Short Communication

Recurrent rhabdomyolysis and exercise intolerance: A new phenotype of late-onset thymidine kinase 2 deficiency

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A B S T R A C T

A 29-year-old man developed, since the age of 18, exercise intolerance and exercise-induced rhabdomyolysis, with myoglobinuria. Muscle biopsy showed ragged-red fibers. Multiple mitochondrial DNA deletions were detected. The previously reported pathogenic homozygous mutation c.323C>T (p.Thr108Met) in *TK2* was identified.

This case expands the phenotypic spectrum of TK2 deficiency and indicates that it should be considered in the differential diagnosis of episodic rhabdomyolysis and exercise intolerance, along with other metabolic and mitochondrial myopathies. Since a new treatment is under development, it is essential improving knowledge of the natural history of TK2 deficiency.

1. Introduction

Thymidine kinase 2 (TK2) is a nuclear-encoded mitochondrial enzyme that phosphorylates the pyrimidine nucleosides thymidine (dT) and deoxycytidine (dC) to generate their nucleoside monophosphates. TK2 is essential in the deoxynucleoside triphosphate *salvage* synthesis pathway in quiescent cells, and its deficiency cause mitochondrial depletion / multiple deletion syndrome [1,2]. Recessive mutations in the *TK2* gene cause primarily a mitochondrial myopathy with a broad spectrum of age of onset and severity [3]: from extremely severe and rapidly progressive infantile-onset forms, with survival under two years, linked to mitochondrial DNA (mtDNA) depletion (MIM# 609560), to less severe forms with later onset and a slower rate of progression, associated to mtDNA multiple deletions. Late-onset patients, previously defined as patients where symptoms starts after the age of 12 [3], display a phenotype consisting of progressive proximal limb, axial, neck flexor and facial muscle weakness, frequently associated with ptosis, ophthalmoparesis and bulbar weakness, along with an early and severe respiratory involvement [4]. In some cases, chronic late-onset progressive external ophthalmoplegia is the main manifestation (MIM# 617069) [5].

We report a patient with TK2 deficiency presenting with recurrent rhabdomyolysis, myoglobinuria and exercise intolerance. Since a new treatment is under development [6] (ClinicalTrials.gov Identifier NCT03845712), it is essential improving knowledge of the natural history of TK2 deficiency.

2. Methods

All procedures were conducted as part of routine clinical care. The patient underwent clinical evaluation, muscle magnetic resonance imaging (MRI), muscle biopsy and genetic analysis by a customized next generation sequencing (NGS) panel. Patient's skeletal muscle mitochondrial DNA (mtDNA) deletions were investigated by Southern blot, and mtDNA copy number was assessed by quantitative polymerase chain reaction (PCR). Written informed consent was obtained for the genetic
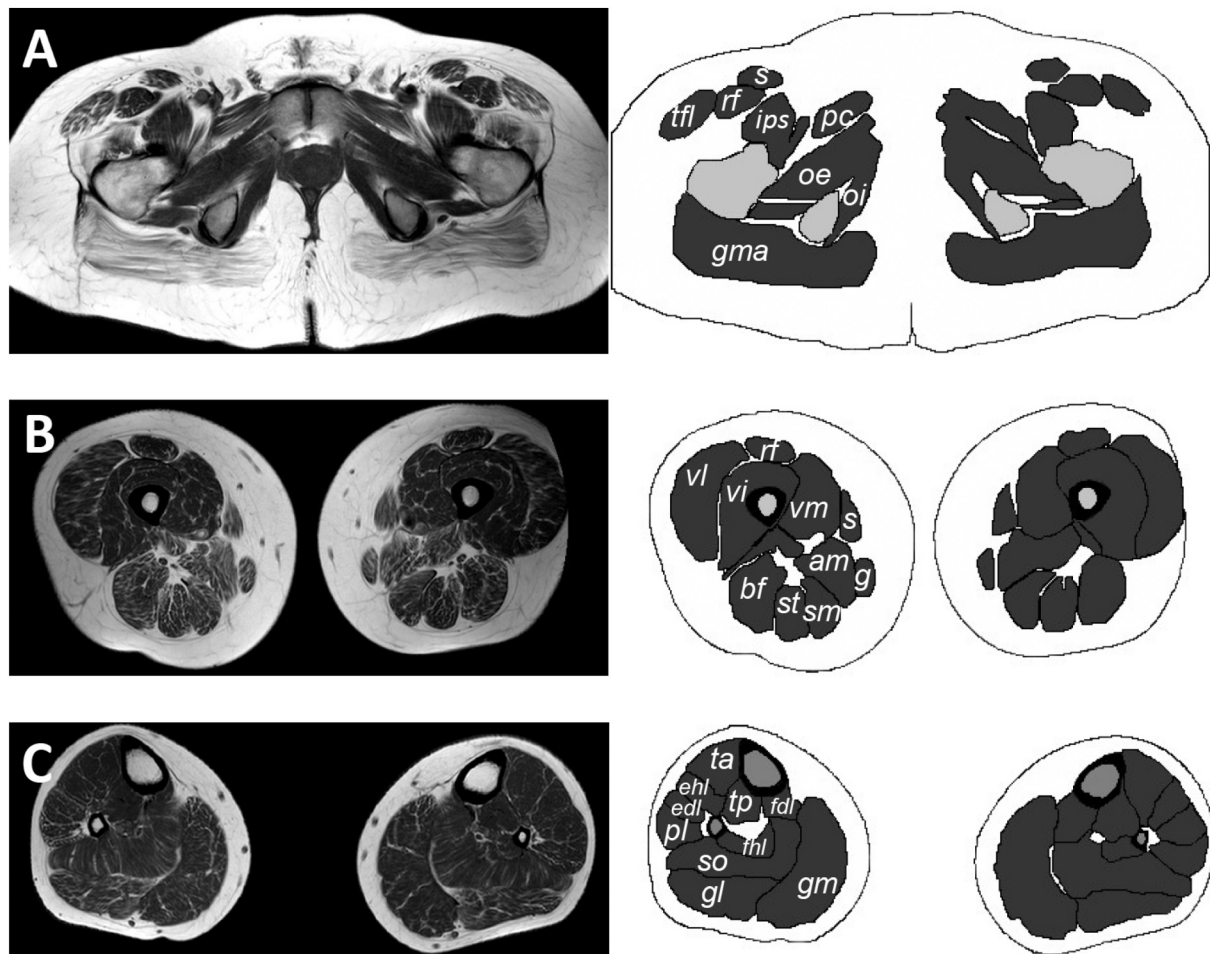
Fig. 1. T1-weighted MRI of lower limbs. A: pelvis. Symmetrical fat infiltration of gluteus maximus (MVS: 3), which is the most severely affected muscle. Bilateral involvement of tensor fascia latae is also observed (MVS: 2). B: thighs. Mild and diffuse fat muscle substitution (MVS: 1–2), which is predominant in posterior compartments. Sartorius is bilaterally affected by fat infiltration (MVS: 2). C: legs. Very mild and diffuse fat muscle substitution.
s: sartorius, rf: rectus femoris, tfl: tensor fascia latae, ips: iliopsoas, pc: pectinius, oe: obturator externus, oi:obturator internus, gma: gluteus maximus, vl: vastus lateralis, vi: vastus intermedius, vm: vastus medialis, g: gracilis, am: adductor magnus, st: semitendinosus, sm: semimembranosus, bf: biceps femoris, ta: tibialis anterior, ehl: extensor hallucis longus, edl: extensor digitorium longus, pl: peroneus longus, tp: tibialis posterior, fdl: flexor digitorium longus, fhl: flexor hallucis longus, so: soleus, gl: gastrocnemialis lateralis, gm: gastrocnemius medialis.

study. Anonymized data from this study will be shared by request from qualified investigators.

3. Results

The patient is a 29-year-old man born from non-consanguineous parents. He had a normal development and had no symptoms during infancy and childhood. HyperCKemia was detected for the first time when the patient was 16 years old. At the age of 18, he had the first exercise-induced rhabdomyolysis episode. Since then, he developed exercise intolerance characterized by disproportionate fatigue during physical activity of any intensity and recurrent episodes of rhabdomyolysis with myoglobinuria.

Although the patient did not complain of weakness, on examination he had mild bilateral facial weakness involving only orbicularis oculi, and muscle strength testing revealed mild bilateral weakness in hip extension and knee flexion (4+/5 on the Medical Research Council scale) in lower limbs, and in fingers extension (4/5) in upper limbs. The rest of the examination was normal. He had no ptosis, no ophthalmoparesis, and no bulbar, axial or neck flexion weakness. There was no muscle atrophy. Deep tendon reflexes were normal.

Basal serum creatine kinase levels were increased (615–931 U/l, normal range: 34–171 U/l). Spirometry, electrocardiogram and echocardiogram were normal. Lower-limb muscle Magnetic Resonance Imaging (Fig. 1) showed a severe and symmetrical fatty infiltration of gluteus maximus (Mercuri visual scale modified by Fisher, MVS: 3), mild and diffuse fat muscle substitution at thighs and legs (MVS: 1–2), which was predominant in posterior compartments, and affectation of sartorius (MVS: 2). Muscle biopsy (quadriceps) showed numerous COX-negative, ragged-red fibers and no dystrophic features. Mild variability in fiber size, and mild nuclear internalization were observed, as well as one isolated necrotic fiber. Respiratory chain complexes activities were normal. Muscle mtDNA analysis revealed multiple mtDNA deletions (Fig. 2) and decreased mtDNA/nuclear DNA copy-number ratio (48% respect to controls; mtDNA depletion cut-off < 30%). Serum levels of growth differentiation factor-15 (GDF-15) were 1503–2525 pg/ml (upper normal limit, 550 pg/ml). A previously reported pathogenic homozygous mutation c.323C>T (p.Thr108Met) in the *TK2* gene was identified by a NGS panel.

4. Discussion

This is the first description of a patient with TK2 deficiency presenting with recurrent rhabdomyolysis and exercise intolerance. They keep being the main manifestations after more than 10 years from the onset of symptoms. It corresponds, therefore, with a milder phenotype
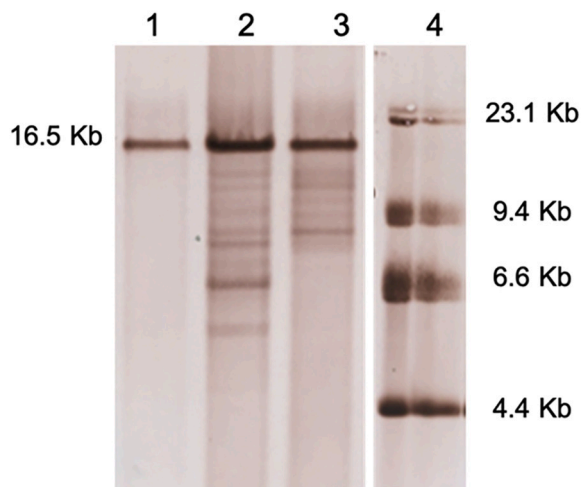
Fig. 2. Patient's skeletal muscle showing multiple mitochondrial DNA deletions. Southern blot analysis of muscle mitochondrial DNA to detect mtDNA reorganizations. Lane 1, control muscle DNA showing the normal 16.5 Kb size of mtDNA; Lane 2, TK2-patient displaying normal sized mtDNA and multiple mtDNA deletions; Lane 3, positive control for mtDNA deletions; Lane 4, molecular weight marker.

than those previously described. GDF-15 is a biomarker which was shown to correlate with severity of TK2 deficiency. It is elevated on average 30-fold in children and 6-fold in adults [7]. The degree of increase in GDF-15 levels in our patient (3-5-fold) is also consistent with a milder phenotype, and supports the idea of the prognostic value of GDF-15 levels in TK2 deficiency. Since initial manifestations in our patient were earlier than in patients with more severe symptoms, the age of onset is not enough to establish, by itself, the prognosis. On lower-limb muscle MRI, gluteus maximus was the most severely affected muscle, and sartorius, a muscle which is usually spared in many genetic muscle diseases (even in late stages), was also affected. Interestingly, these MRI fits with a pattern previously described in late-onset TK2 patients [4].

In conclusion, this case expands the phenotypic spectrum of TK2 deficiency, which should be considered in the differential diagnosis of episodic rhabdomyolysis and exercise intolerance, along with other metabolic and mitochondrial myopathies, and it suggests that the muscle MRI could help to guide the differential diagnosis in a patient with a metabolic myopathy.

Author statement

CPFF, CDG, MAM were responsable for the conception and design of this report. CPFF, GM, CJM, CB, AH and ABE acquired and analyzed data. CPFF, MAM and CDG drafted the manuscript and figures.

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