Clinical Neurology and Neurosurgery 150 (2016) 41–45

Contents lists available at ScienceDirect

Clinical Neurology and Neurosurgery

journal homepage: www.elsevier.com/locate/clineuro

Limb-girdle congenital myasthenic syndrome in a Chinese family with novel mutations in *MUSK* gene and literature review

Xinghua Luan [1], Wotu Tian [1], Li Cao *

Department of Neurology and Institute of Neurology, Ruijin Hospital affiliated to Shanghai JiaoTong University School of Medicine, Shanghai, 200025, China

ARTICLE INFO

Article history:
Received 12 June 2016
Received in revised form 4 August 2016
Accepted 21 August 2016
Available online 22 August 2016

Keywords:
Congenital myasthenic syndrome
Neuromuscular transmission
MUSK gene
Limb-girdle CMS
Missense mutations

ABSTRACT

Objectives: To describe the clinical and genetic features of a Chinese congenital myasthenic syndromes (CMS) patient with two novel missense mutations in muscle specific receptor tyrosine kinase (*MUSK*) gene and review 15 *MUSK*-related CMS patients from 8 countries.
Methods: The patient was a 30-year-old man with chronic progressively proximal limb weakness for 22 years and diagnosed as muscular dystrophy before. Serum creatine kinase (CK) was normal. Repetitive nerve stimulation (RNS) test showed decrements at low rate stimulation. Weakness became worse after conventional doses of pyridostigmine. Mild multiple atrophy of thigh and leg muscle was observed in MRI. Open muscle biopsy and genetic analysis were performed. One hundred healthy individuals were set for control.
Results: Muscle biopsy showed mild variation in fiber size. Two missense mutations in *MUSK* gene (p.P650T and p.I795S) were identified in the patient. The mutation of p.I795S was identified in his father and p.P650T in his mother. Both of them were not detected among the healthy controls and predicted to be damaging or disease causing by prediction tools.
Conclusion: In this study, we identified a limb-girdle CMS (LG-CMS) patient carrying two novel heterozygous missense mutations in *MUSK* gene. CMS related genes should be analyzed in patients with limb-girdle weakness, normal CK, decrement of CMAP at RNS and mild change in muscle biopsy or MRI.

1. Introduction

The congenital myasthenic syndromes (CMS) are a diverse group of genetic disorders caused by abnormal signal transmission at the motor endplate by one or more specific mechanisms resulting in fatigable weakness [1]. Initially, CMS were classified according to the location of the mutant protein as presynaptic, synaptic basal lamina-associated and post synaptic. The prominent features of this pathological condition are exercise-induced weakness and fatigability, usually presenting from birth to childhood. To date, twenty three CMS disease genes have been identified [2–5]. The most common cause of CMS is the defects in the acetylcholine receptor. The second most common cause is mutations that affect endplate development and maintenance.

Muscle associated receptor tyrosine kinase (MuSK) is a single-pass transmembrane protein that plays a critical role in signaling between motor neurons and skeletal muscle [1]. Mutations that impair MuSK kinase activity or the signal pathway cause congenital myasthenia as a result of structurally and functionally defective synapses [6]. Structural studies in human beings and animal models show general remodeling of the endplates as a result of denervation and reinnervation [7]. MuSK deficiency affects the proximal limb, ocular, facial muscles, and in some kinships the bulbar and respiratory muscles as well [8]. In the past decade, seven unrelated families with a recessive CMS caused by *MUSK* gene mutations have been described, characterized by limb-girdle weakness, eyelid ptosis, or respiratory distress presenting at birth or early life.

Here, we describe a Chinese CMS patient with novel mutations in exon 15 of *MUSK*, characterized by limb-girdle weakness without ptosis, external ophthalmoplegia, facial, bulbar or respiratory involvement.

2. Materials and methods

2.1. Patient

We identified a Han Chinese patient fulfilled the generic diagnosis of a CMS on the basis of fatigable weakness and a decremental

* Corresponding author.
E-mail address: caoli2000@yeah.net (L. Cao).
[1] These authors contributed equally to this work.

http://dx.doi.org/10.1016/j.clineuro.2016.08.021
0303-8467/

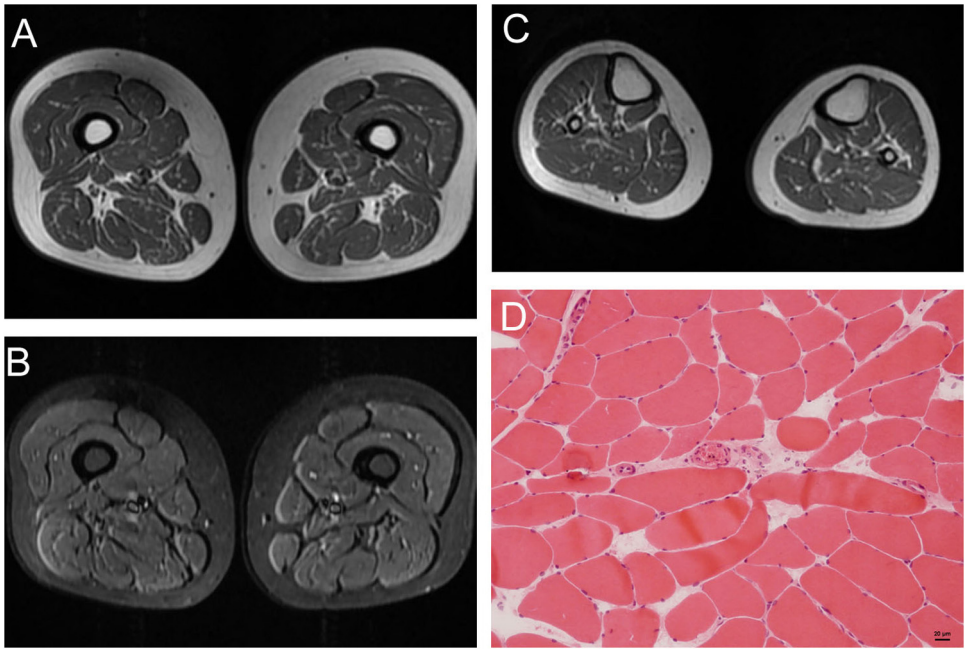

Fig. 1. Mild change in muscle MRI and biopsy.
Axial T1-weighted MRof the legs showed minimal fatty infiltration and atrophy in most of thigh muscle (A) and soleus (C). There was no evidence of edema or inflammation in short-tau inversion recovery (STIR) image (B). Muscle biopsy (D) of the biceps showed mild variation in fiber size with scattered atrophic fibres (HE, bar = 20 μm).

EMG response [1]. After an informed consent, the patient and his parents were clinically examined.

2.2. Muscle biopsy

An open muscle biopsy was performed to obtain muscle tissue from the left biceps brachii in the patient. The tissue was frozen and then cut at 8-μm sections. These sections were stained according to standard histological and enzyme histochemical procedures with hematoxylin and eosin (HE), modified Gomori Trichrome, periodic acidic Schiff, oil red O, nicotinamide adenine dinucleotide tetrazolium reductase, succinate dehydrogenease, cytochrome C oxidase, and esterase.

2.3. Mutation analysis

Blood samples were collected from the patient and his parents after obtaining informed consent. Unaffected individuals (n = 100) of matched geographic ancestry were also included as healthy controls. The protocols were all approved by the Ruijin Hospital Ethics Committee, Shanghai Jiao Tong University School of Medicine. Genomic DNA was extracted from whole blood using the standardized phenol-chloroform extraction method. CMS panel was performed on the patient and his parents.

3. Results

3.1. Clinical findings

This Chinese non-consanguineous family comprised of one affected individual and the healthy parents. He was a 30-year-old man, born of full term spontaneous vaginal delivery with unremarkable neonatal problems. His age at achieving sitting and standing alone was 8 months and 12 months respectively. He started to walk at 14 months. Though the patient did not run as fast as the peers before 8 years old, he had no trouble doing normal daily activities. Initial signs of muscle weakness were first noted at the age of 8, demonstrated by a difficulty in climbing stairs. Over the next few years his muscle weakness progressed slowly to be proximal upper limbs. Once at the age of 11, he was unable to stand up after riding a bike for 6–7 km. When he was 12, he became unable to run. There was clear fatigability with more pronounced weakness after exercise. Upon evaluation, no evidence was discovered about impaired extra ocular movements, mastication, dysphagia, dysarthria, or other cranial muscle involvement. It was found that he had weakness in neck flexion (4/5 on a medical research council scale graded 0–5), reduced strength in the proximal muscle of all extremities (3/5), and distal muscles were almost normal (5/5). Muscle tone in four limbs was normal. His quantitative myasthenia gravis score (QMG) was 13 [9]. There was neither obvious atrophy of proximal upper and lower limb muscles nor hypertrophy of calf muscles. Deep tendon reflexes were normal in all limbs. The patient walked slowly with a duck like walk. Respiration involvement and cognitive decline were not noted. CK was normal. An electromyogram showed motor unit potential (MUP) spontaneous activity with small, short and multi-phase in tibialis anterior muscle, gastrocnemius, and biceps. Significant decrements on low (3–5 Hz) rates of repetitive nerve stimulation (RNS) of a bilateral deltoid at baseline were noticed. Nerve conduction studies (NCS) were normal. MRI of the lower limbs showed mild atrophy in most of the thigh muscle and soleus (Fig. 1A–C). Muscle biopsy was applied in a left biceps, showing mild variation in fiber size (Fig. 1D). Oral pyridostigmine (30 mg three times daily) led to a worsening of the symptoms and was stopped after 20 days of treatment. Then he was treated with salbutamol (2 mg three times daily) showing mild improvement of limb weakness.

3.2. Genetic findings

The patient was identified to have compound heterozygous mutations c.1948C > A (p.P650T), c.2384T > G (p.I795S) and c.2573G > A (p.R858H) in *MUSK* gene, of which p.P650T and p.R858H were inherited from his mother and p.I795S from his father (Fig. 2). However, c.2573G > A (p.R858H) has been reported in the 1000 Genome Project (http://browser.1000genomes.org) as SNP rs34115159. The other two variants were not found in 100 healthy