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● ● ● MYELOID NEOPLASIA

Comment on Malcovati et al, page 6239

It dices, it splices!

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A novel class of molecular lesions that affect components of the splicing machinery is the focus of the work by Malcovati et al in this issue of *Blood*.¹ Their data confirm the strong correlation between splicing factor SF3B1 mutations and the presence of ringed sideroblasts in patients with myelodysplastic syndromes (MDS). In addition, they show an improved survival for patients with SF3B1 mutations independent of the International Prognostic Scoring System (IPSS).

The usual introduction for an MDS article includes a description of MDS as a group of heterogeneous clonal marrow disorders, characterized by cytopenias, dysplasia, and a variable propensity for progression to AML. While this statement remains true today, refinements to the MDS IPSS² and the World Health Organization (WHO) classification criteria are expected as molecularly defined MDS subtypes emerge to replace the current “heterogeneous” characterization. Progress toward this goal includes the large-scale effort reported by Bejar et al³ demonstrating that point mutations in *TP53*, *EZH2*, *ETV6*, *RUNX1*, and *ASXL1* were significantly associated with poor survival in MDS independent of IPSS risk category.

In recent months data from different groups have been published that describe novel mutations in elements of the RNA splicing machinery in MDS. These spliceosome components are required for normal constitutive and alternative splicing, which can be compromised by *cis*- or *trans*-acting alterations to cause or modulate human disease and cancer.⁴ An important contribution to this body of literature came from Yoshida et al, who performed whole exome sequencing of DNA from 29 MDS patient samples and found recurrent novel mutations in 6 components of the RNA splicing machinery.⁵ In their subsequent analysis of samples from 582 patients

with myeloid malignancies, mutually exclusive mutations of the splicing machinery were commonly found in MDS with (84.9%) or without (43.9%) ringed sideroblasts, in CMML (54.5%), and in therapy-related MDS or AML with myelodysplasia (25.8%). Mutations in *SF3B1*, a core component of the RNA splicing machinery located at the U2-snrNP spliceosome catalytic site, were most common in RARS (82.6%) and RCMD-RS (76%) WHO subtypes.

Visconte et al also discovered recurrent splicing factor *SF3B1* mutations in MDS patient samples using high-throughput next-generation sequencing.⁶ In their sample set, 9 of 14 (64%) RARS and 13 of 18 (72%) RARS-T patients had mutations in *SF3B1*. This group also noted a correlation between RARS patients with *SF3B1* mutations, a normal or elevated platelet count, and a higher incidence of thrombotic events.

Using massively parallel sequencing technology, Papaemmanuil et al identified somatic *SF3B1* mutations in 6/9 MDS patient samples.⁷ *SF3B1* heterozygous substitution mutations were then found in 20% (n = 72) of 354 MDS patient samples, including 53 of 82 patients (65%) with the RARS and RCMD-RS subtype, compared with 5% to 10% in other MDS WHO subtypes. In their analysis of 123 MDS patients for whom clinical data were available, *SF3B1* mutations

were reported in 34 patients, and were associated with the ringed sideroblast phenotype, higher white blood cell and platelet counts, lower bone marrow blast counts, and longer overall survival (OS) and leukemia-free survival (LFS).

In this issue of *Blood*, Malcovati and colleagues confirm their previous report of the association between *SF3B1* mutations and WHO ringed sideroblast subtypes⁷ in 533 MDS patient samples (including 354 patients from their prior study), showing a 72% frequency of *SF3B1* mutations in the RARS and RCMD-RS subtypes.¹ They then carefully enumerate ringed sideroblasts in marrow specimens from an annotated 325-patient MDS cohort. The concordance between those with *SF3B1* mutations (n = 101) and any percentage of ringed sideroblasts was 97.7%; conversely, 1 in 4 patients with a ringed sideroblast WHO subtype had wild-type *SF3B1*. Survival end points appeared improved for patients with *SF3B1* mutations compared with nonmutated patients in multivariate analyses that included all IPSS elements and ringed sideroblasts, and when IPSS low and intermediate-1 risk patients were stratified by mutation status to compare Kaplan-Meier estimates of event-free survival (EFS) and OS. However, because of the close association between *SF3B1* mutations and the ringed sideroblast phenotype, LFS (in univariate analysis) and OS (in multivariate analysis) were not improved by the presence of an *SF3B1* mutation when RARS and RCMD-RS subtypes were analyzed alone.

The current findings by Malcovati et al and other recent reports discussed above are remarkable for several reasons. First, the *SF3B1* mutation is the first point mutation linked to a specific morphologic MDS subtype. Second, the proportion of MDS patients affected by spliceosome mutations appears to exceed frequencies for most other known point mutations in MDS. Third, the discovery that the spliceosome mutations are mutually exclusive⁵ suggests the importance of such mutations in MDS pathogenesis, and therefore the potential for spliceosome-directed therapeutic interventions.

Whether *SF3B1* mutations will alter current prognostic models and the WHO classification system is unknown. The correlation of *SF3B1* mutation with ringed sideroblasts suggests the arbitrariness of a 15% cutoff to define

a ringed sideroblast subtype, and also begs the question of whether the prognostic value of an *SF3B1* mutation is equivalent to a careful and consistently performed (and less expensive) ringed sideroblast stain, as suggested with the apparent loss of a protective effect when *SF3B1* mutation status is assessed in RARS/RCMD-RS subtypes alone. This question will be answered in future efforts to validate the independent prognostic value of *SF3B1* in other large sample sets.

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

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