

Neointima formation after vascular injury: Is it all about CD39?

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Since the pioneering era of interventional cardiology, platelet deposition after vascular injury has been regarded of central importance for neointima formation and subsequent restenosis (1–3). Depending on the type of injury, i.e. how many layers of the vessel wall are affected, surface-adherent platelets can be distinguished from mural thrombus extending into the torn media (4). Whereas organised thrombi can sometimes cause occlusion and restenosis, platelets adherent to the subendothelial matrix constitute the main adhesive substrate for the recruitment of leukocytes and smooth muscle progenitor cells to the injury site driving neointimal growth (5–8). P-selectin expressed on activated platelets, rather than platelet aggregation, is decisively involved through mediating adhesive interactions and the deposition of pro-inflammatory chemokines in a PSGL-1-dependent manner (9–12). In contrast to the compelling experimental evidence, however, most antiplatelet compounds have hitherto failed to prevent neointimal hyperplasia in the clinical setting (13–15). This mandates a search for new antiplatelet strategies, which specifically target platelet functions involved in the response to vascular injury (16, 17).

In the current issue of *Thrombosis and Haemostasis*, Drosopoulos et al. evaluated the role of a recombinant and soluble form of human CD39 (solCD39) in a murine model of neointima formation after wire-induced denudation of the femoral artery

by targeting platelet function (18). CD39 is a cell surface ADPase expressed on leukocytes and endothelial cells, which degrades nucleotides tri- and/or diphosphates and thereby terminates the aggregation response of platelets to adenosine diphosphate (19). The isolation of a 439 amino acid polypeptide from the extracellular domain of CD39 yielded an injectable protein which retained ATPase activity *in vivo* (20). The most astonishing finding of Drosopoulos et al. is the complete suppression of neointima formation and the absence of vascular inflammation, as evident by VCAM-1 expression and macrophage recruitment, after 19 days in solCD39-treated mice, indicating a highly active pharmacological compound in this disease model (18). Moreover, Drosopoulos et al. found that solCD39 treatment basically preserved the vessel wall architecture enclosing re-endothelialisation. These findings are in line with reports demonstrating a protective role of CD39 locally over-expressed at the injury site in neointima formation (21, 22). The infusion of solCD39, however, obviates gene transfer as a therapeutic approach making it much more attractive. Of note, CD39^{-/-} mice have recently been shown to be also protected from neointimal hyperplasia (23), which might be explained by purinergic type P2Y1 receptor desensitisation responsible for platelet hypofunction in CD39^{-/-} mice (24).

As one might have deduced from the well established antithrombotic activity of CD39, Drosopoulos et al. reported an absence of mural thrombus formation in solCD39-treated mice, although platelets lining the luminal wall were still present. In addition, platelet P-selectin expression was markedly reduced by solCD39, which might account for the diminished recruitment of monocytes/macrophages to the injured artery. However, even in P-selectin^{-/-} mice, neointima formation is detectable at least to some degree, suggesting additional CD39-dependent mechanisms (10). Apoptosis of medial smooth muscle cells is a very

early event after vascular injury (25). Recently, caspase-dependent release of ATP has been shown to induce monocyte recruitment, which can be prevented by ectopic CD39 (26). In mice deficient in CD73/ecto-5'-nucleotidase, which sequentially catalyses the conversion of AMP to adenosine, neointima formation is significantly increased due to constitutive NF-κB activation in the absence of adenosine (27). Furthermore, the absence of the A2 adenosine receptors on bone marrow cells exacerbated neointimal growth after femoral wire-injury probably by controlling CXCR4 expression (28). In this regard, it would be interesting to examine whether the beneficial effect of solCD39 on neointima formation is impeded in CD73^{-/-} mice. Some caveats remain in the study by Drosopoulos et al. and need to be further explored. This includes the decline of the ADPase activity after 17 days of treatment, which was assigned to a neutralizing immune response by the authors. Since the mice in the control group were only treated with saline, but not with inactive solCD39, the contribution of an elicited anti-CD39 immune response to the observed effect is a matter of debate.

Thus, solCD39 infusion appears to be a valuable new anti-platelet strategy in the prevention of restenosis potentially harbouring some collateral effects associated with its potent anti-inflammatory properties. Nevertheless, future studies are warranted to fully address the mechanisms of the “holistic” impact of solCD39, before it can reach the potential status of a novel “magic bullet” in restenosis research.

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