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Non-Alloimmune Mechanisms of Thrombocytopenia and Refractoriness to Platelet Transfusion

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ABSTRACT

Refractoriness to platelet transfusion is a common clinical problem encountered by the transfusion medicine specialist. It is well recognized that most causes of refractoriness to platelet transfusion are not a consequence of alloimmunization to human leukocyte, platelet-specific, or ABO antigens, but are a consequence of platelet sequestration and consumption. This review summarizes the clinical factors that result in platelet refractoriness and highlights recent data describing novel biological mechanisms that contribute to this clinical problem.

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Platelet refractoriness describes the circumstance in which platelet transfusion fails to yield an adequate increase in the platelet count. Definitions for platelet refractoriness depend upon the timing of the post-transfusion platelet count and the metric used to assess the increase in platelet count (Table 1). One definition for platelet refractoriness is a corrected count increment less than $5X10^9/L/m^2$ on two sequential occasions after transfusion of ABO-compatible platelets [1]. Although alloimmunization to human leukocytes antigens (HLA) or platelet-specific antigens is a well understood mechanism of platelet refractoriness that can be ameliorated through specific product selection strategies, it is observed in <10% of cases of platelet refractoriness [2]. Thus, the majority of platelet refractoriness is not a result of alloimmunization but is instead a result of clinical factors in the patient [3]. Indeed, in patients with both clinical and alloimmune factors for platelet refractoriness, use of HLA-selected platelet components may not substantially improve the observed post-transfusion platelet increment [4]. The purpose of this review is to summarize the clinical factors associated with platelet refractoriness and highlight recent data describing biological mechanisms that contribute to this phenomenon.

Many different factors related to the patient's clinical state have been implicated as causes of platelet refractoriness (Table 2) [5-7]. In multivariate analyses conducted with different patient cohorts, splenomegaly, hematopoietic stem cell transplantation, disseminated intravascular coagulation, fever, bleeding, and use of antimicrobial agents have all been implicated in platelet refractoriness [8-12]. A subsequent analysis of the Trial to Reduce Alloimmunization to Platelets (TRAP), which enrolled 533 AML patients receiving induction chemotherapy and captured data from 6379 platelet transfusions, quantified the

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Table 1
Definitions of platelet refractoriness.

Variable	Formulae	Suggested cut-offs defining clinical refractoriness	
		1 h	16-24 h
Platelet Increment (PI)	Post-Pre	$<10 \times 10^9$/L	$<10 \times 10^9$/L
Percentage Platelet Recovery (PPR)	[PI × TBV* × 100%]/PD†	<20%	<10%
Corrected Count Increment (CCI)	[PI × BSA‡]/PD†	$<5 \times 10^9$/L/m^2	$<2.5 \times 10^9$/L/m^2

*Total blood volume (L); †platelet dose, typically 4×10^{11}/component; ‡body surface area (m^2)

impact of various non-immune clinical factors on the 1- and 18-24-hour post-transfusion increment. Aside from parity and gender, the most important factors that independently predicted a decrease in the post-transfusion platelet count at either time point were splenomegaly, exposure to amphotericin, bleeding, fever, and infection [12]. Interestingly, transfusion sequence number was an independent, albeit minor, predictor of a reduced platelet count increment at both 1- and 18- to 24-hour time points; the effect was most pronounced in the earliest transfusions and was independent of factors such as fever and infection. The authors hypothesized that chemotherapy-induced endothelial damage might mediate increased platelet adhesion and loss from the circulation [12]. Therefore, clinical studies have reproducibly identified a series of non-immune factors that can impact the outcome of platelet transfusion. With the exception of splenomegaly, the factors identified by these studies are presumed to either directly or indirectly reflect pathophysiologic processes that lead to increased platelet consumption in the patient.

Splenomegaly and Splenic Sequestration

The spleen influences post-transfusion platelet counts via sequestration. Seminal studies by Aster in which normal subjects and patients with splenomegaly were infused with ^{51}Cr-labeled platelets followed by quantitation of radioactivity in organs via surface scintillation scanning demonstrated that the spleen was the primary organ in which labeled platelets accumulated [13-15]. The accumulation of radiolabeled platelets in the spleen following platelet transfusion was rapid, occurring within approximately 10 minutes. With increasing splenomegaly, the proportion of labeled platelets detectable in the peripheral blood decreased while increasing in the spleen. It was estimated that at any given time, 30% of total platelet mass resided in the spleen of normal subjects and 50% to 90% of platelet mass in patients with splenomegaly [15]. Interestingly, epinephrine infusion increased circulating platelet levels in individuals with spleens but not in asplenic individuals. Furthermore, in patients with splenomegaly, the percent increase in platelet count from baseline was almost double that observed in normal individuals. Thus, platelet sequestration was reversible, and platelets could be mobilized by stress [15], distinguishing splenomegaly from consumptive processes in which transfused platelets are destroyed. Indeed, in many patients with splenomegaly, satisfactory post-transfusion increments may be observed [9]. However, among the subset of patients with massive splenomegaly resulting in repeatedly inadequate post-transfusion increments, the efficacy of prophylactic platelet transfusion is uncertain.

Table 2
Causes of platelet refractoriness

Antibody-mediated	Non-antibody-mediated
Platelet alloantibodies	Splenomegaly
Class I human leukocyte antigens	Infection
Human platelet glycoproteins	Fever
ABO	Disseminated intravascular coagulation
Platelet autoantibodies	Amphotericin B
Drug-dependent platelet antibodies (eg, quinine, β-lactam antibiotics, vancomycin)	Heparin
Immune complexes	Bleeding
	Graft versus host disease
	Veno-occlusive disease
	Transfusion sequence number
	Male sex
	Increasing weight

Novel Mechanisms of Platelet Consumption

The systemic inflammatory response and sepsis may prompt platelet consumption and thrombocytopenia through disseminated intravascular coagulation. However, recent work has highlighted multiple additional pathways through which infection and inflammation may lead to thrombocytopenia, including through direct interactions between platelets and pathogens, neutrophils and neutrophil extracellular traps, histone proteins, and the activated endothelium. See Fig. 1. Additionally, new insights suggest mechanisms through which platelet activation may result in increased clearance of platelets via apoptosis and desialylation.

Pathogen Recognition Receptor Signaling in Platelets

A series of studies over the last two decades have reported that platelets express pathogen recognition receptors (PRRs), including Toll-like receptors (TLRs) and NOD-like receptors. Prior to the discovery of PRRs, Davis et al observed that intravenous injection of purified lipopolysaccharide (LPS) from *E. coli* was associated with rapid platelet activation and profound thrombocytopenia [16]. More recently, several independent groups showed that the receptor required for LPS-mediated intracellular signaling, TLR4, is expressed on both human and mouse platelets [17-19]. In mouse models, platelet expression of TLR4 was involved in several LPS-induced platelet phenotypes, including thrombocytopenia, adhesion to fibrinogen, P-selectin expression, and potentiation of thrombin- or collagen-induced aggregation [17,18,20]. In addition to TLR4, it has been reported that platelets express TLR2 [18,19,21,22], TLR7 [23], TLR9 [24,25], and NOD2 [26], suggesting that platelets are also potentially responsive to a wide range of pathogen-associated molecular patterns. The bacterial cell wall mimetic, Pam3CSK4, stimulated platelet aggregation that was significantly reduced by TLR2 genetic deletion or a TLR2-blocking antibody [22]. Thrombocytopenia that developed in mice treated with the guanosine analog loxoribine or infected with encephalomyocarditis virus required TLR7 [23]. Thon et al reported that platelets exposed to unmethylated CpG DNA showed an increase in aggregation and P-selectin expression that was reduced in the absence of TLR9 [25]. Finally, NOD2 was essential for the muramyl dipeptide-mediated potentiation of platelet aggregation and ATP release after stimulation with collagen or thrombin [26].

Despite these data, the role of PRR signaling in platelet activation remains controversial. Several studies did not observe a significant change in platelet aggregation or P-selectin expression after treatment with agonists of TLR2, TLR4, or TLR9 [19,27-31]. In addition, platelet-specific deletion of the MyD88 signaling adaptor, which is required for signaling downstream of most TLRs, had a minimal effect on platelet counts and P-selectin expression in mouse models of *S. pneumoniae* and *K. pneumoniae* sepsis, implying that platelet TLR signaling may have a limited role in the platelet activation and thrombocytopenia observed

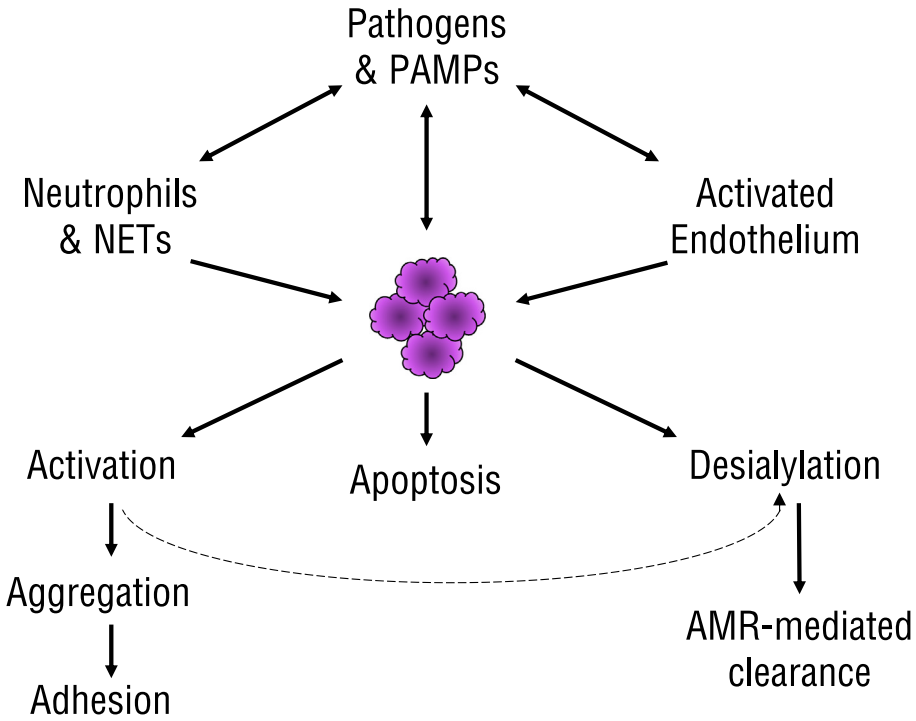

Fig. 1. Non-alloimmune mechanisms of thrombocytopenia and refractoriness to platelet transfusion. Platelet interactions with neutrophils, microbial pathogens, and activated endothelium all promote biological processes that lead to consumption and/or receptor-mediated clearance of endogenously produced and transfused platelets. AMR, Ashwell-Morell Receptor. NETs, neutrophil extracellular traps. PAMPs, pathogen-associated molecular patterns.

in systemic bacterial infections [30,32]. Whether engagement of TLRs modulates platelet biology via "non-canonical" mechanisms [28] remains an open question and deserves further investigation. However, the existence of such mechanisms is suggested by multiple studies in which binding of TLR4 by LPS alters platelet function in a TLR-dependent manner, including mitochondrial function [28], fibrinogen binding [17], neutrophil binding [17,27], and cytokine secretion [29].

Platelet Interactions with Bacteria

In addition to recognition of bacterial molecules via PRRs, platelet binding to whole bacterial pathogens has been widely described [33]. Platelet-bacterium interactions can promote platelet activation and aggregation through multiple mechanisms, including indirect binding to a plasma protein that is subsequently bound by a platelet receptor or direct bacterial protein binding to a platelet receptor. For example, *S. aureus* ClfA and FnbpA/B proteins both bind to fibronectin and fibrinogen, which facilitates interactions with the GPIIb/IIIa receptor on platelets [34]; another *S. aureus* surface protein, IsdB, promotes platelet-bacterium interaction via direct binding to glycoprotein (GP) IIb/IIIa [35].

Numerous other platelet-bacterium interactions involving staphylococcal and streptococcal proteins have been characterized and are presumed to play important roles in infective endocarditis and other pathologic states [36]. Platelet activation driven by bacterial interactions with GPIIb/IIIa and GPIbα receptors most likely contributes to platelet aggregation and consumption in sepsis. Interestingly, *H pylori* binding to von Willebrand factor (vWF) promotes GPIbα-mediated interactions with platelets [37], resulting in platelet activation, aggregation, and thrombocytopenia in a subset of patients. Remarkably, antibiotic therapy for *H pylori* that effectively resolves infection also normalizes platelet counts [38-41], suggesting that platelet-bacterium interaction alone could be sufficient to cause thrombocytopenia [42].

Neutrophil Extracellular Traps and Histone Proteins

In response to infection, neutrophils release neutrophil extracellular traps (NETs), which are composed of DNA, histones, and granule proteins [43]. NET release is a host defense mechanism employed by neutrophils to trap and kill bacteria, however exuberant NET formation or reduced NET clearance are implicated in numerous pathologic processes including autoimmunity, thrombosis, ischemia-reperfusion injury, and cancer progression [44-46]. Platelets appear to facilitate NET formation and to bind NETs. Infusion of LPS in mice caused thrombocytopenia and pulmonary sequestration of platelets through a mechanism that required both neutrophils and platelet TLR4 [17]. LPS was subsequently shown to bind platelet TLR4, leading to platelet activation and binding to neutrophils. The interaction between activated platelets and neutrophils resulted in neutrophil activation, degranulation, and NET formation. Plasma from severely septic, thrombocytopenic patients also stimulated NET release via TLR4-dependent platelet-neutrophil interactions [27]. Platelets may stimulate NET release via signaling between platelet P-selectin and neutrophil PSGL-1[47]. Platelets can bind directly to histone/DNA complexes within NETs or via plasma proteins that decorate NETs [46]. NET binding results in platelet adhesion, activation, and aggregation *in vitro* [48]. Histone H3 and H4 are able to directly bind to platelet TLR2 and TLR4 to mediate platelet aggregation [48,49]; platelet aggregation requires both histone-induced calcium signaling to activate GPIIb/IIIa and fibrinogen crosslinking of histone-coated platelets [49]. The pathophysiologic relevance of these findings is suggested by murine models in which NETs promote deep venous thrombosis [50,51] and histone infusion induces profound thrombocytopenia *in vivo* [49].

In addition to NETs, cell injury and death are other sources of circulating DNA and histones. High levels of circulating histones may be observed in patients with sepsis [52-54] or following trauma [55] and may contribute to end-organ injury [52,54,55]. In a murine model, infusion of a sublethal dose of purified histones resulted in pulmonary injury characterized by accumulation of neutrophils and platelet-rich thrombi in the alveolar microvessels, vacuolation of endothelial and epithelial

cells, intra-alveolar hemorrhage with accumulation of fibrin as well as platelet-rich microthrombi, and deposition of fibrin and collagen in the interalveolar septae [52]. A relationship between circulating histones and thrombocytopenia in the clinical setting is suggested by the observation that high levels of plasma histones detected in critically ill patients was associated with subsequent development of moderate to severe thrombocytopenia [56].

Platelet Apoptosis

Apoptosis comprises an ordered series of biochemical and morphologic changes that ultimately result in cell death [57]. Though nuclear condensation is one of the original hallmark features of apoptosis [58], it is now well-established that platelets, which lack a nucleus, can undergo apoptosis [59]. In addition, several lines of evidence indicate that apoptosis plays a significant role in determining platelet lifespan. In this context, it is possible that clinical factors (eg, drugs, inflammation, infection) could affect the lifespan of endogenously-produced and transfused platelets via accelerated apoptosis.

Oltersdorf et al reported a reduction in platelet counts in mice treated with ABT-737, a small molecule antagonist targeting the antiapoptotic proteins BCL2, BCL2L1 (aka BCL-xL), and BCL2L2 (aka BCL-w) [60]; similarly, patients with lymphoid malignancies who received a related compound with improved oral bioavailability, ABT-263, also developed dose-limiting thrombocytopenia [61,62]. One would predict that ABT-737 and ABT-263 also affect transfused platelets, though there are currently no published data on platelet lifespan after transfusion in mice or humans treated with either of these drugs.

Using a genome-wide screening approach in mice, Mason et al identified *Bcl2l1* as a key determinant of platelet lifespan [63]. Moreover, *in vitro* analyses demonstrated that both human and mouse platelets express BCL2, BCL2L1, and BCL2L2, suggesting that inhibition of antiapoptotic pathways promotes platelet death and removal from the circulation [64-66]. Genetic ablation or targeted inhibition of BCL2L1 alone was sufficient to recapitulate the thrombocytopenic phenotype observed with ABT-737, demonstrating that the activity of BCL2L1 is essential to prevent platelet apoptosis [64,67,68]. Interestingly, human platelets exposed to *E. coli* or *S. aureus* showed rapid BCL2L1 degradation, mitochondrial depolarization, and cytoplasmic condensation, indicating that bacteria or bacterial products have the capacity to induce platelet apoptosis [69]; it is likely that both endogenously-produced and transfused platelets would be affected by this pro-apoptotic mechanism.

Inhibition of protein kinase A (PKA) may promote apoptosis in platelets. A murine model in which PKA was knocked out only in the megakaryocytic lineage demonstrated that platelets in mice homozygous for the deletion had a significantly shortened life span and the mice themselves were thrombocytopenic. Interestingly, apoptosis was observed in platelets from thrombocytopenic patients with immune thrombocytopenic purpura (ITP), diabetes, and sepsis and was accompanied by markedly reduced PKA activity. Reduced PKA activity and apoptosis was detected in normal platelets following incubation in the plasma from patients with ITP or diabetes [70]. Antibodies directed against glycoprotein Ibα (GPIbα) can induce platelet apoptosis in vitro, which can be prevented by genetic or chemical inhibition of the AKT, a serine/threonine kinase that functions upstream of PKA [71] . Thus humoral factors may activate apoptotic pathways in platelets, shortening their lifespan and resulting in thrombocytopenia.

Examination of mice lacking proapoptotic proteins also supports a role for apoptosis in the regulation of platelet lifespan. Combined loss of *Bak* and *Bax*, which drive apoptosis via mitochondrial permeabilization, was associated with a significant increase in platelet number and survival in circulation [72]. In addition, thrombocytopenia in the setting of BCL2L1 loss or inhibition by ABT-737 was completely rescued in *Bak*/*Bax* double knockout mice, indicating that the equilibrium between pro- and antiapoptotic proteins is a key determinant of platelet lifespan [63,64,72]. Interestingly, platelet numbers and survival were minimally affected in mice lacking upstream activator(s) of BAK and BAX, including BAD, BBC3 (aka PUMA), BID, and BIM [64,73,74]; the absence of a robust platelet phenotype in these mice could reflect functional redundancy among these proteins or the presence of a novel BAK/BAX activation pathway.

Platelets also undergo phenotypic changes consistent with apoptosis, including mitochondrial depolarization, cytochrome c release, membrane blebbing and phosphatidylserine (PS) exposure [63,66,75]. In nucleated cells, exposed PS is recognized by several receptors and secreted proteins, serving as a canonical “eat me” signal that promotes cell clearance via phagocytosis [76]. Notably, PS exposure on platelets occurs after activation with physiologic agonists and promotes the procoagulant function of platelets by facilitating the assembly of tenase and prothrombinase complexes [77,78]. The precise role of PS exposure in platelet clearance is uncertain, though there is evidence from patients with acute myocardial infarction, bacteremia, dengue fever, and essential thrombocythemia suggesting that increased PS exposure is associated with platelet phagocytosis by neutrophils, macrophages, and endothelial cells [79-82]. Activation of Akt by antibodies against GPIbα results in exposure of PS on platelets and their phagocytosis by macrophages in the liver; conversely, inhibition of Akt signaling or prevention of PS exposure rescues platelets from phagocytosis [71]. Interestingly, platelet PS exposure due to activation and apoptosis appear to occur via distinct molecular mechanisms [75,83]; whether thrombocytopenia and platelet transfusion refractoriness in critically ill patients are predominantly linked to one or both of these mechanisms remains to be determined.

Platelet Desialylation

O- and N-linked glycans decorating platelet glycoproteins, particularly GPIbα, terminate in sialic acid residues [84]. Removal of sialic acids, or desialylation, exposes β-galactose moieties to which the sialic acids were linked. Exposed β-galactose on the surface of the platelet can lead to its binding and clearance from the blood via the Ashwell-Morrell receptor (AMR, also known as the asialoglycoprotein receptor) on hepatocytes and Kupffer cells [85-87]. Desialylation occurs as platelets age and may be a mechanism for removal of senescent platelets [86,88]. In a murine model of *S. pneumoniae* sepsis, marked thrombocytopenia results not from disseminated intravascular coagulation but is instead the result of platelet desialylation by the bacterial NanA neuraminidase, leading to platelet clearance by the AMR [89,90]. Similarly, Jansen et al recently reported that binding of influenza virus to platelet sialoglycans was associated with platelet desialylation by the viral neuraminidase [91], providing a possible mechanistic explanation for the thrombocytopenia observed in influenza-infected patients [92].

Interestingly, platelet activation also leads to desialylation. In mice, translocation and surface expression of endogenous platelet lysosomal neuraminidase occurs after platelet activation with antibodies directed against GPIbα, leading to platelet desialylation. Desialylation results in platelet clearance and thrombocytopenia in an Fc receptor (FcR)-independent mechanism that depends upon the AMR [93]. In patients with immune thrombocytopenic purpura, detection of autoantibodies targeting GPIbα predicts refractoriness to therapies that inhibit clearance by the FcR (ie, steroids and intravenous immunoglobulin), suggesting that platelet activation, desialylation, and AMR-mediated platelet clearance may be a key driver of thrombocytopenia in these patients [94,95]. In this context, increased platelet activation observed in SARS-CoV2 infection may underlie the thrombocytopenia and thromboembolic complications observed in patients with severe COVID-19 [96-98], though platelet sialylation in COVID-19 patients has yet to be examined. Binding of soluble vWF to platelet GPIbα under shear stress also results in platelet signaling, activation, desialylation and clearance. The mechanism of platelet activation involves shear stress-induced unfolding of a mechanosensory domain of GPIbα that occurs when

vWF binds, leading to platelet signaling and activation [99]. This mechanism may explain why increased binding of vWF to platelets, as in type 2B von Willebrand disease or following administration of ristocetin, results in thrombocytopenia [84]. Interestingly, platelet desialylation resulting from a marked increase in vWF binding to platelets is thought to mediate the thrombocytopenia observed in patients with acute dengue infection [100].

The clinical relevance of desialylation as a mechanism of thrombocytopenia in critically ill patients is supported by a recent prospective study [101]. Patients meeting the clinical definitions of sepsis, severe sepsis, and septic shock were stratified by the presence of thrombocytopenia, defined as a platelet count $<100X10^9$/L. The degree of platelet desialylation was compared between septic patients with and without thrombocytopenia and was found to be significantly greater among the group with thrombocytopenia. The patients enrolled in this study who met clinical criteria for severe sepsis with a platelet count $\leq 50X10^9$/L were further enrolled in a clinical trial in which they were randomized to receive standard antimicrobial therapy versus antimicrobial therapy combined with oseltamivir, a neuraminidase inhibitor that has clinical utility for the treatment and prevention of influenza. In comparison with patients receiving standard of care, a larger proportion of patients receiving oseltamivir increased their platelet count to at least $100X10^9$/L during the trial. Additionally, patients in the oseltamivir arm had a shorter duration of thrombocytopenia and received fewer platelet transfusions. Nevertheless, there was no impact of treatment with oseltamivir on overall 28-day mortality [101].

Endothelial activation and ADAMTS13 activity

Under healthy, steady-state conditions, platelet interactions with endothelial cells are limited by the endothelial glycocalyx, which serves the dual purposes of electrostatic repulsion and masking of platelet adhesion receptors [102]. Endothelial nitric oxide, prostacyclin, and CD39 ecto-ATPase activity also regulate platelet-endothelium interactions by inhibiting the surface expression of adhesion receptors on both platelets and endothelial cells [103]. Impairment of the endothelial mechanisms that prevent platelet adhesion occurs in numerous pathologic states, including ischemia [104], chronic kidney disease [105], hyperglycemia [106], dyslipidemia[107], trauma[108], inflammation [109], and sepsis [110]; it has also been suggested that platelet-endothelial interactions can initiate the development of atherosclerotic lesions [111,112]. These pathologic states are often associated with inflammatory cytokine production that leads to endothelial activation [113-115], which is marked by E-selectin, P-selectin, and vWF exposure on the endothelial luminal surface [116]. Endothelial P-selectin engages P-selectin glycoprotein ligand-1 (PSGL-1) or GPIbα on platelets to support platelet rolling along the endothelium [117-121]; platelet GPIbα binding to endothelial vWF also enables platelet rolling [122,123]. With additional inflammation and platelet activation, fibrinogen can facilitate firm adhesion of platelets to the endothelial surface by bridging platelet integrin αIIbβ3 with endothelial intercellular adhesion molecule-1 (ICAM-1) or αVβ3 [124]. Finally, activated platelets can promote endothelial cell activation [125,126], indicating that platelets have the capacity to initiate and sustain platelet-endothelial interactions.

It is likely that enhanced platelet-endothelial interaction contributes directly to thrombocytopenia in critically ill patients. Gawaz et al observed a significant increase in platelet-endothelial interaction *in vitro* when normal donor platelets were treated with plasma from septic patients compared to plasma from healthy individuals [127]. ADAMTS13 (*a d*isintegrin-like *a*nd *m*etalloprotease with *t*hrombo*s*pondin type I repeats 13) activity, which is required for the cleavage of vWF that releases platelets from endothelial interaction, is reduced in a significant fraction of critically ill, thrombocytopenic adults and children [128]. Notably, these patients are distinct from patients with thrombotic thrombocytopenic purpura (TTP): they typically have higher platelet counts and measurable ADAMTS13 activity (11-40% of normal); they do not harbor the autoantibodies that directly inhibit ADAMTS13 function, which are pathognomonic for TTP; and they do not respond to plasma exchange [129-131]. Not surprisingly, ADAMTS13 deficiency associated with critical illness and thrombocytopenia has been detected in the setting of sepsis with consumptive coagulopathy [130,132-143]; thrombocytopenic patients with non-infectious systemic inflammation have also been described to have below normal ADAMTS13 activity, though levels are typically higher in these patients compared to those with sepsis [132,134].

Conclusions

Splenic sequestration and disease processes that result in increased platelet consumption can result in refractoriness to platelet transfusion. Myriad mechanisms have been defined that potentially explain why thrombocytopenia and refractoriness to platelet transfusion are reproducibly observed in the setting of infection and inflammation. Understanding these pathophysiologic processes may lead to novel therapeutic interventions in the future, such as the use of neuraminidase inhibitors in patients with sepsis [101]. However, readers of this review are most probably confronted with the dilemma of what to offer now to the severely thrombocytopenic patient who is bleeding and unresponsive to platelet transfusion. On this question, there is little clinical guidance. After exonerating antibody-mediated clearance as a cause of platelet refractoriness, there is no product selection strategy available that will temper other mechanisms that significantly reduce platelet lifespan in the circulation. Attempting to exceed a defined platelet count threshold, which is itself anecdotally determined, with repeated platelet transfusions is unproven as a therapeutic intervention to treat bleeding, risks volume overload in the patient and contributes to local, regional, and national platelet shortages. Treatment decisions should be guided by careful assessment of the patient and the nature of the patient's bleeding. Where possible, local bleeding should be addressed through local measures, such as packing for a nosebleed, rather than platelet transfusion. Additionally, consideration should be given to administration of antifibrinolytic agents such as epsilon aminocaproic acid or tranexamic acid [7,144]; multiple clinical case series suggest benefit in bleeding patients with thrombocytopenia [145-147]. Randomized controlled trials using tranexamic acid in diverse clinical settings have demonstrated safety and observed no to minimal risk of thrombosis [148-150]. Prophylactic use of tranexamic acid in patients with hematologic malignancies receiving chemotherapy or stem cell transplant is currently being studied in a double-blind randomized controlled trial, the TREATT Trial [151]. In conclusion, platelet transfusion refractoriness due to clinical factors associated with critical illness are often unavoidable and unmodifiable, representing a significant therapeutic challenge and opportunity. As in many areas of transfusion medicine, well-designed clinical studies are needed to inform treatment decisions in this setting.

Conflict of interest

The authors have disclosed no conflicts of interest.

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