal, cardiac, hepatic, neurologic, or other dominant organ involvement. This clinical classification was performed independently blind to clonal VL gene usage. Patients with more than one organ involved were categorized according to the most prominent and symptomatic affected organ. Because several patients had both cardiac and renal dysfunction due to amyloid deposition, information on dominant or codominant involvement was collected for these patients. Dominant gastrointestinal, pulmonary, or soft tissue AL amyloidosis (excluding cardiac, renal, hepatic, or neurologic dominant involvement) were all classified in the category "other" to simplify analysis.

Dominant cardiac involvement was defined as having positive results on endomyocardial biopsy or abnormally elevated cardiac biomarkers, including brain natriuretic peptide (BNP), and cardiac troponin T (cTnT), on the basis of our previous observations.[3,12] Additionally, echocardiographic parameters, such as mean LV wall thickness (half the sum of the anteroseptal and posterior wall thickness in the parasternal long-axis view) >12 mm and diastolic dysfunction with a grade of 3 or 4 (restrictive pattern), were also used to define dominant cardiac involvement.[13,14] Dominant renal involvement was defined as having positive results on kidney biopsy and proteinuria >0.5 g/dL, dialysis dependence, or creatinine clearance <10 mL/min.

Classification of enrolled patients followed a three-step algorithm. First, patients were categorized on the basis of clonal VL germline gene usage, and thus two groups were generated: group Vκ, including patients using Vκ I family genes, and group Vλ, including patients using Vλ I, Vλ II, Vλ III, and Vλ VI families. Second, group Vλ patients were further subdivided on the basis of cardiac function by standard echocardiography (LV wall thickness, mass index, and ejection fraction) at baseline (before auto-SCT): group Vλ I-II-III and group Vλ VI. Third, patients in group Vλ I-II-III were subdivided on the basis of hematologic response[11]; all group Vλ I patients ($n = 7$) had either partial or complete response at day 100 and were therefore considered independently from those in group Vλ II-III, who had heterogeneous hematologic responses. In summary, four different groups were used in the analysis for comparison: groups Vκ, Vλ I, Vλ II-III, and Vλ VI.

Specimen Preparation and Cloning of Ig VL Genes

Bone marrow samples from all 53 patients with AL amyloidosis were processed to collect plasma cells and clonal Ig gene analysis was performed as per protocol (see the online Appendix for methodologic details).

Definition of Clonal Ig Gene Usage

The term "Ig gene usage" refers to the concept that antibodies or Ig molecules are generated in the immune system as a result of selection and molecular combining of three or four individual gene segments (three for Ig light chain and four for Ig heavy chain, light and heavy chain being essential for the formation of a complete antibody molecule). This process of selection and molecular recombination is a stochastic event whereby individual gene segments are apparently randomly "chosen" or "used" out of a larger array of such gene "families." In a normal immune response, the diversity of antibody molecules produced by a terminally differentiated B cell (plasma cell) is large (polyclonal response) in contrast to what is observed during a neoplastic process, such as in AL amyloidosis, in which a single plasma cell undergoes expansion to produce a population of plasma cells (monoclonal or clonal), all secreting exactly the same antibody (Ig), from which the light chain forms amyloid fibrils and undergoes tissue deposition.

Biomarkers

BNP, N-terminal pro-BNP, cTnT, creatinine, and glomerular filtration rate were measured and collected at the time of the