Hypoxia and erythropoietin (EPO) were a couple from the very beginning. Hypoxia induces the production of EPO, which became the paradigm for oxygen-regulated gene expression [1]. Dissecting EPO gene regulation has led to the discovery of the transcription factor complex hypoxia inducible factor-1 (HIF-1), the widespread “master regulator of O₂ homeostasis” in the tissue, and the identification of cellular oxygen sensors in control of HIF-1 (reviewed by [2]). EPO as a erythropoietic hormone is mainly synthesized by the kidneys in the adult, and in the liver before birth [1]. However, EPO is also expressed in the brain, in the gonads and female reproductive tract, in the bone marrow, and in tumor cells [1] due to tissue specific transcription factors, e.g. WT1 [3], which are most likely integrated into oxygen-dependent regulation.

It is now well recognized that EPO has a much broader range of action than its well-known role as an erythropoiesis-stimulating hormone [4]. EPO inhibits apoptosis in neurons, endothelial smooth muscle cells, and cardiomyocytes, and EPO expression in organs other than the kidneys appears to act in a paracrine way [5]. While signaling of EPO through erythropoietin receptor (EPOR) activation in erythroid progenitors is well characterized, knowledge of the signal transduction in other EPO-responsive cells is still fragmentary. EPO-induced signaling in neurons was suggested to serve as a paradigm to better understand cardioprotection by EPO [5]. However, extensive studies of the signaling pathways responsible for neuroprotective effects have not provided a consensus whether activation of the classical EPOR alone is responsible for neuroprotection [6,7].

In this issue, Burger et al. contribute a new piece to the puzzle of how EPO protects the heart from hypoxia-induced apoptosis [8]. EPO has been found to reduce apoptotic cell death of cardiomyocytes in vitro and in vivo (references in [8]). Burger et al. induced cardiomyocyte apoptosis in vitro by norepinephrine treatment and in vivo by myocardial ischemia and reperfusion. In both cases, apoptosis was prevented by co- and pre-treatment with recombinant EPO. As an important control experiment, the authors applied heat-inactivated EPO to exclude cardioprotective effects of other ingredients in the EPO preparations. Central to the protective effects was the induction of endothelial NO synthase (eNOS) in cardiomyocytes. EPO induced the expression eNOS mRNA and protein as well as activity of the enzyme demonstrated by increased eNOS phosphorylation and NO synthesis in vitro. Pharmacological inhibition of eNOS by L-NAME in vitro abrogated the protective effect. Likewise, and importantly, eNOS knockout mice were less protected by EPO than wild-type animals upon cardiac ischemia and reperfusion [8].

How does EPO exert these effects? As pointed out above, for neuroprotection the activation of a heterodimeric receptor consisting of the classical EPOR and the common β-chain from the IL-3 receptor has been proposed [6]. This may hold true for therapy using high doses of recombinant EPO but not for much lower concentrations of the endogenous hormone. Tissue-specific knockout of the EPOR revealed the importance of endogenous EPO and signaling through EPOR for post-stroke neurogenesis [7]. A similar role for the endogenous EPO/EPOR system in cardioprotection has recently been suggested [9].

The present article by Burger et al. may close the gap between EPO-induced signaling and protection of the heart [8]. EPO signaling is known to protect cardiomyocytes through the PI3-kinase/Akt- as well as the ERK-dependent pathways [10]. Burger et al. now show that at least in vitro...
signaling through PI3-kinase and Akt is important for EPO-induced eNOS activation and that NO is the key anti-apoptotic factor. Similarly, it was recently found that EPO prevents doxorubicin-induced cardiomyopathy through Akt signaling, underlining the importance of these signaling pathways for EPO’s cardioprotective effects [11]. Of note, Li et al. reported a significant reduction of GATA-4 by doxorubicin that was completely reversed by EPO treatment [11]. Since GATA-4 is also an important transcription factor for tissue-specific expression of the EPO gene [12], it is tempting to speculate whether the endogenous EPO/EPOR system is affected in other disease models through effects on GATA-4.

Effects of EPO on NO synthase expression and activity have been described almost as long as EPO has been used for treatment. Hypertensive effects of EPO therapy were attributed to the selective suppression of eNOS in endothelial cells [13]. In contrast, in a mouse model of chronic polycythemia due to transgenic EPO overexpression, blood pressure was within the normal range [14] due to EPO-induced overexpression of eNOS in the vessel wall [15]. Thus, EPO potentially targets the cardiovascular system in a tissue-specific manner, which needs to be considered when EPO treatment for cardiovascular ischemia is envisioned [4]. This may, in particular, require meticulous studies on the hemostatic system in patients with cardiovascular disease and underlying risk factors. As promising as the study by Burger et al. is, questions from a previous review [5] on the cardioprotective effects of EPO may be readdressed: (1) Does EPO only effectively protect cardiomyocytes when eNOS is induced before or simultaneously with the apoptotic stimulus? (2) Does the protective effect of eNOS induction extend over the acute phase, or are additional mechanisms responsible for longer-lasting protection by EPO? (3) Is cardiomyocyte eNOS the key to EPO protection or, as suggested by Burger et al., is it one mechanism of a combination of effects?

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References