

Case report

A Novel Heterozygous *ACAN* Variant in a Short Patient Born Small for Gestational Age with Recurrent Patellar Dislocation: A Case Report

Short title: Short Stature due to a Novel *ACAN* Variant

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What is already known on this topic?

ACAN is located on chromosome 15q26.1 and variants in this gene impair chondrogenesis at the growth plate, resulting in defects in longitudinal bone growth and skeletal development. Heterozygous *ACAN* variants can induce several clinical characteristics in patients, including variable degrees of short stature and mild skeletal dysplasia such as midface hypoplasia, joint problems, brachydactyly, broad great toes, and lumbar lordosis.

What this study adds?

We report a Korean patient with a novel nonsense variant c.1968C>G, p.(Tyr656*) of *ACAN*. The patient born small for gestational age (SGA) presented with short stature and displayed several dysmorphic features including genu valgum, cubitus valgus, and recurrent patellar dislocations. Patellar dislocation is a rare clinical feature of patients with *ACAN* defects. *ACAN* variants should be considered in short stature patients born SGA with joint problems, particularly those with recurrent patellar dislocation and genu valgum.

Abstract

ACAN variants can manifest as various clinical features, including short stature, advanced bone age (BA), and skeletal defects. Here, we report rare clinical manifestations of *ACAN* defects in a 9 year, 5 month-old girl born small for gestational age (SGA), presented with short stature, and was initially diagnosed with idiopathic growth hormone deficiency. She displayed several dysmorphic features including genu valgum, cubitus valgus, and recurrent patellar dislocations. She presented with progressive advancement of BA compared to that for chronological age and whole exome sequencing confirmed the presence of a novel heterozygous nonsense variant c.1968C>G, p.(Tyr656*) of *ACAN*. *ACAN* variants should be considered in short stature patients born SGA with joint problems, particularly those with recurrent patellar dislocation and genu valgum.

Keywords: *ACAN*; short stature; patellar dislocation; small for gestational age

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25.03.2021

14.06.2021

Published: 2 June 2021

Introduction

Aggrecan is a component of the extracellular matrix around the cartilage growth plate and provides a hydrated gel structure for the load-bearing properties of joints. The aggrecan gene, *ACAN*, is located on chromosome 15q26.1 and variants of this gene impair chondrogenesis at the growth plate, resulting in longitudinal bone growth and skeletal development defects (1). Heterozygous *ACAN* variants can induce several clinical characteristics in patients, including variable degrees of short stature and mild skeletal dysplasia such as midface hypoplasia, joint problems, brachydactyly, broad great toes, and lumbar lordosis (2,3,4). Short patients with *ACAN* variants often present with advanced bone age (BA), which is different from the short stature associated with endocrine diseases such as idiopathic growth hormone deficiency (IGHD) and hypothyroidism, which are related to delayed BA. Therefore, advanced BA may lead clinicians to test specifically for *ACAN* variants in patients with short stature and other clinical features such as skeletal defects (5,6). However, as *ACAN* variant patients without advanced BA have also been reported, advanced BA may suggest the presence of *ACAN* variant but cannot be referred to as a specific characteristic of *ACAN* variants (7). In clinical practice, *ACAN* variants are difficult to diagnose because their phenotype-genotype relationships are inconsistent.

Here, we report a case of a patient with a novel *ACAN* variant presenting with patellar dislocation and suggest that this rare clinical feature of *ACAN* variant may be a definitive clue for diagnosis.

Case Report

A girl aged 9 years and 5 months was referred to a pediatric endocrinology clinic for evaluation when she presented with abnormally reduced stature. Her height was 115 cm [-3.93 Korean standard deviation score (SDS)] and weight 35.2 kg (0.60

SDS) (Figure 1A). According to the Tanner stage for puberty, her breast development was at stage 3 and pubic hair development was at stage 1. She was born at 40 weeks through normal vaginal delivery with birth weight 2800 g (-1.53 SDS; $\leq 10^{\text{th}}$ percentile); she was diagnosed with small for gestational age (SGA). Her mother's height was 161 cm (-0.01 SDS) and the father's height was 175 cm (0.10 SDS), yielding a predicted adult height—calculated using mid-parental height—of 161.5 cm (0.09 SDS) (Fig 1A). Family history also included short stature, with a recorded height of 150 cm (-2.36 SDS) in her paternal grandmother (Figure 1B). The subject exhibited various skeletal defects, including cubitus valgus, genu valgum, and recurrent patellar dislocation. BA was 10 years (1.37 SDS) and within the normal range compared to that for her chronological age (CA). No abnormalities were identified in either chemistry or thyroid function and the karyotype of the subject was normal (46, XX). The levels of basal luteinizing hormone, follicle-stimulating hormone, and estradiol were 2.02 mIU/mL, 6.16 mIU/mL, and 25 pg/mL, respectively, confirmed that the subject was in the pubertal stage. We performed the growth hormone provocation tests using arginine and L-dopamine without sex steroid priming, and the peak levels were 0.29 ng/mL and 2.3 ng/mL, respectively. No abnormal findings were identified using sellar magnetic resonance imaging and the patient was treated with 0.23 mg/kg/day of recombinant human growth hormone (rhGH) to combat IGHD. One year after initiation of treatment, the age of the subject was 10 years and 6 months, and her height increased to 124.1 cm (-2.97 SDS). Serum IGF-1 levels were in the upper half of the reference range (0.69 SDS) after rhGH treatment. BA was shown to be advanced by 1 year and 6 months (2.31 SDS). Gonadotropin-releasing hormone agonist (GnRHa) treatment was added to the rhGH treatment to suppress bone maturation. She developed knee deformity over the 4 years, with recurrent intermittent pain during walking. She showed no improvement in her knee deformity and pain with conservative treatment. At the age of 10 years and 10 months, she underwent knee realignment surgery to correct a progressive bilateral genu valgum deformity and address her recurrent patellar dislocations.

DNA was extracted from EDTA blood samples and the entire exome was captured using SureSelectXT Human A11 Exon v5 (Agilent), followed by sequencing using NextSeq (Illumina). The reported variant was confirmed using Sanger sequencing. WES revealed a novel heterozygous variant, NM_013227.3:c.1968C>G, p.(Tyr656*), in exon 10 of *ACAN* (Figure 2). The pathogenicity of variant was evaluated in accordance with the American Society of College of Genetics and Genomics (8). This variant was considered pathogenic in accordance with the PSV1, PM2, and PP3 criteria, and was not found in the Genome Aggregation Database (gnomAD) or the Korean Reference Genome Database (KRGDB). Thus, we concluded that it represents a novel pathogenic variant that results in a nonsense variant of the tyrosine residue to a stop codon.

After 3 years and 6 months of rhGH treatment, the patient was receiving rhGH (0.3 mg/kg/day) and her height was 137.9 cm (-2.91 SDS). She has since been rehabilitated and her walking has stabilized.

Discussion

In this study, we confirmed a novel heterozygous *ACAN* variant using WES. The subject presented with short stature, SGA, IGHD, progressive advanced BA, and various skeletal defects including cubitus valgus, genu valgum, and recurrent patellar dislocations.

The increase in genetic testing to determine the underlying cause of idiopathic short stature (ISS) has highlighted the importance of *ACAN* variants. Recently, the *ACAN* variant has been shown to be one of the most common causes of ISS with a prevalence of 1–6% (9,10). In addition, *ACAN* presents with autosomal dominant (AD) inheritance, i.e., *ACAN* variants are also a common cause of familial ISS (11).

Aggrecan is a major proteoglycan component of the extracellular matrix in joints and intervertebral disc cartilage and provides the hydrated gel structure necessary to facilitate the load-bearing properties of skeletal joints (1). Therefore, aggrecanopathy resulting from *ACAN* variant has been linked to several degrees of skeletal defects and presents with some consistent clinical characteristics such as advanced BA and early growth cessation (12). However, *ACAN* variants with genotype–phenotype correlation are not always consistent, resulting in a high degree of clinical heterogeneity.

Advanced BA is considered a hallmark of *ACAN* variant; however, some patients develop delayed BA. *ACAN* variant with short stature and delayed BA was first reported in 2017 (7). A study showed that 79 out of 110 affected patients (72%) experienced advanced BA, suggesting that accelerated skeletal maturation may be a useful indicator of *ACAN* variant, but is insufficient as a clear clinical feature (2,4,9). The initial BA of the subject was within the normal range compared to that for her CA, but her BA progressed to more than 12 months in relation to CA after 1 year of rhGH treatment. It can be assumed that IGHD influences the progression of BA. Previous studies have reported that GnRHa treatment was successfully applied to prevent bone aging (2,4), as was also proven in this case.

It has been suggested that the possibility of *ACAN* variant in short-statured patients born SGA should be considered (4). A flowchart to determine the possibility of having an *ACAN* variant was presented; the clinical features of midface hypoplasia, joint problems, and broad great toes among short-statured patients born SGA were considered to be linked to the possibility of having *ACAN* variant. In this earlier study, since only 13.8% of patients with *ACAN* variant with advanced BA were found, the researchers reported that advanced BA was not sufficient as an indicator of *ACAN* variant. Among the major clinical symptoms presented in literature (midface hypoplasia, joint problems, and broad great toes), the subject of this case had only one characteristic feature, i.e., joint problems. According to the presented flowchart, the probability of *ACAN* variant was rare in this subject.

Skeletal defects from *ACAN* variants diagnosed in ISS do not present with serious defects. Osteoarthritis is a representative feature of the joint problems associated with the *ACAN* variant; however, the subject in this case did not experience any osteoarthritis, but developed genu valgum and recurrent knee dislocations at a young age. In a previous study, knee dislocation was found to be a relatively rare clinical feature reported in only a few affected patients (3). Knee dislocation is a very rare symptom and considered a hallmark of suspected *ACAN* variant when accompanied by short stature.

ACAN variant is inherited as an AD feature, but the parents of the subject were not short in stature. The grandmother of the subject was short in stature, but she grew up in a period of poor nutrition and hence environmental factors cannot be excluded. The parents of the subject did not want to undergo genetic testing, hence, we could not test them for genetic abnormalities. Owing to these limitations, we assumed that the patient had a *de novo* heterozygous *ACAN* variant.

Conclusion

We report a novel heterozygous variant of *ACAN*. If short statured children born SGA without catch-up growth exhibit joint problems, particularly recurrent patellar dislocation and genu valgum, we would suggest that these patients be immediately referred for genetic testing as there is a possibility that they may harbor *ACAN* variants.

Ethics

Ethics Committee Approval: This study was approved by the Institutional Review Board at the Hallym University Kangdong Sacred Heart Hospital (No. 2021-02-001).

Informed Consent: A written informed consent was obtained from her parents.

Peer-review: Externally and internally peer-reviewed.

Author Contributions: Conceptualization: Kim SJ, Yoon JS, Hwang IT; Data curation: Kim SJ; Methodology: Yoon JS and Hwang IT; Writing - original draft: Kim SJ; Writing - review and editing: Yoon JS and Hwang IT. All authors have accepted responsibility for the entire content of this submitted manuscript and approved its submission.

Conflict of Interest: The authors declare no conflict of interest.

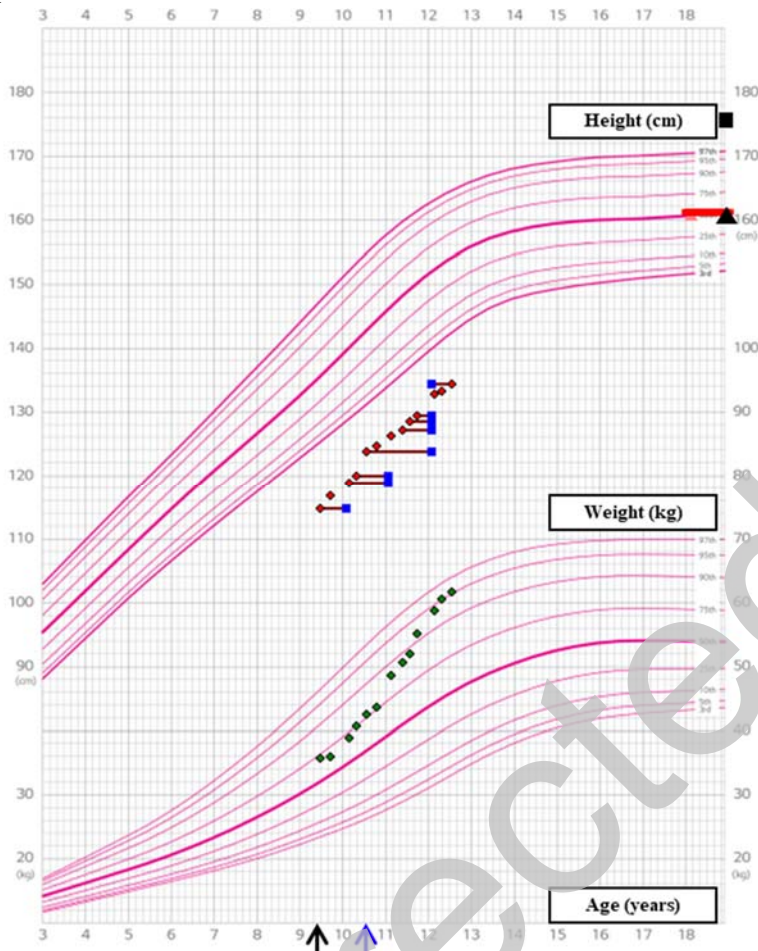
Financial Disclosure: None

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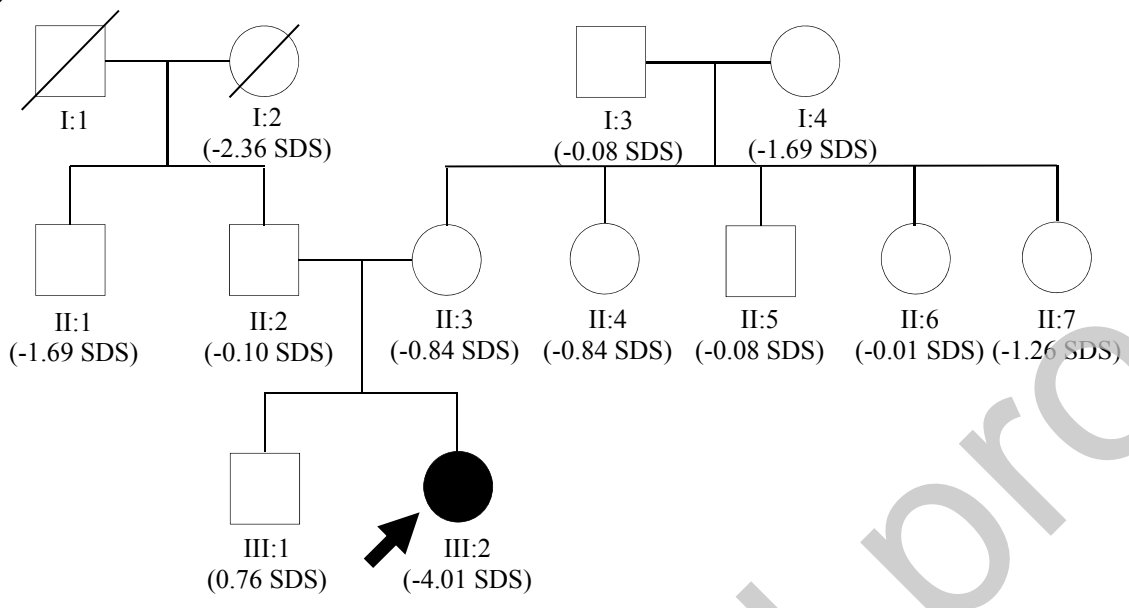
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Figure 1. (A) Reference growth chart for Korean females (3-18 years) (13). Heights and weights are shown as red and green dots, respectively. BA is represented as blue dots. Black squares and triangles indicate the height of the father and the mother, respectively. Mid-parental height is shown as the red line. The time point at which the patient started treatment with rhGH is indicated by a black arrow, and the time point at which GnRH α treatment was started is indicated by a blue arrow.

A



B



Uncorrected proof

Figure 2. Results of *ACAN* sanger sequencing in the patient.
NM_013227.3:c.1968C>G, p.(Tyr656*), Heterozygote, Nonsense

