LETTER TO THE EDITOR

Rare mutations in DNMT3A in myeloproliferative neoplasms and myelodysplastic syndromes

Blood Cancer Journal (2011), e18; doi:10.1038/bcj.2011.15; published online 13 May 2011;

Alterations of epigenetic marks are thought to play an important role in myeloid malignancies. In particular, aberrant DNA methylation is a hallmark of these diseases. DNMT3A and DNMT3B methyltransferases have predominant role in de novo methylation of DNA. Mutations in DNMT3A have been found in roughly 20% of acute myeloid leukemia (AML),1–3 The precise mechanism by which DNMT3A may affect DNA methylation is not known. The TET2 gene encodes an enzyme that favors the transformation of 5-methylcytosines in 5-hydroxymethylcytosines. TET2 function requires alpha-ketoglutarate (αKG). TET2 is frequently mutated in myeloid diseases. Mutation in IDH1 and IDH2 changes their enzymatic activity and induces an hypermethylation of AML DNA.4 Mutated IDH1/2 enzymes catalyze αKG into 2-hydroxyglutarate (2HG). Production of 2HG impairs TET2 function. This explains why mutations in TET2 and in IDH1/2 are mutually exclusive.4 In contrast, mutations in IDH1/2 are more frequent in AML cases with DNMT3A mutations.2

We searched for mutations and deletions of DNMT3A, TET2, and IDH1/2 in a series of 201 chronic myeloid diseases including 135 myeloproliferative neoplasms (MPNs) and 66 myelodysplastic syndromes (MDS). The MPN cases comprised 33 polycythemia vera (PV) and 5 post-PV myelofibrosis (MF), 56 essential thrombocythemia (ET) and 10 post-ET MF, 25 primary myelofibrosis (PMF), 3 MPN- unclassifiable and 3 MDS/MPN cases. The MDS comprised 5 refractory anemia (RA), 13 RA with ring sideroblasts (RARS), 7 refractory cytopenia with multilineage dysplasia, 16 RA with excess blasts (RAEB) type 1, 20 RAEB type 2 and 5 MDS-unclassifiable cases.

We determined the sequence of all exons of DNMT3A, TET2, and IDH1/2 of which had two mutations), 0 mutations in IDH2. TET2 functions in 12 patients (2 PV, 1 in a JAK2 V617F-positive PV, 1 in a JAK2 V617F-negative PMF) (see Table 1). The two mutations in DNMT3A were missense (c.2245C>T, p.Arg749Cys in the PV; c.2644G>A, p.Arg882Ser in the PMF). All mutations were heterozygous.

In MDSs, we found 12 mutations and 1 deletion of TET2 (all heterozygous), 5 mutations of IDH1/2, and 4 mutations (6%) and 1 deletion of DNMT3A (all heterozygous) (see Table 1). Mutations in DNMT3A were 1 nonsense (c.1681G>T, p.Glu561Stop), 1 frameshift (c.1872del, p.Pro625Leu632X26) and 2 missense (c.1723G>C, p.Ala575Pro; c.2141C>G, p.Ser714Cys). Mutations in TET2, IDH1/2 and DNMT3A were all mutually exclusive.

Thus, 23 MDS cases out of 66 (roughly one-third) showed one alteration (mutation or deletion) in either DNA methylation-associated gene. Strikingly, the 4 DNMT3A-mutated cases were 1 RA and 3 RARS. One RARS case had a trisomy 8.

DNMT3A mutations were very recently reported in two series of MDSs, including 62 RAEB cases5 and 150 cases of various subclasses. In the RAEB series,6 3 cases (4.8%) were mutated. In the second series,7 12 patients had DNMT3A mutations (8%). These results show that, in chronic myeloid diseases, TET2 mutations are prominent, whereas IDH1/2 and DNMT3A are less frequent. In MPNs, we did not find any IDH1 mutation; previous works had found that only 4% of PMF cases and few PV and ET were mutated in IDH1/2.8,9 IDH1/2 mutations are also rare in MDSs, except in some subclasses such as MDSs with del(5q) or trisomy 8.5,10 Only six cases were mutated in DNMT3A in our whole series of chronic cases. Overall, IDH1/2 and DNMT3A mutations are therefore more a feature of AMLs, especially primary AMLs with normal karyotype and intermediate prognosis.2,3 This suggests that mutations in TET2, IDH1/2 and DNMT3A, although potentially all functionally linked to DNA methylation, may not be equivalent events in the initiation of leukemogenesis; TET2 mutation could be more efficient in triggering the process. In our series, mutations of the three genes were mutually exclusive, whereas DNMT3A mutations have been found to be associated with TET2 or IDH1/2 mutations in AMLs.2 This may just be because of a low number of mutated samples in chronic cases. However, this may also suggest that IDH1/2 and DNMT3A mutations may participate, although less frequently than TET2, to the initial phases of the disease. This may be in collaboration with specific cooperating alterations such as trisomy 8 or del(5q).

All our DNMT3A-mutated MDSs were low-risk RA/RARS cases. The DNMT3A Arg882 amino-acid residue, which is a mutation hotspot in AMLs,1–3 was only mutated once in our series of MPNs (in a PMF) and it was not mutated in our series of MDSs. In the reported RAEB series,6 the three mutations affected the Arg882 residue. In the other published series,7 three out of the four Arg882-mutated MDSs were RAEB/RAEB-T cases. The DNMT3A mutations can occur in the various subclasses of MDS. However, the Arg882 mutation may be more specific of RAEB and/or aggressive cases, whereas mutations at the other residues may have a different function and may be associated with a different (milder?) phenotype.

Table 1 Mutations in three DNA methylation-associated genes in patients with chronic myeloid diseases

| Gene    | TET2 | IDH1/2 | DNMT3A | Total |
|---------|------|--------|--------|-------|
| MPNs (N=135) | 12 (8.9) | 0 | 2 (1.5) | 14 (10.4) |
| MDSs (N=66)   | 12 (18.2) | 6 (7.6) | 4 (6) | 21 (31.8) |
| Total (N=201) | 24 (11.9) | 6 (2.5) | 6 (3) | 31 (17.4) |

Abbreviations: IDH1, isocitrate dehydrogenase 1; MDS, myelodysplastic syndrome; MPN, myeloproliferative neoplasm.

Conflict of interest
The authors declare no conflict of interest.
Acknowledgements

This work was supported by Inserm, Institut Paoli-Calmettes and grants from Association pour la Recherche contre le Cancer (AM, no. 4929, 2008) and Association Laurette Fugain (MJM 2010).

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