Article published online: 2024-10-28

Editorial

Be Curious. Try New Things

Johannes Thaler[1]

[1] Division of Haematology and Haemostaseology, Department of Medicine I, Medical University of Vienna, Vienna, Austria

Hamostaseologie 2024;44:342–344.

Not much is needed to get started in experimental hemostasis research. If you have not done so yet, just go to your institution's lab and ask whether you can use some of their equipment: pipette, centrifuge, microplate reader, and $CaCl_2$, that's basically all you need. Then, let someone draw a vial of your blood and here you go, with your first blood clotting experiment. But, wait! One thing is still missing. To perform your first assay, you are advised to use a reagent that initiates clotting of your blood sample. Interested in contact activation? Wondering whether the presence of coagulation factor (F) XII really is unimportant (as indicated by the fact that a complete deficiency of FXII does not lead to bleeding)? Then do it the way Christian Kastrup and his research group did.[1] Grab your backpack and collect soil and clay in the nearby countryside. Back in the lab, crush the collected soil and clay samples and add a small amount of each sample to your aliquoted blood plasma sample, then add $CaCl_2$, and measure the plasma clotting time on the microplate reader (do not forget the negative control, i.e., the addition of buffer to plasma instead of soil or clay sample). Likely, you will detect a massive reduction of the plasma clotting time when adding soil or clay (which you may also call "dirt") to the recalcified aliquoted plasma samples. Doing so, you just have found further evidence for the *dirty-wound hypothesis of coagulation function* as proposed by Brian Cooley recently.[2] He explained that dirt is an unavoidable ingredient of open wounds of land-based animals, and that coagulation is activated by dirt via FXII. Still believe that FXII is irrelevant? Interesting, isn't it? Before you swing the pipette, you might also be inspired by the exciting research presented in this theme issue of *Hämostaseologie* entitled "*Coagulation Diagnostics beyond Routine*."

Up to 30% of persons with hemophilia receiving factor (F) VIII replacement therapy develop neutralizing antibodies specific for FVIII.[3] These FVIII neutralizing antibodies are a major complication in the replacement therapy of persons with hemophilia A. In this theme issue, **A. Schmidt and colleagues**[4] report on a method of acid denaturation for FVIII-containing plasma samples of persons with hemophilia A. Acid treatment resulted in a temporary denaturation of antibody–FVIII complexes, which allowed the detection of most FVIII-neutralizing antibodies via ELISA. Acid treatment for the detection of anti-FVIII antibodies in FVIII-containing plasma samples seemed to be superior compared to heat treatment, because of lower background signals in the ELISA and less antigen destruction. Persons with hemophilia A may benefit from this improved method, because it allows a more reliable detection of FVIII-specific antibodies and therefore may better guide treatment decisions.

In their comprehensive review, **D. Mehic and colleagues**[5] explore the utility of global hemostatic assays in elucidating the pathophysiology of bleeding disorders of unknown cause (BDUC), which is a diagnosis of exclusion after exhaustive evaluation of plasmatic coagulation and platelet function. Global hemostatic assays such as viscoelastic assays and thrombin generation can be performed in plasma and whole blood. These assays provide a more comprehensive understanding of hemostasis than routine assays. The clinical utility of most assays in the workup of BDUC, however, seems not to be clear at the moment. Global assays are therefore currently not recommended for clinical decision-making. Nevertheless, global assays have the potential to serve as confirmatory tests for BDUC patients and they can shed light on aspects of hemostasis that are overlooked by traditional laboratory analyses. It is concluded by the authors that global assays could serve as promising research tools for the elucidation of different pathophysiological mechanisms underlying BDUC.

Extravascular tissue factor (TF) initiates blood coagulation by forming a complex with circulating FVII(a) in the event of vascular damage. Functional TF can also circulate in blood either as a component of blood cells or exposed on extracellular vesicles (EVs). Results from several clinical studies suggest that EV-associated TF activity could be a biomarker for thrombosis and survival in cancer patients.[6] In their review, **A. Bonifay and colleagues**[7] first provide a brief

Address for correspondence
Johannes Thaler, MD, PhD,
Division of Haematology and
Haemostaseology, Department
of Medicine I, Medical University
of Vienna, Währinger Gürtel
18-20, A-1090 Vienna, Austria
(e-mail: johannes.thaler@meduniwien.ac.at).

© 2024. Thieme. All rights reserved.
Georg Thieme Verlag KG,
Rüdigerstraße 14,
70469 Stuttgart, Germany

DOI https://doi.org/
10.1055/a-2316-4116.
ISSN 0720-9355.

This document was downloaded for personal use only. Unauthorized distribution is strictly prohibited.

overview of different forms of TF and mechanisms of TF biogenesis. Then different methods to measure TF in blood and plasma samples are discussed and the results of the first multicentric study that compares the analytical performance of 27 different EV-TF activity assays are summarized and interpreted. Finally, seven steps to further improve EV-TF activity determination are proposed, which (the authors suggest) are necessary to standardize and automate assays for clinical practice.

In their review, **J. Thaler and colleagues**[8] summarize the evidence that physiological body fluids such as mother's milk, saliva, urine, semen, and amniotic fluid trigger coagulation. The studies discussed go up to a century back and seem to have been almost completely forgotten in recent years. The ability of physiological body fluids to trigger coagulation is explained by the presence of EVs that expose extrinsic tenase complexes, that is, complexes of TF and activated factor VII (FVIIa). Why these body fluids share this coagulant potential, however, is unknown. Possible explanations are that these body fluids contribute to hemostatic protection and/or to the regulation of the barrier function of mucosal tissues. The authors conclude that interactions between TF/VII(a) complex exposing EVs from body fluids and epithelia may represent an important protective mechanism.

Conflict of Interest

The author declares that he has no conflict of interest.

References

1 Juang LJ, Mazinani N, Novakowski SK, et al. Coagulation factor XII contributes to hemostasis when activated by soil in wounds. Blood Adv 2020;4(08):1737–1745
2 Cooley BC. The dirty side of the intrinsic pathway of coagulation. Thromb Res 2016;145:159–160
3 Iorio A, Halimeh S, Holzhauer S, et al. Rate of inhibitor development in previously untreated hemophilia A patients treated with plasma-derived or recombinant factor VIII concentrates: a systematic review. J Thromb Haemost 2010;8(06):1256–1265
4 Schmidt A, Stichel D, Salzmann-Manrique E, Königs C. Acid treatment of FVIII-containing plasma samples unmasks a broad spectrum of FVIII-specific antibodies in ELISA. Hamostaseologie 2024;44:350–357
5 Mehic D, Assinger A, Gebhart J. Utility of global hemostatic assays in patients with bleeding disorders of unknown cause. Hamostaseologie 2024;44:358–367
6 Hisada Y, Sachetto ATA, Mackman N. Circulating tissue factor-positive extracellular vesicles and their association with thrombosis in different diseases. Immunol Rev 2022;312(01):61–75
7 Bonifay A, Cointe S, Plantureux L, et al. Update on Tissue Factor Detection in Blood in 2024: A Narrative Review. Hamostaseologie 2024;44:368–376
8 Thaler J, Tripisciano C, Nieuwland R. Investigations on the Hemostatic Potential of Physiological Body Fluids. Hamostaseologie 2024;44:377–385

This document was downloaded for personal use only. Unauthorized distribution is strictly prohibited.

 © 2024. Thieme. All rights reserved.