Received: 22 January 2024 | Revised: 5 February 2024 | Accepted: 13 February 2024

https://doi.org/10.1016/j.rpth.2024.102395

ILLUSTRATED REVIEW

Coagulation and platelet biology at the intersection of health and disease: illustrated capsules of the 11th Symposium on Hemostasis at the University of North Carolina

Maria Aleman[1] | Gowthami M. Arepally[2] | Trevor Baglin[3] | Lorena Buitrago[4] | Pavel Davizon-Castillo[5] | Sanjana Dayal[6] | Matthew J. Flick[7] | Gloria Gerber[8] | Yohei Hisada[9] | Krasimir Kolev[10] | Ana O'Loghlen[11] | Alireza R. Rezaie[12] | Erica M. Sparkenbaugh[7,9] | Evi X. Stavrou[13,14] | Betül Ünlü[15] | Gregory M. Vercellotti[16]

[1]Blood Research Center, Department of Pharmacology, University of North Carolina at Chapel Hill, Chapel Hill, North Carolina, USA

[2]Division of Hematology, Duke University Medical Center, Durham, North Carolina, USA

[3]Centessa Pharmaceuticals plc, Cheshire, United Kingdom

[4]Allen and Frances Adler Laboratory of Blood and Vascular Biology, Rockefeller University, New York, New York, USA

[5]Department of Pediatrics Hematology/Oncology and Bone Marrow Transplantation, University of Colorado Anschutz Medical Campus, Aurora, Colorado, USA

[6]Department of Internal Medicine, University of Iowa Carver College of Medicine, Iowa City, Iowa, USA

[7]Blood Research Center, Department of Pathology and Laboratory Medicine, University of North Carolina at Chapel Hill, Chapel Hill, North Carolina, USA

[8]Division of Hematology, Department of Medicine, School of Medicine, Johns Hopkins University, Baltimore, Maryland, USA

[9]Blood Research Center, Division of Hematology, Department of Medicine, University of North Carolina at Chapel Hill, Chapel Hill, North Carolina, USA

[10]Department of Biochemistry, Institute of Biochemistry and Molecular Biology, Semmelweis University, Budapest, Hungary

[11]Epigenetics & Cellular Senescence Group, Blizard Institute, Barts and the London School of Medicine and Dentistry, Queen Mary University of London, London, United Kingdom

[12]Department of Biochemistry and Molecular Biology, Cardiovascular Biology Research Program, Oklahoma Medical Research Foundation, Oklahoma City, Oklahoma, USA

[13]Department of Medicine, Hematology and Oncology Division, Case Western Reserve University School of Medicine, Cleveland, Ohio, USA

[14]Medicine Service, Section of Hematology-Oncology, Louise Stokes Cleveland Veterans Affairs Medical Center, Cleveland, Ohio, USA

[15]Einthoven Laboratory for Experimental Vascular Medicine, Department of Internal Medicine, Leiden University Medical Center, Leiden, The Netherlands

[16]University of Minnesota School of Medicine, Minneapolis, Minnesota, USA

Correspondence
Erica M. Sparkenbaugh, Department of Medicine, Division of Hematology, The University of North Carolina at Chapel Hill, Mary Ellen Jones building, 116 Manning Drive, Chapel Hill, NC 27599, USA.
Email: erica_sparkenbaugh@med.unc.edu

Handling Editor: Michelle Sholzberg

Conference Organizers Chairs: Matthew J. Flick and Evi X. Stavrou matthew_flick@med.unc.edu;

Abstract

The University of North Carolina Symposia on Hemostasis began in 2002, with The First Symposium on Hemostasis with a Special Focus on FVIIa and Tissue Factor. They have occurred biannually since and have maintained the primary goal of establishing a forum for the sharing of outstanding advances made in the basic sciences of hemostasis. The 2024 11th Symposium on Hemostasis will bring together leading scientists from around the globe to present and discuss the latest research related to coagulation factors and platelet biology. In keeping with the tradition of the conference, we expect

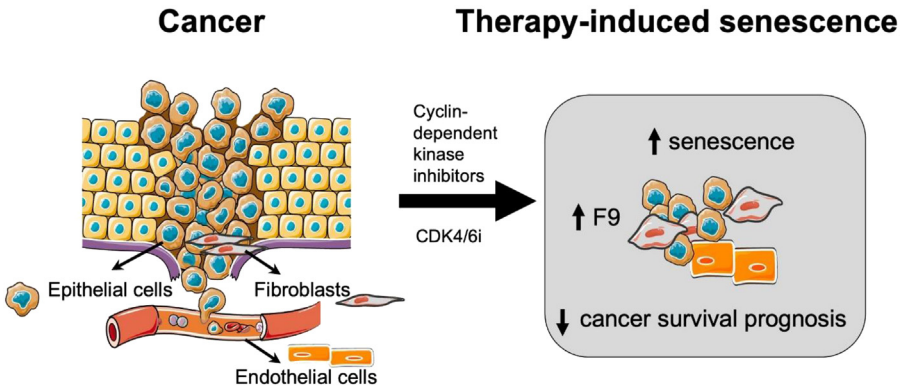
exs003@case.edu Vice-Chair: Erica M. Sparkenbaugh (and editor of RPTH Graphical Abstracts) Erica_sparkenbaugh@med.unc.edu Symposium Committee: Wolfgang Bergmeier, Maureane Hoffman, Nigel S. Key, Nigel Mackman, Dougald Monroe, Rafal Pawlinski, and Alisa Wolberg

novel cross-disciplinary collaborations to result from bringing together fundamental scientists and physician-scientists from different backgrounds and perspectives. The aim of these collaborations is to springboard the next generation of important advances in the field. This year's program was designed to discuss Coagulation and Platelet Biology at the Intersection of Health and Disease. The goal is to develop a better understanding of the pathophysiologic mechanisms leading to hemostatic and thrombotic disorders as this understanding is critical for the continued development of safe and efficacious therapeutics. Included in this review article are illustrated capsules provided by our speakers that highlight the main conclusions of the invited talks.

KEYWORDS
cancer-associated thrombosis, complement cascade, hemophilia, hemostasis, heparin-induced thrombocytopenia, intrinsic coagulation, neutrophil extracellular traps, platelet, protease-activated receptors, sickle cell disease, thrombosis

COAGULATION FACTOR IX AS A REGULATOR OF SENESCENCE

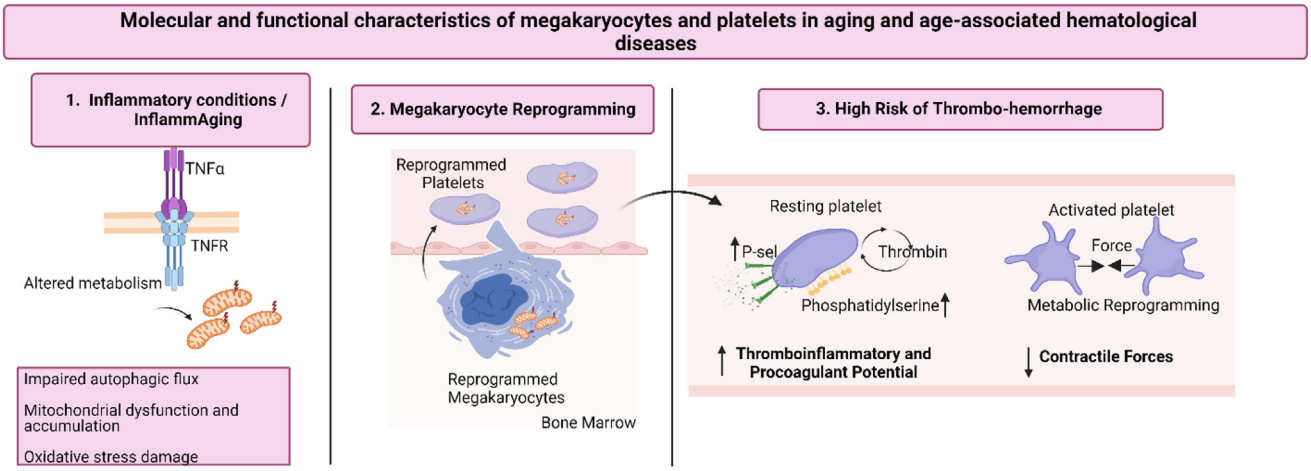
Ana O'Loghlen



Cellular senescence is a known tumor suppression mechanism [1,2]. However, genes regulating senescence in this context are still unknown. Here, using a functional genome-wide CRISPR/Cas9 genetic screen, we found several genes within the coagulation pathway that participate in preventing senescence induced by cyclin-dependent inhibitors (CDK4/6i). We found that downregulation of the coagulation factor IX (F9) using single-guide RNA and short hairpin RNA prevents the senescent-like phenotype induced in different breast tumor cells. These findings were validated using alternative CDK4/6 inhibitors and in a panel of cancer cells. While F9 knockout prevents senescence, treatment with a recombinant F9 protein was sufficient to induce a senescence-like state. In addition, endogenous F9 is upregulated and released in different human primary cell cultures undergoing senescence. Bioinformatics analysis of cancer datasets suggests a role for F9 in human tumors. These data collectively propose key genes useful for designing new therapeutic strategies in personalized medicine in oncology [3].

MOLECULAR AND FUNCTIONAL CHARACTERISTICS OF MEGAKARYOCYTES AND PLATELETS IN AGING

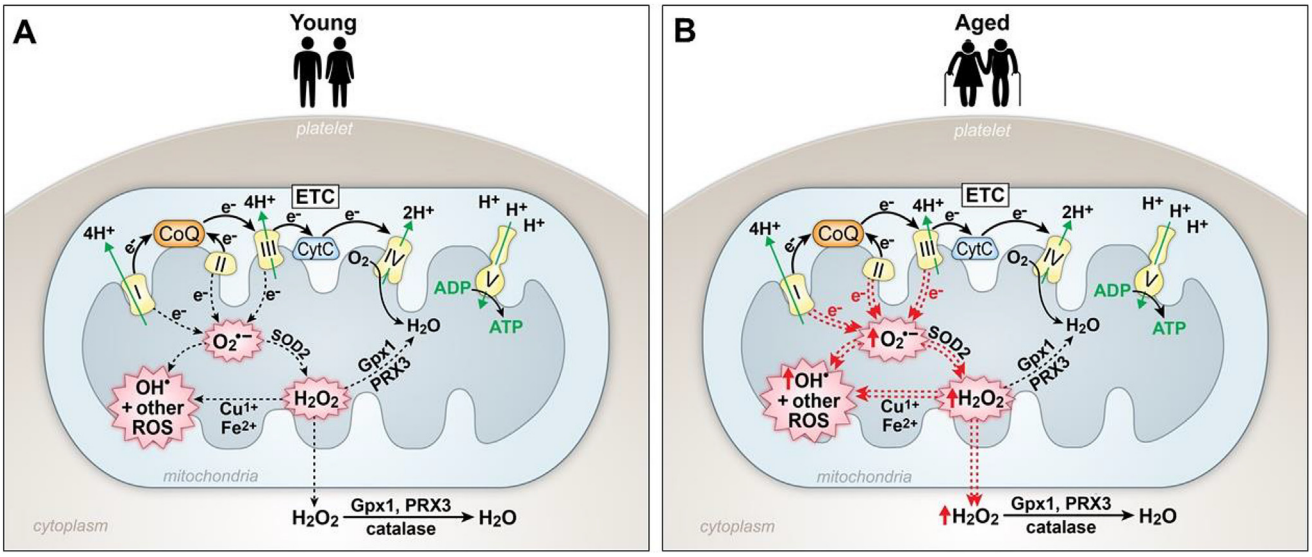
Guadalupe Rojas-Sanchez, PhD and Pavel Davizon-Castillo, PhD



Aging-associated inflammation is associated with alterations of mitochondrial mass in megakaryocytes and platelets [1] and an overall metabolic reprogramming of megakaryocytes [2]. These processes promote the formation of platelets with high thromboinflammatory and procoagulant potentials and significant alterations of platelet contractile forces [3]. The dysregulation of platelet contractile forces by aging-associated inflammation may be an important element of the physiopathology of thrombohemorrhagic events of older individuals. TNFα, tumor necrosis factor alfa; TNFR, tumor necrosis factor receptor.

ENDOGENOUS ANTIOXIDANTS IN BLOOD CELLS, AGING, AND THROMBOSIS

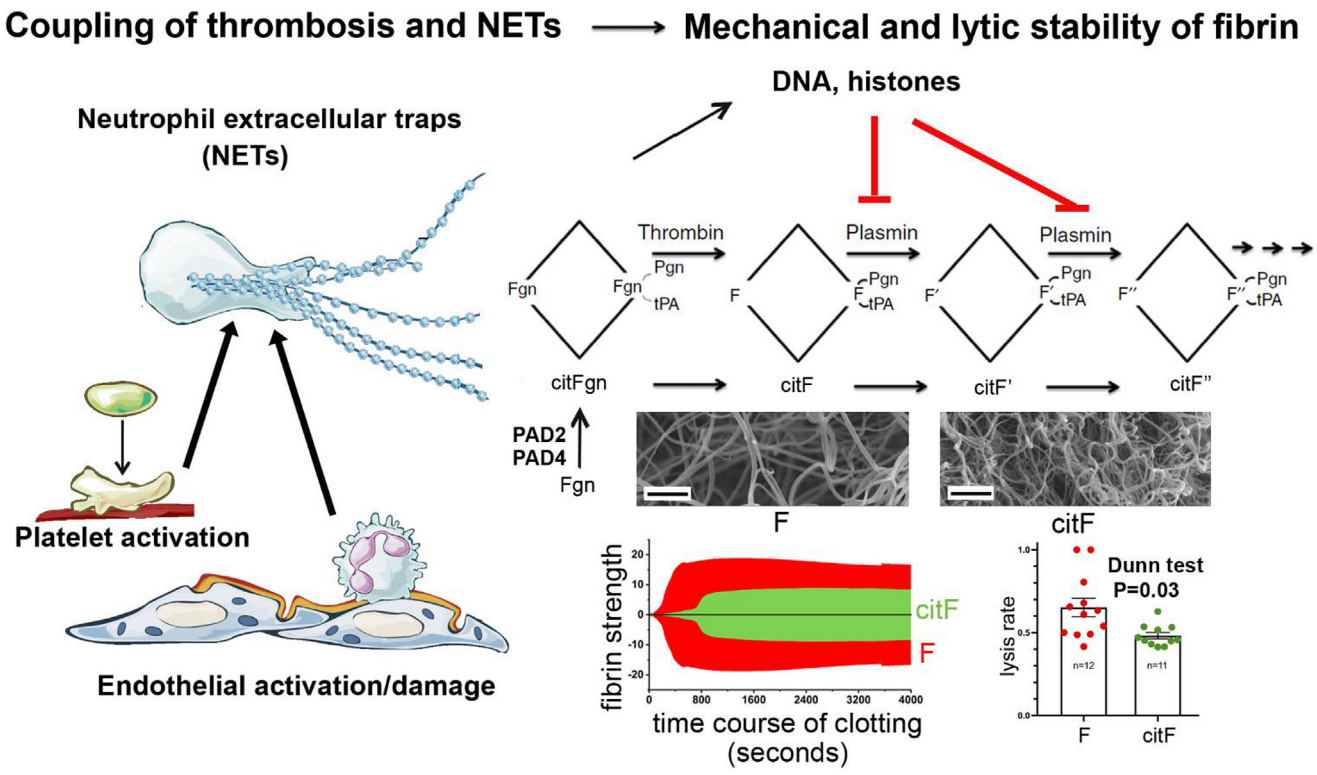
Sanjana Dayal



Platelet redox regulation: Reactive oxygen species (ROS) are important mediators of platelet activation. (A) Under physiological state, mitochondrial-superoxide ($O_2^{\cdot-}$) is generated by electron transport chain (ETC) through reductions of O_2 at complexes I, II, and III. Antioxidant enzymes regulate ROS levels to sublethal concentrations. For instance, superoxide dismutase 2 (SOD2) converts $O_2^{\cdot-}$ to hydrogen peroxide (H_2O_2), which is further catalyzed to H_2O by antioxidant enzymes glutathione peroxidase 1 (Gpx1) or peroxiredoxin 3 (PRX3). Since H_2O_2 is permeable, it can leak out from mitochondria and will be catalyzed to H_2O by cytoplasmic Gpx1, PRX3, or catalase. (B) During aging, ETC assembly may be disrupted, increasing residence time of electrons to complexes I, II, or III and excess 1 electron reductions of O_2, leading to increased steady-state levels of $O_2^{\cdot-}$ and H_2O_2. If the excess H_2O_2 is not completely neutralized into water by Gpx1 and/or PRX3, it will cause lipid peroxidation or, in presence of labile iron (Fe^{2+}) or copper (Cu^{1+}), will generate other ROS such as hydroxyl radicals. $O_2^{\cdot-}$ can also react at diffusion-limited rates with nitric oxide to form peroxynitrite and other reactive nitrogen species, outcompeting other reactions kinetically. ADP, adenosine diphosphate; ATP, adenosine triphosphate; CytC, cytochrome C; CoQ, coenzyme Q.

NET-RELATED ASPECTS OF THROMBUS RESOLUTION

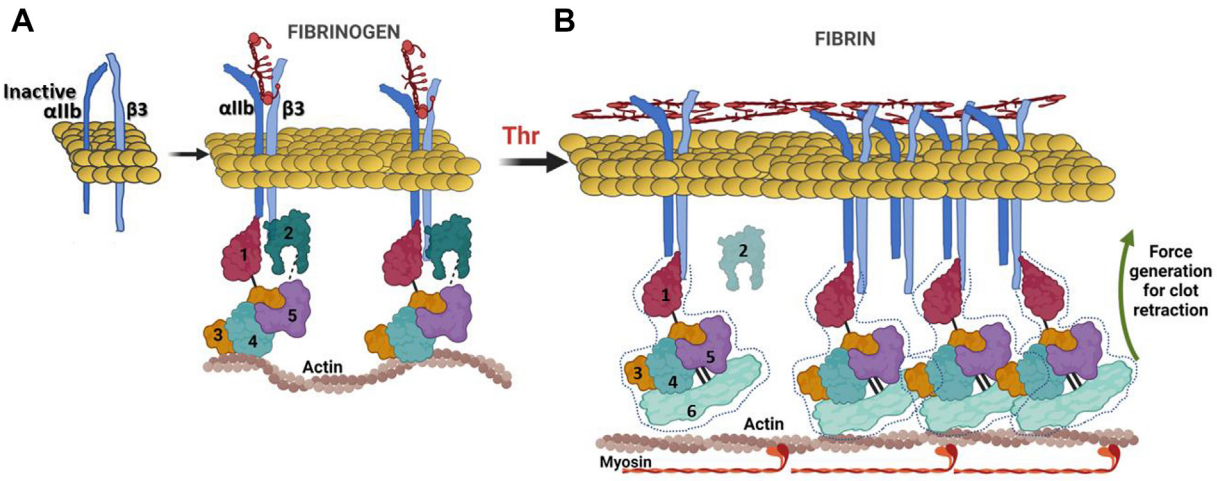
Krasimir Kolev



At sites of intravascular thrombus formation, the interplay of activated platelets and endothelium triggers the release of neutrophil extracellular traps (NETs), whose 2 major components are DNA and histones (reviewed in [4]). When fibrinogen (Fgn) is converted to fibrin (F), the copolymerization of F with NET-components results in an altered F structure with modified functionality as a template of plasminogen (Pgn) activation by tissue-type plasminogen activator (tPA) and susceptibility to plasmin-mediated proteolysis to generate various polymeric (F', F'') or soluble (→ → →) degradation products. NET formation is accompanied by the release of peptidyl-arginyl deiminase isoforms 2 and 4 (PAD2 and PAD4), which convert arginine residues in a number of proteins, including Fgn, to citrulline. Citrullinated Fgn (citFgn) is converted to citrullinated F (citF) and is present in the structure of murine model of venous thrombi [5]. The citF is composed of thinner fibers and larger pores [6]. The modified structure of citF results in impaired mechanical stability and enhanced lytic resistance compared with noncitrullinated F [5].

PLATELET-FIBRIN INTERACTIONS, AND CLOT CONTRACTION

Lorena Buitrago

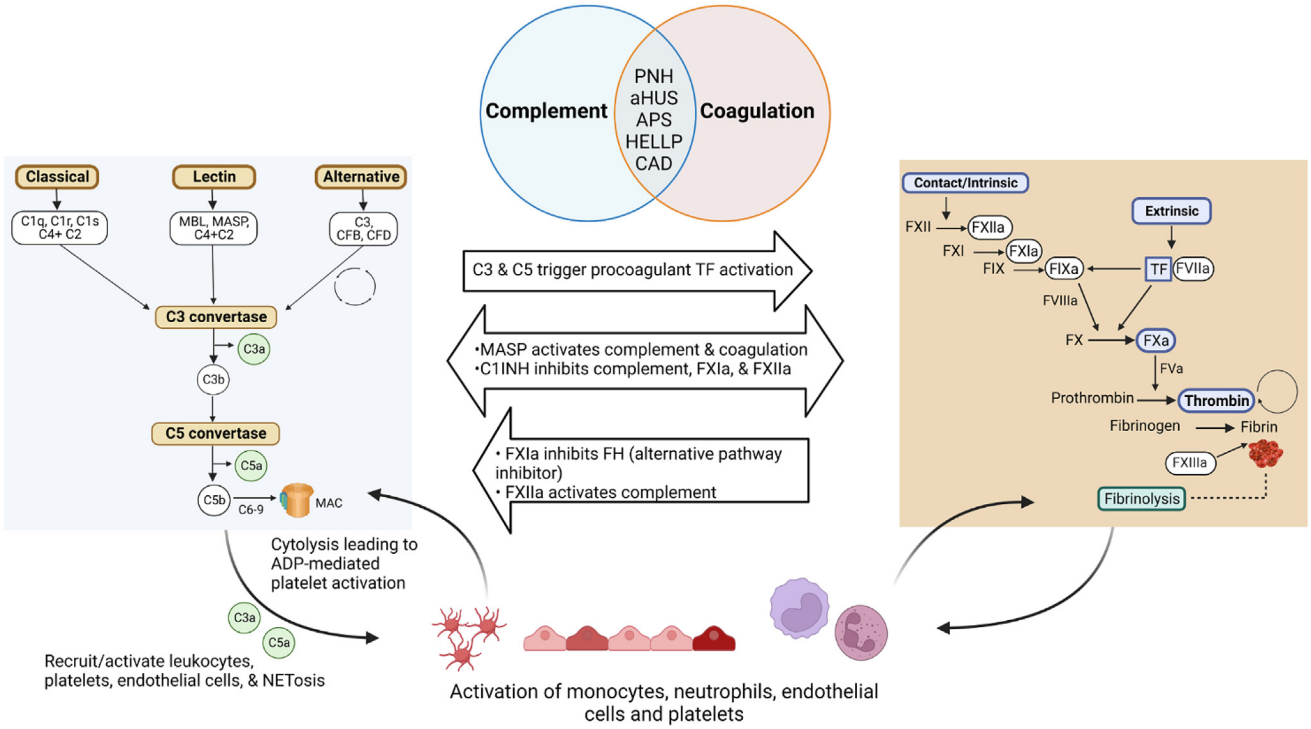


Upon platelet activation, $\alpha_{IIb}\beta_3$ undergoes a conformational change, increasing its affinity for fibrinogen. (A) Fibrinogen binding initiates cytoplasmic signaling, forming protein complexes (eg, hypothetical proteins 1-2-3-4-5) that transiently link extracellular fibrinogen to the actin cytoskeleton. (B) Within 20 seconds of initiation of vascular injury, thrombin (Thr) is generated, and fibrinogen polymerizes into insoluble fibrin. Polymerized fibrin binds multiple $\alpha_{IIb}\beta_3$ receptors simultaneously with similar affinity but by a mechanism that differs subtly from binding to fibrinogen [7]. We hypothesize that the greater avidity and/or receptor clustering results in a more robust signaling, leading to strong molecular interactions and the formation of stable protein complexes (eg, hypothetical proteins 1-3-4-5-6) that lead to a stable linkage to the actin cytoskeleton and initiation of actin-myosin–mediated clot retraction and clot stabilization.

The $\alpha_{IIb}\beta_3$-fibrinogen interaction is a validated therapeutic target, with 3 Food and Drug Administration–approved drugs that inhibit the interaction. Despite the recognized physiological and clinical significance, the biochemistry underlying platelet-fibrin interactions remains less well understood. Our research focuses on identifying the structural differences in $\alpha_{IIb}\beta_3$ binding to fibrin compared with fibrinogen. We seek to understand how fibrin binding triggers contraction of the fibrin strands and results in retraction, consolidation, and stabilization of the clot.

THE INTERPLAY OF COMPLEMENT AND COAGULATION IN THROMBOSIS

Gloria Gerber

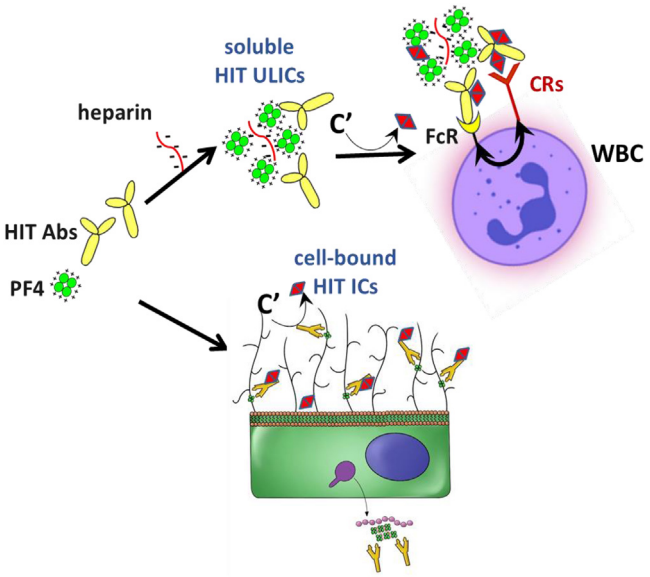


CFB, complement factor B; CFD, complement factor D; MAC, membrane attack complex; NETosis, release of neutrophil extracellular traps.

Complement and coagulation have a complex and multifactorial relationship in affecting innate immunity and hemostasis. *In vitro* and *ex vivo* studies have identified a multitude of interactions between these 2 evolutionarily-linked cascades [8,9]. In some cases, studies have yielded conflicting results or not translated *in vivo* [10]. The close interplay of complement and coagulation is most convincingly underscored by their central role in diseases such as paroxysmal nocturnal hemoglobinuria (PNH); atypical hemolytic uremic syndrome (aHUS); antiphospholipid syndrome (APS); hemolysis, elevated liver enzymes, low platelets (HELLP) syndrome; and cold agglutinin disease (CAD). In PNH, the prototypical complementopathy, thrombosis was the leading cause of mortality despite anticoagulation. The risk of thrombosis has decreased significantly since the advent of the C5 complement inhibitor, eculizumab, and mortality is now comparable with age-matched controls. Similarly, in aHUS, eculizumab led to improvement in hematologic thrombotic microangiopathy and renal function. ADP, adenosine Diphosphate; MBL, mannan-binding lectin; MASP, mannan-binding lectin-associated serine protease; TF, tissue factor.

COMPLEMENT ACTIVATION AND HEPARIN-INDUCED THROMBOCYTOPENIA

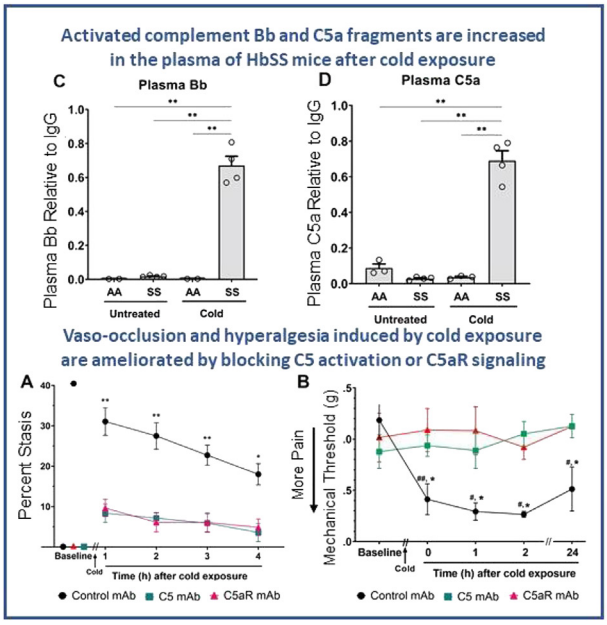
Gowthami M. Arepally

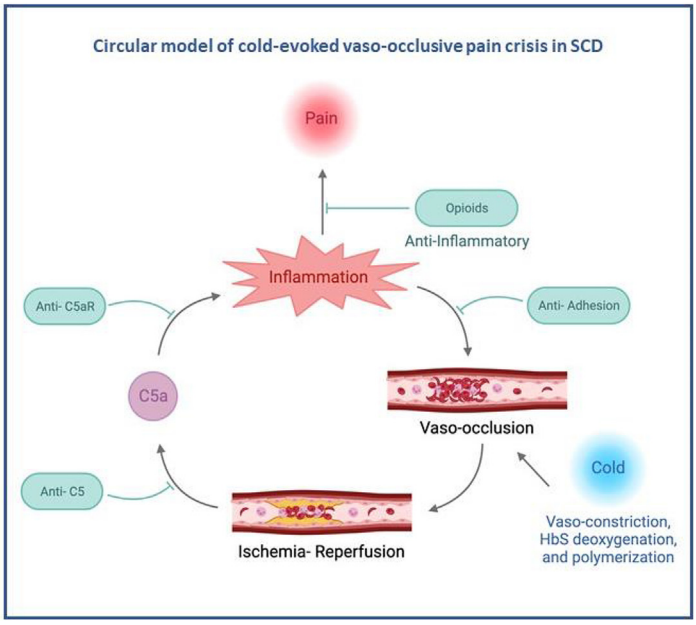


Heparin-induced thrombocytopenia (HIT) is a potentially life-threatening thrombotic disorder caused by antibodies (Abs) to platelet factor 4 (PF4) and heparin. Thrombotic risk in HIT remains high long after heparin discontinuation. In published studies [11], we demonstrated a prominent role of complement in the pathogenesis of HIT. In these studies, we showed that ultralarge immune complexes (HIT ULICs) robustly activate complement, initiating complement-dependent binding of immune complexes (ICs) to circulating neutrophils (polymorphonuclear leukocytes) and monocytes. In new studies, we show that complement-activating properties of HIT ULICs are, in large part, dictated by heparin itself. Circulating heparin (at disease initiation) causes PF4 to dissociate from cellular glycosaminoglycans to generate circulating multivalent PF4/H complexes that bind HIT Abs and form soluble HIT ULICs. In the absence of heparin (during disease propagation), HIT Abs binds to PF4/glycosaminoglycan antigenic complexes on platelets and endothelial cells, forming cell-bound HIT ICs. The consequences of heparin-dependent/independent complement activation on thrombotic manifestations in HIT using *ex vivo* and *in vivo* data will be discussed. CRs, complement receptors; WBC, white blood cell.

COLD EXPOSURE INDUCES VASO-OCCLUSION AND PAIN IN A MOUSE MODEL OF SICKLE CELL DISEASE THAT DEPENDS ON COMPLEMENT ACTIVATION

Gregory M. Vercellotti

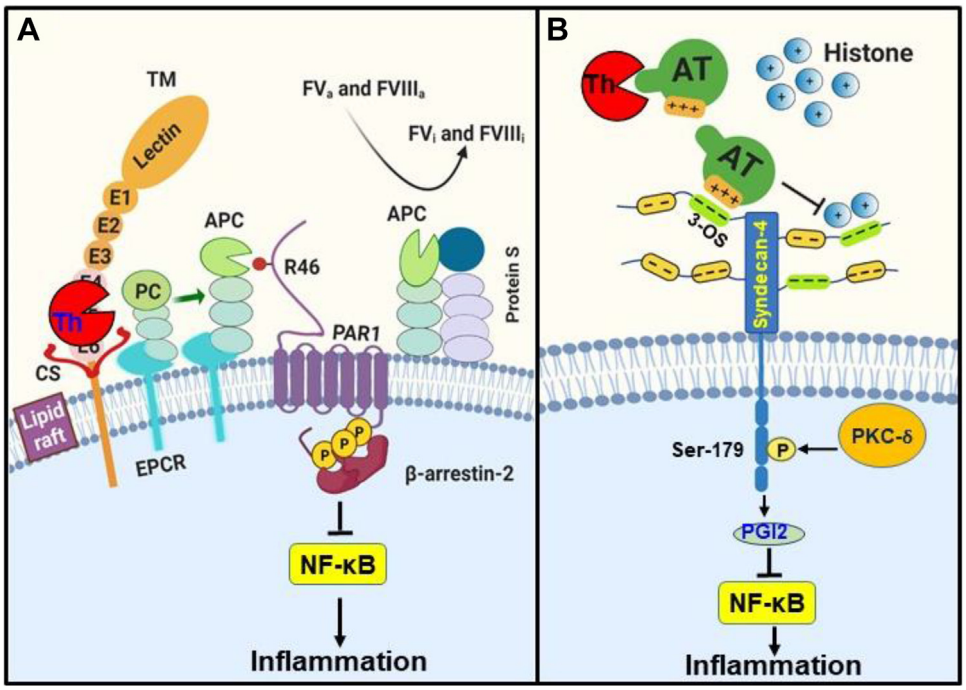




Patients with sickle cell disease (SCD) experience debilitating vaso-occlusive pain episodes (VOE) with ischemia/reperfusion and complement activation [12]. We hypothesized that complement activation is linked to VOE and pain. We induced VOE in Townes sickle (HbSS) mice with cold exposure and measured vaso-occlusion in dorsal skinfold chambers. Cold exposure (10 °C/50 °F, 1 hour) caused more vaso-occlusion in non-hyperalgesic HbSS mice (33%) than in HbAA mice (11%) or HbSS mice left at room temperature (1%). Cold exposure also produced mechanical hyperalgesia—assessed by hind paw withdrawal from von Frey filaments—in HbSS mice compared with HbAA mice or HbSS mice at room temperature. Cold exposure increased complement activation fragments Bb and C5a and increased hepatic proinflammatory nuclear factor-κB activation and VCAM-1 and ICAM-1. Pretreatment of nonhyperalgesic HbSS mice before cold exposure with anti-C5 or anti-C5aR monoclonal antibody (mAb) decreased vaso-occlusion, mechanical hyperalgesia, complement activation, and liver inflammatory markers as compared with pretreatment with control mAb. Anti-C5 or C5aR mAb infusion also abrogated mechanical hyperalgesia in HbSS mice with ongoing hyperalgesia at baseline [13]. These findings suggest that C5a promotes vaso-occlusion, pain, and inflammation during VOE and may play a role in chronic pain. IgG, immunoglobulin G.

ACTIVATED PROTEIN C AND ANTITHROMBIN IN IMMUNE REGULATION

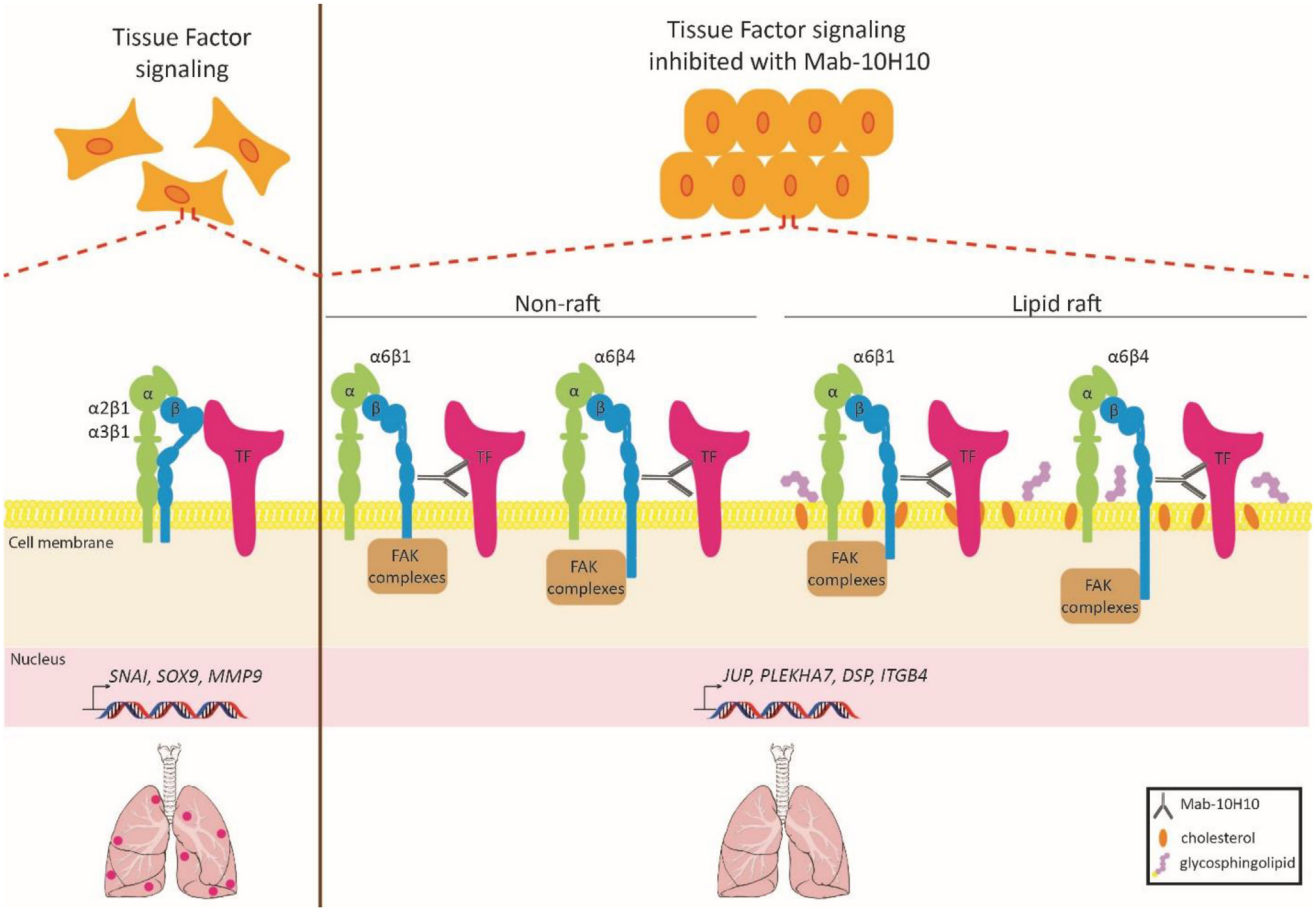
Alireza R. Rezaie



(A) The 3 receptors thrombomodulin (TM), endothelial protein C (PC) receptor (EPCR), and protease-activated receptor 1 (PAR1) are colocalized in lipid rafts of endothelial cells [14]. Thrombin (Th) binds TM and activates EPCR-bound PC to activated PC (APC). APC, in association with EPCR, functions in the anti-inflammatory pathway by activating PAR1, inducing a β-arrestin-2 biased signaling and inhibiting the nuclear factor-κB (NF-κB) pathway [15]. APC, upon dissociation from EPCR, binds protein S and inactivates procoagulant cofactors FVa and FVIIIa (FVi/FVIIIi) in the anticoagulant pathway. (B) Antithrombin (AT) is a plasma serine protease inhibitor that has a basic surface loop called D-helix that binds to 3-O-sulfate (3-OS) containing vascular heparan sulfate proteoglycans (primarily syndecan-4) to regulate inflammation and coagulation [19]. The interaction with AT culminates in phosphorylation of the cytoplasmic domain of syndecan-4 by protein kinase C (PKC)-δ, induction of prostacyclin (PGI2) synthesis, and inhibition of the NF-κB pathway [3,16]. AT also inhibits interaction of histones with overlapping AT-binding sites in a competitive manner. In addition to its anti-inflammatory signaling function, AT also regulates the activity of coagulation proteases by directly binding to their active sites via its reactive-center loop, thereby trapping them (shown for Th only) in the form of inactive covalent complexes [16]. CS, chondroitin sulfate; R46, Arg46; TAT, thrombin-AT complex.

INTEGRIN REGULATION BY TISSUE FACTOR PROMOTES CANCER STEMNESS AND METASTATIC DISSEMINATION IN BREAST CANCER

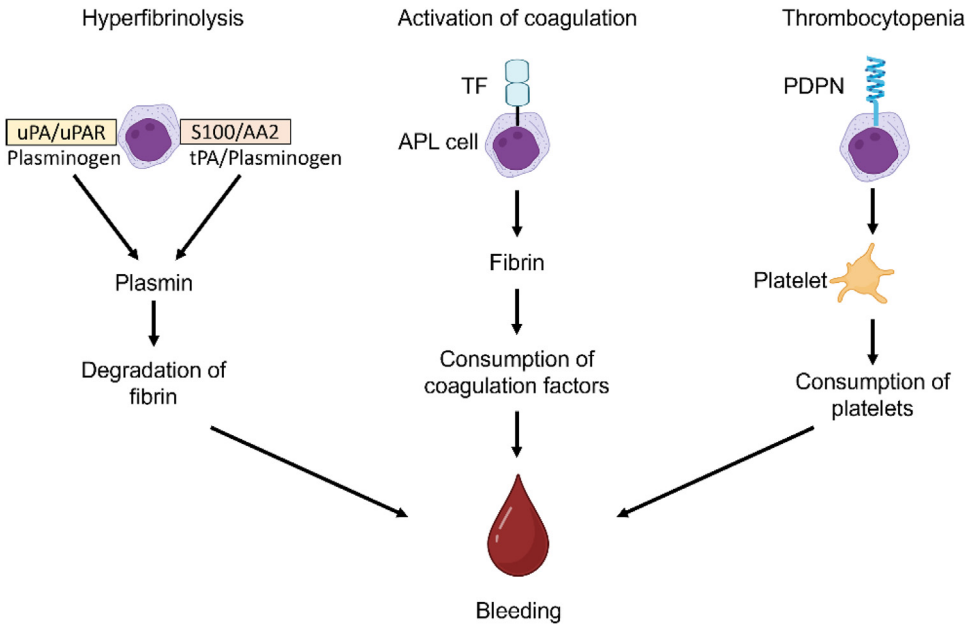
Betül Ünlü



Tissue factor (TF) expression associates with enhanced metastasis in patients with estrogen-receptor–negative tumors. Establishment of breast tumors in mice resulted in lung metastasis that required co-operation between TF and β_1 integrin signaling to promote an invasive phenotype. When TF signaling was inhibited with a TF-blocking antibody, less invasion, cancer stemness, and metastasis were observed. Mechanistically, blockade of TF signaling shifts the shape of the cells toward an endothelial morphology with increased expression of adherent junction genes. In addition, increased formation of focal adhesion complexes by integrins was observed, suggesting an activated integrin conformation. Furthermore, TF was found to be in complex with active $\alpha_6\beta_4$ integrins, a negative cancer stem cell marker. Thus, TF affects early onset of metastasis by affecting cancer stemness via regulation of the expression and function of β_1 and β_4 integrins. For details, see [17]. FAK, focal adhesion kinase; mAb, monoclonal antibody.

COAGULOPATHY AND BLEEDING IN ACUTE PROMYELOCYTIC LEUKEMIA

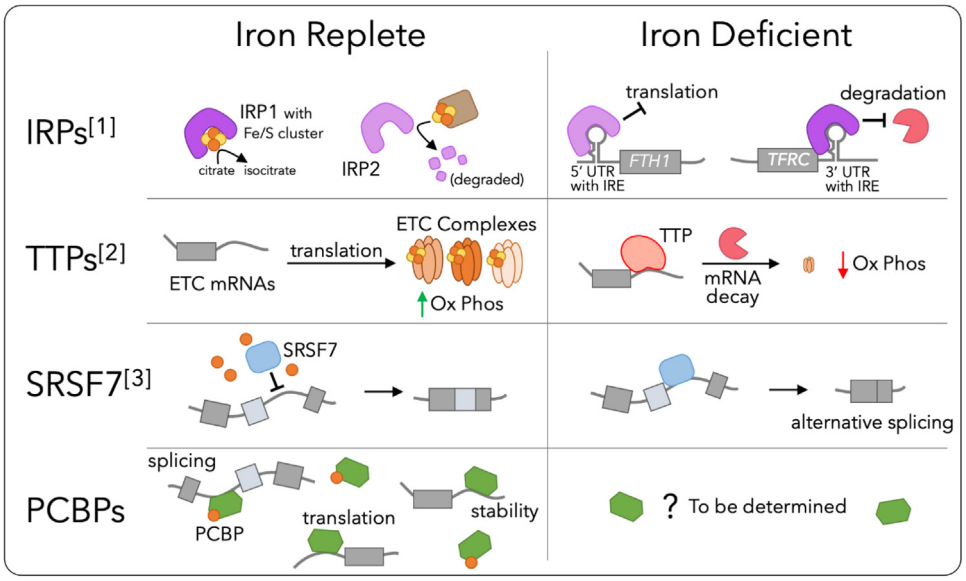
Yohei Hisada



Acute promyelocytic leukemia (APL) is a subtype of acute myeloid leukemia (AML) and accounts for 5% to 20% of AML cases. APL is associated with a high incidence of disseminated intravascular coagulation (60%-85%) and fatal bleeding, particularly intracranial hemorrhage. Several pathways that induce coagulopathy and associated bleeding were proposed. Tissue factor (TF) is a transmembrane protein and receptor for factor VII/FVIIa. The TF/FVIIa complex triggers activation of coagulation. APL cells express high levels of TF, which induces overactivation of coagulation that leads to consumption of coagulation factors [18]. In addition, APL cells express both S100A10 (S100) and annexin A2 (AA2). They form a heterotetrameric complex that serves as a receptor for both tissue plasminogen activator (tPA) and plasminogen. APL cells also express urokinase plasminogen activator (uPA) and uPA receptor (uPAR). Both S100/AA2 and uPA/uPAR complexes cleave plasminogen to plasmin, which degrades fibrin. Because of high expression of both S100/AA2 and uPA/uPAR complexes, APL induces hyperfibrinolysis [19]. A recent study showed that APL cells express higher levels of podoplanin (PDPN) compared with AML cells [19]. PDPN contributes to activation and consumption of platelets that lead to bleeding in APL.

IRON-SENSITIVE RNA REGULATION

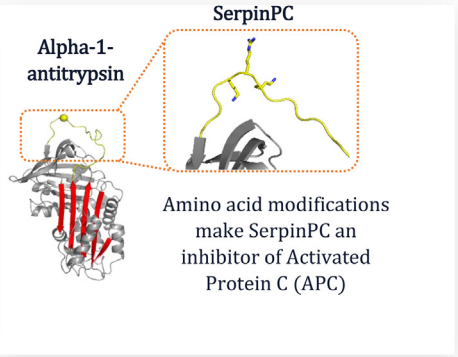
Maria M. Aleman

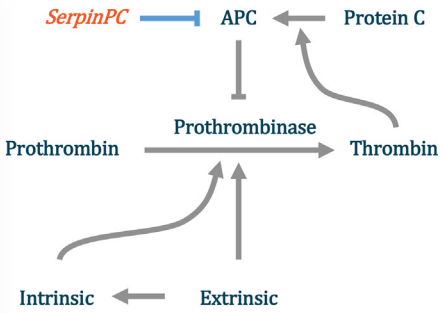


Iron deficiency remains the most common nutritional deficit worldwide and is associated with anemia, fatigue, and cognitive defects. Intracellular iron homeostasis has long been recognized to be regulated posttranscriptionally by iron regulatory proteins (IRPs) that moonlight as RNA-binding proteins. IRPs bind to iron regulatory elements (IREs) in untranslated regions (UTRs) of iron homeostasis messenger RNAs (mRNAs) when iron levels are low (reviewed in [20]). However, other RNA-binding proteins RNAs in an iron-sensitive manner, suggesting iron deficiency may cause broad changes in the transcriptome. Tristetraprolin (TTP) and paralogs are upregulated during iron deficiency and promote degradation of electron transport chain (ETC) component mRNAs to free up iron for use in more essential pathways [21]. Serine/arginine-rich splicing factor 7 (SRSF7) is a splicing factor demonstrated to have increased RNA regulatory activity, such as control of alternative splicing, with low iron levels [18]. Lastly, Poly C binding proteins (PCBPs), which regulate RNA splicing, stability, and translation also chaperone iron, suggest that PCBPs have the potential for iron-sensitive RNA regulatory activity and warrant further investigation. Ox Phos, oxidative phosphorylation.

MODIFIED ALPHA1 ANTITRYPSIN IN HEMOPHILIA

Speaker/Author: Trevor Baglin, MedScD, PhD





SerpinPC, a covalent inhibitor of activated protein C (APC) [22,23], designed with 3 substitution mutations in the reactive-center loop of α1AT, selectively targets APC.

APC, a serine protease, plays a key role in anticoagulation by degrading factor Va, the essential cofactor in prothrombinase. Under normal conditions, APC rapidly degrades the initial prothrombinase formed by extrinsic tenase. Consequently, activation of X by intrinsic tenase (VIIIa/IXa) is necessary to sustain prothrombinase activity at sites of injury. In persons with hemophilia lacking intrinsic tenase, thrombin generation due to prothrombinase produced by extrinsic tenase alone is insufficient for hemostasis. By inhibiting circulating APC, SerpinPC is able to preserve the activity of prothrombinase formed by extrinsic tenase.

Subcutaneous SerpinPC administration in knockout hemophilia mouse models eliminates bleeding. A phase 2a study demonstrates continued favorable safety and tolerability profile for SperinPC, as well as evidence of sustained efficacy, as measured by a reduction in the all-bleeds annualized bleeding rates. No thromboembolic events or treatment-related sustained elevations of D-dimer were observed in the study to date.

FUNDING

The authors received no funding for this study.

AUTHOR CONTRIBUTIONS

M.F.J., E.X.S., and E.M.S. organized the conference and invited the speakers. All remaining authors prepared a written and graphical abstract based on the contents of their invited seminars.

RELATIONSHIP DISCLOSURE

There are no competing interests to disclose.

X, FORMERLY KNOWN AS TWITTER

Maria Aleman 𝕏 @scientistaleman
Trevor Baglin 𝕏 @trevorbaglin
Pavel Davizon-Castillo 𝕏 @davizonpavel
Matthew J. Flick 𝕏 @fib390-396A
Erica M. Sparkenbaugh 𝕏 @sparkenbaughlab
Evi X. Stavrou 𝕏 @evistavrou
Betül Ünlü 𝕏 @betulunlu86

REFERENCES

[1] Fafian-Labora JA, O'Loghlen A. Classical and nonclassical intercellular communication in senescence and ageing. *Trends Cell Biol*. 2020;30:628–39.
[2] Mylonas A, O'Loghlen A. Cellular senescence and ageing: mechanisms and interventions. *Front Aging*. 2022;3:866718.
[3] Carpintero-Fernández P, Borghesan M, Eleftheriadou O, Pan-Castillo B, Fafián-Labora JA, Mitchell TP, et al. Genome wide CRISPR/Cas9 screen identifies the coagulation factor IX (F9) as a regulator of senescence. *Cell Death Dis*. 2022;13:163.
[4] Varju I, Kolev K. Networks that stop the flow: a fresh look at fibrin and neutrophil extracellular traps. *Thromb Res*. 2019;182:1–11.
[5] Varjú I, Sorvillo N, Cherpokova D, Farkas ÁZ, Farkas VJ, Komorowicz E, et al. Citrullinated fibrinogen renders clots mechanically less stable, but lysis-resistant. *Circ Res*. 2021;129:342–4.
[6] Varjú I, Tóth E, Farkas ÁZ, Farkas VJ, Komorowicz E, Feller T, et al. Citrullinated fibrinogen forms densely packed clots with decreased permeability. *J Thromb Haemost*. 2022;20:2862–72.
[7] Buitrago L, Lefkowitz S, Bentur O, Padovan J, Coller B. Platelet binding to polymerizing fibrin is avidity driven and requires activated alphaIIbbeta3 but not fibrin cross-linking. *Blood Adv*. 2021;5:3986–4002.
[8] Puy C, Pang J, Reitsma SE, Lorentz CU, Tucker EI, Gailani D, et al. Cross-talk between the complement pathway and the contact activation system of coagulation: activated factor XI neutralizes complement factor H. *J Immunol*. 2021;206:1784–92.
[9] Grover SP, Kawano T, Wan J, Tanratana P, Polai Z, Shim YJ, et al. C1 inhibitor deficiency enhances contact pathway-mediated activation of coagulation and venous thrombosis. *Blood*. 2023;141:2390–401.
[10] Keshari RS, Silasi R, Lupu C, Taylor Jr FB, Lupu F. In vivo-generated thrombin and plasmin do not activate the complement system in baboons. *Blood*. 2017;130:2678–81.
[11] Khandelwal S, Barnes A, Rauova L, Sarkar A, Rux AH, Yarovoi SV, et al. Complement mediates binding and procoagulant effects of ultralarge HIT immune complexes. *Blood*. 2021;138:2106–16.
[12] Vercellotti GM, Dalmasso AP, Schaid Jr TR, Nguyen J, Chen C, Ericson ME, et al. Critical role of C5a in sickle cell disease. *Am J Hematol*. 2019;94:327–37.
[13] Ivy ZK, Belcher JD, Khasabova IA, Chen C, Juliette JP, Abdulla F, et al. Cold exposure induces vaso-occlusion and pain in sickle mice that depend on complement activation. *Blood*. 2023;142:1918–27.
[14] Bae JS, Yang L, Rezaie AR. Receptors of the protein C activation and activated protein C signaling pathways are colocalized in lipid rafts of endothelial cells. *Proc Natl Acad Sci U S A*. 2007;104:2867–72.
[15] Mosnier LO, Sinha RK, Burnier L, Bouwens EA, Griffin JH. Biased agonism of protease-activated receptor 1 by activated protein C caused by noncanonical cleavage at Arg46. *Blood*. 2012;120:5237–46.
[16] Rezaie AR, Giri H. Anticoagulant and signaling functions of antithrombin. *J Thromb Haemost*. 2020;18:3142–53.
[17] Ünlü B, Kocatürk B, Rondon AMR, Lewis CS, Swier N, van den Akker RFP, et al. Integrin regulation by tissue factor promotes cancer stemness and metastatic dissemination in breast cancer. *Oncogene*. 2022;41:5176–85.
[18] Hambley BC, Tomuleasa C, Ghiaur G. Coagulopathy in acute promyelocytic leukemia: can we go beyond supportive care? *Front Med (Lausanne)*. 2021;8:722614.
[19] Lavallée VP, Chagraoui J, MacRae T, Marquis M, Bonnefoy A, Krosl J, et al. Transcriptomic landscape of acute promyelocytic leukemia reveals aberrant surface expression of the platelet aggregation agonist Podoplanin. *Leukemia*. 2018;32:1349–57.
[20] Andrews NC. Forging a field: the golden age of iron biology. *Blood*. 2008;112:219–30.
[21] Sato T, Chang HC, Bayeva M, Shapiro JS, Ramos-Alonso L, Kouzu H, et al. mRNA-binding protein tristetraprolin is essential for cardiac response to iron deficiency by regulating mitochondrial function. *Proc Natl Acad Sci U S A*. 2018;115:E6291–300.
[22] Polderdijk SG, Adams TE, Ivanciu L, Camire RM, Baglin TP, Huntington JA. Design and characterization of an APC-specific serpin for the treatment of hemophilia. *Blood*. 2017;129:105–13.
[23] Polderdijk SGI, Baglin TP, Huntington JA. Targeting activated protein C to treat hemophilia. *Curr Opin Hematol*. 2017;24:446–52.