procoagulant platelet formation intensifies coagulation; VWF multimers released by damaged endothelium further recruit platelets, and engagement of the innate immune system by virus and by the host's response initiate and intensify the terrifying "perfect storm" of COVID-19.

Intriguingly, the authors find that the quantity of IgG to the spike-protein of SARS-CoV-2 significantly correlates with the patient serum's ability to induce procoagulant platelet formation. In a seeming paradox, stronger antibody response to SARS-CoV-2 and its spike protein has been associated with increased disease severity following SARS-CoV-2 infection.[7] Higher viral load may elicit both more severe disease and a stronger antibody response. Alternatively, the current study suggests that the specific and productive anti–SARS-CoV-2 antibody response may overlap with a dysfunctional antibody response in severe COVID-19. Similar prothrombotic and autoreactive antibodies, including antiphospholipid antibodies and anti-heparin/PF4 antibodies akin to those occurring in heparin-induced thrombocytopenia, have also been associated with severe COVID-19.[8] A robust extrafollicular B-cell response occurs in severe COVID-19, possessing cellular, repertoire, and serological characteristics resembling processes mediating pathogenic autoantibody development in systemic lupus erythematous.[9] In the current study, the precise origin and nature of the procoagulant platelet initiating antibodies are not defined. However, an intriguing hypothesis is that, in severe COVID-19, a dysfunctional and overly robust extrafollicular anti–SARS-CoV-2 B-cell response generates autoreactive and prothrombotic antibodies that, in the context of the local immune response, drive a dysfunctional and autodestructive response within the vasculature. Understanding how these different prothrombotic antibodies are elicited, the association between such antibodies and antiviral immunity, and the distinction between these prothrombotic antibodies and their contribution to disease severity will impact COVID-19 diagnosis and treatment by guiding risk stratification and educating vaccine development based on the nature of the B-cell response produced.

Excitingly, the studies presented here encourage the development of therapeutic approaches in COVID-19 targeting FcγRIIA-mediated–platelet activation and procoagulant platelet formation. Fostamatinib and ibrutinib, US Food and Drug Administration–approved inhibitors of spleen tyrosine kinase and Bruton tyrosine kinase, respectively, limit both FcγRIIA-mediated platelet and B-cell activation and are currently in phase 2/3 studies in COVID-19. How these agents impact the local and systemic thrombotic manifestations of COVID-19 and their impact on bleeding risk in this setting will be of interest. In the setting of increased bleeding risk, targeting procoagulant platelet formation may be particularly beneficial, as procoagulant platelet formation can be specifically abrogated, without inhibiting the platelet aggregatory response or increasing bleeding risk. In this regard, inhibitors of mitochondrial calcium entry and of the mitochondrial permeability transition, among these, cyclosporine, specifically abrogate procoagulant platelet formation without limiting other aspects of platelet activation, including aggregation and granule release.[10] The impact of anticoagulation and classical antiplatelet therapies is the subject of ongoing trials, and the results of these studies are eagerly awaited. Encouragingly, the studies presented here by Althaus et al offer a new beachhead in the scientific community's efforts to mitigate and defeat the thromboinflammatory storm induced by SARS-CoV-2.

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THROMBOSIS AND HEMOSTASIS

Comment on Vollack-Hesse et al, page 1072

Getting under the skin: a new route for factor VIII?

Thomas A. J. McKinnon | Imperial College London

In this issue of *Blood*, Vollack-Hesse et al present an elegant study demonstrating a possible new path for administering factor VIII (FVIII) via subcutaneous injection.[1]

The past few decades have seen major advances in the treatment of hemophilia, perhaps most notably the advent of gene therapy approaches that have led to sustained expression of both FVIII and FXI in patients with hemophilia A and B, respectively.[2] But the mainstay of hemophilia treatment is still repeated with IV administration of recombinant protein concentrates. Although repeated IV injections are clinically effective, they are not pleasant, they can be particularly challenging in patients with poor vein access, and they are especially difficult for parents who have to inject small children.

Previous attempts to deliver FVIII via the subcutaneous route have been unsuccessful because FVIII binds to phospholipids, which results in proteolytic cleavage and poor absorption into the circulation.[3] However, Vollack-Hesse et al overcame this pitfall by coadministering FVIII alongside a small recombinant fragment of its binding partner, von Willebrand factor (VWF) in hemophilia A mice. The authors expressed a dimer of the VWF-D'D3 domains with a double repeat of the C-terminal 1238-1268 region that contains a series of *O*-linked glycosylation sites. This fragment, which they name VWF-12, contains the FVIII binding site and has been previously shown to be the minimal fragment required to protect FVIII in the circulation.[4] The authors show that although VWF-12 does not interfere with the function of full-length VWF, it prevents FVIII from binding to phospholipids and components of the subendothelial matrix and protects against proteolytic cleavage. The net result is that when VWF-12 is subcutaneously injected alongside FVIII into hemophilia A mice, absorption of FVIII into the circulation is enhanced. Not surprisingly, the subcutaneous route resulted in a slower time to peak FVIII concentration; however, the half-life of FVIII was increased 2.5-fold over FVIII injected into mice via the IV route (7.2 vs 2.8 hours) and with greater bioavailability. Significantly, it offered up to 24 hours of protection from bleeding. It should be noted that in this study, the concentration of FVIII injected via the subcutaneous route was 5 times higher than that via the intravenous route, but even so, the therapeutic goal was successfully achieved.

The concept of codelivering VWF alongside FVIII has been widely regarded as a means to enhance the half-life of FVIII. Modifications to FIX have resulted in molecules with extended half-lives, but this has proved to be more difficult to achieve with FVIII because its half-life is extrinsically linked to that of VWF, the so called "VWF ceiling."[5] BIVV001 is a novel fusion protein of FVIII and VWF that overcomes the VWF ceiling and has been shown to have a significantly extended half-life.[6] Although VWF-12 does not directly prolong the half-life of FVIII, because endogenous full-length VWF will outcompete it once it is in the circulation, these data from Vollack-Hesse et al clearly show the advantage of exploiting VWF fragments to protect FVIII. In the future, it may be interesting to see how BIVV001 performs after subcutaneous injection.

Subcutaneous delivery of a hemophilia A therapeutic agent has already been achieved by the bispecific antibody emicizumab, which mimics activated FVIII.[7] There are no long-term data on the safety of emicizumab, but there is still a strong rationale for developing regular FVIII products. There are also concerns over the immunogenicity of therapeutics delivered via the subcutaneous route,[8] but Vollack-Hesse et al show in their study that the FVIII/VWF-12 combination was no more immunogenic than FVIII delivered via IV and, in fact, was marginally less immunogenic, which indicates another potential advantage of this delivery route. Many questions still need to be addressed, but these data have great promise and clearly warrant further investigation. Detailed dissection of how this particular VWF fragment protects FVIII will be of scientific interest, and studies to enhance the affinity of VWF-12 for FVIII would also be useful. Ultimately, if successful in humans, this may offer a more patient-friendly alternative to repeated intravenous injections without compromising safety and efficacy.

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Comment on Samuelson Bannow et al, page 1082

Can HIT testing lose its radioactivity?

Grace M. Lee | Duke University

In this issue of *Blood*, Samuelson Bannow et al describe the results of a multicenter, prospective, blinded study that compared the performance of the serotonin release assay (SRA) against the platelet factor 4 (PF4)-dependent P-selectin expression assay (PEA) for the diagnosis of heparin-induced thrombocytopenia (HIT).[1]

HIT is a potentially life-threatening, prothrombotic, immune complication of heparin caused by immunoglobulin G antibodies that recognize complexes of PF4 and heparin. Thrombocytopenia and thrombosis in HIT are caused by a subset of anti-PF4/heparin antibodies that elicit cellular activation by binding and cross-linking platelet FcγRIIa.[2] Recent studies suggest that antibody binding to FcγRIIa on monocytes[3] and on neutrophils[4] contributes significantly to thrombosis in HIT.

Central to the diagnosis of HIT is laboratory identification of heparin-dependent, platelet-activating antibodies. In most circumstances, HIT testing begins with an immunoassay to detect the presence of