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Insights from experimental post-thrombotic syndrome and potential for novel therapies

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Clinical Background and the Problem

Post-thrombotic syndrome (PTS) is a common sequela after lower extremity deep vein thrombosis (DVT),^{1, 2} and like atherosclerosis, is both driven by inflammation and generates inflammation as well.^{3, 4} PTS affects 6 to 7 million patients,^{5, 6} and between 400,000 and 500,000 patients have skin ulcerations, the most severe and costly manifestation of the disease.^{3, 7} Quality of life is negatively impacted in patients with PTS.⁸ The severity of PTS is related to the extent and location of the DVT⁹ as well as other clinical factors, including patient's sex, weight, and prior thrombotic history.^{10, 11} PTS is a challenging problem because the vein wall becomes irreversibly fibrosed by the resolving DVT, often with complete luminal obstruction, and externally manifests with skin changes, sometimes leading to ulceration in the lower limbs.¹² The estimated cost of treating PTS is significant, and amounts to billions of dollars yearly.¹³ Few therapies directly address PTS, and once established, therapy is primarily palliative.¹⁴ While impaired thrombus resolution in humans correlates with PTS,^{7, 15, 16} no therapies reverse the damage to the vein wall.^{6, 17} This review will focus primarily on the experimental insights into PTS, and will not review venous thrombosis basics.

Current Medical and Interventional Therapy

Human studies of vein segments by duplex ultrasound after DVT show changes in the vein wall architecture, including focal thickening, even with full thrombus resolution.¹⁸ Beyond obstruction there are no specific duplex findings that correlate with the magnitude of PTS,¹⁹ such as valvular reflux. Pathological specimens from chronically occluded femoral vein show extensive collagen and neovessels,²⁰ similar to what is found in scar tissue, and these changes correlate with the chronicity of thrombus age. These human data suggest a complex, multi-factorial pathobiology of PTS.

Primary therapies to prevent PTS include rapid and therapeutic anticoagulation to promote dissolution of, and prevent recurrence of, DVT.^{6, 12} The most common anticoagulation regimens are low molecular weight heparin (LMWH) followed by a direct oral anticoagulant

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(DOAC), a DOAC alone as monotherapy at higher and then lower doses, or less commonly, LMWH or unfractionated heparin (UFH) followed by Warfarin.²¹ Limitations of compliance with Warfarin dosing result in sub-therapeutic levels,²² and sub-therapeutic Warfarin is associated with a 78% increased PTS risk.²³ As compared with the Warfarin, the DOAC's don't have such constraints in part due to more consistent anticoagulation levels. DOAC's are associated with improved deep venous recanalization and a lower incidence of PTS.^{24, 25} It is not known yet known if one DOAC is better than another in terms of the decrease in PTS observed from these studies. Further prospective studies may provide better evidence, but is likely moot given the increased DOAC use for most all VTE patients. However, the DOACs direct effect on the vein wall remains unknown.

In addition to pharmacologic therapy, rapidly applied graded compression (at least 30–40 mm Hg) within 24 hours after DVT diagnosis may reduce residual venous obstruction, which contributes to PTS.²⁶ However, results from a randomized controlled trial of a placebo controlled compression failed to show a decrease in incident PTS, as measured by the Villalta score and the Ginsberg PTS score.²⁷ Thus, it is reasonable to use compression for acute DVT, and likely most effective when placed within 24 hours of diagnosis. Further studies may better clarify timing and compression strength, but this is unlikely to significantly impact the disease process,²⁶ particularly given patient compliance issues.

Active pharmacomechanical thrombolysis has mixed results in preventing PTS. For example, the CaVenT study showed an early and late statistical reduction in PTS, but with an increased bleeding risk.^{28, 29} Conversely, the ATTRACT trial showed no reduction of PTS, although a modest benefit signal was observed in those patients with iliofemoral DVT.³⁰ Thus, rapidly opening a recently thrombosed deep vein may not prevent PTS, suggesting early intrinsic vein wall damage, possibly due to alteration in venous compliance, damage to venous valves resulting in reflux, and institution of cellular injury programs. Consistently, residual or non-dissolved thrombus contributes to PTS, post lysis.³¹ In fact, 'residual thrombus' observed by venogram or intravascular ultrasound, may be scar tissue which will not lyse, and correlates with PTS risk in humans.^{16, 31} Experimentally, the physical thrombus adjacent to the vein wall also directly affects physiological function, with impaired venous relaxation, but not vein wall contraction, with just 2 days of exposure.³² This suggests one mechanism whereby PTS may develop even in the case of thrombus resolution, and maybe due to prolonged endothelial dysfunction. Further experimental studies will better define this mechanism.

Late manifestations of PTS are primarily swelling and skin changes, including ulceration. Ablation of refluxing superficial veins in the ipsilateral vascular bed should be done for venous leg ulcer treatment, if the deep system is patent, and is supported by level I evidence.^{33, 34} However, it is not clear there is any benefit in treating superficial reflux to prevent PTS prior to ulceration. Endoluminal therapies for proximal venous obstructive disease may be beneficial in improving venous ulcer healing as well as symptomatology.^{12, 35} However, there is no level I evidence that venous angioplasty and stenting improves long term PTS outcomes.³⁶ A current trial addresses this question, called the C-TRACT trial ([NCT #3250247](#)). Beyond these measures, there is no distinct pharmacologic therapy that decreases PTS. Use of venotonics such as micronized purified flavonoid fraction³⁷ (MPFF)

as well as pentoxifylline¹² do aid in venous ulcer healing, and rutosides may improve edema and symptoms such as leg restlessness, but strong evidence is lacking.³⁸ MPFF may improve pain and swelling to some degree, but this effect is non-specific and observed amongst patients with and without venous disease. Lastly, structured exercise may decrease PTS associated quality of life measures, and is highly recommended in general.³⁹

PTS pathobiology from experimental models of VT

It is beyond the scope of this review to detail the clotting cascade specifics, but in general, DVT is thought to start at the inner area of venous valve cusps⁴⁰ that physically thickens with aging.⁴¹ At these high risk locations, thrombus propagation can occur, likely promoted by impaired oscillatory shear stress, and loss of antithrombotic endothelial surface phenotype. The antithrombotic phenotype is maintained by high levels of the antithrombotic proteins thrombomodulin, endothelial protein C receptor, and tissue factor pathway inhibitor, and low levels of prothrombotic proteins von Willebrand factor, P-selectin, and intracellular adhesion molecule (ICAM)-1.⁴² Furthermore, the hypoxemic micro-environment present in the valve cusp may also drive changes in the endothelial phenotype, thereby promoting a procoagulant state.⁴³ What actually tips the balance to the formation of a clinically detectable DVT is not known.

Human DVT and murine models

Imaging of DVT in humans suggests that there are regions of total stasis and non-stasis within the same affected venous segment, such as the femoral or iliofemoral vein.^{9, 18, 44} Thus, two models of venous thrombosis (VT), stasis and non-stasis, are most representative for what occurs in humans. Human specimen evaluation of chronically occluded femoral vein sections has shown a collagen rich and cellular histology, similar to the experimental histological appearance noted in chronic mouse models of late VT.^{45, 46}

Venous thrombosis (VT) and its resolution is an inflammatory process.^{47–51} Experimental DVT models suggest the vein wall injury is dependent on the mechanism of thrombogenesis (stasis or non-stasis), the duration of thrombus-vein wall contact, and other factors such as pro-inflammatory cytokines, chemokines, and matrix metalloproteinases (MMPs).^{47, 52} Specifically, vein wall fibrotic injury is modulated *in part* by plasminogen activator inhibitor (PAI-1), urokinase-type plasminogen activator (uPA), matrix metalloproteinase (MMP) –2 and –9, Toll-Like Receptor-9 (TLR9), and cysteine-cysteine receptor-7 (CCR7).^{45, 53–60} However, while vein wall fibrosis was variably altered with global genetic deletions of the above mediators and receptors, concurrent increase in thrombus size often occurred and makes these targets less than ideal therapeutically. Interestingly, the size of the VT has less impact on the vein wall response than the mechanism of thrombogenesis and cellular milieu;^{57, 58, 61} suggesting the critical importance of leukocyte-vein wall-matrix interactions.

Recurrent DVT is a strong risk factor for PTS.¹² A recently published model was developed that may allow insights into therapies – and suggest the vein wall is primed for a fibrotic phenotype, with increased levels of transforming growth factor-beta (TGFb), interleukin-6 (IL-6), and MMPs in the secondary (e.g. recurrent) VT post- thrombotic vein walls.⁶² This

study also delineated the incorporation of the primary thrombus into the vein wall, contributing to the thickness and architecture of the post- thrombotic vein.

In-depth assessment using a rat model with several inferior vena cava permutations, including full venous stasis (ligation of IVC), limited venous stasis (clip on and removed after 2 days), non-stasis thrombosis, or non-thrombotic IVC occlusion using an inert silicone plug clarified several temporal and mechanistic aspects of vein wall remodeling.⁶¹ First, the vein wall stiffness was greatest with prolonged thrombus apposition, as compared to limited stasis. Collagenolysis was greater with 7 days of stasis injury, but neither MMP-2 or MMP-9 activity directly correlated with these injury mechanisms in the rat, suggesting the action of yet to be delineated mediators of collagen remodeling. Vein wall cellular proliferation paralleled stasis thrombotic injury, and was greatest in prolonged stasis. Non-thrombotic IVC occlusions showed lesser inflammatory responses, suggesting that the thrombus directs the vein wall injury via multiple mechanisms.

Leukocyte Roles

Venous thrombus resolution resembles sterile wound healing,^{63, 64} with well- regulated phases of neutrophil (polymorphonuclear leukocytes, PMN) and monocyte/macrophage (Mo/MΦ) influx, followed by later fibrosis. Concurrently, the post-thrombotic vein walls stiffen and thicken with interstitial matrix and myofibroblast accumulation, likely mediated by these leukocyte and other vein wall cellular interactions (Figure).^{57, 58, 61, 65, 66}

Polymorphonuclear cells (PMN, Neutrophils) are the first leukocyte to invade the VT after it forms. The role of the PMN is multifactorial and dictated by the thrombotic model, and the environment, as PMN depletion may not affect thrombogenesis, may increase VT size, or may be associated with smaller VT, depending on the experimental model and setting.^{67–69} Neutrophils release several factors, including plasminogen activators that may promote thrombus resolution, but in a different rodent model may promote thrombosis, via neutrophil extracellular traps (NETs).⁷⁰ PMNs may also affect the vein wall injury response, and interestingly, with PMN depletion, later time point vein walls were thicker and stiffer.⁶⁷ This suggests that early PMN processes in the thrombus are important for normal vein wall healing.

Monocytes/macrophages are the critical second leukocyte type to infiltrate the thrombus, beginning at day 4 experimentally, and these cells peak at day 8 in a mouse model of complete stasis thrombosis.^{71, 72} However, Mo/MØ are not required for early stasis or non-stasis thrombogenesis, as suggested from conditional Mo/MØ (CD11b-DTr) conditional depletion model.⁷³ The micro-environment of the VT seems to determine whether the Mo/MΦ is pro-inflammatory that drives tissue damage or pro-resolution/healing that promotes tissue homeostasis.^{74–80}

Mo/MΦ drive VT resolution by clearance of apoptotic and necrotic cells and matrix debris,^{64, 70, 80–82} as well as having pro-fibrinolytic activity and promoting thrombus neovascularization.^{61, 83–86} These processes may directly affect the vein wall injury response. Mo/MΦ are classified by their inflammatory or anti-inflammatory functions.^{87–89} For example, interleukin-1 (IL-1), IL-12 secreting and cell surface Ly6C^{hi}, CCR2⁺⁺,

CX₃CR1⁺ antigen expression characterize classically activated, or pro-inflammatory Mo/MΦ. Conversely, IL-10 secreting, *Nr4a1* transcription factor dependent, and cell surface Ly6C^{lo}, CCR2⁻, CX₃CR1⁺⁺ antigen expression, characterize alternatively activated anti-inflammatory Mo/MΦ, with pro-healing and inflammation-resolving activities. It is important to note that this process is highly dynamic in tissues, and a pure M₁ (pro-inflammatory) or M₂ (anti-inflammatory) phenotype derived in cell culture is not replicated *in vivo*.^{76, 78, 90, 91} Nonetheless, early proinflammatory Mo/MΦ activities such as uPA release,⁸³ and factors such as p53 mediated downstream⁹² effects are likely important early for resolution and set the stage for later healing.

A recent study in a VT non-stasis model showed that Mo/MØ peak by 6 to 10 days.⁹³ Utilizing a lysM⁺ leukocyte conditional depletion strategy, thrombosis resolution was enhanced when this cell line was depleted, with decreased PMNs and increased IL-4 and IL-10, suggesting a shift to a pro-healing response. To further investigate this, Tbx21^{-/-} mice, lacking ability to produce interferon gamma (IFN-γ), are skewed toward a TH₂ immune response. Post-thrombosis, these mice had a decreased thrombus collagen, increased neovascular channels, and decreased IL-12 and CCL-2 at day 14 in the same non-stasis model. The response of the vein walls was not directly examined.

Other work *indirectly* supports the context dependent role of Mo/MΦ in VT resolution and vein wall injury, including: A). Both CXCR2 and CCR2 signaling in VT resolution are important for Mo/MΦ recruitment and activation in VT resolution;^{71, 72} B). P-selectin inhibition is associated with decreased vein wall fibrosis and increased Mo/MΦ in the vein wall;⁹⁴ C). Genetic deletion of the plasmin-MMP-2/9 system decreases vein wall injury, but inhibits VT resolution, and is associated with decreased vein wall Mo/MΦ;^{66, 84, 95} D). Late vein wall remodeling is dependent on TLR9 signaling in Mo/MΦ, supporting their role in cell debris clearance and VT resolution;⁹⁶ E). Early and mid-time-point Mo/MΦ phenotypes vary depending on the VT models, including circulating Mo markers in humans.⁹⁷

The role of lymphocytes in VT resolution is now a bit clearer using a non-stasis model of VT.⁹⁸ Memory T-cells play a role in post-thrombotic vein wall inflammation with T-cells peaking late, at 21 days. Reducing primarily effector T-cells, post-thrombosis, showed reduced IL-1, IL-6, and IL-18, all inducible by IFN-γ. The T-cell depleted mice showed decreased thrombus size, decreased IFN-γ, and increased neovascular channels at 21 days. The direct role of the cells on vein wall thickening or injury was not explicitly examined.

Cellular Factors

The local venous environment post-thrombosis contains growth factors such as TGFβ, pro-inflammatory cytokines such as IL-1⁹⁹ and activation of the uPA/PAI-1/MMP axis.^{58, 95, 100, 101} Matrix remodeling enzymes have been shown through multiple studies to modulate vein wall fibrosis, primarily in the stasis VT model.⁶¹ For example, it is clear that MMP-9 regulates vein wall biomechanical function post-thrombosis. Using a stasis VT model, MMP-9^{-/-} mice had less vein wall stiffness post thrombosis, with less mononuclear cell infiltration, less collagen, yet an increased VT size (corrected for body weight).¹⁰² Deatrick et al also showed similar findings with MMP-9^{-/-} mice, with less vein wall collagen, decreased pro-inflammatory mediators (such as IL-1b), but no change in thrombus

size.⁹⁵ Consistently, the effects were both observed at the later time points of 8 to 14 day. Moreover, MMP-2 deletion was also shown in an 8d stasis VT model to have decreased procollagen-III, decreased histologically measured fibrosis, decreased TNF, and IL-1b but increased mononuclear cell influx. When combined genetic mice were tested (MMP-2/9^{-/-}), the vein wall was remarkably thinner with minimal collagen noted.⁶⁶ Conversely, the VT were larger, suggesting the role of MMP-2/9 in VT resolution, in addition to the vein wall effects. These data suggest that the MMP axis plays a role in the post-thrombotic vein wall remodeling.

Interestingly, plasmin over expression, using a global PAI-1^{-/-} mouse, was associated with increased vein wall injury despite smaller VT size.⁵⁸ This finding emphasizes the point that it is not merely the size of the thrombus that regulates the vein wall response. In PAI-1^{-/-} mice, administration of low molecular weight heparin post-thrombosis showed VT resolution was increased, with no impact on thrombogenesis. Importantly, 14-day assessment showed decreased fibrosis with LMWH and this has been recapitulated at 21 days (unpublished observation). There was no change in PAI-1 levels, but PAI-1^{-/-} mice had increased MMP-2, TIMP-1, IL-13, and increased vein wall Mo/MØ influx. This study was further expanded using PAI-1 over expressing mice that had the opposite effect, with decreased fibrosis, decreased Mo/MØ influx, and decreased MMP-2 and TIMP-1 with significantly reduced vein wall injury.

Deletion of Toll-like receptor (TLR)-9, an important receptor for sterile inflammatory processes, promotes venous thrombogenesis, and impairs VT resolution at early and mid time points (eg. 8d).¹⁰³ TLR9^{-/-} mice had decreased late endothelial recovery with increased fibrotic markers, suggesting its role in late vein wall injury.⁹⁶ Specifically, in TLR9^{-/-} mice, at mid to later time points, thrombus matrix cellular breakdown products such as uric acid, cathepsin-G, HMGB₁ were all increased as compared with controls. Thrombus size at 21 days was similar, but increased makers of fibrosis and decreased endothelial markers of the post-thrombotic late vein segment suggest that this pathway is important for normal vein wall healing.

Endothelial to mesenchymal transformation (EndMT) is well documented in certain models of arteriopathies, but may also be important in the modulation of vein wall fibrosis. Deletion of CCR7 signaling may accelerate EndMT in a non-stasis VT model, while having no significant effect in the more common stasis model.⁴⁵ Others have shown, using a non-stasis VT model, that endothelin-1 may also modulate TGFβ₁ – TGFβ₁-R signaling, driving later vein wall fibrosis, as well as decreased VT resolution at 21 days.¹⁰⁴ Whether these targets would allow therapeutic decrease in fibrosis is not known yet.

Selectins are cell adhesion molecules that are involved with early VT genesis and may be involved with vein wall injury. Selectin inhibition, such as P-selectin inhibition, is associated with decreased vein fibrosis experimentally in rats, as well in primates.^{101, 105} In mice genetically deleted for E-selectin, P-selectin, and E/P-selectin together, showed a significant correlation in decreased fibrin deposition and decreased thrombus mass in a stasis VT model.¹⁰⁶ Exogenous inhibition of P-selectin via rPSGL-1 was shown to be associated with less vein wall fibrosis in a rat model,⁹⁴ but with increased vein wall Mo/MØ, suggesting

potentially a pro-healing phenotype of these leukocytes. However, the exact monocyte/macrophage phenotype has not been defined with selectin inhibition in the venous milieu.

Translational Avenues

Several potential avenues to decrease vein wall fibrosis have been shown experimentally to be effective. A currently used anticoagulant, LMWH, may have anti-inflammatory effects beyond the anticoagulant mechanism as stated previously. Consistently, LMWH may be more efficacious than Warfarin in preventing PTS, and is recommended in those with iliofemoral DVT.¹⁰⁷ The downside for patients is the need for daily injections rather than an oral route. There is no experimental data regarding DOAC effects on vein wall fibrosis or inflammation, although as stated, clinical data is emerging, suggestive of a protective benefit.

HMG-Co reductase inhibitors (statins) are perhaps the more appealing and translatable potential therapy to decrease the severity of PTS.¹⁰⁸ Statin use has been associated with decreased incident DVT.¹⁰⁹ Modulating some of the aforementioned vein wall injury factors underlies the beneficial effect of statins on vein wall injury in mice.¹¹⁰ Statins administered after VT in full and partial stasis models decreased vein wall collagen at 8 day and 21 day time points,¹¹⁰ and statin administered pre-thrombosis was associated with reduced venous thrombogenesis,¹¹¹ decreased circulating PAI-1, IL-6, and thrombus PMNs. Currently, a small ongoing trial will address whether statins can inhibit PTS development (NCT #02679664).

Stimulation of certain cellular pathways such as TLR9 or CCR7 signaling may allow modulation of vein wall injury. However, off target effects make these targets difficult to employ. Nanoparticles targeting fibrin or other thrombus matrix components, is appealing strategy to concentrate the agent at the active injury site. Similarly, modulating leukocyte function locally is promising. For example, direct CCR2 antagonism is possible with a nano-targeted agent,¹¹² and using CCR2 blockade (to decrease pro-inflammatory Mo/MΦ influx) has strong precedent in promoting a pro-healing response with significantly reduced tissue damage in experimental models.^{80, 113–117}

Doxycycline, which inhibits MMPs, and a direct MMP-2 inhibitor, was associated with less vein wall stiffness, but overall no difference in collagen levels in a rat model of stasis VT.^{66, 118} However, clinical trials of doxycycline in abdominal aortic aneurysm patients make this agent less appealing due to lack of efficacy¹¹⁹ and intolerance to the medication. However, a direct MMP2/9 inhibitor given at the proper timing may be useful to mitigate vein wall injury in conjunction with an anticoagulant to allow thrombus resolution.

The role of IL-6 and sICAM-1 as potential biomarkers for PTS has been suggested by the BioSOX trial.¹²⁰ Anti-IL-6 antibodies in stasis VT were associated with reduced vein wall fibrosis at 14 days.¹²¹ Pro-inflammatory cytokines that act by both a cis and trans signaling routes are likely involved with thrombus resolution and/or vein wall injury. IL-6 globally depleted mice have reduced vein wall fibrosis in the stasis VT model at day 21, but not at earlier time points, and was not recapitulated with the non-stasis model (unpublished

observations). Similarly, exogenous ICAM-1 inhibition is associated with decreased Mo/MØ vein wall infiltration, but has unclear effects on the vein wall.¹²² More work needs to be done with exogenous agents to see if this strategy has translational potential.

Major Gaps in PTS Research

Several areas are ripe for both experimental and clinical investigation as suggested in this review. The role of leukocytes, particularly the Mo/MØ in directly modulating the fibrotic response is not fully understood to the degree they are in other fibrotic diseases. Several of the studies just discussed showed either increased or decreased vein wall Mo/MØ, correlating with increased or decreased vein wall injury. Thus, much biology remains to be learned. The thrombus-vein environment is quite different from the lung or liver, as well as being relatively hypoxic. It is likely the Mo/MØ functions change over time after thrombosis.

While the endothelial layer is critical for antithrombotic protection, early thrombus endothelialization may prevent complete native fibrinolysis. For example, an experimental study showed by detailed intra-vital imaging that the fibrinolytic efficacy markedly decreased between days 2 and 6 in the mouse model of stasis and non-stasis VT. At the same time, CD31⁺ endothelial cells line the abluminal surface of the resolving DVT.¹²³ Balanced against this is the need for thrombus-vein wall adherence to prevent PE, and few mouse models of PE yet exist to explore the trade-off between benefit of fibrosis reduction compared to risk of embolism. Thus, defining the timing of reversible and non-reversible vein wall fibrosis is a critical gap in knowledge.

The interplay between leukocytes, including mediators of recruitment, phenotype differentiation as well as the downstream cytokine/chemokine signaling that drives the fibrotic and fibrinolytic response remains a ripe area for investigation. With the advantage of being able to direct therapy either via targeted drug delivery (utilizing fibrin or a clotting factor as the molecular target) or using intravenous catheters at the site of thrombosis, the ability to directly deliver a medication that affects any of the pathological processes is certainly plausible (Table).

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Subacute to Chronic

Hypothesized cartoon of vein wall responses post thrombosis, in the sub-acute to late time points (after 8 days in mice, 14 days in humans). Note that the plasmin – MMP axis, and the endothelin-TGFb, and CCR7 via endothelial to mesenchymal transformation may all play a role, but at different stages of the resolving thrombus-vein wall integration. Leukocytes are critical for driving these processes, including early PMN influx, followed later by monocyte/macrophages via CCR2 and CXCR2 chemokine receptors. Note that the arrows point to the ultimate process of collagen deposition and fibrosis. + = positive feedback loops; - = inhibition;

Table:

Potential avenues for novel therapies to reduce development of or treat PTS.

Pathways involved in development of PTS	Molecular Target(s)	Current State of Investigation	Mechanisms involved
Endothelial to mesenchymal transformation	CCR7, Endothelin-1, TGF β	mice	Cellular response to injury, fibrosis
Leukocyte recruitment/venous inflammation	CAMs, CCR2, CXCR2	Mice, primates	Cellular trafficking, cellular activation
Inflammatory cytokine signaling	TLR-9, IL-6, IL-6R α	Mice, rats	Cellular activation
Fibrinolysis	Fibrin, uPA, plasmin, PAI-1	mice	Thrombus clearance, maturation
Vein wall matrix metabolism	MMP-2 MMP-9	Mice, rats	Fibrosis

CCR7 = cystine-cysteine receptor7; CCR2 = cystine-cysteine receptor2; CAMs = cell adhesion molecules;

CXCR = cysteine - x - cysteine receptor2; TLR = Toll-like receptor;

IL6 = interleukin6; uPA = urokinase plasminogen activator;

PAI-1 = plasminogen activator inhibitor-1; MMP = matrix metalloproteinase