RESEARCH LETTER

blood advances

TO THE EDITOR:

Clinical and prognostic impact of *STAG2* mutations in myeloid neoplasms: the Mayo Clinic experience

Bahga Katamesh,[1] Ahmad Nanaa,[1,2] Rong He,[3] David Viswanatha,[3] Phuong Nguyen,[3] Patricia Greipp,[3] Kurt Bessonen,[4] Naseema Gangat,[1] Kebede Begna,[1] Abhishek Mangaonkar,[1] Mrinal Patnaik,[1] William J. Hogan,[1] Ayalew Tefferi,[1] Mark Litzow,[1] Mithun Vinod Shah,[1] Cecilia Arana Yi,[5] James Foran,[6] Talha Badar,[6] Hassan B. Alkhateeb,[1] and Aref Al-Kali[1]

[1]Division of Hematology, Mayo Clinic, Rochester, MN; [2]John H. Stroger, Jr. Hospital of Cook County, Chicago, IL; [3]Division of Hematopathology, and [4]Division of Molecular Hematology, Mayo Clinic, Rochester, MN; [5]Division of Hematology, Mayo Clinic, Scottsdale, AZ; and [6]Division of Hematology, Mayo Clinic, Jacksonville, FL

The stromal antigen 2 (*STAG2*) gene, located on chromosome Xq25, is a core component of the cohesin complex that functions on chromatin organization, transcriptional regulation, and postreplicative DNA repair.[1-3] *STAG2* mutations (*STAG2*ms) are reported in 5% to 10% of myeloid neoplasms (MNs), mostly high-risk myelodysplastic syndrome (MDS) and acute myeloid leukemia (AML).[4,5] Although few data are available on the frequency of *STAG2*ms alone, collectively, cohesin complex mutations are present in 8% of MDS, 12% of AML, and 10% of chronic myelomonocytic leukemia cases. It is considered 1 of 8 secondary-type mutations and are specific to secondary AML (sAML).[6] *STAG2*ms correlate with MDS response to hypomethylating agents (HMAs)[3,5] but are linked to poor prognosis in MDS,[3,7,8] with conflicting data on AML outcome[8-11] and limited data on effects of hematopoietic stem cell transplantation (HSCT).

Our single-institute retrospective study included 91 patients with *STAG2*m MN, whose charts were reviewed for clinical information after institutional review board approval. Patients were included at date of in-house *STAG2*-harboring next-generation sequencing (NGS) (performed at diagnosis or progression). Diagnosis was rendered according to World Health Organization classification[12,13] and MDS risk stratification according to the Revised International Prognostic Scoring System.[14] BlueSky Software V7.40 was used for statistical analysis.

Most patients were older (median age, 72 years) males (78%). MDS was the most common diagnosis (55%), followed by AML (29%), MDS/myeloproliferative neoplasm (MPN) overlap, and MPN. Of the MDS cases, 36 (72%) belonged to the excess blast subtype (MDS-EB) (including 17 MDS-EB1 and 19 MDS-EB2) and 23 (46%) were high or very high risk. None were of the ring sideroblast subtype (including the 8 patients carrying *SF3B1*m). Ten patients with AML (38%) had sAML, whereas 14 patients (15%) had therapy-related MN (t-MN) (including 10 and 8 who received prior chemotherapy and radiotherapy, respectively). By the European LeukemiaNet risk stratification, 18 of the AML cases were adverse risk, 7 intermediate, and 1 favorable risk. Twenty-eight patients (31%) had abnormal cytogenetics, with trisomy 8 being the most common (Table 1; supplemental Tables 2-4). None of the females had X-chromosome deletion.

The median VAF of *STAG2*ms was 50% (range, 5% to 100%) and was significantly higher in males ($P < .001$), as expected of an X-linked gene. Corrected median VAF (accounting for X chromosome) was 29.5% and 27% in males and females ($P = .5$), respectively. There was no difference in VAF between diagnostic groups ($P = .6$), MDS subtypes ($P = .7$), high and non–high-risk MDS ($P = .6$), de novo AML (dnAML) ($P = .2$), and sAML or between t-MN and de novo MN (dnMN) ($P = .1$). There was no correlation between VAF and bone marrow (BM) or peripheral blasts. Most *STAG2*ms were in the N-terminus (64%) and 15 (16%) were in the STAG domain. Within the STAG domain, there were

Submitted 5 May 2022; accepted 23 November 2022; prepublished online on *Blood Advances* First Edition 7 December 2022. https://doi.org/10.1182/bloodadvances.2022007937.

Data are available on request from the corresponding author, Aref Al-Kali (alkali.aref@mayo.edu).

The full-text version of this article contains a data supplement.

Table 1. *STAG2*-mutated patients' characteristics

Variable	Value
Characteristics and hematologic features of patients with *STAG2*m	
Total no. of patients	91
Race, n (%)	
White	85 (93)
Latino/Hispanic	4 (4)
African American	1 (1)
Other	1 (1)
Sex (male), n (%)	71 (78)
Age (range), y	72 (25-91)
Hemoglobin, median (Q1, Q3), g/dL	8.7 (7.9, 9.8)
Leukocytes, median (Q1, Q3), $\times 10^9$/L	2.8 (1.8, 6.0)
Platelets, median (Q1, Q3), $\times 10^9$/L	88 (36, 129)
BM blasts (range), %	7 (0-86)
Circulating blasts (range), %	3 (0-71)
Cytogenetics, n (%)	
Abnormal	28 (31)
Normal	59 (65)
Diagnosis, n (%)	
MDS	50 (55)
AML	26 (29)
MDS/MPN	9 (10)
MPN	3 (3)
CCUS	2 (2)
Aplastic anemia	1 (1)
t-MN	14 (15)
STAG2m characteristics	
VAF of *STAG2* (range), %	50 (5-100)
Median VAF, %	
Males	59
Females	27
AML	51
MDS	50
MDS/MPN	41
MPN	31
Mutation type, n (%)	
Nonsense	49 (54)
Frameshift	33 (36)
Splice site	9 (10)
Comutational pattern	
No. of comutations, median (range)	3 (0-6)
Isolated *STAG2*, n (%)	6 (7)
Major comutations, n (%)	
ASXL1	59 (65)
SRSF2	33 (36)
TET2	33 (36)
RUNX1	27 (30)
BCOR	18 (20)
IDH2	16 (18)
U2AF1	12 (13)
Treatment, n (%)	
Received at least 1 line of treatment	64 (70.3)
HMA	30 (47)
HMA + VEN	17 (27)
Low-dose cytarabine + VEN	3 (5)
Chemotherapy	6 (9)
TK inhibitor	4 (6)
IDH inhibitor	1 (2)
Immunomodulator	1 (2)
Study therapy	2 (3)
HSCT, n (%)	25 (28)

Table 1 (continued)

	Response to therapy		
	HMA, n (%)	HMA + VEN, n (%)	Chemotherapy, n (%)*
CR	1 (3.8)	2 (11.8)	3 (50.0)
CRi	6 (23.1)	7 (41.2)	2 (33.3)
PR	0 (0)	1 (5.9)	0 (0)
HI	5 (19.2)	0 (0)	0 (0)
No response	14 (53.8)	7 (41.2)	1 (16.7)
Relapse			
Yes	3 (25)	6 (60)	2 (40)
No	9 (75)	4 (40)	3 (60)

CCUS, clonal cytopenia of unknown significance; CR, complete remission; CRi, complete remission with incomplete hematologic recovery; HI, hematologic improvement; IDH, isocitrate dehydrogenase; PR, partial response; TK inhibitor, tyrosine kinase inhibitor; VEN, venetoclax.

*Chemotherapy regimens used were 7 + 3 with or without midostaurin.

6 (p.Arg259) and 4 (p.Arg216) nonsense mutations, representing mutational hotspots (Figure 1). Nonsense mutations were the most common (54%), followed by frameshift (36%) and splice site (10%) (supplemental Table 5). There was no correlation between mutation type or site and MN classification, BM blasts, or VAF.

The median number of comutations was 3 (range, 0-6) (supplemental Table 6). The most common were *ASXL1* (65%), *TET2* (36%), *SRSF2* (36%), *RUNX1* (30%), and *BCOR* (20%), whereas *TP53* was uncommon (1%) (supplemental Figure 1; supplemental Tables 7 and 8). There was no correlation between the number of comutations and MN classification. Higher risk MDS had more *BCOR* mutations ($P = .01$) and *RUNX1* mutations ($P = .2$) than lower risk MDS. There was no difference in the comutational pattern between dnAML and sAML or between dnMN and t-MN. t-MN cases carried fewer comutations than dnMN (2 vs 3, $P = .02$), whereas sAML cases and those that progressed to AML carried 4. No single comutation was associated with progression to AML and none of those that progressed carried *KRAS* mutations/*NRAS* mutations. Six cases (7%) had isolated *STAG2*ms, including 3 MDS, 2 AML, and 1 CCUS. There was no difference in

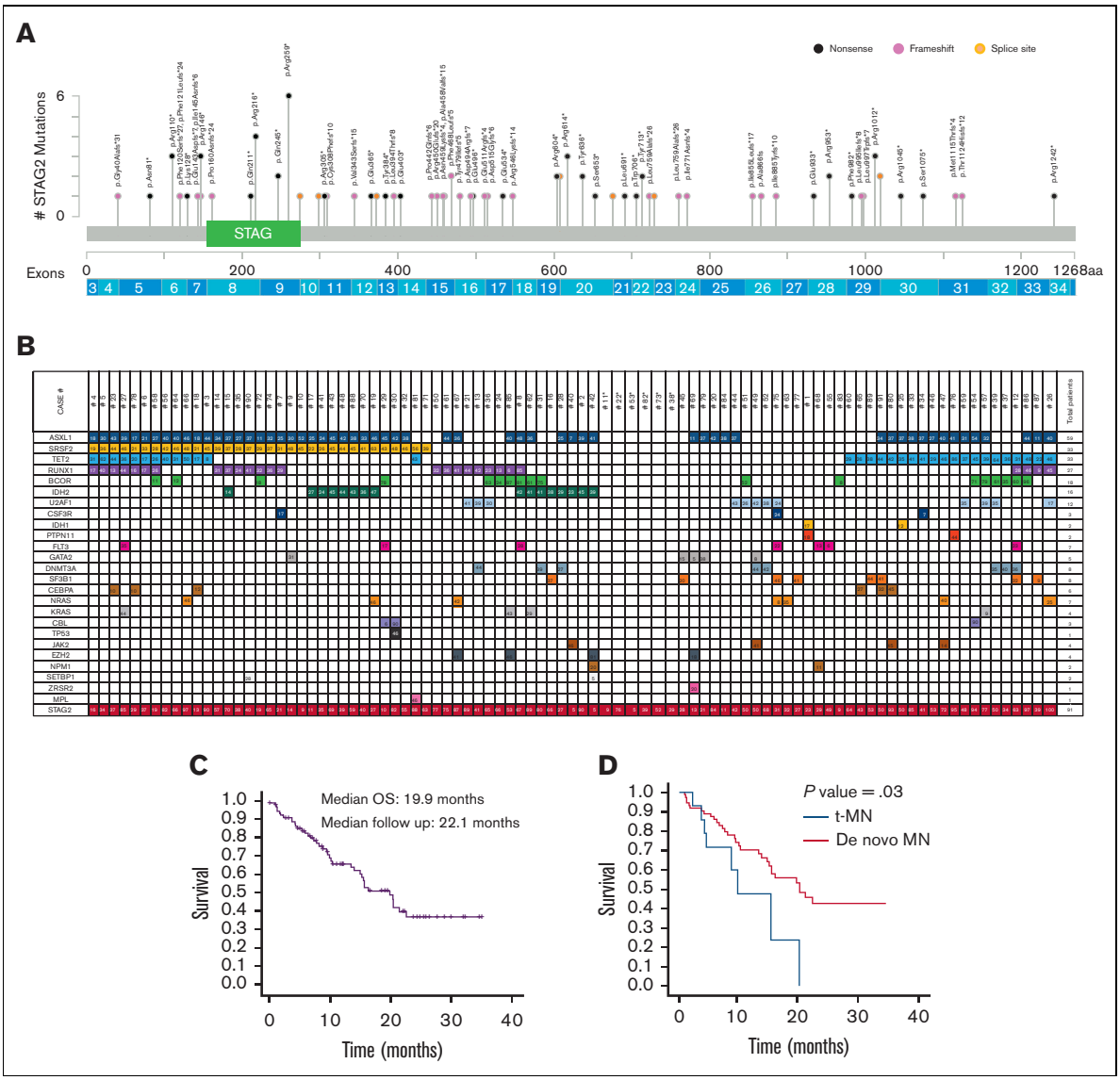

Figure 1. *STAG2*m characteristics in patients with MN. (A) Representation of *STAG2* variants detected, positioned on the *STAG2* protein and its functional domains. The protein alterations are indicated on top of the corresponding mutation location, exon locations are indicated below the protein. (B) Comutational pattern in 91 patients with *STAG2*m MN. Each patient is represented by a column. The number reported in the box represents the VAF of each mutation. (C) OS for 88 patients with *STAG2*m MN. (D) OS for tMN vs dnMN in patients with *STAG2*m. STAG, STAG domain; aa, amino acid.

VAF ($P = .2$), BM blasts ($P = .5$), or cytogenetics in them ($P = .3$) compared with those in the comutated cases.

Sixty-four patients (70%) received treatment, including 33 patients with MDS, 21 with AML, 6 with MDS/MPN, and 3 with MPN. Among these, 47 (73%) received HMA either alone or in combination with venetoclax (VEN), and 6 (10%) received intensive chemotherapy (IC). Those who received HMA and those who received HMA + VEN had response rates of 46% and 59%, respectively ($P = .4$) (Table 1). There was no difference in the response rate between those receiving HMA and those receiving IC (83%) ($P = .2$) or between those receiving HMA + VEN and those receiving IC ($P = .3$). Median overall survival (mOS) for HMA responders was not reached, and for HMA nonresponders, it was 20.4 months ($P = .06$). Twenty-five patients (28%) received HSCT including 11 with AML, 12 with MDS, and 2 with MDS/MPN. Before NGS, 3 patients had received HSCT. The median time from NGS to HSCT was 4.9 months.

Fifty-four percent and 19% of AML and MDS cases, respectively, relapsed after remission. There was no difference between those who received HMA and those who received IC ($P = .6$); and cytogenetics, VAF, and BM/peripheral blasts did not affect relapse. Nine MDS (18%) and 2 MDS/MPN (22%) cases progressed to AML. By competing risk analysis, time to progression in patients with MDS was 10.4 months. Median event-free survival for patients with MDS was 16.3 months. Higher risk MDS and those with higher BM blasts were more likely to progress ($P = .04$ and $P = .008$, respectively), whereas no other factors predicted progression.

mOS among 88 patients (after excluding CCUS and aplastic anemia) was 19.9 months (number of deaths = 39). Median 2-year survival in patients with AML was 40%. There was no difference in survival based on sex ($P = .2$) or cytogenetics ($P = .08$). There was no difference between diagnostic groups ($P = .3$), sAML and dnAML ($P = .2$), or higher and lower risk MDS ($P = .6$). Patients with t-MN had lower mOS than dnMN (9.9 vs 20.5 months, $P = .03$). Those with circulating blasts ≥5% had lower mOS than those with <5% (10.1 vs 21.4 months, $P = .01$). On univariate Cox regression analysis with VAF as a continuous variable, mOS worsened with increasing VAF (hazard ratio [HR], 1.01; $P = .03$). t-MN and circulating blasts ≥5% remained significant on multivariate analysis (supplemental Table 9), whereas higher VAF worsened OS (HR, 1.01; $P = .09$). A higher VAF worsened OS in males alone with HR, 1.01 and $P = .1$. Analysis of OS against corrected VAF among all patients yielded HR, 1.02 and $P = .1$. Patients who received HSCT after NGS had longer mOS than those who without HSCT (HR, 0.4; $P = .06$; by time-dependent variable analysis). Mutations of *KRAS*, *PTPN11*, and *CBL* led to lower mOS ($P < .001$, $P < .001$, and $P = .04$, respectively), whereas *IDH2* mutations improved mOS ($P = .04$). Neither *ASXL1* mutations nor comutational burden affected mOS (Figure 1; supplemental Figures 2-11).

Of 91 patients, 47 (52%) were diagnosed with MN before NGS (supplemental Figure 12), with a median time of 8.4 months from initial diagnosis to in-house NGS. Among 37 non-AML cases, 7 (19%) had progressed to AML at time of NGS. mOS did not differ between those with and without a prior diagnosis ($P = .9$). Thirty-one of 91 patients had a subsequent NGS (S-NGS). Sixteen patients (52%) continued to harbor *STAG2*m in S-NGS and had poor mOS compared with those who did not (19.9 months vs not reached, $P = .03$) (supplemental Figure 13). Where the *STAG2*m was lost on S-NGS, the median number of comutations became 2. Of 11 patients with AML who had a S-NGS, 7 had lost *STAG2*m. Four of these 7 patients had shown response to therapy, whereas the remaining 3 had not.

Our study included 91 patients, which is the largest cohort of patients with *STAG2*m MN in current literature. We found a high cooccurrence of *STAG2*ms and *ASXL1* mutations, as previously alluded to by Kon et al.[15,16] We demonstrated the prevalence of higher risk disease among patients with *STAG2*m MDS, as previously suggested.[4,5] Eighteen percent of MDS cases progressed to sAML, with an increased comutational burden on progression, as previous studies suggest.[17-19] It is possible that *STAG2*ms, therefore, favor transformation through induction of genetic instability and acquisition of new mutations. In our cohort, 31% patients had abnormal karyotype. Thota et al[5] had found that the prevalence of abnormal karyotype was similar among cohesin-mutated and wild-type MN. Our study found that karyotype did not impact survival or disease progression, which previous studies had not alluded to.

Our study also showed *STAG2*ms to be associated with poor prognosis in MN. There was possible worsening of OS with higher VAF, which may be highlighted with a larger cohort. This, combined with the lack of association between MN classification and OS, suggests an adverse impact of *STAG2*ms on survival regardless of phenotypic features. There was also no correlation of VAF with clinical features (age, MN phenotype, and blast counts); hence, there was no clear explanation for an impact of VAF. Because none of the females had X-chromosome deletion, the impact of the loss-of-heterogeneity could not be assessed. *STAG2*m sAML had comparable clinical characteristics and survival to dnAML, unlike what others have suggested.[20] This supports the notion that *STAG2*ms are secondary type and define a subset of dnAML cases with worse clinical outcome, comparable to that of clinically defined sAML.[6] Our data also suggest a positive response to HSCT among patients with *STAG2*m MN, albeit a larger sample size is needed to confirm this.

Our study is limited by the relatively small sample size, retrospective nature, patient heterogeneity, and the lack of long-term follow-up. To our knowledge, we present novel findings on the role of *STAG2*ms in MN progression. A larger cohort can provide further insight.

Contribution: B.K., A.N., and A.A.-K. planned the study, reviewed data, completed statistical analysis, and wrote the manuscript; R.H. and D.V. performed molecular analysis and reviewed the manuscript; P.N. reviewed BM slides and manuscript; P.G. performed cytogenetic analysis and reviewed the analysis; K. Bessonen coordinated NGS data collection; and N.G., K. Begna, A.T., A.M., M.P., W.J.H., M.L., M.V.S., C.A.Y., J.F., T.B., and H.B.A. reviewed the manuscript and contributed patients.

Conflict-of-interest disclosure: The authors declare no competing financial interests.

ORCID profiles: B.K., 0000-0002-3651-1764; A.N., 0000-0001-5247-5065; R.H., 0000-0001-6116-8163; P.G., 0000-0002-5536-9011; N.G., 0000-0002-9104-6172; K.B., 0000-0003-2730-8593; M.P., 0000-0001-6998-662X; M.L., 0000-0002-9816-6302; M.V.S., 0000-0002-5359-336X; C.A.Y., 0000-0001-5420-3514; T.B., 0000-0003-1548-918X; A.A.-K., 0000-0002-0824-3715.

Correspondence: Aref Al-Kali, Division of Hematology, Mayo Clinic, 200 First St SW, Rochester, MN 55905; email: alkali.aref@mayo.edu.

References

1. Hill VK, Kim JS, Waldman T. Cohesin mutations in human cancer. *Biochim Biophys Acta*. 2016;1866(1):1-11.
2. Jann JC, Tothova Z. Cohesin mutations in myeloid malignancies. *Blood*. 2021;138(8):649-661.
3. Viny AD, Levine RL. Cohesin mutations in myeloid malignancies made simple. *Curr Opin Hematol*. 2018;25(2):61-66.
4. Makishima H, Yoshizato T, Yoshida K, et al. Dynamics of clonal evolution in myelodysplastic syndromes. *Nat Genet*. 2017;49(2):204-212.
5. Thota S, Viny AD, Makishima H, et al. Genetic alterations of the cohesin complex genes in myeloid malignancies. *Blood*. 2014; 124(11):1790-1798.
6. Lindsley RC, Mar BG, Mazzola E, et al. Acute myeloid leukemia ontogeny is defined by distinct somatic mutations. *Blood*. 2015; 125(9):1367-1376.
7. Ganguly BB, Kadam NN. Mutations of myelodysplastic syndromes (MDS): an update. *Mutat Res Rev Mutat Res*. 2016;769:47-62.

8. Xu F, Wu L-Y, He Q, et al. Exploration of the role of gene mutations in myelodysplastic syndromes through a sequencing design involving a small number of target genes. *Sci Rep*. 2017;7:43113.
9. Cuartero S, Innes AJ, Merkenschlager M. Towards a better understanding of cohesin mutations in AML. *Front Oncol*. 2019; 9:867.
10. Thol F, Bollin R, Gehlhaar M, et al. Mutations in the cohesin complex in acute myeloid leukemia: clinical and prognostic implications. *Blood*. 2014;123(6):914-920.
11. Zhang L, Padron E, Lancet J. The molecular basis and clinical significance of genetic mutations identified in myelodysplastic syndromes. *Leuk Res*. 2015;39(1):6-17.
12. Arber DA, Orazi A, Hasserjian R, et al. The 2016 revision to the World Health Organization classification of myeloid neoplasms and acute leukemia. *Blood*. 2016;127(20):2391-2405.
13. Vardiman JW, Harris NL, Brunning RD. The World Health Organization (WHO) classification of the myeloid neoplasms. *Blood*. 2002;100(7): 2292-2302.
14. Greenberg PL, Tuechler H, Schanz J, et al. Revised international prognostic scoring system for myelodysplastic syndromes. *Blood*. 2012;120(12):2454-2465.
15. Kon A, Shih L-Y, Minamino M, et al. Recurrent mutations in multiple components of the cohesin complex in myeloid neoplasms. *Nat Genet*. 2013;45(10):1232-1237.
16. Li Z, Zhang P, Yan A, et al. ASXL1 interacts with the cohesin complex to maintain chromatid separation and gene expression for normal hematopoiesis. *Sci Adv*. 2017;3(1):e1601602.
17. Martín-Izquierdo M, Abaigar M, Hernandez-Sanchez JM, et al. Co-occurrence of cohesin complex and Ras signaling mutations during progression from myelodysplastic syndromes to secondary acute myeloid leukemia. *Haematologica*. 2021;106(8): 2215-2223.
18. Tsai CH, Hou H-A, Tang J-L, et al. Prognostic impacts and dynamic changes of cohesin complex gene mutations in de novo acute myeloid leukemia. *Blood Cancer J*. 2017;7(12):663.
19. Walter MJ, Shen D, Ding L, et al. Clonal architecture of secondary acute myeloid leukemia. *N Engl J Med*. 2012;366(12): 1090-1098.
20. Granfeldt Østgård LS, Medeiros B, Sengelov H, et al. Epidemiology and clinical significance of secondary and therapy-related acute myeloid leukemia: a national population-based cohort study. *J Clin Oncol*. 2015;33(31):3641-3649.