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Gain in translation: heme oxygenase-1 induced by activated protein C promotes thrombus resolution

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The protein C pathway functions to maintain a regulated balance between homeostasis and host defense systems in response to vascular and inflammatory injury. The anticoagulant protein C pathway regulates coagulation and prevents thrombosis, whereas the cytoprotective protein C pathway employs cell signaling initiated by activated protein C (APC) to prevent cellular and organ vascular injuries. APC-initiated signaling induces up and down regulation of many genes which is translated into cytoprotection and reduction of inflammation [1–3].

In this issue, Gabre and colleagues [4] have investigated the effects of a repeated daily bolus dose of APC on thrombus resolution in a mouse model of deep vein thrombosis. Following establishment of a thrombus due to ligation of the inferior vena cava, animals were treated with APC for 8 days (from day 4 to day 11 post ligation). This treatment with APC promoted resolution of the venous thrombus as quantified at day 12. Moreover, APC significantly induced heme oxygenase-1 (HO-1) in macrophages as measured at day 12, and inhibition of HO-1 ablated the observed ability of APC treatment to promote thrombus resolution [4].

How might HO-1 be related to resolution of thrombus? HO-1 is an inducible enzyme that is expressed in most mammalian tissues, and HO-1 exerts potent anti-oxidative and anti-inflammatory effects [5]. HO-1 degrades free heme and generates carbon monoxide (CO), biliverdin, and iron [5]. These reaction products can provide anti-inflammatory and anti-oxidative functions. When a venous thrombus is formed, it is thought that reactive oxygen produced by leukocytes and/or vessel walls could oxidize hemoglobin in fibrin-trapped RBCs and release free heme [6, 7]. The free heme, possessing pro-inflammatory and oxidative properties, could contribute to the reciprocally reinforcing dyad of inflammation and coagulation, thereby promoting clotting on the thrombus surface and delaying resolution [7]. The oxidative, pro-inflammatory properties of heme would be neutralized by HO-1 activity. Thus, while HO-1 beneficial activities have been shown in many settings, HO-1 anti-inflammatory action may contribute to thrombus resolution.

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Significant roles for HO-1 in the setting of venous thrombosis have emerged in two clinical studies and in another murine venous thrombosis study. Genetic variations in the human HO-1 gene, HMOX1, were linked to increased risk for recurrence of venous thrombosis in Austrian patients [8] and for first occurrence of venous thrombosis in Black Americans although not in Caucasian Americans [9]. In a murine venous thrombosis study complementary to that of Gabre et al [4], it was shown that knock-out of HO-1 resulted in impaired venous thrombus resolution and in an exaggerated inflammatory response [10].

New questions for clinical research arise based on the preclinical and clinical data linking HO-1 to enhanced thrombus resolution or altered risk of venous thrombosis [8–10]. One wonders whether efforts to achieve more effective thrombus resolution, especially for the patients with low HO-1 levels, would help to reduce the risk of recurrent thrombosis. Further, might new knowledge about factors that enhance thrombus resolution in patients yield novel knowledge about post thrombotic syndrome which is a clinically significant disorder with morbidity for the patient and a substantial expense for the health care system [11]. Since increased residual thrombosis is associated with an increased risk of recurrent venous thrombosis [12], knowledge about post thrombotic syndrome and risk for recurrence could ultimately impact the frequency and duration of secondary treatment for prevention of recurrence with systemic anticoagulation therapy, which carries a risk of bleeding for the patient. Careful long-term clinical studies with follow-up for residual venous thrombosis, post thrombotic syndrome, and recurrent venous thrombosis would be required to answer important questions about HO-1 and thrombosis in man.

In a number of preclinical studies, APC and HO-1 are similarly engaged in homeostasis and cytoprotection and they appear to speak the same language of host protection. Each protein is linked to the anti-inflammatory cytokine, IL-10 [13, 14]. Each protein reduces organ damage caused by ischemia/reperfusion injury of the brain [15–18], heart [19–21], and kidney [22, 23], and each protein reduces morbidity and mortality in one or more models of sepsis [24–28]. Based on the present study in which APC induce HO-1 in macrophages, the question arises of whether some dimension of APC's protective effects that resemble those of HO-1 might be mediated, in part, through an up-regulation of HO-1 by APC. However, since Gabre and colleagues only tested macrophages for HO-1 up-regulation by APC [4], it is not clear if APC induction of HO-1 in other cells contributes to APC's therapeutic promise. Other inducers of HO-1 often pose the problem of off-target deleterious side effects [29]. The major deleterious side effect of recombinant APC is serious bleeding, and risk for serious bleeding has been reduced by engineering an APC variant with three amino acid changes (3K3A-APC) that selectively reduces anticoagulant activity but not cytoprotective signaling. This APC variant is fully neuroprotective in preclinical studies [18] and it is under development for clinical trials in man [30]. It will be interesting to determine the extent to which this 3K3A-APC variant induces HO-1 and then whether HO-1 induction contributes to APC's pharmacologic benefits.

In summary, Gabre and colleagues have discovered that APC promotes venous thrombus resolution and that up-regulation of HO-1 contributes to this effect [4]. This new APC function as an HO-1 inducer may represent a significant therapeutic aspect of APC pharmacology.

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