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ORIGINAL ARTICLE

Clinical and Genetic Spectrum of Titinopathy: A Turkish Pediatric Case Series

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Abstract

OBJECTIVES: Our aim is to contribute to the literature by presenting pediatric titinopathy cases with different clinical and genetic profiles, including the newly identified homozygous *TTN* mutation.

METHODS: We retrospectively evaluated five pediatric patients with genetically confirmed titinopathy who presented to the Cukurova University Pediatric Neurology Department between January 2015 and May 2025. Clinical features, pattern of muscle weakness, electromyography (EMG), creatine kinase (CK) levels, muscle biopsy findings, cardiac and respiratory involvement, and genetic analyses via next-generation sequencing (NGS) were documented.

RESULTS: Five pediatric patients from three consanguineous families were diagnosed with titinopathy. Patients' median age was 14 years (30 months-17 years). The mean age of walking in the patients was 28.5 months. Four patients from two consanguineous families carried a novel homozygous c.15218-2A>G mutation in the *TTN* gene, not previously reported as pathogenic. These cases exhibited a wide spectrum of muscle involvement, variable facial, respiratory, and cardiac manifestations, and histopathological features of myotubular or centronuclear myopathy. One unrelated patient had a known pathogenic homozygous c.35296G>A mutation, associated with limb-girdle and facial muscle weakness and mild cardiomyopathy. Despite genetic similarities, phenotypic expression varied even within families. All CK levels were normal.

CONCLUSION: This study expands the clinical and molecular understanding of titinopathies by identifying a novel *TTN* mutation associated with marked phenotypic variability, even within the same family. The findings underscore the significant heterogeneity of *TTN*-related myopathies and reinforce the necessity of comprehensive clinical and genetic evaluations for accurate diagnosis and effective genetic counseling.

Abbreviations:
EMG - Electromyography
CK - Creatine kinase
NGS - Next-generation sequencing
TTN - Titin gene

INTRODUCTION

Titin, encoded by the titin gene (*TTN*), which includes 364 exons, is one of the largest proteins and spans the half-sarcomere from the Z-disk to the M-band, and is an important protein involved in muscle development, structure, elasticity, and signalling. The gene product protein titin is traditionally divided into three main isoforms, N2A, N2B, and N2AB. N2A isoforms are mainly expressed in skeletal muscles, while N2AB isoforms are mainly expressed in the heart and contain both N2A and N2B elements. Mutations in the titin gene cause clinical manifestations involving cardiac or skeletal muscle or both (Huang *et al.* 2021; Savarese *et al.* 2020). *TTN* mutations may present with different inheritance, pathological and clinical findings. These include: dilated cardiomyopathy, familial hypertrophic cardiomyopathy, congenital myopathy with cardiomyopathy, limb girdle muscular dystrophy R10 (2J), myofibrillar myopathy with early respiratory failure, and tibial muscular dystrophy (Savarese *et al.* 2020; Huang *et al.* 2021). This heterogeneity highlights the complex genotype-phenotype correlations specific to titinopathies.

In this article, we present five cases that were followed up in our clinic with a diagnosis of titinopathy and presented with different clinical and genetic findings. Four of these patients carried a novel homozygous c.15218-2A>G mutation in the *TTN* gene, not previously reported as pathogenic.

MATERIALS AND METHODS

Five patients who were admitted to the Cukurova University Pediatric Neurology Clinic between January 2015 and May 2025 with a genetically established diagnosis of titinopathy were retrospectively evaluated. The diagnosis of titinopathy was based on clinical symptomatology and molecular genetic confirmation. Current age, age at presentation to the clinic, symptoms, age at onset of the symptoms, presence of consanguinity, family history, motor milestone delay, ophthalmoparesis, muscle weakness, facial, bulbar, cardiac involvement, EMG, CK levels, muscle biopsy findings and genetic analyses via NGS results of the patients were reviewed. Peripheral blood samples were isolated from leukocytes using QIAsymphony DSP DNA Midi Kit (Qiagen, Hilden, Germany) according to the manufacturer's instructions. The quality and quantity of the DNA samples were determined by Qubit™ Fluorometric Quantitation (Thermo Fisher Scientific, Waltham, MA, USA). Whole exome sequencing as a total workflow covers several steps, which are fragmentation, ligation, enrichment, hybridization, and sequencing. Finally, next-generation sequencing was performed via Illumina Next-Seq NGS system (Illumina, California, USA) platform with a minimum coverage of 100x. The data sizes of FASTQ files and variant qualities were checked. Bioinformatics analyses were carried out using QCI-A and QCI-I (Qiagen, Hilden, Germany). Detected variants were interpreted comparatively with at least 18 databases including HGMD (Human Gene Mutation Database), ClinVar, VarSome (The Human Genomic Variant Search Engine), NCBI (National Center for Biotechnology Information), 1000 Genome Frequency, ExAC (The Exome Aggregation Consortium), Ancestry, Ingenuity Knowledge Base, ESP (Exome Sequencing Project) and OMIM (Online Mendelian Inheritance in Man). Additionally, multiple in-silico tools such as CADD, MutationTaster, PolyPhen, and DANN were used during in-silico analysis. All identified genetic alterations were categorized based on their pathogenicity according to the American College of Medical Genetics (ACMG) criteria.

Statistical Analysis

All analyses were performed using IBM SPSS Statistics Version 20.0 statistical software package. Categorical variables were expressed as numbers and percentages, whereas continuous variables were summarized as mean and standard deviation, and as median and minimum–maximum where appropriate (IBM Corp. Released 2011. IBM SPSS Statistics for Windows, Version 20.0. Armonk, NY: IBM Corp.).

RESULTS

Five pediatric patients from three consanguineous families were diagnosed with titinopathy. Their median age was 14 years (30 months-17 years). Patients achieved independent walking at a mean age of 28.5 months. Four patients (Cases 1–4) carried a novel homozygous c.15218-2A>G mutation in the *TTN* gene, while one patient (Case 5) had a homozygous c.35296G>A mutation previously reported as pathogenic. All patients exhibited symmetrical muscle weakness predominantly involving the lower limbs, with either proximal or combined proximal-distal distribution. Facial weakness was observed in three patients, and two patients had signs of respiratory involvement, particularly among those with the novel mutation. Cardiac findings ranged from normal to mild mitral insufficiency and were present in both mutation groups. CK levels were within normal range in all patients. EMG findings indicated myogenic involvement in most cases, while one patient showed neurogenic features. Muscle biopsies, performed in two patients with the novel mutation, revealed findings consistent with myotubular or centronuclear myopathy. Despite genetic similarities, phenotypic expression varied even within the same family. Despite

Tab. 1. Genetic and demographic characteristics of patients with titinopathy

Feature / Case	Case 1	Case 2	Case 3	Case 4	Case 5
Sex / Age (year)	F / 17	M / 7	M / 2.5	F / 17	F / 14
Initial Symptoms	Inability to walk and lumbar scoliosis	Frequent pulmonary infections	Floppiness, muscle weakness, frequent pulmonary infections.	Muscle weakness	Difficulty in walking, and stair climbing
Family history/ Consanguinity	Yes/Yes	Yes/Yes	Yes	Yes	Yes
Age at Walking (months)	35	Never walked	22	36	12
Loss of Ambulation	12 years	Never ambulatory	No	5 years	No
Mental Status	Normal	Normal	Normal	Normal	Normal
Facial Muscle Involvement	No	Mild	No	Mild	Mild
Ptosis	No	Bilateral	No	No	No
Ophthalmoparesis	No	No	No	No	No
Muscle Weakness	Lower extremities and proximal muscles; generalised muscle atrophy	Lower extremities and proximal muscles; generalised muscle atrophy	Lower extremities (proximal and distal muscles); generalised hypotonia	Lower extremities and proximal muscles; generalised muscle atrophy	Lower extremities proximal muscles
Contractures	Elbows, knees	Knees	No	Knees, ankles	No
Scoliosis	Severe lumbar	Mild	No	Mild and rigid spine	No
Respiratory Involvement	Nocturnal hypoventilation	Restrictive respiratory disease	Mild	No	No
Cardiac Involvement	No	Mild mitral insufficiency	No	No	Mitral insufficiency
Bulbar Involvement	No	No	Yes	No	No
CK Level	Normal	Normal	Normal	Normal	Normal
EMG Findings	Myogenic	Myogenic	Mild neurogenic	Myogenic	Myogenic
Muscle Biopsy	Myotubular myopathy	Not done	Not done	Centronuclear myopathy	Not done
Genetic Mutation	*TTN* c.15218-2A>G homozygous	*TTN* c.15218-2A>G homozygous	*TTN* c.15218-2A>G homozygous	*TTN* c.15218-2A>G homozygous	*TTN* c.35296G>A homozygous

genetic similarities, clinical severity and progression varied significantly among family members, suggesting phenotypic heterogeneity. All patients were presumed to have autosomal recessive inheritance based on family history and consanguinity. The clinical findings and genetic results of the patients are summarized in Table 1.

DISCUSSION

Titinopathies are a heterogeneous group of inherited neuromuscular disorders varying in terms of inheritance, age of onset, degree of muscle involvement, and rate of progression. Muscle weakness may affect distal or proximal muscles or both. Cardiac and/or respiratory system involvement may be absent or may occur at a very early age. While some cases present with severe clinical findings at an early age, there may be cases in which the findings are not recognised until adolescence or adulthood. Different clinical findings may be observed even within the same family. Also, behavioural or mental problems have been reported in some mutations in the gene (Huang *et al.* 2021; Tang *et al.* 2017).

Among our patients, we had four patients from 2 different families carrying a mutation that has not been previously reported to be pathogenic. The first two patients were siblings and one of them walked late, and she had been non-ambulatory since the age of 12 years, while her brother never gained the ability to walk. In the first case, respiratory failure started to develop in adolescence. There was no facial and cardiac involvement. In contrast, her sibling (case 2) developed facial, cardiac and respiratory involvement

findings at an early age. In the third case, belonging to a different family with the same mutation, there were findings of skeletal muscle involvement with a milder course, such as mild motor retardation and hypotonia, as well as findings of bulbar and respiratory system involvement with early onset, and he was able to walk at 22 months of age. The second-degree cousin of this case (case 4) also walked late, had mild facial weakness, but cardiac and respiratory system findings were normal at the age of 17 years, and there was no bulbar involvement. While muscle weakness was prominent in proximal muscles in the first family, both proximal and distal muscle involvement were present in the second family carrying the same mutation. In two of these four patients, respiratory failure symptoms developed at an early age. But in case 4, there was no respiratory involvement despite severe muscle weakness at the age of 17 years. In the other patient of the same age, respiratory system involvement occurred in adolescence.

In all patients, limb weakness was predominantly symmetrical. But muscle weakness severity varies from mild to severe. In patients with c.15218-2A>G mutation, clinical findings started at an early age. Half of the patients had proximal and the other half had proximal and distal muscle involvement. Facial muscles were also involved in two of the four patients. In addition, the 4th case showed Emery-Dreifuss muscular dystrophy clinic with proximal-distal muscle weakness, joint contractures, scoliosis and rigid spine, but without cardiomyopathy. Similar cases are reported in the literature (Savarese *et al.* 2020; Oates *et al.* 2018; De Cid *et al.* 2015). In contrast, the patient with c.35296G>A mutation had normal gait age, muscle involvement was of the limb girdle type, but there was also facial muscle involvement. In the literature, this mutation has been associated with Salih myopathy (congenital myopathy type 5 and cardiomyopathy). It is characterised by hypotonia, delayed motor development, and distal contractures, which usually start in the neonatal period (Carmignac *et al.* 2007). Cardiac involvement may be congenital defects, dilated or hypertrophic cardiomyopathy or conduction defects (Herman *et al.* 2012; Martinez-Thompson *et al.* 2019). Muscle involvement is generalised distal and proximal involvement, and facial weakness and ptosis may also be present. Our patient had limb girdle type muscle involvement and mild facial weakness, her early motor development was normal. Cardiac examination was consistent with mild dilated cardiomyopathy and mitral valve prolapsus.

Respiratory system involvement was seen especially in patients with c.15218-2A>G mutation. Early cardiac involvement was found in both mutation types. However, cardiac involvement was absent in some cases despite severe muscle involvement findings.

None of the cases had asymmetric involvement findings or behavioural problems and mental involvement. We found parental consanguinity in all families. Parental consanguinity and family history suggested that the genetic transmission pattern may be autosomal recessive.

The most common histopathological abnormalities found in patients with titinopathy are non-specific changes such as increased fibre size variation, increased central nuclei, muscle atrophy and hypertrophy. In addition, cores, centronuclear or myofibrillar myopathy findings may also be detected (Huang *et al.* 2021; Oates *et al.* 2018; Martinez-Thompson *et al.* 2019; Ceyhan-Birsoy *et al.* 2013). Our two patients with the same mutation who underwent biopsy had myotubular/centronuclear myopathy findings. Although EMNGs are generally compatible with myogenic involvement, neurogenic involvement findings were found in 1 patient. It has been reported in the literature that neurogenic involvement findings may be present, especially in the early period (Huang *et al.* 2021). CK levels were normal in all patients.

The titin protein is organized into four structurally and functionally distinct regions that correlate with the muscle sarcomere. These regions, located from the amino terminus to the carboxy terminus of the protein, include the Z-disk, I-band, A-band, and M-line. Different variants are responsible for the heterogeneous phenotype (Baban *et al.* 2023; Baskar *et al.* 2025). There is an expanding panel of mutations in *TTN* associated with titinopathy. We found a new homozygous mutation in four patients from two different families, which has not been previously reported in the literature. In those patients, variants of other genes were also detected. The findings suggested that the genetic inheritance was autosomal recessive. We have seen that clinical findings can be different even within the same family.

LIMITATIONS

This study is limited by the small number of patients, which restricts the generalizability of the findings. As a retrospective case series, it lacks longitudinal follow-up data for all patients and does not include functional outcome measures or standardized clinical scales to assess disease severity. Another important limitation is that segregation analysis in extended family members was not performed. This would have provided stronger evidence to support the pathogenicity of the identified variants. Muscle biopsies were not performed in all patients, which may have limited the correlation of clinical findings with histopathology.

In conclusion, titinopathy patients may present at different ages of onset, with different clinical and histopathological findings and inheritance patterns. Reporting different cases will contribute to the literature for phenotype-genotype correlation.

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ETHICS APPROVAL AND CONSENT TO PARTICIPATE

This study was conducted in accordance with the Declaration of Helsinki and approved by the Ethical Review Board for Non-interventional Studies of Cukurova University Faculty of Medicine, Adana, Türkiye (Date 18 July 2025 - Meeting No:157).

CONSENT FOR PUBLICATION

Not applicable.

COMPETING INTERESTS

No conflict of interest was declared by the authors.

REFERENCES

1 Baban A, Cicenia M, Magliozzi M, Parlapiano G, Cirillo M, Pascolini G, Fattori F, Gnazzo M, et al. (2023). Biallelic truncating variants in children with titinopathy represent a recognizable condition with distinctive muscular and cardiac characteristics: a report on five patients. Front Cardiovasc Med. **10**: 1210378.

2 Baskar D, Vengalil S, Polavarapu K, Preethish-Kumar V, Nashi S, Arunachal G, Srivastava K, Desai V, et al. (2025). Titinopathies: Phenotype - genotype heterogeneity in an Indian cohort. J Neuromuscul Dis. **12**: 364–371.

3 Carmignac V, Salih MA, Quijano-Roy S, Marchand S, Al Rayess MM, Mukhtar MM, Urtizberea JA, Labeit S, et al. (2007). C-terminal titin deletions cause a novel early-onset myopathy with fatal cardiomyopathy. Ann Neurol. **61**: 340–351.

4 Ceyhan-Birsoy O, Agrawal PB, Hidalgo C, Schmitz-Abe K, Dechene ET, Swanson LC, Soemedi R, Vasli N, et al. (2013). Recessive truncating titin gene, *TTN*, mutations presenting as centronuclear myopathy. Neurology. **81**: 1205–1214.

5 De Cid R, Ben Yaou R, Roudaut C, Charton K, Baulande S, Leturcq F, Romero NB, Malfatti E, et al. (2015). A new titinopathy: Childhood-juvenile onset Emery-Dreifuss-like phenotype without cardiomyopathy. Neurology. **85**: 2126–2135.

6 Herman DS, Lam L, Taylor MR, Wang L, Teekakirikul P, Christodoulou D, Conner L, Depalma SR, et al. (2012). Truncations of titin causing dilated cardiomyopathy. The N Engl J Med. **366**: 619–628.

7 Huang K, Duan HQ, Li QX, Luo YB, Bi FF, Yang H (2021). Clinicopathological features of titinopathy from a Chinese neuromuscular center. Neuropathology. **41**: 349–356.

8 Martinez-Thompson JM, Winder TL, Liewluck T (2019). Centronuclear myopathy with cardiomyopathy due to recessive titinopathy. Muscle Nerve. **59**: E26–e27.

9 Oates EC, Jones KJ, Donkervoort S, Charlton A, Brammah S, Smith JE, 3rd, Ware JS, Yau KS, et al. (2018). Congenital Titinopathy: Comprehensive characterization and pathogenic insights. Ann Neurol. **83**: 1105–1124.

10 Savarese M, Vihola A, Oates EC, Barresi R, Fiorillo C, Tasca G, Jokela M, Sarkozy A, et al. (2020). Genotype-phenotype correlations in recessive titinopathies. Genet Med. **22**: 2029–2040.

11 Tang J, Fan Y, Li H, Xiang Q, Zhang DF, Li Z, He Y, Liao Y, et al. (2017). Whole-genome sequencing of monozygotic twins discordant for schizophrenia indicates multiple genetic risk factors for schizophrenia. J Genet Genomics. **44**: 295–306.