To properly integrate preclinical observations into clinical practice, correlative studies of large patient cohorts are essential. To that end, Müller et al evaluated more than 800 patients enrolled in clinical studies with dasatinib following imatinib failure, and in 384 patients with detectable mutations at baseline, 63 different mutations were identified. Of mutations detected in at least 5 patients, 3 were associated with a particularly low probability of likelihood of CCyR achievement on dasatinib: *T315I* (0/21 cases), *F317L* (1/14 cases), and *Q252H* (1/6 cases). Similarly, a recent publication of imatinib-resistant CP-CML patients treated with nilotinib[8] has revealed a handful of problematic mutations in addition to *T315I* (*Y253H*, *E255K*, *E255V*, *F359C*, *F359V*). Again, it is scientifically satisfying that these mutations are the most relatively resistant to nilotinib after the *T315I* mutation.[5] Although neither report demonstrated a significant difference in outcome based on the presence or absence of any mutation at baseline, it is becoming clear that patients who harbor specific mutations may, in fact, be better served by treatment with a particular second-line agent.

A notable finding of the Müller et al study is the relative incidence of mutations that are potentially problematic to second-line therapies. Of the 384 patients with mutations, 42 patients had 1 of the 4 mutations documented to respond poorly to dasatinib, while 103 had mutations that do not respond well to nilotinib, a finding that is in general agreement with other studies that have assessed the relative frequencies of imatinib-resistant mutations in large cohorts of patients.[9] This higher degree of cross-resistance between imatinib and nilotinib (relative to dasatinib) is not surprising given the degree of structural similarity between imatinib and nilotinib. However, it must be noted that no randomized controlled studies comparing dasatinib and nilotinib in patients with imatinib failure have been performed, and both agents achieve CCyR in a substantial proportion of CP-CML patients with imatinib resistance.

Nonetheless, the findings of Müller et al[1] and Hughes et al[8] provide clinicians with guidance regarding the preferential choice of a second-line agent. For imatinib-intolerant patients, there is no clinical evidence at this time to support either dasatinib or nilotinib, and weighing individual comorbidities with drug side effect profiles can be important. It is interesting that both Müller et al and Hughes et al identified *BCR-ABL* mutations in a small but significant proportion of imatinib-intolerant patients (8% and 10%, respectively), suggesting that some patients with imatinib-"intolerant" disease have evolved subclinical imatinib resistance, and screening imatinib-intolerant patients, in addition to imatinib-resistant patients, for *BCR-ABL* kinase domain mutations may further guide clinicians in pursuit of the optimal second-line therapy.

Because CML represented the very first human malignancy to be treated with small-molecule tyrosine kinase inhibitors, it is hoped that the clinical experience with imatinib, dasatinib, and nilotinib will establish a paradigm that can be applied to this burgeoning class of molecules. With the recent work of Müller et al and Hughes et al, the era of personalized medicine for CML has arrived.

Conflict-of-interest disclosure: The author declares no competing financial interests. ■

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● ● ● PLATELETS & THROMBOPOIESIS

Comment on Salles et al, page 5044

Ready to analyze genetically modified human platelets

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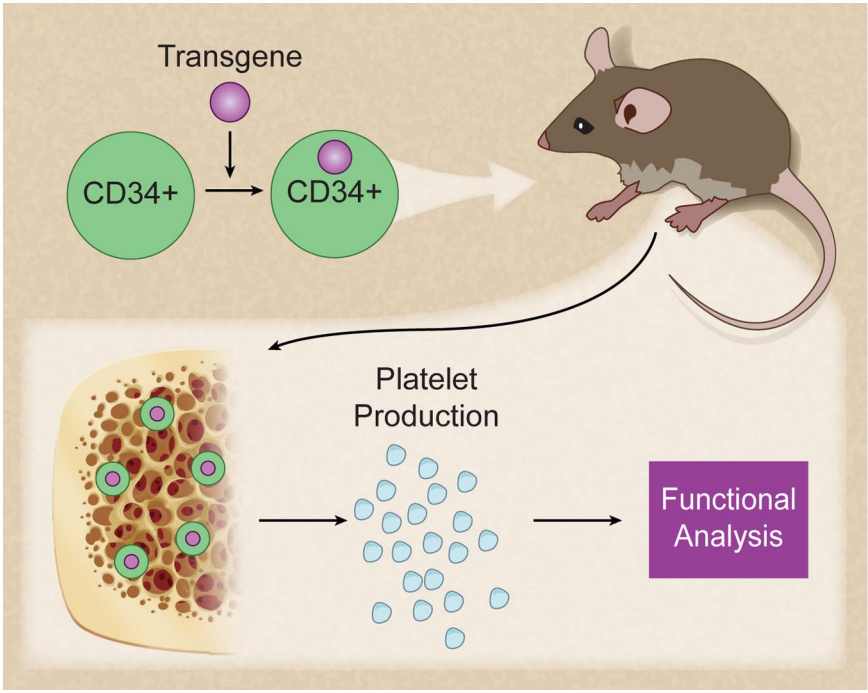
Platelets play a critical role in hemostasis and thrombosis. In this issue of *Blood*, a new study presented by Salles and colleagues demonstrates that functional human platelets can be generated in mice after transplantation of human hematopoietic stem cell progenitors.[1] This method opens up new avenues to study thrombopoiesis and platelet function in vivo, and the effect of distinct genetic modifications.

Platelets are anucleated cells that play a central role in hemostasis and thrombosis. In the past, considerable interest has focused on the identification and characterization of pivotal platelet proteins to further evaluate platelet physiology and new pharmacologic strategies. That these proteins cannot be easily accessed hampers the study of the role of proteins in platelet function. Genetic modification of mature platelets is not possible, thus evaluation of the function of transgenes in platelets requires the generation of platelets from nucleated progenitor cells in vitro.

Several studies have shown that platelets can be generated from CD34-positive progenitor cells or from megakaryocytes in vitro, and that these culture-derived (CD) platelets have some characteristic morphologic features of mature platelets.[2,3] The development of CD platelets permits expression of any protein of interest in these platelets and analysis of its function in the natural environment of primary mature platelets.[4,5] However, this method previously has been limited to in vitro analysis.

The current article by Salles et al[1] describes a mouse model that allows the study of the



In vivo production of genetically modified human platelets. Professional illustration by Debra T. Dartez.

function of human platelets in vivo (see figure). The authors convincingly demonstrate for the first time that human platelets can be produced in nonobese diabetic/severe combined immunodeficient mice after xenotransplantation of CD34-positive cord blood cells. Generation of human platelets occurred rapidly approximately 10 days after transplantation with a second peak at 5 weeks. Salles et al further show that human platelets generated in mice are functional as assessed by flow cytometry and thrombus formation in flow chamber experiments.

In summary, this intriguing new method opens an avenue for the exploration of the function of any protein in the environment of mature platelets in vivo. Defining the functional role of transgenes in platelets may disclose so far hidden targets to control platelet function and to evaluate new mechanisms in megakaryopoiesis.

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