CASE REPORTS

4G/4G PAI-1 gene variant in a patient with non-healing ulcers

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ABSTRACT

Plasminogen activator inhibitor is a serine protease inhibitor from the serpin gene family that modulates fibrin clot breakdown. PAI-1 irreversibly inhibits tissue plasminogen activator (t-PA) and urokinase plasminogen activator (u-PA) from activating plasminogen. PAI-1 also inhibits integrin-vitronectin and vitronectin-vitronectin interactions that are essential for cell migration, adhesion, and angiogenesis. We describe a patient, who developed chronic non-healing ulcers after minimal trauma to several areas of his body. Genetic testing revealed the 4G/4G homozygous genotype for the polymorphism in the promoter region of the PAI-1 gene. Increased PAI-1 activity prevents the breakdown of the fibrin clot and cell migration to remodel damaged tissue. A combination of poor clot fibrinolysis and cell recruitment to the site of injury may explain our patient's non-healing ulcers following minor traumatic injury. Early treatment with excision and skin grafting may benefit patients presenting with non-healing ulcers and the homozygous 4G/4G PAI-1 variant. To our knowledge, there have been no reports in the literature associating PAI-1 overexpression and chronic non-healing wounds.

Key Words: Plasminogen activator inhibitor-1, 4G/4G PAI-1 variant, Chronic wounds, t-PA, Skin graft

1. CASE PRESENTATION

A 56-year-old white male presented with scattered, chronic non-healing wounds. The wounds were located in multiple sites. The depth extended to dermis (left arm) to muscle (plantar surface right foot and second right phalange) (see Figure 1) to bone (see Figure 2).

Wound culture and biopsies were unhelpful. Serology was significant for elevated plasma levels of PAI-1. Genetic testing through polymerase chain reaction amplification with capillary electrophoresis revealed a homozygosity for the 4G variant of the polymorphism in the promoter region of the PAI-1 gene. The patient received multiple treatments at multiple medical institutions, including topical and oral antibiotics, steroids, anticoagulation, intravenous t-PA, and hyperbaric oxygen. His past history is significant for pulmonary fibrosis.

His wounds were excised and autografted in a two staged procedure (see Figure 3). At his 6-month follow-up, the patient's wounds remained healed with no new lesions.

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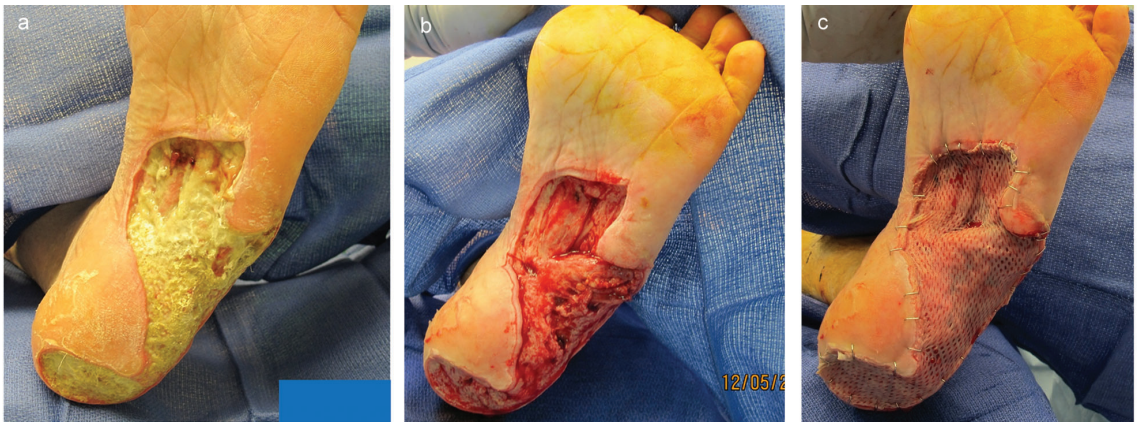

Figure 1. a. Plantar surface wound after excision; b. Plantar surface wound at presentation; c. Plantar surface wound after skin grafting

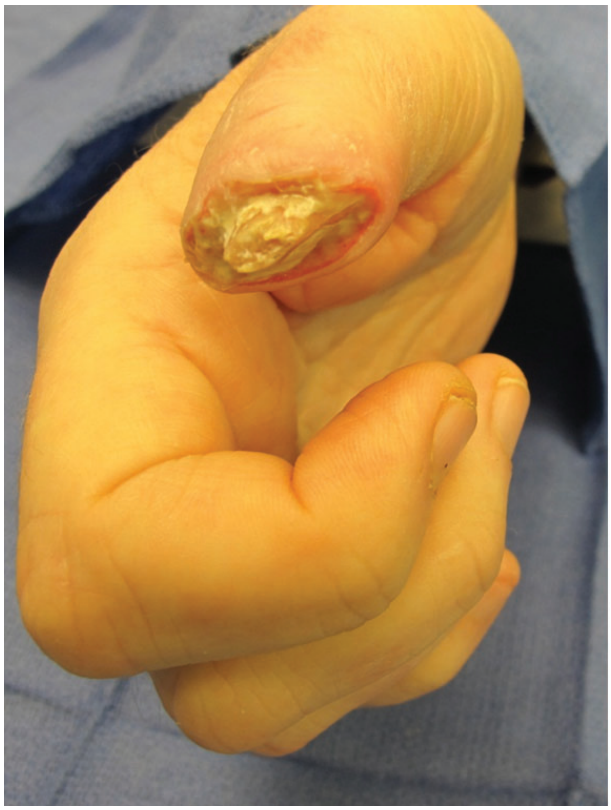

Figure 2. Right thumb wound with exposed bone

2. Discussion

Plasminogen activator inhibitor (PAI-1) is a serine protease inhibitor from the serpin gene family that modulates fibrin clot breakdown. PAI-1 is released from platelets, hepatocytes, macrophages, vascular endothelial cells and smooth muscle cells, and is secreted in response to cytokines, specifically interleukin 1, tumor necrosis factor-alpha, and transforming growth factor-beta.[1–3] PAI-1 binds irreversibly t-PA and u-PA in circulation which inhibits their binding with and conversion of plasminogen into plasmin on the fibrin clot surface.[1,4] Plasmin promotes fibrinolysis by directly degrading the cross-linked fibrin into fibrin degradation products. PAI-1 can also inhibit fibrinolysis by binding to and protecting the fibrin clot from degradation while complexed with t-PA or u-PA[1,2] (see Figure 4).

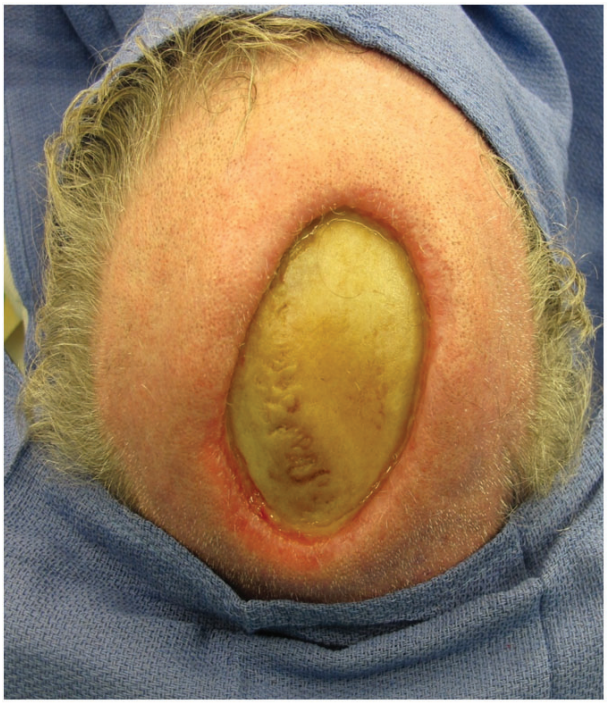

Figure 3. Scalp wound at presentation

Single nucleotide insertion or deletions at the PAI-1 promoter sequence create PAI-1 gene polymorphisms with varying phenotypes.[1,5] For example, the insertion/ deletion located 675 bp upstream of the transcription site in the promoter region of the PAI-1 gene: a five guanine nucleotide tract is the 5G allele, whereas a deletion of one G nucleotide results in the 4G variant allele.[6] The homozygous 4G allele mutation

results in PAI-1 overexpression and therefore elevated levels of PAI-1 in plasma.[1,3,5]

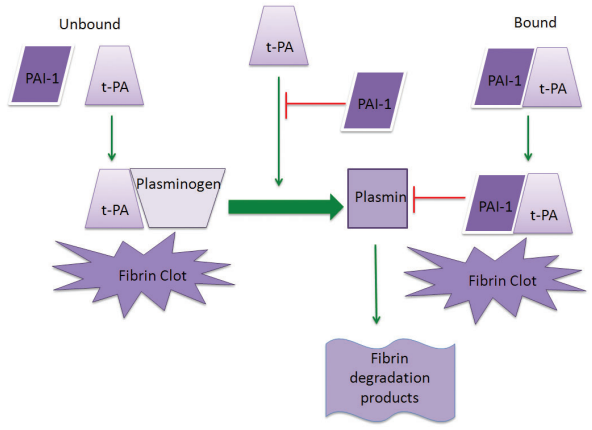

Figure 4. t-PA (or u-PA) catalyzes the conversion of plasminogen to plasmin. Free t-PA can bind to plasminogen on the fibrin clot surface for increased stability. When in circulation PAI-1 and t-PA form a reversible complex. Unbound t-PA readily binds to the surface of the fibrin clot to activate fibrinolysis. PAI-1 bound to the fibrin clot surface irreversibly binds t-PA and inhibits its effects

Decreased levels of PAI-1 are associated with increased clot degradation and varying degrees of bleeding impairment.[1,7] Elevated levels of PAI-1 have been implicated in thrombosis, recurrent otitis media in children, and poor wound healing. Increased PAI-1 has been implicated in both coronary arteries disease and venous thromboembolic events, by promoting plaque formation and venous thrombosis.[2,3,7,8] PAI-1 has also been shown to delay wound healing in vitro mostly through impairing angiogenesis and by inhibiting integrin-vitronectin and vitronectin-vitronectin which are essential for cell migration and angiogenesis though activation of vascular endothelial growth factor receptor 2.[9–11]

Our patient presents with multiple, chronic, non-healing wounds of varying locations and depths. An extensive work up was only significant for the homozygous 4G/4G mutation in the promoter sequence of PAI-1, resulting in increased PAI-1 levels. His hematologic workup was negative for other clotting disorders, vasculitis, or vasculopathy that could otherwise explain his symptoms. Our patient may represent the first patient documented to develop multiple non-healing skin ulcers caused by the 4G/4G PAI-1 mutation.

PAI-1 has both an active form and a latent form, however PAI-1 detection assays cannot distinguish between active and latent forms.[1,2] It is possible that our patient has normal PAI-1 activity, and elevated levels of PAI-1. Considering his vast array of non-healing wounds and otherwise negative coagulation workup, it is unlikely that PAI-1 is uninvolved in his poor wound healing. In addition, previous studies have demonstrated increased PAI-1 activity and decreased levels of plasmin in lung fibrosis.[2,4,7] Our patient was diagnosed with pulmonary fibrosis 6 years before he developed non-healing ulcers. Increased PAI-1 activity enhances collagen formation and scarring in pulmonary fibrosis through inhibition of t-PA and u-PA in response to injury to lung tissue.[3] It is likely that his 4G/4G PAI-1 variant contributed to his pulmonary fibrosis.

Our case report presents novel information on the clinical association of PAI-1 level and chronic non-healing wounds. In addition, it presents a therapeutic modality that was successful in healing the patient's wounds short-term and for a follow up period of 6 months. Standard treatment modalities for chronic, non-healing wounds were attempted and found to be unsuccessful in the context of PAI-1 overexpression, being refractory to antibiotics, steroids, hyperbaric oxygen and anticoagulation/t-PA infusion. Excision and skin grafting is a practical solution in the management of non-healing wounds secondary to PA-1 overexpression, and its use should be explored in studies involving larger numbers of patients.

REFERENCES

[1] Mehta R, Shapiro A. Plasminogen activator inhibitor type 1 deficiency. Hemophilia. 2008; 14: 1255-1260. PMid:19141166 http://dx.doi.org/10.1111/j.1365-2516.2008.01834.x

[2] Ghosh A, Vaughan D. PAI-1 in tissue fibrosis. J. Cell. Physiol. 2012; 227: 493-507. PMid:21465481 http://dx.doi.org/10.1002/jcp.22783

[3] Huber K, Christ G, Wojta J, *et al*. Plasminogen activator inhibitor type-1 in cardiovascular disease. Status report 2001. Thrombosis Research. 2001; 103(Suppl. 1): S7-S19. http://dx.doi.org/10.1016/S0049-3848(01)00293-6

[4] Eitzman D. Mc Coy R, Zheng X, *et al*. Bleomycin-induced pulmonary fibrosis in transgenic mice that either lack or overexpress the murine plasminogen activator inhibitor-1 gene. J Clin Invest. 1996; 97: 232-237. PMid:8550840 http://dx.doi.org/10.1172/JCI118396

[5] Ebrahimian T, Squiban C, Roque T. Plasminogen activator inhibitor-1 controls bone marrow-derived cells therapeutic effect through MMP9 signaling: role in physiological and pathological wound healing. Stem Cells. 2012; 30: 1436-1446. PMid:22570200 http://dx.doi.org/10.1002/stem.1126

[6] Eriksson P, Kallin B, van't Hooft FM, *et al*. Allele-specific increase in basal transcription of the plasminogen-activator inhibitor 1 gene is associated with myocardial infarction. Proc Natl Acad Sci USA. 1995; 92: 1851-1855. PMid:7892190 http://dx.doi.org/10.1073/pnas.92.6.1851

[7] Stevens P, Kicic A, Sutanto E, *et al*. Dysregulated repair in asthmatic

paediatric airway epithelial cells: the role of plasminogen activator inhibitor-1. Clinical & Experimental Allergy [serial online]. 2008; 38(12): 1901-1910. Available from: AcademicSearchPremier,Ipswich,MA.AccessedMay3,2015

[8] Sobel B, Woodcock-Mitchell J, Schneider D, *et al*. Increased plasminogen activator inhibitor type 1 in coronary artery atherectomy specimens from type 2 diabetic compared with nondiabetic patients: a potential factor predisposing to thrombosis and its persistence. Circulation. 1998; 97: 2213-2221. PMid:9631870 http://dx.doi.org/10.1161/01.CIR.97.22.2213

[9] Devy L, Blacher S, Grignet-Debrus C, *et al*. The pro-or antiangiogenic effect of plasminogen activator inhibitor 1 is dose dependent. FASEB J. 2002; 16: 147-154. PMid:11818362 http://dx.doi.org/10.1096/fj.01-0552com

[10] Wu J, Strawn T, Luo M, *et al*. Plasminogen activator inhibitor-1 inhibits angiogenic signaling by uncoupling vascular endothelial growth factor receptor-2-alpha v, beta 3 integrin cross talk. Arterioslcer Thromb Vasc Biol. 2015; 35: 111-120. PMid:25378411 http://dx.doi.org/10.1161/ATVBAHA.114.304554

[11] McBride J, Jenkins A, Liu X, *et al*. Elevated circulation levels of an antiangiogenic SERPIN in patients with diabetic microvascular complications impair wound healing through suppression of Wnt signaling. Society for Investigative Dermatology. 2014; 134: 1725-1734. PMid:24463424 http://dx.doi.org/10.1038/jid.2014.40