highly potent small-molecule Jak inhibitors, targeting the Jak1 and Tyk2 kinases associated with the IL-4Rα/IL-13Rα1 complex is also a consideration. In principle, STAT6 may also be an attractive target given the relative specificity of its activation by IL-4 and IL-13. However, clinical-grade STAT6 inhibitors are still under development. It could be argued that the selective targeting of IL-13/IL-4–mediated cell activation as a single therapeutic modality should be the most effective in erythroderma/SS and the MF cases with the strongest Th2 bias. It may, however, be insufficient in other cases and its effects may be transient even in the responding patients, unless combined with other therapeutic approaches.

In summary, identification of IL-13 as an important factor in CTCL expands our understanding of the pathogenesis of this complex lymphoproliferative disorder and has potential therapeutic implications.

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

REFERENCES

1. Geskin LJ, Viragova S, Stolz DB, Fuschiotti P. Interleukin-13 is overexpressed in cutaneous T-cell lymphoma cells and regulates their proliferation. *Blood.* 2015;125(18):2798-2805.

2. Vaqué JP, Gómez-López G, Monsálvez V, et al. *PLCG1* mutations in cutaneous T-cell lymphomas. *Blood.* 2014;123(13):2034-2043.

3. Zhang Q, Nowak I, Vonderheid EC, et al. Activation of Jak/STAT proteins involved in signal transduction pathway mediated by receptor for interleukin 2 in malignant T lymphocytes derived from cutaneous anaplastic large T-cell lymphoma and Sezary syndrome. *Proc Natl Acad Sci USA.* 1996;93(17):9148-9153.

4. Abraham RM, Zhang Q, Odum N, Wasik MA. The role of cytokine signaling in the pathogenesis of cutaneous T-cell lymphoma. *Cancer Biol Ther.* 2011;12(12): 1019-1022.

5. Netchiporouk E, Litvinov IV, Moreau L, Gilbert M, Sasseville D, Duvic M. Deregulation in STAT signaling is important for cutaneous T-cell lymphoma (CTCL) pathogenesis and cancer progression. *Cell Cycle.* 2014; 13(21):3331-3335.

6. Willerslev-Olsen A, Litvinov IV, Fredholm SM, et al. IL-15 and IL-17F are differentially regulated and expressed in mycosis fungoides (MF). *Cell Cycle.* 2014; 13(8):1306-1312.

7. Wolk K, Mitsui H, Witte K, et al. Deficient cutaneous antibacterial competence in cutaneous T-cell lymphomas: role of Th2-mediated biased Th17 function. *Clin Cancer Res.* 2014;20(21):5507-5516.

8. Ohmatsu H, Humme D, Gulati N, et al. IL32 is progressively expressed in mycosis fungoides independent of helper T-cell 2 and helper T-cell 9 polarization. *Cancer Immunol Res.* 2014;2(9):890-900.

9. Singer EM, Shin DB, Nattkemper LA, et al. IL-31 is produced by the malignant T-cell population in cutaneous T-cell lymphoma and correlates with CTCL pruritus. *J Invest Dermatol.* 2013;133(12):2783-2785.

10. Guenova E, Watanabe R, Teague JE, et al. TH2 cytokines from malignant cells suppress TH1 responses and enforce a global TH2 bias in leukemic cutaneous T-cell lymphoma. *Clin Cancer Res.* 2013;19(14):3755-3763.

© 2015 by The American Society of Hematology

● ● ● MYELOID NEOPLASIA

Comment on Rijal et al, page 2815

INPP4B, a new player in the chemoresistance of AML

Christian Recher   INSTITUT UNIVERSITAIRE DU CANCER DE TOULOUSE ONCOPOLE

In this issue of *Blood*, Rijal et al have identified the inositol polyphosphate 4-phosphatase II (INPP4B) as a poor prognostic marker in acute myeloid leukemia (AML), driving chemoresistance through a novel gain-of-function mechanism independent of its phosphatase functions.[1]

Although there has been no therapeutic breakthrough for the treatment of AML over the last 20 years, the cytogenetic and molecular characterization of this disease has allowed stratifying of risk groups with very different prognoses in terms of disease-free and overall survival (OS), and has identified relevant therapeutic targets currently under investigation, such as Fms-like tyrosine kinase 3-internal tandem duplication (*FLT3*-ITD), IDH1/2, or epigenetic regulators.[2,3] Furthermore, the analysis of residual disease following chemotherapy also helps to stratify postremission therapy. Thus, do we need another new prognostic marker to manage our patients? The answer is "yes" if this marker predicts early response to chemotherapy (ie, primary induction failure), and more importantly, if this new marker is linked to chemoresistance. Rijal et al investigated the expression profile of several phosphatases involved in the phosphoinositide-3 kinase (PI3K) pathway and found that INPP4B can fill in these 2 requirements.

Class I PI3K that produces the D3-phosphoinositide second messenger phosphatidylinositol 3,4,5-trisphosphate in response to membrane receptor activation plays a critical role in cell proliferation, survival, metabolism, and motility. These lipid kinases and the phosphatases regulating the level of D3-phosphoinositides have been an intense area of research in the field of cancer for the past 2 decades.[4] Most AML display a constitutive activation of the PI3K pathway that contributes to cell proliferation and chemoresistance.[5] Several mechanisms converge to activate the PI3K pathway in AML, including mutated receptor tyrosine kinases (ie, *FLT3*-ITD), growth factors (ie, insulin-like growth factor 1), or microenvironment players (ie, SDF1α/ CXCR4 or fibronectin/VLA-4 modules). Much less is known regarding the negative regulators of this pathway. Rijal et al first performed a quantitative gene expression screen of 11 phosphoinositide phosphatases using a mass spectrometry detection system in 22 AML samples and found a reduction of the 5-phosphatase (SH2 domain-containing inositol 5'-phosphatase 2) expression, whereas *INPP4B* and *INPP5B* were significantly overexpressed compared with normal bone marrow (BM) cells. However, high *INPP4B* expression solely emerged as a potential indicator of poor AML survival. Having verified good concordance between *INPP4B* gene expression and protein levels, they assessed INPP4B protein expression in a panel of 205 BM biopsies from patients treated with intensive chemotherapy and eventually allogeneic stem cell transplantation as postremission therapy. They identified 25 patients (12%) with high INPP4B expression and poor outcome both in terms of leukemia-free and OS. There was no significant association between high INPP4B expression and other classical poor risk features (age, secondary AML, leukocytosis, cytogenetic risk, or *FLT3*-ITD). However,

among patients with intermediate risk cytogenetics, most patients with high INPP4B expression were FLT3 and nucleophosmin 1(NPM1) wild-type (WT). Importantly, in patients <65 years of age, a higher INPP4B expression predicted a lower complete response rate (39% vs 71% in low expressers). Moreover, high INPP4B remained an independent factor of poor OS in multivariate analysis performed with or without censoring at transplantation.

Rijal et al went on to explain why INPP4B overexpression impacted on prognosis focusing on chemoresistance, since high expressers more often experienced primary induction failure. First, they demonstrated that overexpressed INPP4B was catalytically active in AML cells. However, a striking observation was the absence of correlation between INPP4B protein levels and the phosphorylation of protein kinase B (AKT) on serine 473, a surrogate marker of PI3K activation, suggesting that although INPP4B has been shown to be a negative regulator of the PI3K pathway, it could play a different role in AML. Next, using ectopically INPP4B overexpression in low-expressing or specific INPP4B depletion in high-expressing AML cells, they provided evidence that INPP4B is an important player in the resistance to drugs used in the management of AML (cytarabine, daunorubicin, and etoposide). They judiciously used a catalytically inactive mutant of INPP4B (INPP4B-C842A) and unexpectedly, showed that it also mediated chemoresistance. Thus, these results demonstrated that INPP4B mediates chemoresistance through a novel gain-of-function mechanism independent of its phosphatase activity in AML. Lastly, they validated in vivo the role of both INPP4B and INPP4B-C842A overexpression using xenografts in immunocompromised mice.

The first question we generally ask over such clinical results concerns its reproducibility in validation cohorts. Interestingly, other investigators very recently reported a similar clinical impact of INPP4B overexpression through the analysis of several publicly available AML datasets, strengthening the findings of Rijal et al.[6] Higher induction failure was also observed in this study, which is of particular interest with respect to our limited ability to predict resistance based on routinely available pretreatment covariates.[7] However, further

studies are needed to address several issues raised by this study: (1) What is the expression level of INPP4B in residual cells after chemotherapy or at relapse after clonal evolution (especially in patients with low expression at diagnosis)? (2) Which molecular mechanisms lead to its overexpression in AML? (3) Are there any correlations between INPP4B expression and other molecular markers since a higher expression was found in patients with WT FLT3 and NPM1? (4) What is the role of INPP4B in normal hematopoiesis? It would be interesting to determine if INPP4B also drives resistance in AML patients treated with hypomethylating agents.

How does INPP4B drive chemoresistance in AML? The field is open. Because INPP4B reduces the activity of drugs with various mechanisms of action, it is likely that it could be involved in apoptotic response rather than in drug-specific metabolisms. INPP4B overexpression in leukemic cells did not impact the phosphorylation of AKT or the expression of antiapoptotic members of the Bcl-2 family. Thus, critical downstream targets of INPP4B remain to be determined. The finding that INPP4B could act independently of its phosphatase activity raises several clues to investigate how cells resist to chemotherapy. Phosphoinositide enzymes have both a catalytic and a molecular adapter activity that are crucial to organizing multimolecular complexes. As discussed by the authors, INPP4B contains an N-terminal C2-lipid binding domain, which interacts with membranes. It also

contains a Nervy homology 2 domain known to mediate oligomerization (ie, AML1-ETO oligomerization) or protein-protein interaction.[8] Thus, the findings of Rijal et al pave the way to perform further molecular studies of the INPP4B interactome in order to identify new therapeutic targets aimed at unlocking chemoresistance in AML.

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

REFERENCES

1. Rijal S, Fleming S, Cummings N, et al. Inositol polyphosphate 4-phosphatase II (INPP4B) is associated with chemoresistance and poor outcome in AML. *Blood.* 2015;125(18):2815-2824.

2. Patel JP, Gönen M, Figueroa ME, et al. Prognostic relevance of integrated genetic profiling in acute myeloid leukemia. *N Engl J Med.* 2012;366(12):1079-1089.

3. Estey E, Levine RL, Löwenberg B. Current challenges in clinical development of "targeted therapies": the case of acute myeloid leukemia. *Blood.* 2015;125(16):2461-2466.

4. Thorpe LM, Yuzugullu H, Zhao JJ. PI3K in cancer: divergent roles of isoforms, modes of activation and therapeutic targeting. *Nat Rev Cancer.* 2015;15(1):7-24.

5. Park S, Chapuis N, Tamburini J, et al. Role of the PI3K/AKT and mTOR signaling pathways in acute myeloid leukemia. *Haematologica.* 2010;95(5):819-828.

6. Dzneladze I, He R, Woolley JF, et al. INPP4B overexpression is associated with poor clinical outcome and therapy resistance in acute myeloid leukemia [published online ahead of print March 4, 2015]. *Leukemia.* doi: 10.1038/leu.2015.51.

7. Walter RB, Othus M, Burnett AK, et al. Resistance prediction in AML: analysis of 4601 patients from MRC/NCRI, HOVON/SAKK, SWOG and MD Anderson Cancer Center. *Leukemia.* 2015;29(2):312-320.

8. Zhang J, Hug BA, Huang EY, et al. Oligomerization of ETO is obligatory for corepressor interaction. *Mol Cell Biol.* 2001;21(1):156-163.

© 2015 by The American Society of Hematology

● ● ● **RED CELLS, IRON, AND ERYTHROPOIESIS**

Comment on Stonestrom et al, page 2825

Toward a BETter grasp of acetyl-lysine readers

Catherine Porcher   UNIVERSITY OF OXFORD

In this issue of *Blood*, Stonestrom et al describe the unanticipated complexity of the distinct yet overlapping activities of acetyl-lysine–binding bromodomain and extraterminal motif (BET) proteins bromodomain-containing 2-4 (BRD2-4) in erythropoiesis, in the context of rising interests in BET pharmacologic inhibitors.[1]

Epigenetic modifications dictate chromatin structure and affect gene expression. It has been long established that aberrant activity

of the enzymes controlling the deposition, recognition, and removal of these chromatin marks often leads to cancer. Indeed, since