Clinical Outcome and Phenotypic Expression in LAMP2 Cardiomyopathy

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METABOLIC MYOCARDIAL storage diseases that mimic the clinical and phenotypic expression of hypertrophic cardiomyopathy (HCM) have recently been reported in young patients, including those diseases due to mutations in the X-linked lysosome-associated membrane protein gene (LAMP2; OMIM 309060; Danon disease). The morphologic expression and the clinical course experienced by patients with this newly identified cardiomyopathy are incompletely resolved. Therefore, it is informative to report our experience with an assessment of the natural history associated with LAMP2 cardiomyopathy.

METHODS

The most current clinical status of 7 previously identified patients with LAMP2 mutations was reexamined as of October 2008. Two-dimensional and Doppler echocardiographic studies were performed according to standard methodology with commercially available instruments. As previously described in detail, sequence analyses of genes that encode sarcomere proteins were performed.

Context Mutations in X-linked lysosome-associated membrane protein gene (LAMP2; Danon disease) produce a cardiomyopathy in young patients that clinically mimics severe hypertrophic cardiomyopathy (HCM) due to sarcomere protein mutations. However, the natural history and phenotypic expression of this newly recognized disease is incompletely resolved and its identification may have important clinical implications.

Objectives To determine the clinical consequences, outcome, and phenotypic expression of LAMP2 cardiomyopathy associated with diagnostic and management strategies.

Design, Setting, and Patients Clinical course and outcome were assessed prospectively in 7 young patients (6 boys) with defined LAMP2 mutations from the time of diagnosis (age 7-17 years; median, 14 years) to October 2008. Phenotypic expression of this disease was assessed both clinically and at autopsy.

Main Outcome Measures Progressive heart failure, cardiac death, and transplant.

Results Over a mean (SD) follow-up of 8.6 (2.6) years, and by age 14 to 24 years, the study patients developed left ventricular systolic dysfunction (mean [SD] ejection fraction, 25% [7%]) and cavity enlargement, as well as particularly adverse clinical consequences, including progressive refractory heart failure and death (n=4), sudden death (n=1), aborted cardiac arrest (n=1), or heart transplantation (n=1). Left ventricular hypertrophy was particularly marked (maximum thickness, 29-65 mm; mean [SD], 44 [15] mm), including 2 patients with massive ventricular septal thickness of 60 mm and 65 mm at ages 23 and 14 years, respectively. In 6 patients, a ventricular preexcitation pattern at study entry was associated with markedly increased voltages of R-wave or S-wave (15-145 mm; mean [SD], 69 [39] mm), and deeply inverted T-waves. Autopsy findings included a combination of histopathologic features that were consistent with a lysosomal storage disease (ie, clusters of vacuolated myocytes) but also typical of HCM due to sarcomere protein mutations (ie, myocyte disarray, small vessel disease, myocardial scarring).

Conclusions LAMP2 cardiomyopathy is a profound disease process characterized by progressive clinical deterioration leading rapidly to cardiac death in young patients (<25 years). These observations underscore the importance of timely molecular diagnosis for predicting prognosis and early consideration of heart transplantation.

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light chain, regulatory myosin light chain, and α-tropomyosin), PRKAG2 (γ subunit of the adenosine mono-
phosphate–activated protein kinase), GLA (α galactosidase), GAA (acid α-1,4-glucosidase), and LAMP2 (lysosome-associated membrane protein 2) were performed in each pro-
band. Only a LAMP2 mutation was

| Table. Clinical, Demographic, and Pathologic Findings in 7 Patients With LAMP2 Cardiomyopathya |
|---------------------------------|--------|--------|--------|--------|--------|--------|--------|
| Patient (Sex)                   | 1 (M)  | 2 (M)  | 3 (F)  | 4 (M)  | 5 (M)  | 6 (M)  | 7 (M)  |
| Age at cardiac diagnosis, y     | 8      | 14     | 11     | 15     | 17     | 7      | 15     |
| Age at last evaluation/death, y | 14     | 23     | 22     | 21     | 24     | 20     | 23     |
| Follow-up duration, y           | 6      | 9      | 11     | 6      | 7      | 13     | 8      |
| Presentation                    | Heart murmur (sports examination) | Syncope | Heart murmur | Family history | Abnormal ECG | Chest pain | AF |
| NYHA functional class Initial   | I      | I      | I      | I      | I      | I      | I      |
| Most recent                     | I      | IV     | I      | III    | IV     | III    | III    |
| Paroxysmal AF/flutter           | Yes    | No     | Yes    | Yes (3 episodes) | Yes | Yes    | Yes |
| Medical treatmentb              | Atenolol, verapamil, amiodarone, warfarin | Sotalol, amiodarone, warfarin, spironolactone | Metoprolol | Atenolol | Spiranolactone, metoprolol, lisinopril, digoxin, diuretics, warfarin | Sotalol, atenolol, diuretics | Atenolol, sotalol, warfarin, diuretics, amiodarone |
| Family history of CM            | 0      | Brother: WPW/LVH; aunt: WPW | 0 | Mother: dilated CM/transplant | 0 | 0      | 0      |
| Electrocardiogram (initial)     | WPW Yesc | Yes | Yes | Yes | 0 | Yes | Yesc |
| Maximum voltage, mm             | 145    | 80    | 75    | 55    | 15    | 55    | 56    |
| PR interval, ms                 | 105    | 80    | 125   | 80    | 154   | 80    | 110   |
| Other                           | T-inversion (11 mm), inferior Qs | T-inversion (30 mm), IVCD | T-inversion (25 mm) | T-inversion (22 mm) | LAD, absent R (V1-V3) | T-inversion (15 mm) | T-inversion (10 mm), LBBB |
| LV outflow gradient (rest), mm Hg1 | 65 | 0 (mild SAM) | 0 | 0 | 0 | 65 | 0 |
| Maximum LV wall thickness, mm2  | 652   | 60    | 30    | 371   | 35    | 529   | 29    |
| Ejection fraction, % Initial    | 70     | Normal | 64     | 70     | 75    | 66    | 68    |
| Most recent                     | 36     | 25     | 35     | 20     | 22    | 15    | 23    |
| LV cavity end diastole, mm     | 25     | 42     | 37     | 40     | 37    | 54    | 55    |
| Most recent                     | 43     | 70     | 53     | 60     | 49    | NA    | 68    |
| Left atrium (initial), mm      | 35     | 39     | 32     | 38     | 41    | 36    | 30    |
| Mitral regurgitation (initial)  | Moderate | Mild | Mild | Mild | 0 | 0 | 0 |
| 24-Hour ambulatory Holter ECG   | 633 PVBs, 8 couplets | NA | 3 PVBs, 1 couplet | Sinus bradycardia | NSVT | NSVT | 127 PVBs, 1 couplet |
| Complications                   | End-stageh | End-stage, embolic stroke | End-stageh | End-stageh | End-stage, acute cardiac/renal failure, syncope | End-stage, pulmonary hypertension, ICD shock for VT | End-stageh |

(continued)
found in the probands, which was confirmed by restriction enzyme digestion. Tissue sections of left ventricular (LV) myocardium were obtained from formalin-fixed hearts, embedded in paraffin, sectioned at 6-µm thickness, and stained with hematoxylin-eosin and Masson trichrome. All participating centers received approval from their institutional review boards.

RESULTS
Clinical Profile
Clinical, demographic, and outcome data were assembled for the 7 affected proband study patients and are summarized in the Table. At cardiac diagnosis, the 1 female and 6 male patients were 7 to 17 years old (median, 14 years). Clinical recognition in 6 patients occurred by virtue of heart murmur, family screening, and findings on routine electrocardiogram (ECG) or by symptoms (chest pain or syncope) and, in 1 patient, by atrial fibrillation. All patients had predominant or isolated cardiac manifestations without mental retardation or the neurological or musculoskeletal deficits associated with Danon disease.

On ECG at diagnosis, 6 patients had ventricular preexcitation patterns with short PR interval (Figure 1). Most patients also showed markedly increased standard lead voltages, precordial lead voltages, or both, with maximum R-wave or S-wave amplitude of 15 to 145 mm (mean [SD], 69 [39] mm), and usually with deep negative T-waves (Figure 1)(Table).

Clinical Course
At cardiac diagnosis, all patients were classified in New York Heart Association (NYHA) functional class I. During the subsequent mean (SD) time of 8.6 (2.6) years, each of the 7 patients experienced serious adverse clinical consequences by 14 to 24 years of age (mean, 21 years). Four patients died of acute or progressive heart failure, and 1 patient underwent heart transplantation. Clinical deterioration was often rapid, with the time interval from clinical stability with little or no symptoms and preserved systolic function to end-stage heart failure as brief as 6 months. Two other patients experienced sudden unexpected major arrhythmic events: 1 patient died suddenly (age 14 years) from ventricular fibrillation refractory to implantable cardioverter-defibrillator (ICD) therapy (Figure 2), and 1 patient received an appropriate defibrillator shock for rapid ventricular tachycardia at age 18 years.

All 7 patients developed marked LV systolic dysfunction (ejection frac-

### Table. Clinical, Demographic, and Pathologic Findings in 7 Patients With LAMP2 Cardiomyopathy

<table>
<thead>
<tr>
<th>Clinical status (most recent)</th>
<th>Patient (Sex)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sudden death (found dead in bed)</td>
<td>1 (M)</td>
</tr>
<tr>
<td>Acute HF death</td>
<td>2 (M)</td>
</tr>
<tr>
<td>Alive (ICD shock for VT [222/min])</td>
<td>3 (F)</td>
</tr>
<tr>
<td>Acute HF death</td>
<td>4 (M)</td>
</tr>
<tr>
<td>Sudden/HF death</td>
<td>5 (M)</td>
</tr>
<tr>
<td>Progressive HF death, liver multi-system failure, pneumonia</td>
<td>6 (M)</td>
</tr>
<tr>
<td>Alive, transplant</td>
<td>7 (M)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>ICD</th>
<th>Yes</th>
<th>Yes</th>
<th>Yes</th>
<th>Yes</th>
<th>Yes</th>
<th>Yes</th>
<th>Yes</th>
<th>0</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum enzymes elevated&lt;sup&gt;6&lt;/sup&gt;</td>
<td>Yes</td>
<td>Yes</td>
<td>0</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>0</td>
</tr>
<tr>
<td>Cognitive issues</td>
<td>Sporadic</td>
<td>Maternal</td>
<td>Sporadic</td>
<td>Maternal</td>
<td>Maternal</td>
<td>Sporadic</td>
<td>Sporadic</td>
<td>0</td>
</tr>
<tr>
<td>Genetic transmission</td>
<td>Y109ter (truncates protein at residue 109)</td>
<td>IVS6 + 1_4 del GTGA (in-frame deletion of exon 6 + 41aa)</td>
<td>IVS6-2A→G (frameshift after 22 aa; no RNA)</td>
<td>928G→A (missense V310I + deletion of exon 7)</td>
<td>IVS1 + 1G→T (deletes 21 aa)</td>
<td>IVS1-2A→G (deletes exon 2 + frameshift)</td>
<td>928G→A&lt;sup&gt;1&lt;/sup&gt; (missense V310I + deletion of exon 7)</td>
<td>0</td>
</tr>
</tbody>
</table>

Abbreviations: ADHD, attention-deficit/hyperactivity disorder; AF, atrial fibrillation; BI, behavioral issues; CM, cardiomyopathy; ECG, electrocardiogram; HF, heart failure; ICD, implantable cardioverter-defibrillator; IVCD, intraventricular conduction defect; LAD, left axis deviation; LBBB, left bundle-branch block; LV, left ventricular; LVH, left ventricular hypertrophy; NA, not available; NSVT, nonsustained ventricular tachycardia; NYHA, New York Heart Association; PVBs, premature ventricular beats; SAM, systolic anterior motion (of mitral valve); VT, ventricular tachycardia; WPW, Wolff-Parkinson-White syndrome (preexcitation pattern).

<sup>1</sup>Selected data from these patients appear in a previous report describing their initial genetic identification.<sup>1,9</sup>

<sup>2</sup>Drugs administered during the follow-up period.

<sup>3</sup>Radiocardiographic tracings at age 8 years in patient 1 and at age 15 years in patient 7.

<sup>4</sup>Maximum value recorded at any time during the follow-up period.

<sup>5</sup>Also, anomalous anterolateral papillary muscle insertion into anterior mitral leaflet.

<sup>6</sup>Left ventricular wall thickness regression to 14 mm at time of death (by echocardiography).

<sup>7</sup>Predominant hypertrophy of posterior LV free wall.

<sup>8</sup>End-stage refers to evolution to systolic dysfunction (ejection fraction <30%)

<sup>9</sup>Creatine kinase and alanine aminotransferase levels elevated by factor of 3.2 and organ-specific enzyme isozymes indicated cardiac, musculoskeletal, and liver involvement; maximum value is reported.

<sup>10</sup>Mosaic; both mutant and wild-type LAMP2 sequences were identified in multiple DNA samples, despite a normal XY karyotype; parental samples showed only normal LAMP2 sequences.

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tion, 20%-35%; mean [SD], 25% [7%]), associated with LV cavity dilatation in 4 patients and enlargement in 2 other patients over the follow-up period (Table). All 7 patients had received ICDs, which ultimately failed to terminate lethal ventricular tachyarrhythmias in 5.

Phenotype

Echocardiography. The most recent echocardiographic studies obtained in these patients demonstrated diffuse and marked LV hypertrophy in each. Maximum wall thickness (usually of ventricular septum) was 29 to 65 mm (mean [SD], 44 [15] mm), including 2 patients with particularly massive septal thickening of 60 mm and 65 mm at age 23 and 14 years, respectively (Table, patients 1 and 2). Left ventricular end-diastolic cavity dimension was documented to have dilated or enlarged over the follow-up period. Left ventricular outflow obstruction due to mitral valve systolic anterior motion was present at rest in 2 patients (gradient, 65 mm Hg).

Autopsy. Postmortem examination of 2 hearts showed massive cardiac hypertrophy; heart weights were 1265 g and 1425 g with asymmetric LV wall thickening (Figure 3). Patient 1 showed, in addition, substantial myocyte disarray, abnormal intramural coronary arteries (with thickened walls and narrowed lumen), and replacement fibrosis including subepicardial distribution (Figure 3).

Notably, both patients showed prominent clusters of numerous myocytes with distinctive and extensive cytosolic vacuolation (Figure 4) and inclusions of amorphous granular material in some cells within areas of scarring.

COMMENT

The clinical course of these 7 patients with LAMP2 mutations provides important insights regarding molecular diagnosis as well as the natural history, pathophysiology, and clinical im-

Figure 1. 12-Lead Electrocardiogram in LAMP2 Cardiomyopathy (Patient 2)

Figure 2. Intracardiac Ventricular Electrocardiogram in a Patient With LAMP2 Cardiomyopathy (Patient 1)
plications of this recently recognized genetic cardiomyopathy. LAMP2 mutations cause a particularly profound and accelerated cardiac disease process characterized by clinical deterioration and early death, perhaps representing one of the most lethal cardiomyopathies in young and usually male patients. Such an outcome occurred in the patients in our study despite application of the most contemporary treatment strategies, including the ICD, which failed to convert ventricular tachyarrhythmias to normal rhythm in 5 patients.

The clinical presentation of LAMP2 cardiomyopathy mimics severe HCM caused by mutations in genes encoding cardiac sarcomere proteins, as both are associated with marked LV hypertrophy. However, even though LAMP2 cardiomyopathy is a phenocopy of HCM, it represents a fundamentally different pathologic process that results from a defect in lysosome function. In 2005, we reported genetic diagnoses in the present 7 patients with LAMP2 mutations, and in the ensuing and relatively brief 3-year period, prospectively recognized that these patients had all experienced adverse and lethal disease consequences. Specifically, each patient evolved into an end-stage phase characterized by LV systolic dysfunction with enlarging cavity size and experienced severe outcome—sudden death, heart failure death, heart transplantation, or an appropriate ICD intervention triggered by rapid ventricular tachyarrhythmia—at age 14 to 24 years. Of note, the patients presented here with LAMP2 cardiomyopathy demonstrated a clinical profile dominated by cardiac manifestations, largely without overt evidence of the multisystem and extracardiac abnormalities (e.g., mental retardation, hepatic involvement, and overt skeletal myopathy) reported in other patients with Danon disease. These observations underscore the heterogeneous clinical expression of LAMP2 mutations.

Reliably predicting future adverse clinical events in HCM by genetic testing for sarcomeric mutations has proved challenging. In contrast, genetic identification of LAMP2 cardiomyopathy is highly informative of prognosis. Although the clinical outcome in the relatively small cohort of patients in this study was uniformly adverse, we recognize that LAMP2 mutations exhibit heterogeneity in disease expression and clinical course, particularly between male and female individuals. Indeed, 7 female LAMP2 obligate carriers (age range, 19-51 years) in 2 of the families remain asymptomatic and at present have not developed LV hypertrophy or systolic dysfunction, under-

Figure 3. Pathology of LAMP2 Cardiomyopathy (Patient 1)

From a 14-year-old boy with sudden death. A, At autopsy, massive asymmetric left ventricular (LV) hypertrophy. Ventricular septal thickness is 65 mm (heart weight, 1425 g), exceeding all hearts reported to date; LV cavity is small. B, Area of LV wall demarcated by white rectangle in A, showing subepicardial necrosis and scarring (arrowheads). C, Disorganized LV myocardium. Adjacent cardiac muscle cells (myocytes), or groups of cells, are arranged at perpendicular or oblique angles (Masson trichrome, original magnification ×100). D, High-power photomicrograph showing an abnormal intramural coronary artery with thickened wall and narrowed lumen (periodic acid–Schiff).
From the same patient shown in Figure 1, with findings consistent with a lysosomal storage disease. A. Small focal scars (stained blue) surrounded by viable myocardium (Masson trichrome, original magnification ×40). B. Similar area of myocardium shows subepicardial distribution of scarring and vacuolated myocytes (Masson trichrome, original magnification ×40). C. Clusters of myocytes with vacuolated sarcoplasm (stained red) embedded in an area of scar (stained blue, Masson trichrome, original magnification ×100). D. High-power photomicrograph showing a large empty myocyte surrounded by smaller vesicles in an area of replacement fibrosis (Masson trichrome).

In conclusion, LAMP2 cardiomyopathy in young patients appears to be a lethal genetic disease. The clinical resemblance of LAMP2 to sarcomeric HCM underscores the necessity and power of timely genetic testing in young patients with substantial LV hypertrophy, for the early molecular identification of this myocardial storage disease characterized by adverse clinical course.

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Study concept and design: Maron, Almquist, Wright, Almquist, Baffa, Saul, C. E. Seidman.
REFERENCES