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Letters to the Editor

Thoracic aortic-aneurysm and dissection in association with significant mitral valve disease caused by mutations in *TGFB2*

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The pathophysiology of thoracic aortic aneurysms and dissections (TAAD) is complex and multifactorial. Classic cardiovascular risk factors play an important role in a majority of patients but genetic factors should always be considered, especially in younger subjects and/or in the presence of a family history of TAAD. To date, several genes have been identified in both syndromic and non-syndromic forms of TAAD, including *FBN1* (Marfan syndrome, MFS), *TGFBR1/2* (Loeys-Dietz syndrome, LDS), *SMAD3* (Aneurysm-Osteoarthritis syndrome, AOS), *ACTA2* (TAA6, TAAD with livedo reticularis and iris flocculi), *MYH11* (TAAD with patent ductus arteriosus), and *MYLK* (TAAD7) [1–7]. Although clinical features show significant overlap, these entities differ in the extent of vascular involvement and clinical course. As a consequence, molecular studies have become pivotal in the evaluation, counselling and management of the patient with TAAD. Moreover, the identification of new genes has led to new insights into the pathogenesis of aneurysm formation.

The crucial role of the transforming-growth-factorβ (TGFβ) pathway in TAAD became evident from both studies on mouse models and the analysis of components of the TGFβ pathway on human aortic tissue of patients with these disorders [3,8–11]. We previously sequenced *TGFB2* as a candidate gene for TAAD, but did not identify mutations in 40 patients with isolated aortic root dilatation (Callewaert et al., unpublished results). The recent findings by Boileau and colleagues identifying the first *TGFB2* mutations leading to familial TAAD in association with cerebrovascular disease and mild systemic features reminiscent of Marfan syndrome [16] have urged us to screen this gene in a patient group associating TAAD with a wider phenotypic spectrum including cerebrovascular disease, arterial tortuosity, marfanoid skeletal features and mitral valve disease. In total, we assessed the prevalence of *TGFB2* mutations in 146 patients.

Using direct sequencing after amplification of all exons and flanking intronic sequences of the TGFB2 gene on genomic DNA level (*TGFB2* NM_001135599.2), we identified 4 heterozygous *TGFB2* mutations in 6 patients: c.475C>T (p.Arg159X), c.979C>T (p.Arg327Trp), c.980G>A (p.Arg327Gln), and c.1125delT (p.Gly376GlufsX17). We found 2 premature truncating mutations and also identified the first missense mutations in *TGFB2*. All mutations are expected to result in a loss-of-function. An overview of the clinical findings is provided in Table 1 and of the mutations in Fig. 1 and Supplemental Table 1.

Aortic pathology was universally present at middle-age (z-scores > 2.5), but aortic dissection was the presenting feature in only one patient at the age of 69 years (patient 1). Importantly, type A dissection occurred in another patient 5 years after initial evaluation and at an aortic root diameter below the classical surgical threshold of 50 mm (patient 6). Significant mitral valve prolapse occurred in 4 patients and was the reason for initial evaluation in two patients (2 and 6) that both required surgical replacement. Patient 4 came to medical attention following a routine echocardiography showing aortic root dilatation and mitral valve prolapse without other manifestations. One patient (patient 3) was evaluated following transient ischemic attacks at the age of 18 years old with underlying tortuosity of the vertebral arteries. Finally, patient 5 was evaluated for skeletal marfanoid features.

Four out of 6 patients had skeletal and/or skin manifestations reminiscent of TGFβ-signalopathies which can be mild; 3 patients had myopia of which one had cataract. Importantly, family history was negative in 3 patients.

Our data indicate a prevalence of *TGFB2* mutations in the examined TAAD cohort of around 4% (6/146), which is significantly lower than the previously reported prevalence of *ACTA2* mutations (16%) [11]. The TGFβ superfamily includes 3 isoforms of TGFβ, TGFβ1, −2, and −3 (for a review [12]). The TGFβs are pleiotropic cytokines, controlling a broad range of biological processes. The 3 TGFβ isoforms exhibit both overlapping and divergent properties as illustrated by the phenotype of the respective knockout mouse models. *Tgfb2* knockout mice die perinatally and display a wide range of developmental defects, including cardiovascular, pulmonary, skeletal, ocular, inner ear

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Table 1
Overview of the clinical findings in all mutation-positive patients.

Nr	Age/ sex	Age at diagnosis presenting symptom	Cardiovascular features		Skeletal	Craniofacial	Skin	Ocular	Cerebro-vascular	Family	Mutation
			Aorta	Mitral valve							
1	69/F	60 y type A dissection	Type A dissection	Normal	Flat feet, hallux valgus	/	/	/	/	Mother died in childbirth, sister died from dissection at age 41	(p.Arg159X)
2	60/M	56 y MVP requiring MVR	TAA 43 mm z 2.7	MVP - MVR	Clubfeet, joint laxity, mild pectus carinatum	High arched palate	Varicose veins	Myopia	/	Negative	(p.Arg159X)
3	53/F	18 y recurrent TIA's	TAA (Dx at 43 y) 40 mm z 5.2	MVP, mild MR	Joint laxity as a child	High palate, retrognatia	Local translucency	Myopia, astigmatism, cataract	Recurrent strokes, corckscrew vertebral Aa	Daughter Marfanoid	(p.Arg327Trp)
4	38/M	38 y TAA	TAA 40 mm z 3.4	MVP	Joint laxity	/	/	Myopia, strabismus	/	Negative	(p.Arg327Trp)
5	60/M	41 y Marfanoid features	TAA 41 mm z 3.5	Trivial MR	Arachnodactyly, camptodactyly, kyphoscoliosis	Hypertelorism, downslanting palpebral fissures, malar hypoplasia, high palate, retrognatia	Atrophic scarring	/	/	Spontaneous pneumothorax in daughter, father sudden death at 43y	(p.Arg327Gln)
6	46/M	32 y MVP requiring MVR	Type A dissection <50 mm	MVP–MVR	Pectus carinatum, arachnodactyly, joint hypermobility	/	Striae	/	/	Negative	(p.Gly376Glu fsX17)

Abbreviations: M, male; F, female; y, years. MVP: mitral valve prolapse, MVR: mitral valve replacement, MR: mitral valve regurgitation, TIA: transient ischemic attack, TAA: thoracic aortic aneurysm, z: z-score of the aortic root, Aa: artery. Dx, diagnosed.

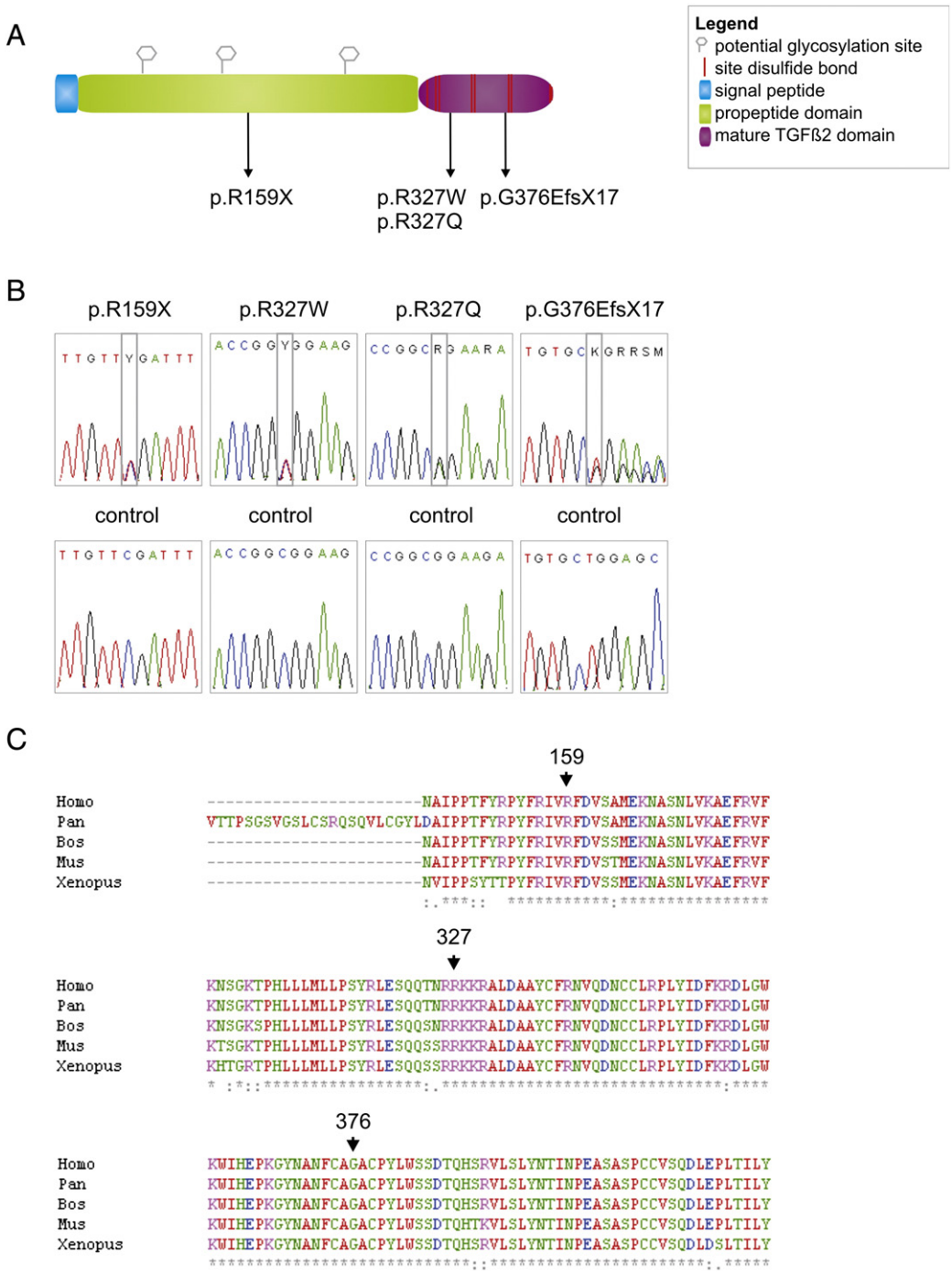

Fig. 1. Four unique *TGFB2* mutations were identified in patients with thoracic aortic aneurysm and/or dissection. (A) Schematic representation TGFβ2 protein and localisation of the mutations found in this study. (B) Sequencing profiles of all unique *TGFB2* mutations compared to the control sequences. (C) Partial alignment of the TGFB2 gene in multiple species showing conservation and position of the affected amino acid residues.

and urogenital manifestations [13,14]. The limited range of phenotypic manifestations seen in human patients is in high contrast to the broad range of developmental problems seen in the *Tgfb2* knockout mouse model.

The phenotype in the patients discussed here shows some overlap with other TGFβ-signalopathies including Marfan syndrome, Loeys-Dietz syndrome and the aneurysm-osteoarthritis syndrome. In congruence, aortic dilatation may result in type A aortic dissection at diameters below classic surgical thresholds. We found significant mitral valve disease, possibly a signature feature that may direct molecular analyses, but this should be confirmed in larger series. Until then, we suggest to exclude *TGFB2* mutations in all patients with TAAD lacking a clear syndromic constellation. The genetic screening should not be restricted to familial cases, as we identified *TGFB2* mutations in patients without a positive family history for TAAD. The identification of *TGFB2* mutations further adds to the considerable genetic heterogeneity in TAAD with 8 genes documented to date in still a limited number of TAAD patients.

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The authors of this manuscript have certified that they comply with the Principles of Ethical Publishing in the International Journal of Cardiology [15].This study was approved by the ethics committee of the Ghent University Hospital and all referring centres, and an appropriate informed consent was obtained from all patients involved in the study.

Appendix A. Supplementary data

Supplementary data to this article can be found online at http://dx.doi.org/10.1016/j.ijcard.2012.09.029.

References

[1] Dietz HC, Cutting GR, Pyeritz RE, et al. Marfan syndrome caused by a recurrent de novo missense mutation in the fibrillin gene. Nature 1991;352:337-9.
[2] Guo DC, Pannu H, Tran-Fadulu V, et al. Mutations in smooth muscle alpha-actin (ACTA2) lead to thoracic aortic aneurysms and dissections. Nat Genet 2007;39:1488-93.
[3] Loeys BL, Chen J, Neptune ER, et al. A syndrome of altered cardiovascular, craniofacial, neurocognitive and skeletal development caused by mutations in TGFBR1 or TGFBR2. Nat Genet 2005;37:275-81.
[4] Pannu H, Tran-Fadulu V, Papke CL, et al. MYH11 mutations result in a distinct vascular pathology driven by insulin-like growth factor 1 and angiotensin II. Hum Mol Genet 2007;16:2453-62.
[5] van de Laar IM, Oldenburg RA, Pals G, et al. Mutations in SMAD3 cause a syndromic form of aortic aneurysms and dissections with early-onset osteoarthritis. Nat Genet 2011;43:121-6.
[6] Zhu L, Vranckx R, Khau Van Kien P, et al. Mutations in myosin heavy chain 11 cause a syndrome associating thoracic aortic aneurysm/aortic dissection and patent ductus arteriosus. Nat Genet 2006;38:343-9.
[7] Wang L, Guo DC, Cao J, et al. Mutations in myosin light chain kinase cause familial aortic dissections. Am J Hum Genet 2010;87:701-7.
[8] Neptune ER, Frischmeyer PA, Arking DE, et al. Dysregulation of TGF-beta activation contributes to pathogenesis in Marfan syndrome. Nat Genet 2003;33:407-11.
[9] Hanada K, Vermeij M, Garinis GA, et al. Perturbations of vascular homeostasis and aortic valve abnormalities in fibulin-4 deficient mice. Circ Res 2007;100:738-46.
[10] Renard M, Holm T, Veith R, et al. Altered TGFbeta signaling and cardiovascular manifestations in patients with autosomal recessive cutis laxa type I caused by fibulin-4 deficiency. Eur J Hum Genet: EJHG 2010;18:895-901.
[11] Renard M, Callewaert B, Baetens M, et al. Novel MYH11 and ACTA2 mutations reveal a role for enhanced TGFbeta signaling in FTAAD. Int J Cardiol 2011 [Electronic publication ahead of print].
[12] Kingsley DM. The TGF-beta superfamily: new members, new receptors, and new genetic tests of function in different organisms. Genes Dev 1994;8:133-46.
[13] Sanford LP, Ormsby I, Gittenberger-de Groot AC, et al. TGFbeta2 knockout mice have multiple developmental defects that are non-overlapping with other TGFbeta knockout phenotypes. Development 1997;124:2659-70.
[14] Bartram U, Molin DG, Wisse LJ, et al. Double-outlet right ventricle and overriding tricuspid valve reflect disturbances of looping, myocardialization, endocardial cushion differentiation, and apoptosis in TGF-beta(2)-knockout mice. Circulation 2001;103:2745-52.
[15] Shewan LG, Coats AJ. Ethics in the authorship and publishing of scientific articles. Int J Cardiol 2010;144:1-2.
[16] Boileau C, Guo DC, Hanna N, et al. TGFB2 mutations cause familial thoracic aortic aneurysms and dissections associated with mild systemic features of Marfan syndrome. Nat Genet 2012;44(8):916-21.

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