Pflügers Archiv - European Journal of Physiology (2019) 471:1245–1246
https://doi.org/10.1007/s00424-019-02308-w

COMMENTARY

Unconventional eNOS in pulmonary artery smooth muscles: why should it be there?

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Received: 13 August 2019 / Revised: 13 August 2019 / Accepted: 29 August 2019 / Published online: 5 September 2019

It is needless to emphasize the fundamental role of endothelial nitric oxide synthase (eNOS, NOS3) in vascular physiology, such as in vascular resistance control and pathophysiology [6, 12]. Ever since the concept of endothelium-dependent relaxing factor (EDRF) was proposed by Furchgott and Zawadzki [7], it was concluded that stimulated endothelial cells produce NO that diffuses to the vascular smooth muscle cells (VSMC) to induce vasorelaxation [13]. Among the three isotypes of NOS (i.e., NOS1–3), eNOS corresponds to NOS3 while the neuronal (nNOS) and inducible NOS (iNOS) correspond to NOS1 and NOS2, respectively. Although the initial nomenclature of NOS isotypes originates from early studies of the tissue-specific expression, such convention is not always valid. Multiple isotypes are co-expressed in the same cells, such as skeletal and cardiac myocytes. The myocardial and skeletal muscle-derived NO participates in the regulation of contractile function and energy production [5, 14, 17].

The expression of NOS isoforms in vascular smooth muscle cells (VSMC) has been relatively rarely investigated: the expression of nNOS (NOS1) or other isotypes in VSMC have been proposed [1, 3, 4]. Regarding their functional implications, an earlier study claimed that the NO released from VSMC appeared in a functionally insignificant amount [16] whereas more recent studies have suggested that the VSMC-derived NO accounts for vasodilation even in endothelium-denuded conditions [2, 8]. Nevertheless, due to the overwhelming reports on the importance of endothelium-derived NO, the physiological implication of VSMC-derived NO has not gained much attention yet.

In this issue of Pflugers Archiv, Kim et al. [9] have proposed an intriguing role of the VSMC eNOS in the pulmonary artery (PA). The rat PA showed only a transient contraction to angiotensin II (AngII), which owes to the concomitant activation of the eNOS in PA myocytes that express significantly higher eNOS than the systemic arteries. Furthermore, recovery from the tachyphylaxis of type 1 AngII receptor (AT1) appears to be reversed by eNOS inhibition in the endothelium-denuded rat PA. The putative physiological role of VSMC eNOS in PA does not seem to be restricted to the modulation of AngII contraction. In their previous study, it was demonstrated that the combined stimulation of PA with increased wall tension (stretch) and thromboxane A2 may have also activated the VSMC eNOS [10].

What is the physiological implication of the eNOS in the medial layer of PA? Since the large amount of pulmonary circulation is operating with distinctively low arterial pressure, the relatively prominent role of VSMC eNOS might be underlying the characteristic low resistance of PA resistance. The activation of intrinsic eNOS by the vasoactive agonists may counterbalance the excessive constriction of PA. Experimentally, the compromised contraction could be acutely revealed with the pharmacological inhibition of eNOS as shown in their studies [9, 10]. Also, Kim et al. demonstrated that the nNOS (NOS1)- and iNOS (NOS2)-specific inhibitors did not affect the contractile response to AngII and thromboxane A2, indicating the specific role of eNOS along with the expression patterns proven by immunohistochemistry [9].

Despite the interesting role of eNOS in VSMC, the pharmacological inhibitor-based approach of endothelium-denuded vessels has always countered several issues questioning the reliability of experimental conditions. Firstly, a kind of contamination occurs from the residual endothelial cells, and secondly, plausible generation of reactive oxygen species and cytokines takes place in response to the partial destruction of vascular wall integrity during endothelial denudation. In fact, the second possibility was raised investigating the role of VSMC eNOS in systemic arteries. Superoxide molecules, known as NO scavengers, appeared to impair the vasodilator responses to endogenous NO in rat systemic and

This article is a Commentary to the original article https://doi.org/10.1007/s00424-019-02305-z

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pulmonary arteries [2]. Thirdly, there is an issue with the specificity of pharmacological agents to dissect the signaling pathways downstream to AngII receptor and eNOS. To overcome this concern, genetic knock-out of smooth muscle-specific eNOS is required. Despite the technical limitation of myography studies using mouse PA with very small diameters, further investigation of such animal models is ardently needed.

Lastly, but most importantly, it remains elusive whether the significant role of VSMC eNOS is also valid in the human PA. Although Kim et al. unequivocally showed eNOS phosphorylation by AngII in the human PA smooth muscle cell-line cells, direct evidence of primary tissue is lacking [9]. Recently, downregulatory changes of VSMC eNOS in systemic hypertension model and histone modification have been reported [11, 15]. Therefore, future studies demonstrating VSMC eNOS in human PA and their disappearance in pathological conditions, e.g., pulmonary arterial hypertension, would be a highly attractive goal.

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