G56S and Arg143Ser mutations in CAV3 and FKRP gene respectively contribute to the expression of phenotypic characteristics of a LGMD patient of South Italy

P. Spadafora,[1] A. Qualtieri [1], N. Romeo [1], A. Bagalà [1], M. De Bartolo[2]

[1] Institute of Neurological Sciences - National Research Council, Cosenza, Italy

[2] Neurofisiopatologia, Distretto Jonio- sud, ASP Cosenza

Introduction

Limb Girdle Muscular Dystrophy (LGMDs) are a heterogeneous group of genetic disorders in which mutations in the same gene display autosomal dominant or recessive inheritance. Moreover, variants can act as a modifier of either a dominant or a recessive mutation[1]. LGMDs are clinically characterized by progressive muscle weakness and atrophy with predominant involvement of scapular and pelvic girdle. Phenotypic variability has been reported even in the same family. Mutations in the human CAV3 gene, located on chromosome 3p25, have been implicated in LGMD1C, Rippling Muscular Dystrophy, hyperCKemia, distal myopathy and cardiomyopathy. Caveolin-3 is a small molecular-weight protein composed of 150 amino acids localized into the sarcolemma. It is the principal component of caveolae, vesicular invaginations of the plasma membrane that regulate endocytosis, cholesterol homeostasis and signal transduction. Pathogenic mutations in FKRP gene, located on chromosome 19q13, were reported in severe forms of Congenital Muscular Dystrophy, in Muscle-Eye-Brain disease, Walker-Warburg Syndrome and LGMD2I[2]. Fukutin Related Protein is a protein involved in the glycosylation of cell surface molecules.

We report a patient from south Italy presenting mild muscle weakness in the first decade, hyperCKemia and dilated cardiomyopathy at age 20.

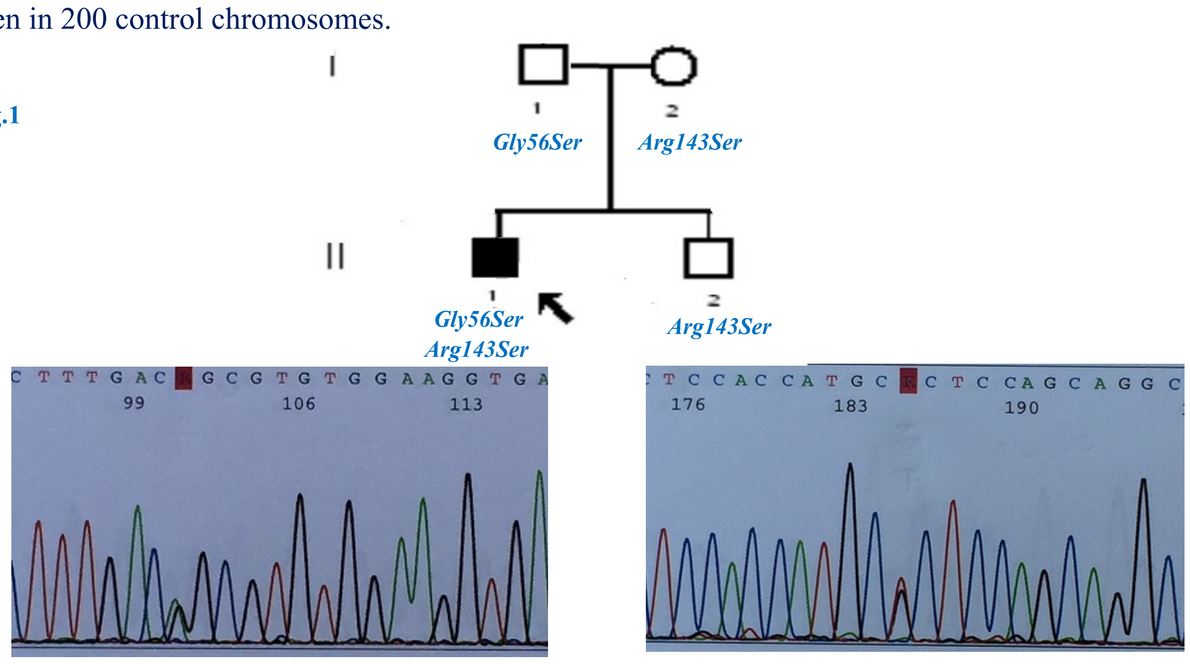
Subjects and Methods

The proband (patient II:1, Fig.1) was a patient from south Italy who at age 20 developed asthenia, proximal and symmetrical muscle weakness in both the upper and the lower limbs, myalgia after muscular efforts. Following hospitalization for chest pain it was identified hyperCKemia (1000U/L). A year later he was diagnosed dilatated cardiomyopathy. Clinical investigation revealed no muscle wasting nor hypertrophy of the calf. Electromyographic exam showed signs of muscle suffering from mild to moderate in all territories explored. Muscle biopsy conducted on the left biceps showed a mild muscular pain of possible primitive nature; immunohistochemical staining for dystrophin, α, β sarcoglycan and CAV3 did not show significant abnormalities. The patient had no cognitive impairment (MMSE 29/30). The Hamilton scale for depression was normal. The other family members were healthy. No consanguinity in the family was reported. All subjects gave informed consent for the molecular tests. Genomic DNA was extracted from peripheral blood leukocytes of the patient and his parents. The two exons and flanking intronic boundaries of CAV3 gene and the entire 1,5kb coding region of FRKP gene (using sets of overlapping primers) were amplified by Polymerase Chain Reaction (PCR). The PCR products were analysed by direct sequencing on ABI 3130XL Genetic Analyzer (Applied Biosystems, Foster City, USA).

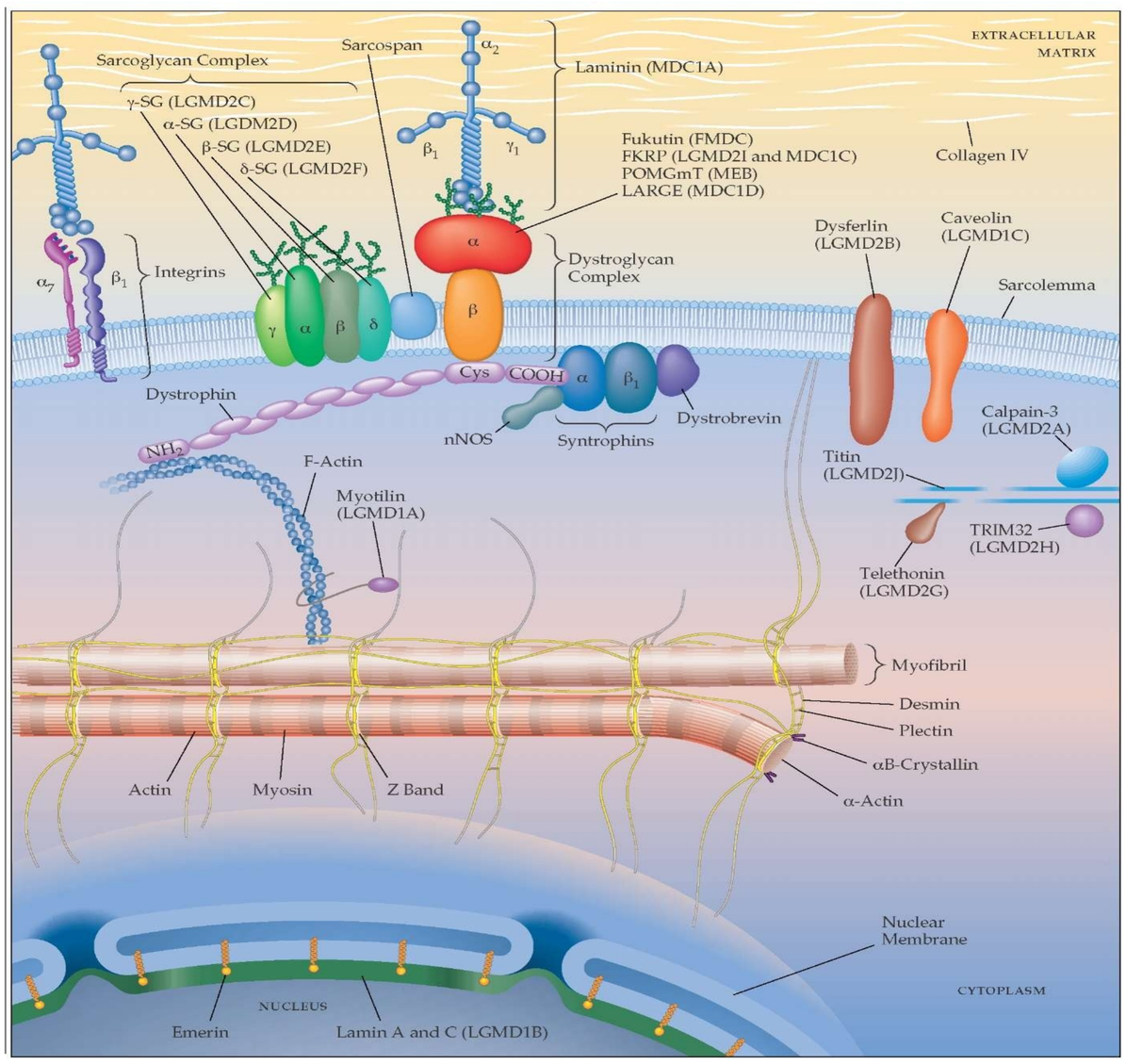
Results

In the proband (II-1), the sequencing analysis allowed the identification of two mutations Gly56Ser (c.166G>A) and Arg143Ser (c.427C>A) in CAV3 and FKRP genes respectively in heterozygosity. Proband's father (I-1) and mother (I-2) were heterozygous for Gly56Ser and Arg143Ser alleles, respectively; his brother (II-2) showed Arg143Ser mutation in one allele. These changes were not seen in 200 control chromosomes.

Fig.1



CAV3 gene, c.166G>A (Gly56Ser) *FKRP gene, c.427C>A (Arg143Ser)*



Discussion and Conclusions

Previous studies have reported that the Gly56Ser mutation is unlikely to cause muscular dystrophy when present in one allele though the amino acid change map to a functional important domain in caveolin3 implicated directly in inhibiting NOS activity. This mutation in homozygosity was identified in a patient with proximal muscle weakness developed in the first decade. Other members of the family (mother and two siblings) carrying the same change in heterozygosity did not show symptoms of muscular dystrophy[3]. Gly56Ser was identified, also, in a patient with hyperCKemia[4]. The possibility that it can act as recessive mutation or interact with other genes involved in dystrophic process was suggested[1]. Moreover, other Authors suggested that Gly56Ser could represent a relatively black specific genetic variant.

Asymptomatic carriers of homozygous FKRP mutations and manifesting carriers seem to be common among patients with LGMD2I[2]. Arg143Ser mutation has been identified in six Italian patients with clinical presentation consistent of LGMD2I. Only in one patient was identified the second mutated allele. Arg143Ser mutation was validate in 200 control chromosomes[5]. This mutation involves an evolutionary conserved residue and the loss of a positively charged amino acid in the mutated protein could have a dramatic effect on protein function.

We report a patient from south Italy presenting mild muscle weakness in the first decade, hyperCKemia and dilated cardiomyopathy at age 20. Molecular analysis of the CAV3 gene revealed the Gly56Ser mutation in compound heterozygosity with the Arg143Ser mutation in FKRP gene. Interesting, his brother and parents carrying one of these mutations did not show symptoms of muscular involvement or hyperCKemia. This finding suggests the possibility that Gly56Ser and Arg143Ser can act as recessive mutations and that both genetic alterations contribute in determining the characteristics of phenotype.

REFERENCES

1) de Paula F, Vainzof M, Bernardino ALF, McNally E, Kunkel LM, Zatz M. Mutations in the Caveolin-3 gene: When are they pathogenic? American Journal of Medical Genetics. (2001) 99: 303-307.

2) Schottlaender LV, Petzold A, Wood N, Houlden H. Diagnostic clues and manifesting carriers in fukutin-related protein (FKRP) limb-girdle muscular dystrophy. Journal of the Neurological Sciences. (2015) 348: 266-268.

3) McNally EM**,** de Sa Moreira E, Duggan DJ, Bonnemann CG, Lisanti MP, Lidov HGW, Vainzof M, Passos Bueno MR et al. Caveolin-3 in muscular dystrophy. Human Molecular Genetics. (1998) 7 (5): 871-877.

4) Reijneveld JC, Ginjaar IB, Frankhuizen WS, and Nothermans NC. CAV3 gene mutation analysis in patients with idiopathic hyper-CK-emia. Muscle & Nerve (2006) 34: 656-658.

5)Boito CA, Melacini P, Vianello A, Prandini P, Gavassini BF, Bagattin A, Siciliano G, Angelini C, Pegoraro E. Clinical and Molecular Characterization of patients with Limb-Girdle Muscular Dystrophy Type 2I. JAMA Neurology. (2005) 62 (12): 1-7.

patrizia.spadafora@cnr.it

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