therapeutic benefit of ticagrelor in reducing the ability of platelets to promote ovarian cancer cell growth.

By identifying a central role for ADP in platelet-induced cancer proliferation and growth, this study then raises several new questions: (1) What is the nature of the molecular signals and underlying pathways that induce cancer cells to release ADP? (2) At what concentration is ADP released by cancer cells sufficient to activate platelets in the bloodstream? (3) What is the nature of the bioactive cargo deriving from ADP-P2Y12-activated platelets that drive and sustain proliferation programs in cancer cells?

Platelets have been shown to trigger an epithelial-mesenchymal–like transition in cancer cells while in the circulation.[4,10] ADP is considered a weak platelet agonist necessary for the amplification and positive feedback required to form a stable platelet clot. It is unclear what minimum number of ovarian cancer cells is required to generate sufficient concentrations of ADP to activate platelets, or whether platelet activation occurs systemically in the circulation or locally at primary or secondary sites of tumor growth. Along these lines, it is unknown whether secretion of ADP by either single circulating tumor cells (CTCs) or CTC clusters is sufficient to promote platelet activation and what effect this might have on the survival and metastatic success of CTCs. Perhaps the rate of ADP secretion by CTCs may serve as a useful biomarker for deciding which patients may benefit from the use of P2Y12 inhibitors as part of their cancer therapy. Alternatively, uncovering the mechanisms underlying the secretion of ADP from tumor cells may help identify novel druggable targets to prevent ADP-mediated platelet activation by targeting the tumor cells rather than systemically inhibiting platelet function, which carries the inherent risk of causing bleeding. In conclusion, the novel findings presented by Cho et al provide rationale for the development of new approaches and therapies specifically designed to prevent platelet-driven cancer proliferation and highlight the key role that platelets play in oncogenesis and metastasis.

Conflict-of-interest disclosure: The authors declare no competing financial interests ■

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● ● ● TRANSFUSION MEDICINE

Comment on Uhl et al, page 1247

Anemia and bleeding in thrombocytopenic patients

Jeffrey L. Carson[1] and Simon J. Stanworth[2] [1]RUTGERS ROBERT WOOD JOHNSON MEDICAL SCHOOL; [2]UNIVERSITY OF OXFORD

One might not have anticipated that lower hematocrit level is associated with bleeding in patients with hypoproliferative thrombocytopenia as suggested in a secondary analysis by Uhl et al in this issue of *Blood*.[1] However, patients with hematocrit ≤25% were reported to have a fivefold higher risk of grade ≥3 bleeding (odds ratio [OR], 5.09; 95% confidence interval [CI], 2.65-9.79) and a 1.2-fold higher risk of grade ≥2 bleeding (OR, 1.20; 95% CI, 1.03-1.39) compared with patients with hematocrit ≥29%.

Patients with hematologic malignancies frequently become thrombocytopenic and bleed as a result of their underlying malignancy or treatments, including cytotoxic chemotherapy and hematopoietic stem cell transplantation (HSCT). Indeed, the PLADO (Effects of Prophylactic Platelet Dose on Transfusion Outcomes) trial and many platelet transfusion trials[2] have reported overall rates of clinically significant bleeding in 50% or more of patients, irrespective of the nature of the policy for prophylactic platelet transfusions being evaluated. Several clinical and laboratory factors have been reported to be associated with an increased risk of bleeding, including the number of days (duration) with a platelet count $<10 \times 10^9$/L, liver disease, uremia, fever and/or sepsis, and medications. A strength of the PLADO study, a randomized clinical trial in which patients undergoing HSCT or chemotherapy for malignancy were randomly assigned to low-dose, medium-dose, or high-dose platelet transfusion strategies when their morning platelet count was $\leq 10 \times 10^9$/L, is the size of the trial. This allowed for factors that might contribute to bleeding, including anemia, to be explored for 16 320 patient-days on or after their first platelet transfusion in 1077 adult patients.

Experimental and clinical data provide additional evidence that the interaction of red blood cells with platelets enhances platelet function.[3] Studies that reduced hematocrit led to a reversible platelet dysfunction as measured by an increase in bleeding time in rabbits[4] and humans.[5,6] One study showed that

erythrocytes were involved with formation of a platelet plug, and a decrease of the platelet count by 50×10^9/L was reversed by a 10% increase in hematocrit.[7] In patients with acute myeloid leukemia, the time to bleeding was delayed in those with higher hemoglobin levels; for each increase of 1 g/dL in hemoglobin level, the relative risk (RR) of clinically significant bleeding was reduced by 22% (RR, 0.78; 95% CI, 0.61-1.00; $P = .048$).[8]

In contrast, a meta-analysis of 6 clinical trials comparing restrictive with liberal transfusion found a nonsignificant trend toward lower risk of re-bleeding in patients receiving transfusion at lower hemoglobin levels (ie, more anemic patients).[9] Of the 1489 patients in the restrictive transfusion group, 14.4% developed re-bleeding compared with 16.3% of the 1619 patients in the liberal transfusion group (RR, 0.75; 95% CI, 0.51-1.10). However, the 2 trials favoring restrictive transfusion were in patients with acute upper gastrointestinal bleeding. Re-bleeding in those patients may be because of higher blood pressure that results from administration of more blood transfusions leading to rupture of the thrombus plug. Thus, the results of these trials may not be applicable to patients with thrombocytopenia in whom the interaction between platelets and red blood cells may be important for normal hemostasis.

Several limitations of the study by Uhl et al deserve emphasis, including those that were described by the authors. The analysis demonstrating that the hematocrit was associated with bleeding did not adjust for partial thromboplastin time and international normalized ratio because these 2 tests were performed in only a minority of patients. The study did not demonstrate that the transfusion of red blood cells or platelets was associated with less re-bleeding the day after transfusion. However, this treatment effect cannot be assessed in an unbiased manner in an observational study because administration of blood products was not based on trial protocol but instead on the clinical judgment of the treating physician, which results in confounding by indication, a common problem in observational studies in transfusion medicine. Another limitation relates to the lack of information on clinical factors, including the common clinical conundrum of whether fever and infection are risk factors for bleeding, which could not be addressed in the PLADO secondary analysis. In a Trial of Prophylactic Platelets (TOPPS) modeling analysis, patients with a temperature of at least 38°C had the highest hazard of a grade 2 to 4 bleed (hazard ratio, 1.7; 95% CI, 1.3-2.4) vs those with a temperature of <37.5°C.[10]

Prospective studies are required to evaluate the relative importance of different risk factors for clinically significant bleeding (which would include the less common but very important groups of intracranial bleeding) and to address how these factors can be used to better target therapies. Different managements could apply for patients considered at higher risk of bleeding (possibly platelet and/or red cell transfusions and/or alternatives such as tranexamic acid), or to withhold prophylactic platelet transfusions (eg, in nonanemic patients undergoing autologous transplantation). Despite the careful secondary analysis of the PLADO trial and other studies cited, we must conclude that it remains unknown whether anemia is a clinically important independent risk factor for bleeding. The clinical question of the impact of hematocrit and red cell transfusion policy can be answered only with a randomized clinical trial that compares the risk of bleeding in a population of patients with high prevalence of thrombocytopenia who have been randomly assigned to a higher hemoglobin level (liberal) vs a lower hemoglobin level (restrictive) transfusion strategy. Most of the trials to date cannot answer this question because thrombocytopenia was uncommon in the study patients. An ideal population would be patients undergoing treatment for acute hematologic malignancies in which thrombocytopenia and bleeding are common. Small pilot studies have been performed that provide a road map for the conduct of such trials, and others are ongoing.[9] Red blood cell transfusion recommendations specific to the thrombocytopenic patient await high-quality evidence from clinical trials.[9]

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