Limb Girdle Muscular Dystrophy Type 2a: Case Report

Limb Girdle Muskuler Distrofi Tip 2a: Vaka Sunumu

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ABSTRACT

Limb Girdle Muscular Disease (LGMD) comprise a group of inherited muscular dystrophy with chronic progressive weakness of hip and shoulder girdles. The inheritance pattern is either autosomal dominant (LGMD1) or autosomal recessive (LGMD2). LGMD 2A is known as calpainopathy in which there was a defect of gene encoding the protein named as calpain. There are three calpainopathy phenotypes according to distribution of muscle weakness and age at onset. In this report, we presented an asymptomatic child with persistant hyper CKemia diagnosed with muscle biopsy and genetic testing. Genetical examination results of the patient showed homozygote mutation of *CAPN3* gene(c.2092C>A) and parents revealed that they were heterozygous unaffected carriers.

Key Words: Biopsy, Calpainopathy, Muscle, Muscular dystrophy, Limb girdle muscular distrophy

ÖZ

Limb Girdle Muskular distrofi (LGMD) kalça ve omuz ekleminde progresif kronik güçsüzlük ile seyreden kalıtımsal bir grup kas distrofisidir. Kalıtım paterni otozomal dominant (LGMD1) ve otozomal resesif (LGMD2) olarak ikiye ayrılır. LGMD tip 2A kalpain proteinini kodlayan gen defektine neden olan kalpainopati olarak bilinmektedir. Kas güçsüzlüğü ve başlangıç yaşına göre 3 çeşit kalpainopati tipi vardır. Bu yazıda asemptomatik persistan kreatin kinaz yüksekliği olan, kas biyopsisi ve genetik analiz ile LGMD tip 2A tanısı alan çocuk hasta sunulmaktadır. Genetik analiz sonucunda *CAPN3* geninde (c.2092C>A) homozigot mutasyonu mevcut olup ebeveynlerin genetik analizinde heterozigot taşıyıcı olarak saptanmıştır.

Anahtar Kelimeler: Biyopsi, Kalpainopati, Kas, Muskular distrofi, Limb girdle muskular distrofi

INTRODUCTION

Muscular dystrophies (MD) are a heterogenous group of disease as a result of defects in genes for normal function of skeletal muscle. Clinical manifestations of MD can range from asymptomatic cases with increase of creatinine kinase (CK) to severe debilitation and death early in life. Autosomal recessive forms of MD produce symptoms early in life and are severe whereas autosomal dominant forms have slower and less debilitating courses (1). Limb-girdle muscular disease (LGMD) comprise a group of inherited muscular dystrophy with chronic progressive weakness of hip and shoulder girdles. The inheritance pattern is either autosomal dominant (LGMD1) or autosomal recessive (LGMD2). There are 30 subtypes of LGMD and the number of disease was given according to the chronology of identification of their genetic loci. The recessive forms also can be divided at molecular level as sarcoglycanopathies and non-sarcoglycanopathies based on the affected mutation in a gene encoding sarcoglycan component of the dystrophin associated complex or not. LGMD 2A is in the group of non-sarcoglycanopathy and known as calpainopathy (2). There is the defect of gene encoding the protein named as calpain and has autosomal recessive inheritance pattern in LGMD 2A.

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Most common mutation is seen on *CAPN3* gene (15q15.1 loci) which encodes calcium sensing proteaz calpain 3 having role in muscle remodelling. Nowadays there are more than 450 pathogenic varient of *CAPN3* protein. There are three calpainopathy phenotypes according to distribution of muscle weakness and age at onset. The most common phenotype is pelvifemoral phenotype in which pelvic involvement occurs firstly. The onset age of this phenotype can be as early as before 12 years old or as late as 30 years old. The second phenotype is scapulohumeral phenotype that is milder form which is infrequent at early ages and shoulder involvement is seen firstly. Third phenotype is hyperCKemia which occurs in chidren and young ages without any symptom (3).

Here we report a case with third phenotype of LGMD and genetically diagnosed as type LGMD 2A to emphasize asymptomatic presentation of LGMD.

CASE REPORT

Twelve years-old female was admitted to pediatric clinic for primary care. She had no complaint of any muscle weakness. She had no sign and symptom of any infection and she had no medication. Her medical history was unremarkable and she had history of long time cycling. There was consanguinity among parents. Her cousin has walking disability with unknown diagnosis. Pedigree chart of family was at Figure 1. On physical examination weight and height percentiles were within 10-25 percentiles and system examination was normal. Muscle strength was 5/5, deep tendon reflexes were normal and she had normal gait examination. Hip and shoulder joint examinations were normal with normal muscle apperance. She was found to have alanine transferase (ALT) of 108 U/L (normal:0-34U/L) and aspartate transferase (AST) of 148 U/L (normal:15-60U/L). Bilirubin, alkalen phosphatase, PT/PTT and albumin levels were all normal. Hepatitis A, B and C screenings were unremarkable. Creatine kinase level was 7186 U/L(normal:<145U/L). We excluded extrahepatic casuses of elevated transaminases (1). She had normal thyroid function, renal function, celiac disease tests and fasting blood glucose. She had no hemolytic disease, adrenal insuffciency and rhabdomyolysis. As she had persistent CK increase, muscle biopsy was done. At muscle biopsy, variation in diameter of muscle fibers, some muscle fibers with internal nucleus, some degenerated muscle fibers and increase of lipid content in some muscle fibers were detected. There were rarely eosinophiles in interstitium but no increase of connective tissue (Figure 2: A-C). Immunohistochemical examination showed merosin, dystrophin, α, β, γ, δ and sarcoglycan antibodies positive. Although dystrophic or inflammatory changes not seen, eosinophilic infiltration can be seen in calpainopathy, genetic study of *CAPN3* gene was done. Genetic study results showed homozygote mutation of *CAPN3* gene (NM_000070.2, c.2092C>A, p. Arg698Ser). Subsequent testing of the patient's parents revealed that they were heterozygous unaffected carriers. Written informed consent form was obtained from the parents of patient.

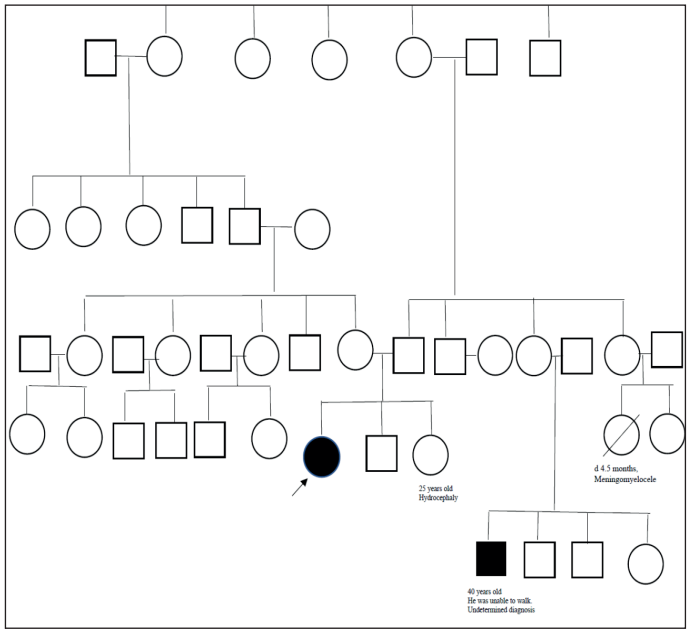

Figure 1: Pedigree chart of family.

DISCUSSION

We presented the asymptomatic child with LGMD type 2A confirmed by genetic study to emphasize asymptomatic presentation of muscle diseases. She was diagnosed when she was admitted to hospital for primary care. After detection of elevated transaminase levels and persistent high CK levels, we performed muscle biopsy and genetic study. Although there is no standard of care, medical therapy for LGMDs and no specific biomarkers for diagnosis other than genetic study, early diagnosis of patients may increase their life quality with supportive therapies. Extensive efforts to develop gene therapies for muscular dystrophies, including LGMD type 2A also increase the importance of early diagnosis.

Our case had no symptom and normal physical examination, patients with LGMD type 2A have heterogenous clinical presentations. Clinically the symptoms of patients with LGMD type 2A are difficulty to run and walk, toe walking, hyperlordosis, proximal muscle weakness (pelvic extensor ve adductor muscles are normal), wing scapula. Muscle atrophy is prominent. Joint contracture, shortness of aschille tendon, scoliosis at presentation can be seen at early period. The onset age can change between 2-40 years old, but the mean onset age of disease is 8-15 years old. Mostly mild and moderate clinic is seen at the beginning. The patient has inability to walk and adherence to wheelchair after 11-28 years of disease. Serum CK levels can increase 5-80 of normal level or can be

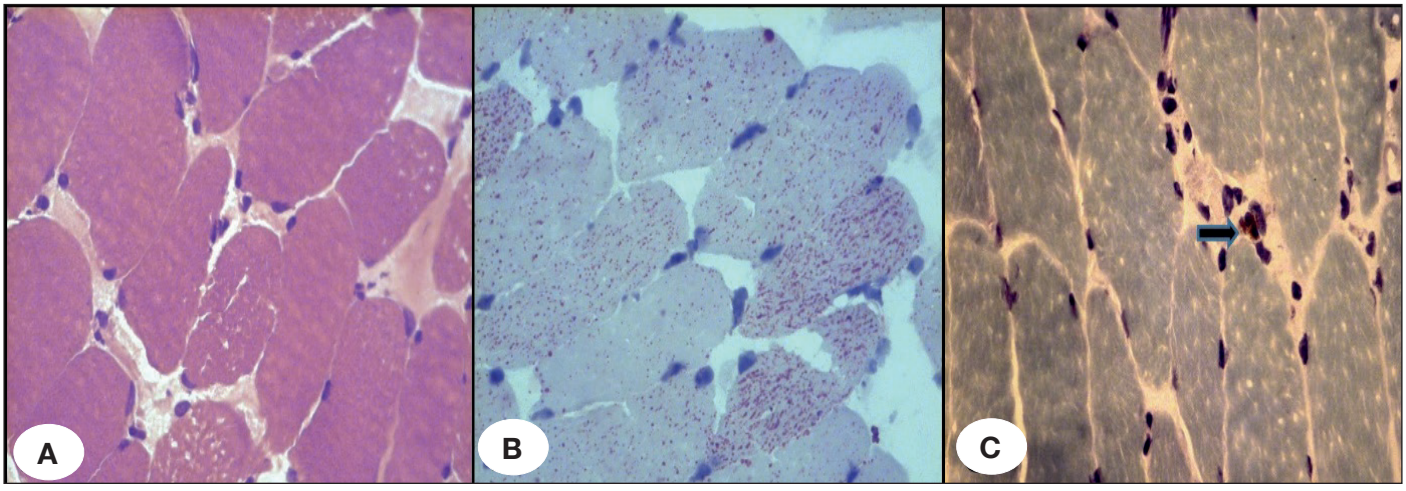

Figure 2: (A) At muscle biopsy, H&E staining, variation in diameters of muscle fibers and some degenarated muscle fibers, **(B)** ORO staining, increase of lipid content in some muscle fibers, **(C)** toluidin staining, eosinophil signed with arrow.

at normal range (4). Muscle biopsy of LGMD type 2A shows mostly a dystrophic pattern like increased fiber size variability, increased fibrosis, regenerating fibers, degenerating and necrotic fibers. Sometimes nonspecific myopathic features like increased central nuclei, fiber splitting, lobulated fibers (misaligned myofibrils that form a lobulated pattern), and type 1 fiber predominance can be seen (5). At diagnoses of MD, biochemical tests help to show deficiency or absence of specific proteins at biopsy material (6). Muscle biopsy should be used when molecular genetic analysis is not conclusive or for diagnostic confirmation. Calpain-3 immunoblot testing is useful in the diagnosis of calpainopathy. Molecular genetic testing of identification of biallelic pathogenic variants in CAPN3 gene or a dominantly acting heterogenic pathological variant for *CAPN3* 21-bp deletion is the gold standard for diagnoses of calpainopathy (3). In our patient we firstly performed muscle biopsy to understand muscle pathology and after result of muscle biopsy genetic testing confirmed diagnosis of LGMD type 2A. In a report of *CAPN3* mutation in Turkey, a total of 15 different *CAPN3* gene mutations were detected, 6 of which were novel (p.K211N, p.D230G, p.Y322H, p.R698S, p.Q738X, c.2257delGinsAA. In this report all LGMD2A-ascertained patients, the onset of symptoms was between 2.5 and 19 years of age. In general, the disease course was mild. Involvement of muscles predominated in the limb girdle and trunk muscles and was usually symmetrical. There was atrophy of the proximal muscles to varying degrees. CK levels were invariably highly elevated and usually measured in several thousands (2000–8000 U/l). None of the presented patients lost ambulation during the time of study. Cardiac functions of all patients were normal. Also, respiratory functions did not show any abnormalities (7).

In an asymptomatic case series with high CK levels (CK>500 IU/L), authors analysed muscle biopsy specimens of 104 patients. In the study, 13 were children (ages 4 to 15 years), 20 were young adults (ages 16 to 30 years), and 71 were adults (ages 31 to 79 years). Fifty patients were completely asymptomatic and 54 patients had myalgia, cramps, or fatigue. CK levels were 500 to 1000 UI/L in 50 patients (mean age 43 years), 1000 to 2000 UI/L in 30 patients (mean age 41 years), >2000 in 24 patients (mean age 30 years). They achieved a definite or probable diagnosis in 55% of cases and concluded that the probability of making a diagnosis was higher in children and when CK level was greater than 2000 UI/L. The most frequently identified diseases were glycogen storage diseases, muscular dystrophies, and inflammatory myopathies. There was 20 patients with diagnosis of muscular dystrohy and one patient was diagnosed as calpainopathy (8). The study also demonstrated that patients with MD may present with asymptomatic or mild clinic as our case.

There are no established drug therapy for LGMDs, but different therapeutic approaches, including gene therapy, cell therapy and pharmacological trials are currently under investigation in animal models and experimental studies (9, 10). After diagnosis of LGMD 2A, control of weight, avoidence of obesity, physiotherapy and support of mobility with stretch exercises, avoidance of joint contracture should be planned. Surgical interventions of orthopedic complications like foot deformity and scoliosis can be required. Follow up for cardiomyopathy should be done although it is uncommon. Baseline cardiac evaluation with echocardiography and respiratory function testing are advised. We can increase life quality of these group of patients with support of social and emotional aspects (6, 9). As we aimed to emphasize asymptomatic presentation of LGMD2A at pediatric age, our report did not mention about prognosis of our case.

As a conclusion, muscle disorders can present with asymptomatic clinic during childhood period. There is no specific biomarker for LGMDs but muscle biopsy helps

diagnosis and molecular genetic analysis makes the exact diagnosis of LGMDs. Early diagnosis is important to increase their life quality with supportive therapies and for future genetic therapies for MD including LGMD type 2A.

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