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The phenotypic heterogeneity of patients with Marfan-related disorders and their variant spectrums

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Abstract

Marfan syndrome (MFS) and Loeys–Dietz syndrome (LDS) are the connective tissue disorders characterized by aortic root aneurysm and/or dissection and various additional features. We evaluated the correlation of these mutations with the phenotypes and determined the clinical applicability of the revised Ghent criteria.

The mutation spectrum and phenotypic heterogeneities of the 83 and 5 Korean patients with suspected MFS and LDS were investigated as a retrospective manner. In patients with suspected MFS patients, genetic testing was conducted in half of 44 patients who met the revised Ghent criteria clinically and half of 39 patients who did not meet these criteria.

Fibrillin1 gene (*FBN1*) variants were detected in all the 22 patients (100%) who met the revised Ghent criteria and in 14 patients (77.8%) who did not meet the revised Ghent criteria ($P = .0205$). Patients with mutations in exons 24–32 were diagnosed at a younger age than those with mutations in other exons. Ectopia lentis was more common in patients with missense mutations than in patients with other mutations. Aortic diameter was greater in patients with missense mutations in cysteine residues than in patients with missense mutations in noncysteine residues. Five LDS patients had either *TGFBR1* or *TGFBR2* variants, of which 1 patient identified *TGFBR1* variant uncertain significance.

The revised Ghent criteria had very high clinical applicability for detecting *FBN1* variants in patients with MFS and might help in selecting patients with suspected MFS for genetic testing.

Abbreviations: cbEGF = calcium-binding epidermal growth factor, EL = ectopia lentis, *FBN1* = fibrillin 1 gene, LDS = Loeys–Dietz syndrome, LTBP = latent transforming growth factor beta binding protein, MFS = Marfan syndrome, SMAD3 = decapentaplegic homolog 3, TGFB2 = transforming growth factor beta 2 ligand, TGFBR1 = transforming growth factor beta receptor I, TGFBR2 = transforming growth factor beta receptor II.

Keywords: Loeys–Dietz syndrome, Marfan syndrome, mutation spectrums

1. Introduction

Marfan syndrome (MFS, MIM 154700) and Loeys–Dietz syndrome (LDS type 1, MIM 609192; LDS type 2, 610168) are the congenital connective tissue disorders.[1,2] MFS is caused by a heterozygous mutation in the fibrillin 1 gene (*FBN1*), and LDS types 1 and 2 are caused by heterozygous mutations in the transforming growth factor beta receptor I and II genes (*TGFBR1* and *TGFBR2*), respectively.[1,2] Mutation in any one of these 3 genes alters TGF-β signaling.[3]

As one of the most common genetic disorders, the prevalence of MFS is estimated as 6.5:100,000 at the end of 2014.[4] The cardinal features of MFS include aortic root aneurysm and/or dissection, ectopia lentis (EL), and skeletal deformities,[1] which highly differ in severity. Since *FBN1* was identified as the causative gene of MFS in 1991,[5] more than 2200 *FBN1* variants have been identified in a recent update.[6] Wild-type *FBN1* is involved in building macromolecules of microfibrils, whereas mutant *FBN1*, especially with mutations in cysteine residues, disrupts the disulfide bond between fibrillins and leads to microfibril misalignment and disintegration.[7,8]

Because of phenotypic variability, the diagnostic confirmation of MFS has been challenging in some patients with mild manifestations, and the positive rates of the genetic testing also have been different among reports.[9,10] The revised Ghent criteria was introduced in 2010, which suggests the MFS

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diagnosis based on the major organ involvements, family history, and *FBN1* gene mutation.[11] With the emphasis on genetic testing, the diagnostic yield of MFS increased.[12]

The hallmarks of LDS include diffuse aortic aneurysm and/or dissection, arterial tortuosity, hypertelorism, and bifid uvula or cleft palate.[2] In addition to the *TGFBR1* or *TGFBR2* mutations, the decapentaplegic homolog 3 (*SMAD3*), transforming growth factor beta 2 ligand (*TGFB2*), and *TGFB3* mutations are recently identified in patients with LDS type 3 (MIM 613795), type 4 (MIM 614816), and type 5 (MIM 615582), respectively.[13–15] Although, LDS are categorized into type 1–5 according to the causative genes, there is no clear genotype and phenotype correlations, and the phenotypes are quite overlapped among subtypes.[13,16–19]

In the present study, we described the clinical and molecular findings of Korean patients with suspected MFS or LDS and identified 34 *FBN1* variants, including 16 novel variants; 3 *TGFBR1* variants and 2 *TGFBR2* variants. In addition, we evaluated the correlation of these variants with the phenotypes of patients and determined the clinical applicability of the revised Ghent criteria as a guideline for selecting patients for genetic testing.

2. Methods

2.1. Patients

A total of 83 and 5 Korean patients with suspected MFS and LDS, respectively, were referred to Medical Genetics Center, Asan Medical Center Children's Hospital, Seoul, Korea, for molecular testing between January 1994 and August 2015. Their detailed clinical characteristics, including gender and age at diagnosis; physical findings; and results of echocardiography, slit-lamp eye examination, radiological studies, and genetic testing, were reviewed retrospectively. Diameter of the aortic root was measured from leading edge to leading edge of sinuses of Valsalva at end diastole by performing transthoracic echocardiography, was adjusted according to patient age and body surface area, and was calculated as *Z*-score.[20,21] EL was determined after inducing maximal dilation of the pupil by performing slit-lamp eye examination. Acetabular protrusion and spinal scoliosis were determined based on radiological findings. The clinical diagnosis of each patient with MFS was reevaluated using the revised Ghent criteria.[11] This study was approved by the Institutional Review Board of the Asan Medical Center, and informed consent was obtained from each patient or his or her parents.

2.2. Genetic testing and variant classification

Genomic DNA was isolated from peripheral blood leukocytes. Sixty-five exons of the *FBN1* gene, 9 exons of the *TGFBR1* gene, and seven exons of the *TGFBR2* gene and their intronic flanking sequences were amplified by PCR with 66, 10, and 9 sets of primers, respectively. After amplification, PCR mixtures were run on 1.2% agarose gel in the presence of ethidium bromide to verify the size and purity of PCR products. After verifying that single specific PCR product was amplified, DNA sequencing was performed using the same primers used in PCR, and BigDye Terminatore V3.1 Cycle Sequencing Ready reaction kit (Applied Biosystems, Foster City, CA). Reaction performed 30 cycles for at 94°C 20 seconds, at 55°C for 20 seconds, and at 72°C 30 seconds with C-1000 PCR machine (Bio-Rad, Hercules, CA) using 10 ng of PCR product, which treated with exonuclease I and Shirmp Alkaline phosphatase (Amersham Pharmacia Biotech, Piscataway, NJ), as template and 10 pmol of proper primer. Electrophoresis and analysis of the reaction mixtures were done with ABI 3130xl Genetic analyzer (Applied Biosystems). All identified sequence variants were analyzed with reference to the *FBN1* mutations database (http://www.umd.be/FBN1/), *TGFBR1* mutations database (http://www.umd.be/TGFBR1/), and *TGFBR2* mutations database (http://www.umd.be/TGFBR2/). The pathogenicity of mutations was assessed by in silico prediction analyses such as PolyPhen-2 (http://genetics.bwh.harvard.edu/pph2) and SIFT (http://sift.jcvi.org). Variants identified were classified according to The American College of Medical Genetics and Genomics.[22]

2.3. Statistical analysis

All statistical analyses were performed using SPSS for Windows (version 21; SPSS, Inc., Chicago, IL). $P < .05$ was considered statistically significant.

3. Results

3.1. Clinical findings of patients with MFS and LDS

Clinical findings of patients with suspected MFS and LDS are presented in Table 1. The median age at diagnosis of clinically suspected 83 unrelated MFS probands and 5 LDS patients were 19.3 ± 14.8 years (range, 0.1–50 years) and 11.8 ± 10 years (range, 1.8–26.3 years), respectively (Table 1). In suspected MFS patients, 23 patients had a positive family history and the other 60 patients were sporadic cases. Aortic root dilation and/or dissection was observed in 59 MFS (71.1%) and 5 LDS (100%) patients, and EL was observed in 36 (43.4%) patients with MFS. Systemic score[11] of ≥7 points was observed in 13 MFS (15.7%) and in 1 LDS (20.0%) patients. The most common finding of systemic features according to the revised Ghent criteria in suspected MFS was wrist or thumb sign (61.4%), followed by scoliosis (44.6%) and mitral valve prolapse (43.4%).

Table 1

Clinical findings of patients with clinically suspected MFS or LDS.

	Patients with MFS (n = 83)	Patients with LDS (n = 5)
Male:female	55:28 patients (66.3%:33.7%)	2:3 patients (40%:60%)
Median age at diagnosis	19.3 ± 14.8 y (1 mo to 58 y)	11.8 ± 10 y (1.8–26.3 y)
Mean height SDS	2.3 ± 1.2	1.2 ± 0.9
Family history	23 (27.7%)	2 (40%)
Aortic root dilation and/or dissection	59 (71.1%)	5 (100%)
Ectopic lentis	36 (43.4%)	0 (0%)
Systemic score of ≥7 points	13 (15.7%)	1 (20%)

LDS = Loeys–Dietz syndrome, MFS = Marfan syndrome, n = number of patients, SDS = standard deviation score.

Table 2

Comparison of clinical features between MFS patients under 18 years and over 18 years.

	<18 y (n=30)	≥18 (n=25)	P
Age at diagnosis, y	7.7±5.3	36.3±12.3	<.001
Aortic root dilation/dissection	23 (76.7%)	25 (100%)	.010
Aortic diameter, *Z*-score	4.0±3.3	6.5±2.9	.006
Ectopic lentis	22 (73.3%)	8 (32%)	.002
Systemic score of ≥7 points	5 (16.7%)	6 (24%)	>.05
Type of variant			
Missense	12/14 (85.7%)	9/19 (47.3%)	.027
Frameshift	1/14 (7.1%)	4/19 (21.1%)	>.05
Nonsense	1/14 (7.1%)	4/19 (21.1%)	>.05

n=number of patients.

When we compared the clinical features according to age, the frequency of aortic root dilatation was significantly higher in patients older than 18 years than those younger than 18 years (100% vs 76.7%, $P=.010$). *Z*-score of aortic diameter were significantly greater in patients older than 18 years than those younger than 18 years (6.5±2.9 vs 4.0±3.3, $P=.006$). EL was more common in patients under 18 years of age (73.3% vs 32%, $P=.002$) (Table 2).

All the 5 patients with LDS showed aortic root dilatation and/or dissection but did not show EL (Table 1). Characteristics of patients with LDS are summarized in Table S1, http://links.lww.com/MD/C245. Various dysmorphic features, including bifid uvula, exophthalmos, hypertelorism, low set ears, plagiocephaly, high arched palate, and pin-point chin, were observed in the 5 patients with LDS. The most common skeletal finding in patients with LDS was scoliosis (80%), followed by wrist and thumb sign (40%), pectus carinatum (40%), pes planus (40%), arachnodactyly (40%), joint laxity (40%), and pectus excavatum (20%). In addition, 1 patient showed cerebral arterial dilatation and tortuosity, as indicated by the results of brain magnetic resonance angiography.

3.2. Diagnostic sensitivity of the revised Ghent criteria and FBN1 mutations

Genetic testing was performed in the 22 out of 44 patients who fulfilled the revised Ghent criteria clinically and in the 18 out of the 39 patients who did not. *FBN1* mutations were detected in all the 22 (100%) patients in the former group, whereas *FBN1* mutations were found in 14 (77.8%) patients in the latter group ($P=.0205$ by Fisher exact test).

A total of 34 *FBN1* different mutations from 36 unrelated Korean families were identified. Of these 34 mutations, 20 (58.8%) were missense mutations, 6 (17.6%) were frameshift mutations, 6 (17.6%) were nonsense mutations, 1 (2.9%) was a deletion mutation, and 1 (2.9%) was a splicing mutation. Most mutations were private, except 1 mutation; c.4588C>T (p.Arg1530Cys) (3/36 alleles). A total of 16 mutations were not previously reported (Table 3).

The majority of variants (23/36 alleles, 66.7%) were located in the calcium-binding epidermal growth factor (cbEGF)-like domain. Further, 6 variants (6/36 alleles, 16.7%) were located in TGF-β protein (Transforming growth factor beta binding protein) domain, 2 (2/36 alleles, 5.6%) were located in hybrid module domain, 2 (2/36 alleles, 5.6%) were located in EGF-like domain, 1 (1/36 alleles, 2.8%) was located in latent TGF-β-binding protein (LTBP) domain, 1 (1/36 alleles, 2.8%) was located in fibulin-like domain, and 1 (1/36 alleles, 2.8%) was located in proline-rich domain (Table 3). Table 3 is placed at the end of the text.

3 *TGFBR1* and 2 *TGFBR2* variants were identified, of which 3 (60%) were missense mutations. The other 2 were an inframe deletion and insertion. Four variants were located in the serine–threonine kinase domain.

3.3. Genotype and phenotype correlations of patients with MFS and LDS

Comparison of clinical findings between patients with MFS with and without *FBN1* mutations did not show any significant difference. Of the 33 patients with *FBN1* mutations, 6 patients (20%) had mutation in exons 24–32. These patients were diagnosed at a younger age than patients with mutations in other exons (8.8±9.5 years vs 23.9±17.6 years, $P=.017$). Of note, 1 patient (case 61, c.3209A>G [p.Asp1070Gly]) was diagnosed in his neonatal period with aortic root dilatation (*z*-score 4.7) and mitral valve prolapse as having neonatal MFS. However, there was no significant difference in other clinical features including aortic root dilatation, EL, and systemic scores between the 2 groups (Table S2, http://links.lww.com/MD/C245).

When comparing the clinical characteristics of 21 patients with missense mutations and 12 patients with other mutations who met the revised Ghent criteria, EL was more common in patients with missense mutations than in those with other types (52.4% vs 16.7%, $P=.043$), whereas dural ectasia were less common in the patients with missense mutations (Fig. 1A and Table S3, http://links.lww.com/MD/C245).

Most missense *FBN1* mutations (76.2%) were located in cysteine residues. Aortic diameter was greater in patients with missense mutations in cysteine residues than in patients with missense mutations in noncysteine residues ($P=.012$; Fig. 1B and Table S4, http://links.lww.com/MD/C245). When the other clinical features were compared between patients with a missense mutation disrupting cysteine residue and a mutation creating de novo cysteine residue, no significant difference was noted.

Of the patients under 18 years of age, 66.7% had missense mutations associated with cysteine residues (8/12) and all were disrupting forms. There were no significantly clinical differences between patients with missense mutations in cysteine residues and those in noncysteine residues. In addition, the clinical features were not significantly different between the patients with mutations in exon 24–32 and those with mutations outside these exons (data not shown).

4. Discussion

In the present study, the wide range of variability of phenotypes in MFS was noted whereas relatively consistent phenotypes were

Table 3

***FBN1* variants spectrum of patients with MFS.**

Ghent criteria*	Exon	Nucleotide change	Amino acid change	Domain	SIFT score	PolyPhen score
(−)	2	**c.224C>G**	**p.Cys68Trp**	LTBP-like	0.02	0.999
(+)	3	**c.295_296ins(A)**	**p.Met99Lysfs*30**	EGF-like #01	n.a.	n.a.
(+)	6	**c.543C>G**	**p.Tyr181***	Hybrid module #01	n.a.	n.a.
(−)	10	c.1285C>T	p.Arg429*	Proline-rich	n.a.	n.a.
(+)	11	c.1421G>A	p.Cys474Tyr	EGF-like #04	0.0	0.995
(+)	14	**c.1759T>G**	**p.Cys587Gly**	cbEGF-like #05	0.0	0.997
(+)	22	**c.2696G>A**	**p.Gly899Glu**	Hybrid module #02	0.0	1.0
(−)	26	c.3209A>G	p.Asp1070Gly	cbEGF-like #12	0.02	0.949
(+)	26	**c.3299G>A**	**p.Gly1100Asp**	cbEGF-like #12	0.02	1.0
(+)	27	c.3349T>C	p.Cys1117Arg	cbEGF-like #13	0.01	0.999
(+)	28	**c.3576delG**	**p.Arg1192Sfs*12**	cbEGF-like #14	n.a.	n.a.
(−)	30	**c.3766A>G**	**p.Asn1256Asp**	cbEGF-like #16	0.14	0.994
(+)	31	**c.3914G>A**	**p.Cys1305Tyr**	cbEGF-like #17	0.00	0.997
(−)	32	c.4061G>A	p.Trp1354*	cbEGF-like #18	n.a.	n.a.
(+)	34	c.4254_4261del	p.Gln1419_Cys1420del	cbEGF-like #20	n.a.	n.a.
(+)	34	**c.4258T>C**	**p.Cys1420Arg**	cbEGF-like #20	0.0	0.999
(+)	35	**c.4412_4415del**	**p.Glu1471Valfs*3**	cbEGF-like #21	n.a.	n.a.
(+)	36	**c.4505G>C**	**p.Cys1502Ser**	cbEGF-like #22	0.01	0.978
(+)	37	c.4588C>T	p.Arg1530Cys	TGFBP #04	0.0	1.0
(+)	37	**c.4684_4697del**	**p.Ser1561fs*3**	TGFBP #04	n.a.	n.a.
(−)	38	c.4786C>T	p.Arg1596*	TGFBP #04	n.a.	n.a.
(+)	39	c.4930C>T	p.Arg1644*	cbEGF-like #23	n.a.	n.a.
(+)	43	c.5338G>A	p.Gly1780Arg	cbEGF-like #25	0.0	1.0
(−)	44	c.5453G>C	p.Cys1818Ser	cbEGF-like #26	0.0	0.997
(−)	44	c.5504G>C	p.Cys1835Ser	cbEGF-like #26	0.0	0.997
(+)		c.5671+1G>A	IVS45(+1)G>A	cbEGF-like #28	n.a.	n.a.
(+)	46	**c.5698T>G**	**p.Cys1900Gly**	cbEGF-like #28	0.0	0.999
(+)	50	c.6254G>A	p.Cys2085Tyr	TGFBP #06	0.03	0.943
(−)	54	c.6658C>T	p.Arg2220*	cbEGF-like #34	n.a.	n.a.
(−)	54	c.6661T>C	p.Cys2221Arg	cbEGF-like #34	0.0	0.999
(−)	55	c.6866G>A	p.Cys2289Tyr	cbEGF-like #35	0.04	1.0
(−)	60	c.7465T>C	p.Cys2489Arg	cbEGF-like #39	0.0	0.999
(−)	63	**c.7839del**	**p.Ser2613fs*69**	cbEGF-like #42	n.a.	n.a.
(−)	65	c.8599del	p.Gln2867Argfs*23	Fibulin-like motif	n.a.	n.a.

SIFT: http://blocks.fhcrc.org/sift/SIFT.html, PolyPhen: http://genetics.bwh.harvard.edu/pph.

Bold characters, novel mutations.

cbEGF = calcium-binding epidermal growth factor, LTBP = latent transforming growth factor beta binding protein, n.a. = not applicable, TGFBP = Transforming growth factor beta binding protein.

* (+) Ghent criteria positive, (−) Ghent criteria negative.

observed in a small number of LDS patients. The overall rates of detecting germline variants were 90% (36/40 alleles) in patients with MFS and 80% (4/5 alleles) in patients with LDS. Notably, the rates of variants detection were very high (as high as 100%) in patients with MFS who met the revised Ghent criteria before undergoing genetic testing. This high rate is consistent to 81% to 85% in the previous studies.[9,10,23] In contrast, rates of mutation detection decreased to 77.8% in patients who did not meet the

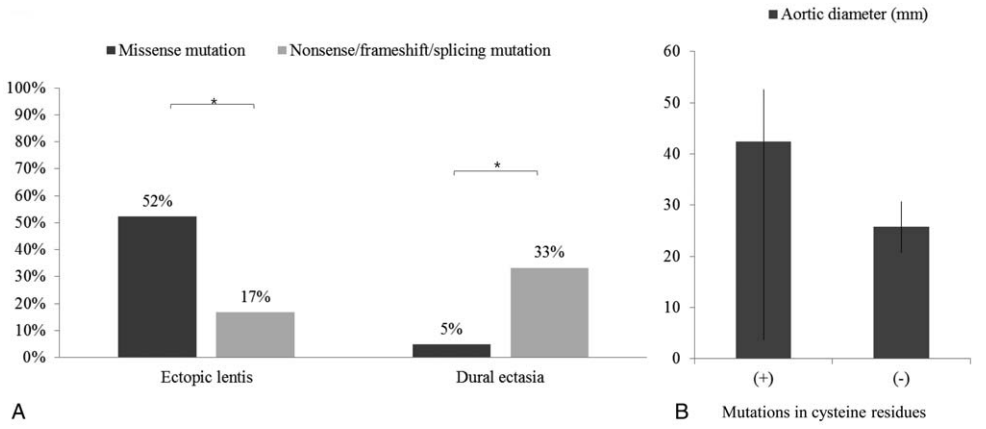

Figure 1. Comparison of clinical features according to *FBN1* mutation types and locations. *$P < .05$.

revised Ghent criteria clinically. Our study suggested that genetic testing should be performed in patients who have strong evidences for MFS by the revised Ghent criteria because of high confirmatory diagnostic yield. However, even for the patients with less clinical evidence, the molecular testing still can be justified because a substantial proportion of the patients do have *FBN1* mutations.[12] Alternatively, exonic deletion or locus heterogeneity may occur in patients without point mutations in *FBN1*, as previously observed in a small subset of patients with MFS.[24]

Approximately 65% of all *FBN1* mutations are missense mutations, of which majority affect one of the 6 highly conserved cysteine residues located in the cbEGF-like domain.[23,25] Consistently, missense variants were the most common (61.1%) in the present study. Overall, 55.9% of the 34 *FBN1* variants were in the cbEGF-like domain, and 14 out of the 20 missense *FBN1* variants (70%) were in cysteine residues. No identical *FBN1* mutations reported and most mutations were private in our study although 1 missense mutation, p. Arg1530Cys, was found recurrently in 3 unrelated families.

The patients with the mutations located in exon 24–32 of the cbEGF-like domain have been identified at earlier ages, experiencing more aggressive clinical courses.[26–28] The region containing these exons plays a major role in the assembly and stability of 10-nm microfibrils in the extracellular matrix.[29] In our present study, patients with a variant in exons 24–32 were diagnosed at an early age, even in the neonatal period. However, no significant difference was observed between these patients and patients with mutations in other exons with respect to other clinical findings. One of the reasons for this result may be attributed to the big differences of the number of cases between the 2 groups (Table S3, http://links.lww.com/MD/C245). In addition, the age at evaluation for the patients with variants in exons 24–32 was 8.8 ± 9.5 years in contrast to 23.9 ± 17.6 years in patients with variants outside exons 24–32.

Indeed, age is an important factor affecting the clinical severity of MFS. In our present study, 54.5% of the patients who met the revised Ghent criteria were under 18 years of age, and cardiac manifestations were less severe in these patients as previously reported.[24] The high prevalence of EL in these patients is attributed to the high prevalence of missense variants.

EL is more common in patients with MFS who have missense mutations in cysteine residues than in patients with missense mutations in noncysteine residues.[28,30,31]. However, no significant difference was observed in the frequency of EL between patients with missense mutations in cysteine residues and patients with missense mutations in noncysteine residues. Frequency of dural ectasia was less common in the patients with missense variants. However, aortic root diameter was significantly greater in patients with missense mutations in cysteine residues than in patients with missense mutations in noncysteine residue. Severe clinical findings such as aortic root dilatation and/or dissection are expected to be more common in patients with severe mutation types, including frameshift, nonsense, deletion, or splicing mutations, or in patients with missense mutations in cysteine residues.[28,32] These results, which are in line with those of previous studies, indicated that aortic root dilation should be carefully monitored in patients with exons 24–32, severe mutations, or missense mutations in cysteine residues.

However, the molecular mechanism of MFS is explained by either haploinsufficiency of *FBN1* gene or dominant negative activity of mutant fibrillin 1 protein.[30] The residual level of fibrillin 1 protein is much lower than 50% level despite the presence of a wild-type copy of *FBN1* in all MFS patients,[30] which might be contributed to inconsistent of genotype–phenotype correlations. In general, missense mutations are associated with a broad spectrum of dysfunctional fibrillin-1 proteins, accounting for the molecular mechanism of two-thirds of MFS patients.[33] Frameshift, nonsense, and splicing mutations usually display haploinsufficiency, leading to reduced amount of normal fibrillin-1 protein in one-third of patients.[34] We did not assay the mRNA level of *FBN1* to categorize the functional type of mutation. However, missense variants are presumed to influence dominant negative effect.

The 5 patients with LDS showed craniofacial features, including hypertelorism, bifid uvula, and plagiocephaly, and skeletal findings including pectus deformities and joint laxity.[16] Clinical findings of patients with LDS are similar to those of patients with MFS but are more aggressive, especially progression of aortic aneurysm and dissection (mean age at death, 26 years).[16] Two patients with a classical triad of LDS showed aortic dissection at an early age, that is, 18 and 26.3 years, respectively. However, recent studies have shown that patients with *TGFBR1* and *TGFBR2* mutations had a wide range of severity in aortic disease, lack of consistent LDS phenotype, or skeletal findings of MFS.[16] About half of LDS have been reported featuring generalized arterial tortuosity and dilatation in head and neck vessels as noted in one of our patients.[15,16,35]

Three *TGFBR1* and 2 *TGFBR2* variants were found in 5 LDS patients. Most previously reported mutations are located in highly conserved amino acids in the serine–threonine kinase domain.[2,16,36] TGF-β binds within the ligand binding domain which subsequently activates the serine–threonine kinase domain of type 1 or type 2 TGF-β receptors and enhance SMADs phosphorylation, which increases TGF-β signaling.[2,37] The four mutations found in this study were also located in this domain.

The present study is a retrospective observational study; hence, it has several limitations that should be addressed. Indeed, only 15.9% of patients showed systemic score of 7 points or more, because some data about the skeletal findings of MFS were missing. In addition, pediatric patients may not show the full systemic MFS features, who consisted 54.5% in our study. There was a wide range of variation in the clinical findings of each patient. Especially, the age at evaluation was different among patients, which is an important factor for assessing the clinical severity. Because genetic testing was performed in only half of the patients, genotype–phenotype correlations could not be assessed in a substantial number of patients in whom genetic testing was not done. Moreover, even after evaluating genotype–phenotype correlations, the total number of patients and their evaluated ages were different among the groups.

5. Conclusions

Due to the high phenotypic heterogeneity of MFS, the genetic testing can be considered for patients whose clinical features meet the revised Ghent criteria. However, still *FBN1* testing can also be performed for the selected cases who are strongly suspected to have MFS on a clinical basis. Further studies are needed to find the genotype–phenotype correlations in MFS and LDS which are important for the postgenetic testing counseling and management for the patients based on the prognosis prediction.

Author contributions

GHS, BHL, and H-WY designed the study. GHS drafted the manuscript. All authors were involved in analyzing and

interpreting data. All authors read and approved the final manuscript.

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