Comment on Pecquet et al, page 4625

JAK2 V617F down-modulates MPL

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Decreased expression of the thrombopoietin receptor (TPOR or MPL) on the cell surface of platelets and megakaryocytes is an established feature of polycythemia vera and myelofibrosis1,2; however, the exact mechanism responsible for this phenomenon has gone largely unexplained. In this issue of Blood, Pecquet and colleagues publish an excellent study revealing that MPL expression is down-regulated in the context of the mutant protein, JAK2V617F3.

JAK2V617F induces enhanced ubiquination of MPL leading to increased proteasomal degradation. The overall consequence of this process is reduced recycling and maturation of the receptor and lower expression of MPL on the cell surface. Notably, this finding is consistent with the previous observation of defective posttranslational processing of MPL and increased susceptibility to endoglycosidase H digestion in megakaryocytes from polycythemia vera patients4, and it is now clear that JAK2V617F plays an important role in this process.

Previous work by Royer et al demonstrated that wild-type JAK2 can stabilize the mature endoglycosidase H-resistant form of MPL, thereby promoting enhanced total levels of the receptor on the cell surface5 (see A and B in figure). In the present study, Pecquet and colleagues make the surprising finding that JAK2V617F has lost this ability. Consequently, cells that express JAK2V617F exhibit an increase in MPL degradation by the proteosome (C in figure). Restoration of cell surface expression of MPL can be achieved with pharmacologic inhibition of either the proteasome or JAK kinase activity (D in figure). These phenomena are consistently demonstrated in a Ba/F3 cell line model system, murine platelets expressing JAK2V617F from the endogenous JAK2 locus, as well as primary cells from myelofibrosis patients.

It has recently been shown that high concentrations of the MPL ligand, thrombopoietin (TPO), can have an antiproliferative effect on cells6 (B in figure). This may serve as a physiologic mechanism to dampen proliferation at late stages of megakaryocyte development. Accordingly, genetically modified mice expressing low levels of MPL at the late megakaryocyte stage develop thrombocytosis7. It is therefore possible that down-regulation of MPL on the surface of platelet progenitors by JAK2V617F allows megakaryocytes to escape this TPO-mediated mechanism of cellular homeostasis, leading to enhanced cell survival and proliferation (C in figure). While it has been reported that patients with essential thrombocythemia do not exhibit decreased MPL expression1, this issue has not been revisited since the discovery of the JAK2V617F mutation. An interesting follow-up to this study would be to distinguish JAK2WT and JAK2V617F.
JAK2V617F patients with essential thrombocythemia and analyze MPL cell surface levels. Decreased MPL surface expression in the context of JAK2V617F would clarify that enhanced MPL degradation is a universal feature of JAK2V617F-positive disease. Conversely, lack of a difference between JAK2V617F and JAK2WT would suggest that the mechanism of excessive platelet production in essential thrombocythemia is distinct from that in polycythemia vera.

There are a variety of implications and future directions that arise from these findings. Considering a clinical standpoint, it is worth pondering whether proteasome inhibitors might yield clinical benefit in the setting of JAK2V617F-positive disease, because inhibition of the proteasome could restore normal levels of MPL on the surface of neoplastic cells and, therefore, restore the quiescent/apoptotic effects of high-concentration TPO signaling. Unfortunately, bortezomib has not been found to demonstrate clinical activity in myelofibrosis as a single agent in a phase 2 clinical trial. As such, it is also worth considering whether combinations of proteasome and JAK2 inhibitors would show synergistic effects on cells. Analysis of surface levels of MPL after kinase/proteasome inhibitor therapy may also serve as a useful biomarker for evaluation of drug efficacy.

Although this study helps explain a long-standing observation in myeloproliferative neoplasms, there are still a variety of mechanistic questions to address. These include the precise nature by which proteasomal degradation of MPL is induced in the context of JAK2V617F. It is possible that this is a passive process whereby JAK2WT innately has the capacity to protect MPL from proteasomal degradation and JAK2V617F has lost this ability. However, the fact that JAK kinase inhibition reverses this phenomenon in the context of JAK2V617F suggests that this is an active process initiated and/or coordinated by signaling from the JAK2V617F mutant protein. Identifying the precise molecular interactions that allow JAK2WT to protect against and JAK2V617F to promote proteasomal degradation will be critical to enhancing our understanding of this process. One useful starting point would be identification of the operationally important ubiquitin ligase. It will also be interesting to study the effect of JAK2WT/V617F heterozygosity versus homozygosity on these signaling events and molecular interactions.

Finally, the observation that TPO can induce either growth or quiescence/apoptosis in a concentration-dependent manner requires further mechanistic exploration. One possibility would be that high concentrations of ligand result in TPO dimers, causing oligomerization of MPL receptors. These aggregated MPL complexes may then form a signalosome with additional receptors and different intracellular components than are normally associated with individual MPL homodimers. Similar mechanisms have been proposed for other ligand/receptor systems. Further interrogation of the precise molecular switches that control this concentration-dependent signaling process will be of great interest and use. In sum, the present study by Pecquet and colleagues has shed light on a long-established feature of myeloproliferative neoplasms. In doing so, their work has suggested novel clinical possibilities as well as a variety of follow-up mechanistic studies to further our understanding of this important process.

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REFERENCES


30. CLINICAL TRIALS

Comment on Leonard et al, page 4597

Hitting the target in MCL

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One of the major challenges in the evaluation of new therapies for lymphoma and other hematologic malignancies has been the ability to demonstrate changes in important biomarkers and pharmacodynamic end points in the tumor cell population. In this issue of Blood, Leonard et al provide an elegant evaluation of the cyclin-dependent kinase (cdk) inhibitor PD0332991 in patients with relapsed mantle cell lymphoma. Their results suggest that the combination of functional imaging using fluorothymidine–positron emission tomography (FLT-PET) and immunohistochemistry can provide important information about target inhibition in tumor cells, and the effects this inhibition has on proliferation and metabolism.

The identification of the signature t(11;14) translocation in mantle cell lymphoma (MCL) has lead to a wealth of information describing the many abnormalities of cell-cycle regulation in this therapeutically challenging subtype of lymphoma, and has accelerated the development and testing of numerous agents that target, at least in vitro, these diverse pathophysiologic processes. Activation and inhibition of signaling proteins, cell-cycle regulatory proteins, and other pathways, however, may be discordant in normal tissues and malignant tumors. Pharmacodynamic assessment that relies on surrogate tissues rather than tumor cells may not be optimal in describing the true effects of targeted therapy. For example, evaluation of the oral BCL-2 inhibitor ABT-263 (navitoclax) in
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