ESC European Society of Cardiology

Cardiovascular Research (2024) **00**, 1–3
https://doi.org/10.1093/cvr/cvae245

EDITORIAL

TGF-β signalling: the Dr Jekyll and Mr Hyde of the aortic aneurysms

Sara Perrotta [1], **Daniela Carnevale** [1,2], **and Giuseppe Lembo** [1,3]*

[1]Department of Angiocardioneurology and Translational Medicine, IRCCS Neuromed, via dell'Elettronica, Pozzilli, IS 86077, Italy; [2]Department of Medical-Surgical Sciences and Biotechnologies, 'Sapienza' University of Rome, Latina 40100, Italy; and [3]Department of Molecular Medicine, 'Sapienza' University of Rome, Rome 00161, Italy

This editorial refers to 'The proprotein convertase *FURIN* is a novel aneurysm predisposition gene impairing TGF-β signalling,' by Z. He *et al.*, https://doi.org/10.1093/cvr/cvae078.

Aortic aneurysm is a potentially fatal disease causing a substantial number of sudden deaths each year worldwide. An aneurysm occurs due to the dilatation of blood vessels at different locations. The mortality is as high as 90% with massive haemorrhages caused by aneurysmatic rupture.

TGF-β signalling has been characterized as a crucial pathway in the pathophysiology of aortic aneurysms.

The early paradoxical evidence of an enhanced TGF-β signalling associated with loss-of-function mutations in the TGF-β receptor type I and type II (TGFBR1 and TGFBR2, respectively), found in aortic aneurysms of patients with Marfan[1] (fibrillin1 mutation) and Loeys–Dietz syndrome,[2] led to the hypothesis that the activation of TGF-β signalling could drive aortic aneurysms' development.

On the other hand, several other evidences in transgenic mouse models and in associative genetic human studies have suggested that an active TGF-β signalling in the vascular wall is a key determinant of a healthy vasculature and helps in counteracting the burden of vascular inflammation.

TGF-β is a ubiquitously expressed member of a superfamily of proteins critical to developmental processes and inflammation. TGF-β activity is highly regulated: it is synthesized as a homodimeric inactive proprotein (proTGF-β) and, in this form, it is secreted into the extracellular space where extracellular matrix (ECM) proteins, like Emilin1,[3] bind to proTGF-β and prevent its processing. The cleavage of proTGF-β is realized by a proteolytic process carried out by FURIN, a proprotein convertase that generates a pro-peptide known as the latency-associated protein (LAP) non-covalently bound to TGF-β in a latent complex. This association prevents TGF-β from interacting with its receptor until the LAP is removed from the complex, releasing active TGF-β through the action of thrombospondin-1, integrins, and other proteins. TGF-β binds to TGFBR2 and this interaction recruits and activates TGFBR1, leading to the phosphorylation of intracellular Smad2/3. Subsequently, phosphorylated Smad2/3 forms a complex with Smad4, which then translocates into the nucleus to regulate gene transcription (*Figure 1*, left panel). The extent of TGF-β tissue availability is controlled by many events at different steps, including protein synthesis, maturation, release in the ECM, and interaction with its receptors. Taken together, these steps modulate the cascade of downstream signalling proteins that control the transcription of TGF-β-regulated genes.

Several mouse models reproducing a condition of defective TGF-β signalling, like TGFBR1 or TGFBR2 deficiency,[4,5] or the absence of Smad4 selectively in smooth muscle cells[6] resulted in spontaneous aortic dilation and aneurysm formation, accompanied by a significant infiltration of pro-inflammatory myeloid cells. To further support this evidence, it has been shown that the inhibition of SLC44A2, a member of the solute carrier series 44 (SLC44) family that interacts with Neuropilin1 to activate TGF-β signalling, facilitated the spontaneous development of aortic aneurysm[7] (*Figure 1*, upper right panel).

In this issue of *Cardiovascular Research*, He *et al.*[8] further support and expand the concept of critical role of TGF-β in the onset of aortic aneurysms focusing on one of the first steps of the chain of events leading the activation of TGF-β: FURIN. It is synthesized as inactive proFURIN and becomes active in the trans-Golgi through autocatalytic cleavages. By whole-exome sequencing of 781 unrelated aortic aneurysm patients, the authors identified rare FURIN genetic variants characterized by the following chain of events: defective protein folding and maturation; impaired proteolytic enzymatic activity that leads to impaired proTGF-β maturation; decreased phosphorylation of the canonical downstream effector SMAD2 and the non-canonical kinases ERK1/2; and decreased transcription of the TGF-β-regulated genes. Thus, He *et al.* add another piece of evidence to the association between TGF-β and aortic aneurysm, identifying genetic FURIN variants impairing TGF-β and predisposing patients to the development of aortic aneurysm.

However, contrasting evidence from experimental animal models in which an inhibition of TGF-β maturation and signalling protects from aortic aneurysms by attenuating arterial dilation and remodelling still survives (*Figure 1*, lower right panel). As an example, it has been shown that the treatment with AGGF1, an angiogenic factor enhancing the interaction between integrin α7 and LAP-TGF-β1 and blunting TGF-β1 maturation and signalling, was as a valid therapeutic strategy to block the development of aortic aneurysm experimentally induced.[9]

What molecular puzzle is under the pathophysiological link between TGF-β signalling and aortic aneurysms?

Could TGF-β signalling play a different mechanistic role in the development of aneurysms depending on the stress conditions imposed on the vessel?

Could reduced TGF-β signalling in unstressed vascular conditions deprive the vessel from a crucial beneficial support? Could a pre-existing vascular challenge dramatically change its mechanistic contribution?

It is noteworthy that the immune system plays a critical role in the onset of aortic aneurysm, and TGF-β is a key modulator of innate and adaptive immunity, acting as a general enforcer of immune tolerance and a suppressor of inflammation.[10] Therefore, a fine-tuning of adaptive and innate immunity, obtained through TGF-β signalling during normal conditions, could be critical to the maintenance of unstressed vascular integrity. Perturbations in this homeostatic system could be the culprit of disease

The opinions expressed in this article are not necessarily those of the Editors of *Cardiovascular Research* or of the European Society of Cardiology.

* Corresponding author. Tel: +39 0865 915225; fax: +39 0865 927575, Email: lembo@neuromed.it

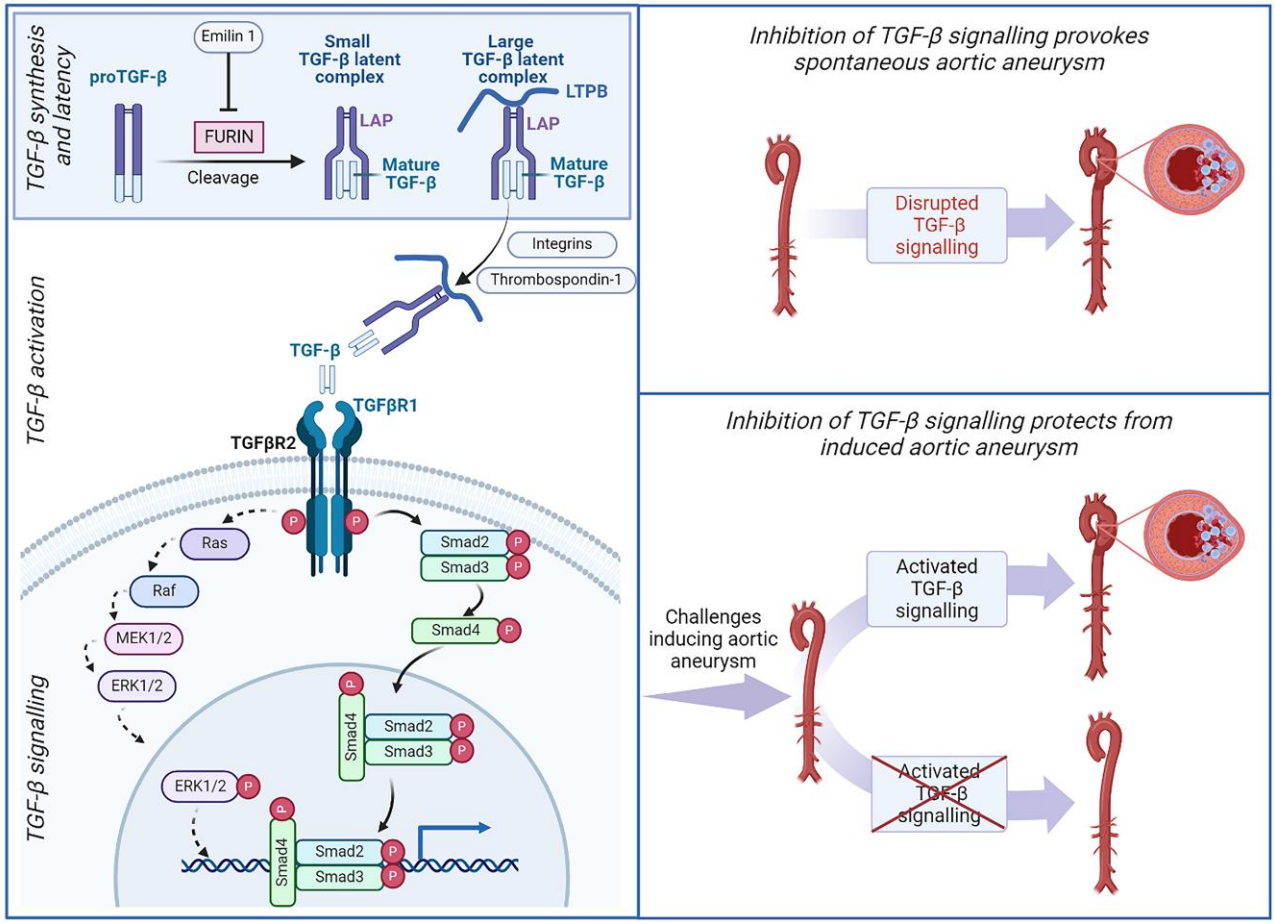

Figure 1 The dual effect of TGF-β signalling in aortic aneurysm. Left panel shows the sequence of TGF-β maturation and downstream signalling activation. TGF-β is synthetized as pro-TGF-β, and the cleavage of the dimeric C-terminal domain is carried out by the proprotein convertase FURIN in the Golgi. After cleavage, mature TGF-β remains sequestered by non-covalent binding to the latency-associated peptide (LAP) forming the small TGF-β latent complex; LAP binds to an isoform of latent TGF-β binding protein to form the large TGF-β latent complex released in the ECM. ECM proteins, like integrins and thrombospondin-1, bind to LAP and generate a TGF-β latent complex conformational change that releases the TGF-β for binding to TGFBR1 and TGFBR2 (TGF-β receptors type I and type II). The binding of TGF-β to its receptors activates SMAD and ERK kinases downstream signalling. The right panels show the Dr Jekyll and Mr Hyde roles of TGF-β. The upper right panel represents the induction of spontaneous aortic dilation and aneurysm formation by inhibition of TGF-β signalling. The lower right panel shows that the inhibition of TGF-β signalling in vessels challenged by thoracic aortic banding, fibrillin1 gene mutation, or β-aminopropionitrile treatment (challenges that induce aortic aneurysm) is beneficial and attenuates arterial dilation. Created in BioRender.com.

onset. On the other hand, when immune mechanisms have been already engaged by other vascular challenges, the activation of TGF-β could be recruited as a consequence of a stress pathway and be a part of more complex molecular interplay further instigating aortic aneurysm's pathogenesis.

Further studies examining the difference between the immune responses participating in normal and in stressed vessels are needed to clarify the disparate effects of this 'Jekyll and Hyde' TGF-β signalling on the pathogenesis of aortic aneurysms.

Conflict of interest: none declared.

Funding

This work has been supported by the Italian Ministry of Health 'Ricerca Corrente' to G.L.

Data availability

No new data were generated or analysed in support of this research.

References

1. Habashi JP, Judge DP, Holm TM, Cohn RD, Loeys BL, Cooper TK, Myers L, Klein EC, Liu G, Calvi C, Podowski M, Neptune ER, Halushka MK, Bedja D, Gabrielson K, Rifkin DB, Carta L, Ramirez F, Huso DL, Dietz HC. Losartan, an AT1 antagonist, prevents aortic aneurysm in a mouse model of Marfan syndrome. *Science* 2006;**312**:117–121.
2. Loeys BL, Schwarze U, Holm T, Callewaert BL, Thomas GH, Pannu H, De Backer JF, Oswald GL, Symoens S, Manouvrier S, Roberts AE, Faravelli F, Greco MA, Pyeritz RE, Milewicz DM, Coucke PJ, Cameron DE, Braverman AC, Byers PH, De Paepe AM, Dietz HC. Aneurysm syndromes caused by mutations in the TGF-beta receptor. *N Engl J Med* 2006;**355**:788–798.
3. Zacchigna L, Vecchione C, Notte A, Cordenonsi M, Dupont S, Maretto S, Cifelli G, Ferrari A, Maffei A, Fabbro C, Braghetta P, Marino G, Selvetella G, Aretini A, Colonnese C, Bettarini U, Russo G, Soligo S, Adorno M, Bonaldo P, Volpin D, Piccolo S, Lembo G, Bressan GM. Emilin1 links TGF-beta maturation to blood pressure homeostasis. *Cell* 2006;**124**:929–942.
4. Li W, Li Q, Jiao Y, Qin L, Ali R, Zhou J, Ferruzzi J, Kim RW, Geirsson A, Dietz HC, Offermanns S, Humphrey JD, Tellides G. Tgfbr2 disruption in postnatal smooth muscle impairs aortic wall homeostasis. *J Clin Invest* 2014;**124**:755–767.
5. Davis FM, Rateri DL, Daugherty A. Mechanisms of aortic aneurysm formation: translating preclinical studies into clinical therapies. *Heart* 2014;**100**:1498–1505.
6. Da Ros F, Carnevale R, Cifelli G, Bizzotto D, Casaburo M, Perrotta M, Carnevale L, Vinciguerra I, Fardella S, Iacobucci R, Bressan GM, Braghetta P, Lembo G, Carnevale D.

Targeting interleukin-1β protects from aortic aneurysms induced by disrupted transforming growth factor β signaling. *Immunity* 2017;**47**:959–973.e9.
7. Song T, Zhao S, Luo S, Chen C, Liu X, Wu X, Sun Z, Cao J, Wang Z, Wang Y, Yu B, Zhang Z, Du X, Li X, Han Z, Chen H, Chen F, Wang L, Wang H, Sun K, Han Y, Xie L, Ji Y. SLC44A2 regulates vascular smooth muscle cell phenotypic switching and aortic aneurysm. *J Clin Invest* 2024;**134**:e173690.
8. He ZS, Ijpma AS, Vreeken D, Heijsman D, Rosier K, Verhagen HJM, de Bruin JL, Brüggenwirth HT, Roos-Hesselink JW, Bekkers JA, Huylebroeck DFE, van Beusekom HMM, Creemers JWM, Majoor-Krakauer D. The proprotein convertase FURIN is a novel aneurysm predisposition gene impairing TGF-β signalling. *Cardiovasc Res* 2024:cvae078. doi:10.1093/cvr/cvae078.
9. Da X, Li Z, Huang X, He Z, Yu Y, Tian T, Xu C, Yao Y, Wang QK. AGGF1 therapy inhibits thoracic aortic aneurysms by enhancing integrin alpha7-mediated inhibition of TGF-beta1 maturation and ERK1/2 signaling. *Nat Commun* 2023;**14**:2265.
10. Massague J, Sheppard D. TGF-beta signaling in health and disease. *Cell* 2023;**186**: 4007–4037.