A novel biallelic mutation in the *COL12A1* gene by a next-generation sequence revealed myopathic EDS. — A case report.

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Introduction

Myopathic Ehlers-Danlos Syndrome (mEDS), caused by mutations in *COL12A1*, was classified as a subtype of EDS according to the 2017 International Classification of the EDS. Major criteria include congenital muscle hypotonia, and/or muscle atrophy, that improves with age; prominent joint contractures (knee, hip, elbow), and hypermobility of distal joints. Minor criteria include soft, doughty skin; and atrophic scarring. At present, 20 patients from 11 families have been reported, with 10 families inherited in an autosomal dominant manner and one family in an autosome recessive manner. We encountered an additional patient with autosome recessive mEDS.

Case report

The patient : 47-years-old Japanese man, the second child of healthy consanguineous parents.
Family history: His brother was healthy. No family members with heritable connective tissue disorders.
Clinical history: After birth, he had hypotonia, scoliosis, weak spontaneous movements, and torticollis. He was diagnosed with Marfan syndrome at age 1 year, and started to have regular cardiovascular check-ups. He underwent surgery for cryptorchidism, scoliosis, and torticollis. Because of hypotonia, he had motor developmental delay and walked unassisted at age 3 years. His cognitive development was normal.
Present status: His weight was 50.5kg, height was 158.4cm, occipitofrontal circumference was 58.0cm, and arm span was 160.0cm.
His craniofacial features included relative macrocephaly, slender facial shape, bilateral mild ptosis, and high-arched palate. He had hypermobile thumbs and wrists, but had contractures of proximal joints. He had soft palms, but had no skin hyperextensibility or fragility. He had muscle weakness and scoliosis, but walked normally.
Skeletal survey revealed S-shaped thoracolumbar scoliosis, acetabular aplasia, and slender long bone. Echocardiography showed normal heart structure and no annualoaortic ectasia. Serum CK value was normal(93U/L).

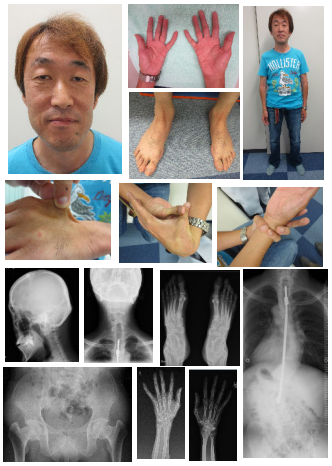

Fig1. Pictures of the patient (We had got his consent in writing on the announcement)

Molecular Investigation

① 17 hereditary connective tissue disease-related genes were searched by next-generation sequencing (ion PGM) : no pathogenic variants in *FBN1, COL1A1, COL1A2, COL3A1, COL5A1,* or *COL5A2* .
② Clinical exome analysis was performed, using TruSight One Sequencing Panel on MiSeq (Illumina, San Diego, CA, USA), including 4813 genes associated with human genetic disorders: A novel *de novo* homozygous variant (*COL12A1*:NM_004370 c.395-1G>A) of splice site of intron 6 (Fig 2A). ③ Expression analysis using mRNA from peripheral leucocytes : Variants resulted in splice abnormalities involving exons 5 and 6 (Fig 2B).

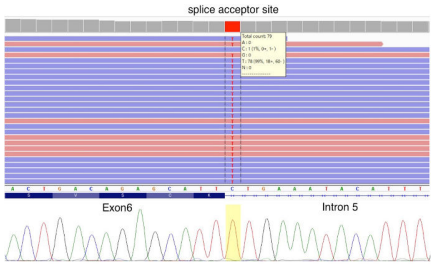

Fig 2A. NGS analysis using TruSight One Sequencing Panel

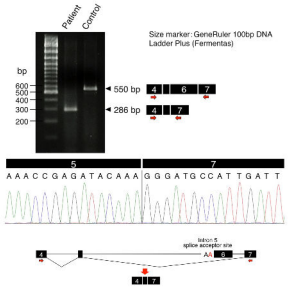

Fig 2B. Expression analysis

Family/Case	Mutation (inheritance)	Age of onset	Physical features at presentation	Course	P.C.	Reference
F1/C1, 2	c.8006+1G>A (AR)	Newborn	Hypotonia, distal joint hyperlaxity, proximal contractures, kyphosis	Able to sit. Unable to walk or stand	+	Zou et al.
F2/C3	c.7167T>C:p.Ile2334Thr (AD)	1 y/o	Hypotonia, distal joint hyperlaxity, proximal contractures	Able to walk	−	Zou et al.
F3/C4, 5	c.8357G>A:p.Gly2786Asp (AD)	Mother: Childhood Daughter: Newborn	Proximal weakness, join hypermobility, hip dislocation Hypotonia in infancy, distal joint hypermobility, left hip subluxation contractures, kyphosis	Able to walk Progressive proximal weakness, hyperkeratosis pilaris	−	Hicks et al.
F4/C6-8	c.5893C>T:p.Arg1965Cys) (AD)	Father: childhood 2 Sons: Adolescence	Poor at sports, unable to squat till 14 years Poor at sports, trouble in going up stairs	Unable to rise from chair Progressive proximal weakness	−	Hicks et al.
F5/C9	c.8329G>C:p.Gly2777Arg (AD)	Newborn	Oligohydramnios; intrauterine growth retardation, hypotonia, joint hyperlaxity, facial dysmorphology	Able to walk, muscle weakness, scoliosis	−	Punetha et al.
F6/C10−13	c.8100+2T>C (in-frame skipping of exon 52) (AD)	Proband, brother, mother, grandmother: newborn	Hypotonic with mild contractures, motor development delay, improvement during childhood	Poor at sports, able to run	−	Witting et al.
F7/C14	del c.8415+1_8415+10 (in-frame skipping of exon 56) (AD)	Childhood	Motor delay, hypotonia, congenital contractures. Joint hypermobility, proximal muscle weakness	Able to walk.	−	Delbaere et al.
F8/C15	c.8265+1G>A (in-frame skipping of exon54) (AD)	Neonatal	Neonatal hypotonia, walked at 17 months, joint hypermobility, scoliosis, pectus deformity	Able to walk	−	Delbaere et al.
F9/C16−18 (C18, mother)	c.8178+3A>C (in-frame skipping of exon 53) (AD)	C16) Neonatal; C17) Childhood; C18) Adult	C16) Walked at 19 months, neonatal hypotonia, joint hypermobility, pectus excavatum; soft and hyperextensible skin with delayed wound healing, facial morphology; C17) Walked at 12 months, joint hypermobility, facial morphology; C18) Normal	C16, 17) Able to walk; C18) Normal	−	Delbaere et al.
F10/C19	c.8100+3_8100+6del GAGT (in-frame skipping of exon52) (AD)	Childhood	Walked at 27months, hypotonia, congenital contractures, distal joint hypermobility	Able to walk	−	Delbaere et al.
F11/C20	c.5587C>T (p.Arg1863Cys) (AD)	Childhood	Poor fine motor skills, progressive contractures, muscle atrophy	Toe walking	−	Delbaere et al.
F12/C21	c.395-1G>A (in-frame skipping of exons 5-6) (AR)	Childhood	Hypotonia, joint hypermobility, scoliosis, facial dysmorphology	Able to walk	+	This report

P.C. parental consanguinity

Discussion & Conclusion

This is the first Japanese patient with myopathic EDS and the second family with autosomal recessive inheritance. Compared with previously reported patients with autosomal recessive inheritance (F1/C1, 2), clinical manifestation of this patient seemed milder. Possibly heterozygous parents in F1/C1, 2 had a mild manifestation: delayed motor development in childhood but normal strength in adult, whereas parents of this patient had no relevant manifestations. A biallelic splice-site mutation in F1/C1, 2 could have resulted in a complete loss of collagen XII, but a biallelic in-frame skipping of exons 5 and 6 in this patient would result in abnormal but functioning collagen XII. Further studies are needed to delineate genotype-phenotype correlation of this disease.

Reference
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The authors declare no conflict of interests.