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Mucopolysaccharidosis Type II in Females and Response to Enzyme Replacement Therapy

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Mucopolysaccharidosis type II (MPS II, Hunter syndrome) is an X-linked lysosomal storage disease caused by a deficiency of iduronate-2-sulfatase (IDS). Two affected girls with moderate and severe forms of MPS II with normal karyotypes and increased urinary dermatan sulphate and heparin sulphate excretion and marked deficiencies of IDS activity are reported. Molecular studies showed that case 1 has a heterozygous mutation c.1568A > G (p.Y523C) associated with almost totally skewed inactivation of the normal maternal X chromosome, and case 2 has a heterozygous deletion that includes exons 1–4 of IDS (minimal deletion range c.1–103_184del). The multi-exon deletion correlated with early onset of the disease and severe phenotype with intellectual disability, whereas the missense mutation was associated with moderate developmental delay. Although genotype–phenotype correlation in MPS II is difficult, gene deletions seem to correlate with more severe clinical manifestation of the disease. Enzyme replacement therapy (ERT) in these two females resulted in disease stabilization in both.

Key words: mucopolysaccharidosis type II; female Hunter syndrome; genotype; phenotype; enzyme replacement therapy

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INTRODUCTION

Mucopolysaccharidosis type II (MPS II, Hunter syndrome, OMIM 309900) is an X-linked recessive disease resulting from deficient activity of lysosomal iduronate-2-sulfatase (IDS; EC 3.1.6.13) [Neufeld and Muenzer, 2001]. Phenotypic expression of X-linked recessive disorders in females may be the result of structural abnormalities of the X chromosome leading to homozygosity for disease-causing mutations or abnormalities in the X-chromosome inactivation (XCI) process. Random inactivation of the X chromosome occurs shortly after the implantation of the blastocyst through silencing of most genes located on the X chromosome, a process that is regulated by the X-inactivation center. Consequently, MPS II heterozygous females are rarely affected unless there is a simultaneous presence of two mutant alleles or if a coincidental genetic defect leads to skewed X-inactivation or hemizygosity in heterozygotes. We describe two affected girls with moderate and severe forms of MPS II, respectively.

Patient 1

The patient was born with a weight of 3,250 g (50th centile), length of 56 cm (90–97th centile), and head circumference of 34 cm (50th centile) as the first child of healthy and non-consanguineous parents of Polish ancestry. The first symptoms of MPS II were

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noticed by the parents at the age of 4. At that time, the girl did not show any characteristic phenotypic features, although there was decreased range of motion (ROM) in multiple joints (especially elbow, hip, and ankle joints), slight hepatomegaly, mild umbilical hernia (not requiring surgical intervention), and discrete cardiac valvular changes with slight left ventricular hypertrophy.

At age 5 years, the diagnosis of MPS II was confirmed by enzymatic and molecular analysis. She exhibited increased urinary GAG excretion (195 mg GAG/1 g of creatinine; control: <66 mg GAG/1 g of creatinine) and very low IDS activity in both serum (35 pmoles/ml/hr; age matched control range: 500–650 pmoles/ml/hr) and cultured fibroblasts (18 pmoles/mg of protein/18 hr; control cells: 622 pmoles/mg/18 hr) (Table I).

Her karyotype was 46, XX. Direct sequencing of the IDS gene showed a heterozygous mutation c.1568A > G (p.Y523C) in exon 9 in association with almost totally skewed XCI of the X-chromosome carrying the wild-type IDS allele from her father. The proband's mother was found to be a heterozygote for this mutation, proband's sister was wild-type homozygote, and proband's father was wild-type hemizygote. Additionally, the proband's brother (one of dizygotic twins) was found to be a mutant hemizygote suffering from Hunter syndrome [Kloska et al., 2010; Tylki-Szymanska et al., 2011].

At 5 years, she presented with delayed mental development with an IQ of 68 (Weschler scale). With time, deterioration was noticed, joint contractures in all joints became more pronounced, and contractures of Achilles tendons led to toe walking. The patient's IQ decreased with time, at the age of 7 years her speech was arrested and her hearing was impaired.

At 9 years, she started enzyme replacement therapy (ERT) with weekly intravenous idursulfase (Elaprase, Shire Human Genetic Therapies, Inc., Cambridge, MA) at the standard dose of 0.5 mg/kg. The study began after it was given the Institutional Ethics Committee's approval and informed consent from the parents. Over the following 2 years, treatment was administered regularly and the compliance with weekly infusions was 100%. ERT was well-tolerated without significant adverse events. Detailed anthropometric features, ultrasound studies of liver and spleen volumes, echocardiography, audiological examinations, psychological tests, joint ROM and skeletal radiographs were monitored (Table II).

Currently, at age 11 years, she exhibits features characteristic of MPS II disease such as intellectual disability (IQ = 24), short stature, significant joint contractures, lumbar lordosis (walking on toes), cardiac disease (left atrial and ventricular hypertrophy and valvular disease—MVR I/II degree, AVR II degree), and slight hepatomegaly (Fig. 1, Table II).

Patient 2

The patient was born with weight of 3,900 g (75–90th centile) and length of 54 cm (75–90th centile) as the second child of healthy and non-consanguineous Latvian parents. Soon after the first year of life, she was recognized to have delayed speech development and chronic nasal discharge. At the age of 2.4 years an adenoidectomy was performed. At age 2 years 9 months, she had coarse facial features, enlarged tongue, stiffness of joints, camptodactyly, hypertrichosis, hoarse voice, and hepatosplenomegaly. She was able to speak some words and maintain good contact with children with only slight developmental delay. She was hearing impaired.

At 2 years 10 months, she was diagnosed with MPS II by GAG analysis and enzyme assay. She had increased urinary GAG excretion (987 mg GAG/1 g of creatinine; age matched control: 116+−70 mg GAG/1 g of creatinine) and very low IDS activity in both serum (2 nmoles/ml/ 4hr, controls 212–660) and leukocytes (0.16 nmoles/mg of protein/4 hr; control cells 30) (Table I).

TABLE I. Summary of Data of 2 MPS II Female Patients

Patient	**1**	**2**
Origin	Polish	Latvian
Age at onset/age of diagnosis	4y/5y	1y 8 mo/2y 10 mo
First symptoms	Developmental delay (speech delay)	Umbilical hernia since birth, developmental delay, chronic nasal discharge
GAG[a] (mg/g creatinine)	195 (control <66)	987 (control 116 + −70)
IDS[b]	35 pmoles/ml/hr	2.0 nmoles/ml/4 hr (controls 212–660)
IDS[c]	nd	0.16 nmoles/mg protein/4 hr (control 30)
IDS[d]	18 pmoles/mg protein/18 hr	nd
Molecular analysis	c.1568A > G (p.Y523C) almost totally skewed XCI in favor of the X-chromosome carrying the wild-type IDS allele of paternal origin	a heterozygous deletion that includes exons 1–4 of the IDS gene (minimal deletion range c.1–103_184del)
Current age (years)	11	7.4
Impaired mobility (age in years)	6	4

y: Years; mo: Months.

[a]GAG in urine (mg/g creatinine), age-matched control.
[b]IDS activity in patient's plasma.
[c]IDS activity in patient's leukocytes.
[d]IDS activity in patient's cultured fibroblasts.

TABLE II. Patient's Assessments at Baseline and After 24 months of ERT With Idursulfase

Patient 1	Baseline (9 years)	Study end-point (24 months of ERT)
Anthropometric measurements	height 122.7 cm (<3rd centile) weight 26.2 kg (<3rd centile) head circumference 56.5 cm (>97th centile)	height 127.4 cm (<3rd centile) weight 32 kg (<3rd centile) head circumference 58.5 cm (90th centile)
Head, eyes, nose, throat:	no symptoms	no symptoms
visual impairment		
hearing loss		
Cardiovascular system	thickness of mitral valve II/III and prolapse with mitral valve regurgitation, LA and LV hypertrophy	thickness of mitral valve II/III and prolapse with mitral valve regurgitation, LA and LV hypertrophy (stable)
Gastrointestinal system (hepatosplenomegaly)	normal volume	normal volume
Musculoskeletal system:		
Spinal deformity	+/−	+
Joint stiffness	++	++
Joint contractures	++	++
Neurological system	seizures developmental delay IQ 48 Termann/Merill	seizures developmental delay, IQ 24 Termann/Merill
GAG in urine (mg/g creatinine)	195mg/g creatinine (control 88+/−77)	45mg/g creatinine (control 88+/−77)

++++ very strong, +++ strong, ++ medium, +slight.

Her karyotype was 46, XX, and MLPA analysis (P164B1) identified a heterozygous deletion of exons 1–4 of IDS (minimal deletion range c.1–103_184del). Her mother and sister have almost totally skewed X inactivation; however, they are not carriers for the deletion present in the patient.

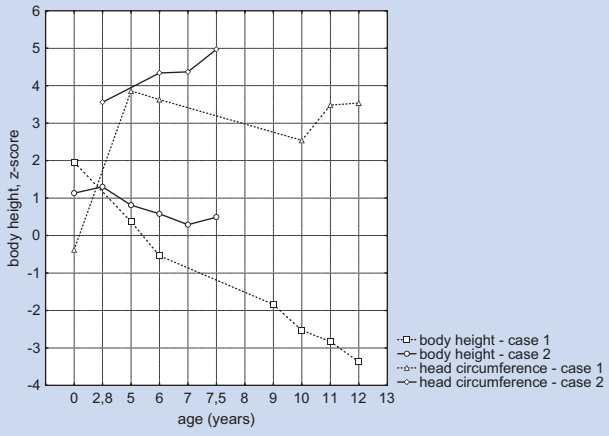

FIG. 1. Z-score values for the body height and the head circumference. The body height for both girls until the 5th year of life was similar (slightly >50th centile). Later the pace of growth of the Polish patient slowed down while for the Latvian patient it remained at similar level. It can be expected, that the rate of growth for the Lativan patient will suddenly slow down around 9th year of life and the body height will be below the 3rd centile [Rozdzynska et al., 2011].

Over time, her coarse facial features, hepatosplenomegaly, and hearing impairment became more pronounced. She developed recurrent airway obstruction, severe intellectual disability, speech arrest, and skeletal disease with dysostosis multiplex. Echocardiography around age 4 years showed a thickened mitral valve with prolapse and regurgitation, pericardial effusion (4 mm), and sinus arrhythmia in ECG.

At 5 years 10 months, she started weekly intravenous infusions of idursulfase (Elaprase, Shire Human Genetic Therapies, Inc, Cambridge, MA) at the standard dose of 0.5 mg/kg. The study began after it was given the Institutional Ethics Committee's approval and informed consent from the parents. Over the following 20 months, treatment was administered regularly and the compliance was 100% and well-tolerated. Detailed anthropometric features, ultrasound studies of liver and spleen volumes, echocardiography, audiological examinations, psychological tests, joint, ROM and skeletal radiographs were monitored (Table III). Currently, at age 7.4 years, her physical endurance increased and she is able to walk longer distances (25–30 m), and hepatosplenomegaly decreased. Her psychomotor development did not change; however, her mother noted improved attention span (Fig. 1, Table III). Nerve conduction studies showed severe carpal tunnel syndrome.

DISCUSSION

A number of female patients with MPS II have been reported [Neufeld et al., 1977; Mossman et al., 1983; Broadhead et al., 1986; Clarke et al., 1991; Winchester et al., 1992; Sukegawa et al., 1997; Sukegawa et al., 1998; Cudry et al., 2000; Neufeld and Muenzer, 2001; Tuschl et al., 2005; Kloska et al., 2010; Sohn et al., 2010; de Camargo Pinto et al., 2011].

TABLE III. Patient's Assessments at Baseline and After 20 months of ERT With Idursulfase

Patient 2	Baseline (5 years 10 months)	Study end-point (20 months of ERT)
Anthropometric measurements	height 120 cm (75th centile) weight 38 kg (>97th centile) head circumference 57.5 cm (>97th centile)	height 126 cm (50th centile) weight 40 kg (>97th centile) head circumference 58.2 cm (>97th centile)
Head, eyes, nose, throat: visual impairment corneal clouding hearing loss	sensoneuronal deafness	hypermetropic astygmatismus, paleness of n.optici/no corneal clouding
Cardiovascular system	thickness of mitral valve and prolapse with mitral valve regurgitation, sinus arrhythmia	thickness of mitral valve and prolapse with mitral valve regurgitation (5mm), no liquid in pericard, sinus arrhythmia
Gastrointestinal system (hepatosplenomegaly)	Liver +4.5, spleen +3	Liver +1, spleen + 0.5
Musculoskeletal system:		claw hands
Spinal deformity	+	+
Joint stiffness	++	+++
Joint contractures	++	+++
Neurological system	moderate development delay and behavioral problems (poor contact and hyperactivity) IQ 30	moderate development delay and behavioral problems (poor contact and hyperactivity) IQ 30 carpal tunnel syndrome ++++
GAG in urine (mg/mM of creatinine)	43.28	18

++++ very strong, +++ strong, ++ medium, +slight.

Clinical manifestations of the disease in these two female patients were a result of non-random inactivation of the X-chromosome carrying the normal allele of IDS.

Correlation between genotype and phenotype in MPS II is difficult because of the heterogeneity of mutations occurring in IDS among patients. Until now, more than 300 different mutations have been described [Froissart et al., 2002]. Large structural alterations of IDS, like gene deletions or rearrangements, correlate with more severe clinical manifestation of the disease [Vafiadaki et al., 1998; Froissart et al., 2002]. This is consistent with data reported in this study. A deletion involving exons 1–4 (detected in Patient 2) correlated with early onset of the disease and a severe phenotype with intellectual disability. In contrast, the missense mutations are difficult to predict clinical severity, and usually depend on the location of the specific mutation. Generally, mutations located in exon 9 of IDS result in a milder form of Hunter syndrome and do not affect highly conserved sequences of the enzyme, thus some residual activity of the enzyme remains. This is in contrast to point mutations affecting catalytic domains, consensus splice sites, or nonsense mutations resulting in extremely low or undetectable catalytic activity of the defective enzyme [Jonsson et al., 1995; Froissart et al., 2002]. Nevertheless, mutations causing severe phenotype have been reported within exon 9 [Vafiadaki et al., 1998]. Mutation p.Y523C (located in exon 9) in case 1 was previously associated with an attenuated form of the disease in a male [Jonsson et al., 1995; Tylki-Szymanska et al., 2011]; however, in this patient it resulted in moderate developmental delay.

It has been suggested that variability in X-inactivation can lead to a milder and more variable clinical and biochemical phenotype in females than in males [Lyon, 1961; Migeon and Pappas, 2007]. Heterozygotes for X-linked diseases could be symptomatic if the mutation confers a proliferative advantage to the mutant cells [Grinzaid et al., 2002] or there is skewed X-inactivation and the mutant is in the active X chromosome [Mossman et al., 1983; Tuschl et al., 2005; Migeon, 2006; Migeon and Pappas, 2007].

Non-genetic factors can also modify the disease phenotype. Recently, a model of correlation between the severity of Hunter syndrome and the residual activity of the deficient enzyme together with the level of GAG synthesis has been proposed [Piotrowska et al., 2009]. According to that study, low- or average-efficiency of GAG synthesis combined with the presence of residual activity of the enzyme results in an attenuated phenotype. Patient 1 in this report was classified as compatible with the model [Piotrowska et al., 2009], but similar studies were not provided for Patient 2.

ERT with recombinant human IDS (Elaprase, Shire Human Genetic Therapies, Inc, Cambridge, MA) has been shown to be a safe and effective therapy across a wide range of ages and disease severity; improving respiratory function, joint mobility, walking ability, and quality of life [Alcalde-Martin et al., 2010; Muenzer et al., 2011a, 2011b, 2007, 2006]. Experience with treating female patients with idursulfase is limited to similar improvement to males, as was demonstrated in these two cases.

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