Biochemical and Biophysical Research Communications 457 (2015) 262–266

Contents lists available at ScienceDirect

Biochemical and Biophysical Research Communications

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

A rare mutation in MYH7 gene occurs with overlapping phenotype

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ARTICLE INFO

Article history:
Received 16 December 2014
Available online 7 January 2015

Keywords:
MYH7
Left ventricular non-compation
Cardiomyopathy
FTD
Laing Distal Myopathy
Overlapping syndrome

ABSTRACT

Mutations in the beta-myosin heavy chain gene (MYH7) cause different muscle disorders. The specific molecular pathobiological processes that cause these different phenotypes remains unexplained. We describe three members of a family with an autosomal dominant mutation in the distal rod of MYH7 [c.5401G> A (p.Glu1801Lys)] displaying a complex phenotype characterized by Laing Distal Myopathy like phenotype, left ventricular non compaction cardiomyopathy and Fiber Type Disproportion picture at muscle biopsy. We suggest that this overlapping presentation confirm the phenotypic variability of *MYH7* myopathy and may be helpful to improve the genotype phenotype correlation.

1. Introduction

Myosin 7 gene (*MYH7*) encodes slow/beta-cardiac myosin heavy chain (MHC-β), a class II myosin expressed primarily in the heart, but also in skeletal muscles and in particular in type I fibers [1]. *MYH7* gene mutations are known to cause three different muscle disorders: Laing Distal Myopathy (LDM), Myosin Storage Myopathy (MSM) and Familial Hypertrophic Cardiomyopathy (FHCM) [2–4]. Clinical phenotypes have been related to the site of mutations; in fact, mutations in the globular head of the protein have been linked to FHCM, mutations in distal rod to MSM, whereas mutations in middle or proximal rod cause LDM [5].

Very recently mutations in distal rod of protein have been associated with a form of Congenital Myopathy with Fiber Type Disproportion (CFTD) [6]. In CFTD cases, the majority of patients have congenital onset and present a LDM phenotype or MSM phenotype without specific cardiac impairment [7,8]. In addition, several families with mutations in the *MYH7 gene* have been described presenting exclusively a heart impairment, consisting in ventricular non compaction (LVNC). LVNC is detected by an echocardiographic examination: the apical region of the heart shows wide trabeculae of the muscular tissue, separated by lacunae. This abnormality leads to contractile dysfunction, as well as arrhythmias and embolic complications, due to the blood stasis within the lacunae [9].

The specific molecular pathobiological processes that cause these different phenotypes remains unexplained [10].

We describe three members of an Italian family with a mutation of the *MYH7* gene c.5401G> A (p.Glu1801Lys) in the distal rod of the protein. This mutation is for the first time described in an italian family whose clinical presentation includes non compaction cardiomyopathy, LDM like phenotype and FTD picture at muscle biopsy. We speculate that the complex phenotype of the family confirms the wide phenotypic variability of MYH7 mutations,

Abbreviations: ACTA1, actin 1; CPK, creatine phosphokinase; CFTD, Congenital Fiber Type Disproportion; COX, cytochrome C oxidase; *DES*, desmin; EMG, electromyography; FHCM, Familial Hypertrophic Cardiomyopathy; IF, immunofluorescence; *ITGA7*, integrin 7; LDM, Laing Distal Myopathy; *LMNA*, lamin; LVNC, left ventricular non compaction; MHC-β, slow/beta-cardiac myosin heavy chain; MSM, Myosin Storage Myopathy; *MYH7*, *Myosin 7* gene; NADH, nicotinamide adenine dinucleotide dehydrogenase; PM, pacemaker; SDH, succinate dehydrogenase.

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http://dx.doi.org/10.1016/j.bbrc.2014.12.098
0006-291X/

which can not be fully explained by mutation sites, and may be helpful to improve the genotype phenotype correlation.

2. Materials and methods

We investigated three members of a family from southern Italy (Fig. 1). The evaluation of patients included neurological examination, blood tests, electrophysiological study, cardiac assessment, muscle biopsy and molecular analysis.

2.1. Cardiac evaluation

Patients underwent a comprehensive cardiac evaluation, including physical examination, search for cardiac symptoms (such as fatigue, dyspnoe and exercise intolerance), a standard 12-lead electrocardiogram and a bidimensional echocardiogram with speckle-tracking analysis of longitudinal myocardial contractile function, as well as a real-time 3D echocardiography. This technique is the only one able to measure the contractile function of the three different layers of myocardium: subendocardial (value of longitudinal strain), midwall (circumferential strain) and subepicardial fibers (radial strain).

2.2. Muscle biopsy

Two patients (case II2 and case III1) had an open biopsy of biceps brachii muscle and specimens were frozen in isopentane cooled in liquid nitrogen. Unfixed cryostat sections (10 μm) were stained with a panel of histological, histochemical and immunohistological techniques according to standard procedures [11].

2.3. Molecular analyses

Genomic DNA was extracted by standard methods from peripheral blood lymphocytes. For mutation screening, primers flanking the intron-exon junctions of each *MYH7* exon, the 5′ and 3′ UTR, were designed for the genes *MYH7* based on published sequences (GenBank accession number: *MYH7* NM_000257.3). PCRs were performed with Mega Mix Double (Microzone, Haywards Heath, West Sussex, UK). The products were purified using microCLEAN (Microzone, Haywards Heath, West Sussex, UK) and sequenced directly with the BigDye Terminator v1.1 Cycle Sequencing Kit (Applied Biosystems, CA, USA). Sequences were analyzed on ABI Prism 3100 Genetic Analyzer (Applied Biosystems, CA, USA).

3. Results

Case 1 (II2): is a 65 year old man, who reported, at the age of 25 years, sudden onset of symptoms with shortness of breath during exertion and orthopnea. A subsequent echocardiographic assessment revealed the presence of a cardiomyopathy, characterized by incomplete compaction of myocardium (Fig. 2) for which the patient started a specific pharmacological treatment. At the age of 60 years, during the routine cardiac follow up, heart rhythm abnormalities were detected and the patient underwent a pacemaker (PM) implantation. Neurological deficits had appeared in the third decade with waddling gait and foot steppage. Over the years, the clinical picture has been slowly progressive and currently the patient requires a support to climb the stairs and to ambulate. Last neurological evaluation showed waddling gait, foot drop (right > left), inability to stand on the heels and tips, hyperlordosis and positive Gowers manouver. In addition, chest muscles, neck flexor, triceps and biceps muscles showed a moderate hypotrophy and weakness, which were marked for the muscles of the shoulder girdle. At lower limbs weakness and hypotrophy were severe in distal muscles and moderate in proximal muscles (quadriceps, iliopsoas and hamstring muscles). Tendon reflex were absent. Creatine phosphokinase (CPK) was increased to 400–500 UI/L [normal value 180 UI/L], electromyography (EMG) showed myopathic changes while the nerve conduction study was within normal limits. For the presence of PM the muscle MRI was not carried out.

A muscle biopsy, performed at the age of 43 years, revealed two separate populations of fibers: hypotrophic and rounded type 1 fibers and normal type 2 fibers. Type 1 fibers were clearly more numerous than type 2 (nearly 80% of all fibers) and their average diameter was 33.8 +/−9.1 μ, while the average diameter of type 2 fibers was 62.5 +/−13.3 μ. Mild increase of nuclear centralizations and splitting phenomena were also found (Fig. 3). COX, SDH, NADH stainings were normal and no accumulation of glycogen and lipids was observed. Immunofluorescence (IF) showed normal expression of dystrophin, sarcoglycans, caveolin-3, dysferlin and merosin. Since the histological findings were consistent with a FTD,

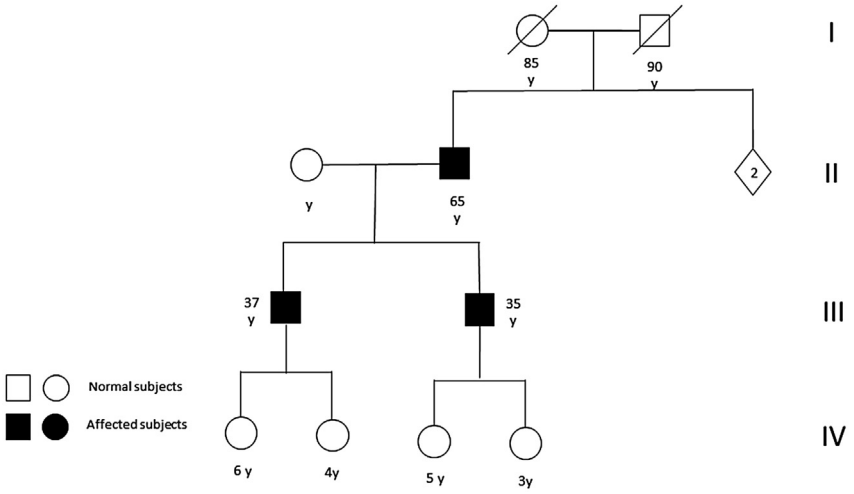

Fig. 1. Family tree Complete pedigree of family with heterozygous mutation p.Glu 1801Lys in *MHY 7 gene*. Squares represent males; circles represent females; blank symbols represent normal subjects; black filled symbols represent affected subjects.