Fabry Disease Female Proband With Clinical Manifestations Similar to Hypertrophic Cardiomyopathy

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SUMMARY

Fabry's disease is an X-linked inborn error of glycosphingolipid catabolism, resulting from a deficiency in α -galactosidase A (α -Gal A). A 56-year-old Japanese woman was at first suspected of having hypertrophic cardiomyopathy. The patient and her son had α -Gal A activity in leukocytes that was remarkably below the limit of controls. DNA analysis of the α -Gal A gene revealed a novel missense mutation at codon 19 in exon 1, resulting in leucine-to-proline substitution. As a result she was confirmed as a classic Fabry heterozygote. Recent advances in enzyme replacement therapy can reverse the storage of glycosphingolipids in Fabry's disease. Thus, in patients with cardiac hypertrophy, it is important to differentiate Fabry's disease from other causes of hypertrophy. Therefore, it is necessary to measure α -Gal A activity in all suspected cases and to analyze genetic abnormalities in heterozygotes. (Jpn Heart J 2004; 45: 685-689)

Key words: Fabry's disease, Hypertrophic cardiomyopathy, Missense mutation

FABRY'S disease is an X-linked inborn disorder caused by deficient activity of the lysosomal hydrolase alpha-galactosidase A (EC 3.2.1.22; α -Gal A).¹⁾ In normal individuals, this enzyme catalyses the hydrolysis of the α -galactosidic linkage at the nonreducing glycosphingolipid terminals. Loss of this activity results in the intralysosomal deposition of neutral glycosphingolipids with terminal α -linked moieties primarily in the plasma and vascular endothelium, leading to angiokeratoma, acroparesthesias, and vascular disease of the heart, kidneys, and brain. In the clinically severe classic form, the onset of disease manifestations usually occurs in childhood or adolescence, while an atypical variant with resid-

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ual α -Gal A activity or late-onset cardiomyopathy without systemic manifestations has been reported.²⁻⁵⁾

Fabry's disease can be diagnosed enzymatically in affected hemizygous males by a deficiency in α -Gal A activity in plasma and leukocytes. In contrast, the identification of heterozygous females is often inconclusive due to the random X-chromosomal inactivation. However, the locus for α -Gal A is assigned to the long arm of the X chromosome within band Xq 22. The entire 12-kb α -Gal A gene, which contains seven exons, is completely sequenced. Thus, genetic analysis is essential for identifying genotype/phenotype correlations and permitting precise heterozygote detection. Moreover, early diagnosis is important and needed since recent clinical trials showed the safety and efficacy of human recombinant α -Gal A.⁶⁾

We describe a female patient with Fabry's disease mimicking hypertrophic cardiomyopathy, confirming a missense mutation in the α -Gal A gene for precise diagnosis in a heterozygous state.

CASE REPORT

A 56-year-old Japanese woman presented at our clinic with dyspnea and bradycardia. On physical examination, her blood pressure was 120/76 mmHg with an irregular pulse rate of 66 beats/min. No heart murmur was heard. Angiokeratoma, acroparesthesia, and hypohydrosis were observed for the first time but corneal opacity and abnormal neurological findings were not found. An electrocardiogram demonstrated atrial fibrillation. Holter ECG recording showed an idioventricular rhythm (34 beats/min) at midnight. An echocardiogram indicated left ventricular hypertrophy (the thicknesses of the interventricular septum and left ventricular posterior wall were 15 mm and 7 mm), dilatation (end-diastolic and end-systolic dimensions, 60 mm and 48 mm), and poor systolic function (left ventricular ejection fraction, 30%). On laboratory examination, proteinuria was detected (Table). Total bilirubin (2.7 mg/dL) and LDH (719 IU/L) levels were high and the platelet count ($13.4 \times 10^4/\mu\text{L}$) was low. Consequently, she was tentatively diagnosed as having a dilated form of hypertrophic cardiomyopathy in conjunction with angiokeratoma and proteinuria.

For the differentiation of Fabry's disease from hypertrophic cardiomyopathy, informed consent was obtained from the patient before the α -Gal A activity and gene analyses. α -Gal A activity was analyzed in the homogenates of peripheral lymphocytes with the substrate of 5mM 4-methylumbelliferyl- α -galactosidase (Sigma-Aldrich, Tokyo) at pH 4.5 by the method of Suzuki.⁷⁾ Genomic DNA was extracted from peripheral lymphocytes by the standard method⁸⁾ and each exon was amplified by polymerase chain reaction according to a previous

Table. Laboratory Data

		Normal range
WBC ($\times 10^2/\mu\text{L}$)	42	43-80
RBC ($\times 10^4/\mu\text{L}$)	451	395-465
Hgb (g/dL)	14.5	11.3-14.9
Hct (%)	42.7	36.0-47.0
Plt ($\times 10^4/\mu\text{L}$)	13.4	18-34
TP (g/dL)	7.6	6.5-8.5
Alb (g/dL)	4.1	3.5-5.0
T-Bil (mg/dL)	2.7	0.2-1.0
Amylase (IU/L)	12	40-200
AST (IU/L)	54	12-40
ALT (IU/L)	20	10-45
ALP (IU/L)	193	80-230
ChE (IU/L)	416	350-750
LDH (IU/L)	719	250-500
CK (IU/L)	126	30-140
BUN (mg/dL)	21	7.0-18
Cre (mg/dL)	0.63	0.4-0.9
Na (mEq/L)	141	137-146
K (mEq/L)	5.0	3.8-5.1
Cl (mEq/L)	103	98-108
FBS (mg/dL)	106	70-105
Urine		
pH	6.0	4.5-7.0
Glucose (mg/dL)	0	0-30
Protein (mg/dL)	30	0-20
Bilirubin (mg/dL)	0	0
Urobilinogen (mg/dL)	0.1	0.2-2.0

method,⁹⁾ with some modifications. PCR products were sequenced by a direct sequencing method using a capillary sequencer (ABI PRISM 310 Genetic Analyzer, Perkin Elmer Japan Co./ABI Inc., Chiba, Japan) with a Rhodamin Terminator Cycle Sequencing Kit from the same company.

α -Gal A activity was considerably low (5.5 nmol/mg/h) compared to the normal control ($n = 26$) value of 18.2 ± 5.9 nmol/mg/h. In the molecular analysis of the α -Gal A gene, an amino acid substitution of leucine (CTG) to proline (CCG) at codon 19 was found to result in a heterozygous state. This was a novel mutation and was located almost immediately downstream to the signal peptide sequence (codon 1-17).

Her 29-year-old son was apparently healthy without any complaints but had angiokeratoma, acroparesthesia, and hypohydrosis. He had a normal ECG. Echocardiographic studies showed normal left ventricular function without hypertrophy. On laboratory examination, proteinuria was detected but the glomerular filtration rate was 93 mL/h. α -Gal A activity was considerably below nor-

CM, cardiomegaly; AK, angiokeratoma; PU, proteinuria.

Figure. Pedigree of the present family. Blackened symbols denote individuals with symptoms related to Fabry's disease.

CM = cardiomegaly; AK = angiokeratoma; PU = proteinuria.

mal (2.6 nmol/mg/h) indicating a hemizygote. In the molecular analysis of the α -Gal A gene, the same mutation was found as in his mother. Meanwhile, her daughter had normal α -Gal A activity (19.0 nmol/mg/h) and no gene mutation, consistent with the results of enzyme analysis. The Figure shows this family pedigree.

DISCUSSION

Precise diagnosis may be delayed or missed since the cardiac manifestation of Fabry's disease often mimics hypertrophic cardiomyopathy.¹⁰⁾ At first, she was tentatively suspected of having hypertrophic cardiomyopathy in the echocardiographic examination. However, the angiokeratoma and proteinuria suggested to us other causes of hypertrophy rather than hypertrophic cardiomyopathy. Therefore, α -Gal A activity was measured and the final diagnosis was confirmed by genetic analysis.

Consequently, we found a Fabry heterozygote with the classic form, identifying a novel missense mutation (L19P) with amino acid substitution in the α -Gal A gene. This was presumed to be a disease-causing mutation and to cause conformational change in the α -Gal A gene.

Enzymatic activity is useful for diagnosing Fabry's disease in hemizygous males, differentiating it from other causes of cardiac hypertrophy. However, this is often inconclusive for the identification of heterozygous females. As a result, it

is important to analyze mutations in Fabry's disease since the detection of these mutations provides a precise diagnosis of the heterozygous carriers of the X-linked recessive disorder and molecular prenatal diagnosis.

Fabry's disease may not be rare in cardiac hypertrophy. When 230 or 153 male patients with cardiac hypertrophy were screened, 7 or 6 cases (3% or 4%) exhibited Fabry's disease as a cardiac variant, respectively.^{4,5)} Cardiac involvement with hypertrophy and valvular abnormalities is also common in heterozygous females.¹¹⁾

In summary, we conclude that it is important to consider Fabry's disease in the differential diagnosis of cardiac hypertrophy since the disease is treatable and the enzyme replacement therapy can clear the capillary endothelial accumulation of glycosphingolipids in the kidneys, heart, and skin.^{6,10)} Therefore, it is necessary to measure α -Gal A activity in all patients suspected of having Fabry's disease and to analyze gene mutations in heterozygotes as well as to confirm the intralysosomal deposition of glycosphingolipids in endomyocardial biopsy specimens.

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