Case Report

LPIN1 deficiency with severe recurrent rhabdomyolysis and persistent elevation of creatine kinase levels due to chromosome 2 maternal isodisomy

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ARTICLE INFO

ABSTRACT

Fatty acid oxidation disorders and lipin-1 deficiency are the commonest genetic causes of rhabdomyolysis in children. We describe a lipin-1-deficient boy with recurrent, severe rhabdomyolytic episodes from the age of 4 years. Analysis of the LPIN1 gene that encodes lipin-1 revealed a novel homozygous frameshift mutation in exon 9, c.1381delC (p.Leu461SerfsX47), and complete uniparental isodisomy of maternal chromosome 2. This mutation is predicted to cause complete lipin-1 deficiency. The patient had six rhabdomyolytic crises, with creatine kinase (CK) levels up to 300,000 U/L (normal, 30 to 200). Plasma CK remained elevated between crises. A treatment protocol was instituted, with early aggressive monitoring, hydration, electrolyte replacement and high caloric, high carbohydrate intake. The patient received dexamethasone during two crises, which was well-tolerated and in these episodes, peak CK values were lower than in preceding episodes. Studies of anti-inflammatory therapy may be indicated in lipin-1 deficiency.

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1. Introduction

Acute rhabdomyolysis is a life-threatening condition. In adults it is typically due to trauma, intoxication or infection, whereas in children with recurrent rhabdomyolysis, inherited muscle disorders are frequent [1,2]. The two commonest causes of early onset recurrent, severe rhabdomyolysis in children are fatty acid oxidation disorders and mutations in the LPIN1 gene that encodes lipin-1. Over 35 lipin-1-deficient patients are reported [3–6]. Clinically, lipin-1 deficiency is an autosomal recessive disorder presenting with episodic myalgia and myoglobinuria, most often triggered by febrile illness and less commonly by prolonged exercise, fasting and anesthesia. Myalgia can precede the increase of the creatine kinase (CK) level [7]. The rhabdomyolytic episodes of lipin-1 deficiency typically begin before 6 years of age [4,8]. Some LPIN1 heterozygotes also experience cramps and exercise-induced myalgia [8]. Heterozygous LPIN1 mutations have been identified in two individuals with statin-induced

myopathy, suggesting that partial lipin-1 deficiency might represent a risk factor for drug-induced myotoxicity [4,9].

Over twenty LPIN1 mutations have been described to date in several ethnic groups but no clear genotype-phenotype correlation has been shown [3–5,8]. The LPIN1 gene spans 19 exons and a deletion mutation spanning exon 18 occurs in 86% of Caucasian families/patients [3,8]. Lipin-1 is an 890 amino acid protein [10], highly expressed in skeletal muscle, adipose tissue, liver and myocardium [11]. Lipin-1 is both an enzyme and a transcriptional regulator. Lipin-1 has phosphatidic acid phosphatase (PAP1) activity [12], converting phosphatidic acid (PA) to diacylglycerol (DAG). DAGs are substrates for the synthesis of triacylglycerols, phosphatidylcholine and phosphatidylethanolamine [13–15] and in mouse cells with severe lipin-1 deficiency, incubation with DAGs enhances autophagy and survival [16]. Lipin-1 is also a co-activator of PPARγ and PPARα, factors that control the transcription of genes of fatty acid oxidation [11,17–19]. Lipin-1 can bind NFATc4 (nuclear factor of activated T-cells) to repress inflammatory gene expression [20,21] and the relationship of rhabdomyolysis, inflammation and lipin-1 deficiency was recently reviewed [22].

We report a child with recurrent episodes of severe rhabdomyolysis, persistent elevation of creatine kinase (CK) between episodes and severe deficiency of lipin-1 caused by complete isodisomy of a

maternal chromosome 2 containing a novel homozygous frameshift *LPIN1* mutation.

2. Case report

The patient presented at 4 years of age with severe rhabdomyolysis during an upper respiratory tract infection with fever and cough, due to a metapneumovirus. His antenatal medical history was unremarkable except for maternal gestational diabetes. Psychomotor development is normal. The parents are non-consanguineous French Canadians. The mother complained of mild unexplained chronic myalgia since childhood.

There was no history of toxin ingestion or trauma. He had a 6-month history of nocturnal lower limb pain, particularly after physical exertion. Physical examination revealed unsteady gait and difficulty rising to a standing position, due to muscle pain. Laboratory evaluation showed marked elevations of creatine kinase (CK, 299,966 U/L, normal range for age, 30–200 U/L), aspartate aminotransferase (5810 U/L, normal for age, 11–43 U/L) and alanine transaminase (1050 U/L, normal for age, 11–25 U/L) but was otherwise normal. His plasma acylcarnitine profile was normal.

LPIN1 gene sequencing was performed after the second rhabdomyolytic episode, revealing homozygosity for a novel mutation in exon 9, c.1381delC (p.Leu461SerfsX47), which is predicted to produce a truncated protein lacking the functionally-essential C-LIP and PAP domains and to result in complete deficiency of lipin-1. Both parents were tested for this mutation and only the mother was found to be a carrier. Because of her history of mild chronic myalgia, physical examination and plasma CK determination were performed and were normal (CK, 90 U/L). Deletion and amplification analysis of the *LPIN1* gene was normal. To rule out non-amplification of a paternal allele (allele drop-out), exon 9 of the *LPIN1* gene was re-amplified from the father's DNA using a different set of primers, and the mutation was absent. The most likely remaining explanations were total or segmental maternal uniparental isodisomy (UPD) of chromosome 2 or nonpaternity. PCR-based analysis of DNA from both parents and the patient was performed to test for UPD of chromosome 2, using 8 microsatellite markers spanning the length of chromosome 2. Seven markers were informative, and they demonstrated that the patient inherited only one set of the maternal alleles for these markers, consistent with maternal isodisomy for all of chromosome 2 (Fig. 1).

At present, the patient has had a total of 6 rhabdomyolytic episodes, three of which occurred during febrile infections. Between episodes of rhabdomyolysis, he has occasionally complained of myalgia, but his neuromuscular examination was normal. His plasma CK levels have never normalized between episodes, typically ranging between 400–700 U/L with a recorded maximum value during an asymptomatic period of 2163 U/L. For each of the rhabdomyolytic episodes, the highest recorded CK values and the duration of the episode (defined as time during which plasma CK levels exceeded 2200 U/L) were as follows: episode 1, 299,966 U/L, 6 days; episode 2, 17,450 U/L, 6 days; episode 3, 140,126 U/L, 7 days; episode 4, 11,148 U/L, 5 days; episode 5, 4434 U/L, 3 days; episode 6, 244,459 U/L, 10 days.

In each episode, high caloric intake was provided orally as high carbohydrate meals and as intravenous dextrose, with lipids contributing ≤30% of total energy intake. Intravenous fluids (10% dextrose, 0.9% sodium chloride) were initially administered at 1.5–2× maintenance rate, with other electrolytes and bicarbonate added as necessary to avoid myoglobinuria-associated renal complications. Cardiac monitoring was performed in the intensive care unit during the acute phase. L-carnitine was also given (≥50 mg/kg/day in 8 doses intravenously). The parents were provided with detailed instructions for prevention of episodes, which include exercise as tolerated, hydration during exercise and prompt consultation to the emergency department in case of myalgia or fever. In the fourth and fifth episodes, in addition to the above mentioned protocol, dexamethasone was given (0.6 mg/kg intravenously, repeated once 24 h later in episode 4 and repeated 4 times every 24 h in episode 5). Episode 6 was atypical, beginning with stridor and respiratory distress, that were treated as bronchospasm with dexamethasone (0.5 mg/kg, one dose) but also salbutamol by nebulisation (800 mcg every 20 min for 4 doses) and one dose of epinephrine (5 mg by nebulisation) following which CK increased markedly.
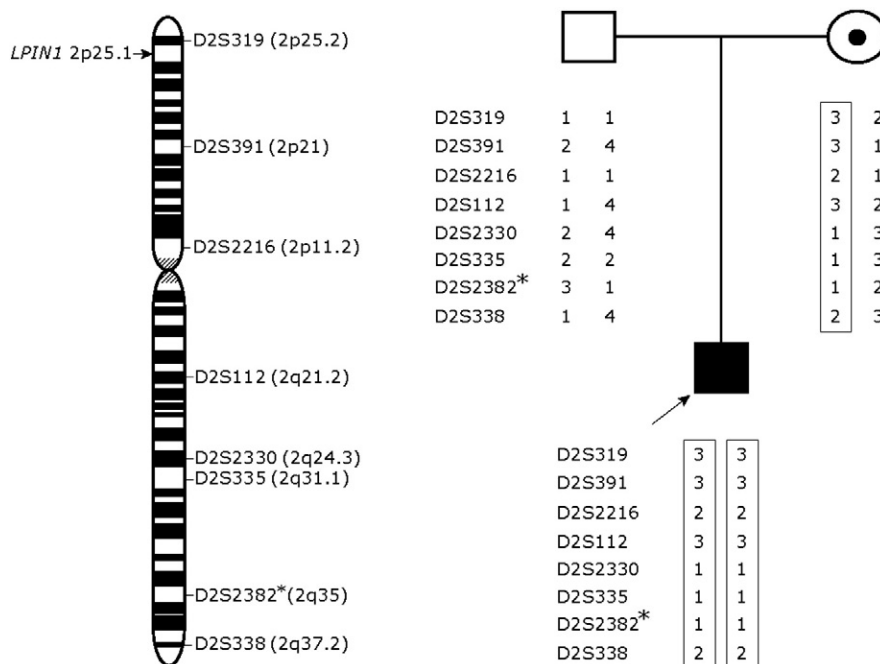

Fig. 1. UPD testing, demonstrating complete maternal isodisomy of chromosome 2 in the proband. Microsatellite markers were used to determine the parental origin of the two chromosome 2 homologs of the proband. All the informative markers (7 out of 8) were inherited from the same maternal chromosome 2. The distribution of the markers along chromosome 2 and the position of the *LPIN1* gene are depicted in the ideogram on the left of the figure. The asterisk indicates the non-informative marker.

3. Discussion

This is the first report of lipin-1 deficiency caused by uniparental disomy. For this patient's family this discovery alters genetic counseling: the risk of recurrence of lipin-1 deficiency by uniparental disomy equals that of the general population [23] i.e. much less than 1%, compared to the 25% risk of autosomal recessive transmission for most families with lipin-1 deficiency.

In lipin-1 deficiency, rhabdomyolytic episodes vary from subclinical elevations of plasma CK level to life-threatening crises. This high clinical variability complicates the interpretation of treatment effects in small numbers of observations and highlights the need for collaborative clinical trials that will allow for evidence-based conclusions. Nonetheless, observations from exceptional patients such as the one described in this report may be useful for designing such trials.

The patient had a severe clinical course, with very high CK levels that diminished slowly and incompletely and interictal elevation of plasma CK. A small fraction of lipin-deficient patients are reported to have interictal exercise intolerance and increased CK levels between crises [5,8], the highest reported value being 500 U/L. Our patient's *LPIN1* mutation is predicted to be severe, producing a truncated protein lacking both the catalytic and the transcriptional domains of lipin-1. Perhaps the severe nature of the patient's mutation may relate to the severity of his rhabdomyolytic episodes.

Regarding the symptomatic treatment of rhabdomyolysis in lipin-1 deficiency, there is general agreement in favor of early detection, aggressive, hydration, high energy intake from carbohydrates and monitoring for hyperkalemia and cardiac arrhythmias [5–8,23]. In contrast, the specific pathophysiology of lipin-1 deficiency is poorly understood. The concept is gaining ground that synergistic interaction among lipid metabolism, lipid signaling and inflammation [22] may underlie the severe rhabdomyolytic crises of lipin-1 deficiency. This perspective suggests new treatment possibilities. Because pro-inflammatory cytokines (TNF1α and IL-1β) exacerbate lipin-1 deficiency [22], inhibition of TNF-α and/or IL-1 with either a recombinant IL-1 receptor antagonist or an anti-IL-1β antibody [24] could be of potential therapeutic value. Dexamethasone, an inducer of PGC-1α, stimulates lipin-1 expression and PAP1 activity in adipocytes and hepatocytes [25], while also acting on inflammation. In addition, dexamethasone was shown to decrease the number of lipid droplets in lipin deficient patients' myoblasts [21]. Interestingly, in our patient, treatment with dexamethasone was well-tolerated and the peak levels of plasma CK during episodes 4 and 5, uncomplicated episodes during which he rapidly received dexamethasone, were lower than the highest CK levels during episodes 2 and 3, during which he did not receive dexamethasone but which were otherwise comparable to episodes 4 and 5. Taken together, these data suggest that dexamethasone treatment merits further study in lipin-1 deficiency.

Compliance with ethics guidelines

Conflict of interest

I. A. Meijer, F. Sasarman, C. Maftei, M.Vanasse, P. Major, E. Rossignol, G. Mitchell, and C. Brunel-Guitton declare that they have no conflict of interest.

This work did not receive funding from a specific grant.

Informed consent

All procedures followed were in accordance with the ethical standards of the responsible committee on human experimentation (institutional and national) and with the Helsinki Declaration of 1975, as revised in 2000. Informed consent was obtained from the family for being included in the study.

Details of the contributions of individual authors

I. A. Meijer was involved in the clinical follow up of the patient and contributed to the conceptualization, planning, writing and revision of the manuscript and figures.

F. Sasarman contributed to the conceptualization, planning, literature review, writing and revision of the manuscript and figures.

C. Maftei was involved in the clinical follow up of the patient and contributed to the conceptualization, planning, literature review, writing and revision of the clinical part and uniparental disomy part of the manuscript and contributed to the figures' conceptualization.

G. Mitchell was involved in the clinical follow up of the patient, in the literature review and in the revision of the manuscript and figures.

E. Rossignol was involved in the planning of the manuscript and the revision.

M. Vanasse was involved in the clinical follow up, and contributed to the conceptualization of the manuscript.

P. Major was involved in the clinical follow up, and contributed to the conceptualization of the manuscript.

C. Brunel-Guitton was involved in the clinical follow up of the patient and contributed to the conceptualization, planning, literature review, writing and revision of the manuscript and figures.

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