Dickkopf-3: a stubborn protector of cardiac hypertrophy

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This editorial refers to ‘Dickkopf-3 attenuates pressure overload-induced cardiac remodelling’ by Y. Zhang et al., pp. 35–45, this issue.

In response to haemodynamic overload under pathological conditions such as hypertension, valvular heart disease, and myocardial infarction, the heart undergoes hypertrophic growth by increasing cell size and protein synthesis as well as changing the transcriptional programme in cardiac myocytes.¹ Although cardiac hypertrophy is beneficial in the way that it reduces ventricular wall tension and maintains pump function, it promotes cardiac structural remodelling and dysfunction, and eventually leads to development of congestive heart failure, arrhythmia, and sudden death. At present, several drugs such as inhibitors of the renin–angiotensin–aldosterone system, β-adrenergic receptor blockers, and calcium channel blockers are clinically available for the management of hypertension, and these drugs have shown significant efficacy in preventing load-induced cardiac hypertrophy and remodelling. However, these pharmacological agents are currently of limited effectiveness, and further discovery and development of novel classes of cardioprotective drugs are in urgent need.² For that purpose, it is important to elucidate the molecular mechanisms underlying the development of cardiac hypertrophy. In the article by Zhang et al.,³ integrating genetic approaches in mice have shed light on Dickkopf-3 (DKK3) as a cardioprotective regulator of cardiac hypertrophy.

DKK3 is a secreted glycoprotein of the Dickkopf family that typically antagonizes the Wnt/β-catenin signalling by interfering with Wnt co-receptors, low-density lipoprotein receptor-related protein and kremen.⁴ ‘Dickkopf’ is a German word for ‘big head’ or ‘stubborn’, and the protein family owes its name to the finding that DKK1 is sufficient and necessary to induce head formation in Xenopus embryo.⁵ Zhang et al.³ first demonstrated that cardiac expression of DKK3 was down-regulated in patients with end-stage heart failure and in mice with pressure-overloaded cardiac hypertrophy. In neonatal rat cardiac myocyte cultures, angiotensin II-induced hypertrophic responses were enhanced by siRNA-mediated knockdown of DKK3, and conversely were attenuated by overexpression of DKK3. Consistently, cardiac hypertrophy following aortic banding in mice was enhanced by genetic disruption of DKK3, and was attenuated by transgenic overexpression of DKK3. These loss- and gain-of-function analyses, both in vitro and in vivo, indicated the regulatory role of DKK3 in protecting the heart from the development of pathological cardiac hypertrophy. The next obvious question is how DKK3 affects the signalling effectors underlying pathological hypertrophy and remodelling of the heart. Zhang et al. demonstrated that DKK3 inhibited the activation of apoptosis signal-regulating kinase 1 (ASK1), and thereby suppressed the activation of its downstream effectors c-Jun N-terminal kinases (JNKs) and p38 mitogen-activated protein kinases (MAPKs) in hearts subjected to hypertrophic stimulation (Figure 1). Importantly, DKK3 overexpression attenuated the activation of ASK1 in pressure-overloaded hearts of mice, and restoration of ASK1 activity by transgenic overexpression abolished the protective effects of DKK3 overexpression against pressure overload-induced cardiac remodelling. Conversely, disruption of DKK3 enhanced the ASK1 activation in pressure-overloaded hearts, and inactivation of ASK1 by transgenic overexpression of a dominant negative mutant prevented exaggeration of pressure overload-induced cardiac remodelling in DKK3-deficient mice.

ASK1 is a key component of a high molecular mass complex, termed the ASK1 signalosome, and is activated in response to a variety of cellular stresses, such as reactive oxygen species (ROS), endoplasmic reticulum (ER) stress, calcium overload, and inflammatory signals mediated by tumour necrosis factor-α (TNF-α) and lipopolysaccharide.⁶ While the ASK1 activity is suppressed by the reduced form of thioredoxin (Trx) (a redox-sensitive protein) within the ASK1 signalosome, oxidation of Trx in response to ROS induced autophosphorylation and oligomerization of ASK1, leading to its activation. Upon ROS-stimulated activation, adaptor proteins, such as TNF-α receptor-associated factor 2 (TRAF2) and TRAF6, and USP9X deubiquitination enzyme are recruited to the signalosome to maintain full activation of ASK1.⁵ The next obvious question is how DKK3 affects the signalling effectors underlying pathological hypertrophy and remodelling of the heart. Although the precise mechanism by which DKK3 inhibits ASK1 activation remains unclear, Zhang et al.³ demonstrated, by co-immunoprecipitation experiments, that exogenously expressed DKK3 and ASK1 formed a complex in HEK293 T cells, and furthermore showed direct interaction between endogenous DKK3 and ASK1 in neonatal rat cardiac myocytes. These results suggest that DKK3 may interfere with ASK1 activation via the physical interaction with ASK1 within the signalosome (Figure 1), but...
clearly leave many open questions: which domains of these two proteins are responsible for mutual binding? How does DKK3 influence the composition of proteins within the ASK1 signalosome in the presence or absence of ROS? As mentioned above, DKK3 is a secreted protein, and possesses an N-terminal signal sequence. Recent studies have shown intracellular localization of DKK3, and such localization can be explained by the fact that the signal sequence is readily recognized by the signal recognition particle that catalyses the transport of the secretory precursor protein into the ER lumen. Indeed, immunofluorescence staining indicated localization of DKK3 at perinuclear structures such as ER and Golgi apparatus, but direct interactions between DKK3 and cytosolic proteins such as βTrCP and dynein light chain Tctex-1 were also reported. Although Zhang et al. showed an intracellular distribution of DKK3 by immunohistochemical analysis of human hearts, further studies are necessary to determine where and how DKK3 forms a complex with the cytosolic protein kinase ASK1 in cardiac myocytes.

It also remains unsolved whether antagonization of the Wnt/β-catenin signalling is involved in the cardioprotection by DKK3 against hypertrophic stimulation. Although there have been conflicting data on the effects of β-catenin stabilization on pathological hypertrophy in mice, Noh et al. recently reported that Wnt-3a negatively regulates the protein level and kinase activity of ASK1 by inhibiting glycogen synthase kinase-3β in murine L929 fibrosarcoma cells. Since the ability of DKK3 to inhibit the Wnt/β-catenin signalling appears to be context dependent, it is of interest to investigate the impact of loss- and gain-of-function of DKK3 on the Wnt/β-catenin signalling in cardiac myocytes under stressed conditions.

Accumulating evidence has indicated that DKK3 is down-regulated in human cancer cells, and DKK3 emerges as a potential tumour biomarker and a promising molecular target for cancer therapy. The study of Zhang et al. provides an initial but important clue towards a new strategy to modulate the DKK3 function for treatment of heart diseases. Of course, further studies are required to explore more precise mechanism of DKK3-mediated cardioprotection and to develop an optimal way to restore the expression and function of DKK3 in stressed hearts.

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