CSF3R mutations and variants in myeloid neoplasms: associated phenotypes, co-mutations, and survival trends

Granulocyte colony-stimulating factor (GCSF) regulates cell proliferation, differentiation and survival through binding to the extracellular domain of the colony-stimulating factor 3 receptor (CSF3R).[1-3] Ligand binding induces formation of CSF3R homodimers, bringing the cytoplasmic domains into proximity.[1] This leads to the activation of signal transduction pathways such as JAK/STAT, MAPK/ERK and PI3K/AKT.[2,4]

Studies have associated *CSF3R* mutations/variants with myeloid neoplasms.[4] The *CSF3R*-T618I mutation has been widely reported in chronic neutrophilic leukemia (CNL).[5,6] This, along with other *CSF3R* mutations/variants, has also been sparsely reported in non-CNL myeloid neoplasms such as acute myeloid leukemia (AML), atypical chronic myeloid leukemia and chronic myelomonocytic leukemia (CMML).[7-9] The current study aims to elucidate the spectrum of *CSF3R* mutations/variants and associated disease phenotypes and genotypes in patients with myeloid neoplasms, and their impact on survival. The study was approved by the Mayo Clinic Institutional Review Board (IRB 12-003574).

We reviewed the Mayo Clinic database to extract records of patients with myeloid neoplasms who had *CSF3R* mutations/variants on next-generation sequencing performed between 2016 to 2024. Mutations/variants were classified as either pathogenic (*CSF3R*^{PATH}) or variants of unknown

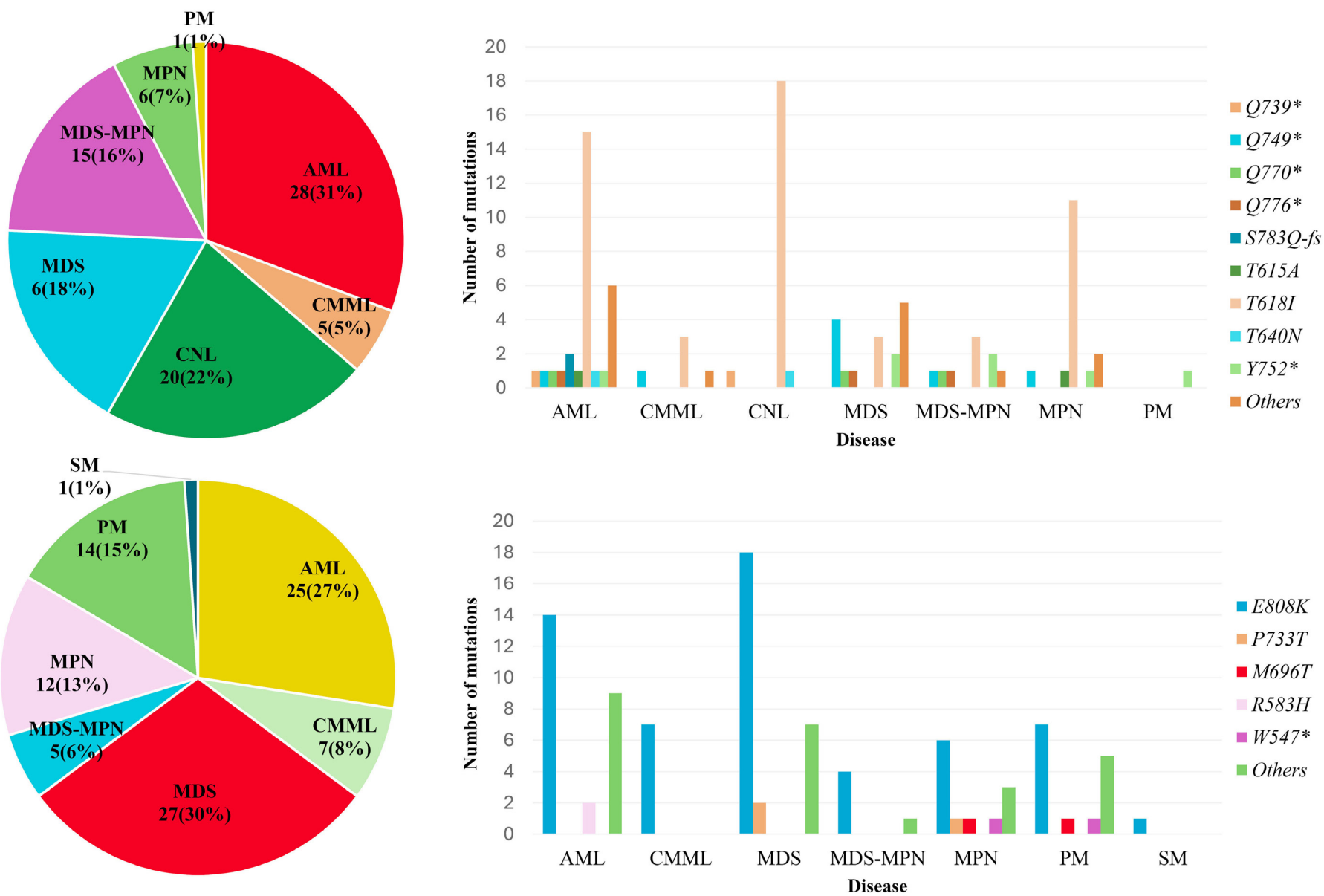


Figure 1. Distribution of myeloid neoplasms and associated mutations/variants. Distribution of myeloid neoplasms in the subcohorts of patients with *CSF3R* pathogenic mutations (*CSF3R*^{PATH}) (top) and variants of unknown significance (*CSF3R*^{VUS}) (bottom). The pie charts (left) show the distribution of the myeloid neoplasms, while the bar charts (right) show the different mutations/variants. AML: acute myeloid leukemia; CMML: chronic myelomonocytic leukemia; CNL: chronic neutrophilic leukemia; MDS: myelodysplastic syndrome; MDS-MPN: myelodysplastic/myeloproliferative neoplasm; MPN: myeloproliferative neoplasms (myelofibrosis, polycythemia vera, chronic myeloid leukemia, and essential thrombocythemia); PM: premalignant conditions; SM: systemic mastocytosis.

significance (*CSF3R*^{VUS}) based on their characterization in the literature at the time of testing. Descriptive and analytical statistical methods were employed, including χ^2 tests, Kaplan-Meier estimates for overall survival (OS) and Cox proportional hazards regression.

We identified 182 patients, including those with myeloid neoplasms (N=167, 91.8%) and premalignant conditions (N=15, 8.2%). The two premalignant conditions were clonal hematopoiesis of indeterminate potential (N=6, 3.3%) and clonal cytopenia of undetermined significance (N=9, 4.9%). The median age at *CSF3R* mutation detection for the entire cohort was 70 years (range, 17-93). Patients with premalignant conditions were younger, with a median age of 66 years (range, 30-92) compared to 71 years (range, 17-93) for patients with myeloid neoplasms.

A total of 52 distinct *CSF3R* mutations/variants were identified in the 182 patients (91 in each *CSF3R*^{PATH} and *CSF3R*^{VUS} subcohort) (*Online Supplementary Figure S1*). *CSF3R*^{PATH} (N=21, 40.4%) included missense (N=5, 9.6%), nonsense (N=12, 23%) and frameshift mutations (N=4, 7.7%). *CSF3R*^{VUS} (N=31, 59.6%) included missense (N=27, 51.9%), nonsense (N=1, 1.9%), frameshift (N=1, 1.9%), synonymous (N=1, 1.9%) and splice site (N=1, 1.9%) variants. There were 12 patients with two *CSF3R* mutations/variants: seven had two *CSF3R*^{PATH}, three had one *CSF3R*^{PATH} and one *CSF3R*^{VUS}, and two had two *CSF3R*^{VUS}.

CSF3R^{PATH} was more prevalent in the cytoplasmic domain (N=17, 81%) than *CSF3R*^{VUS} (N=15, 48%; *P*=0.02). The overall median variant allele frequency (VAF) was 47% (interquartile range [IQR], 25-49). *CSF3R*^{PATH} had significantly

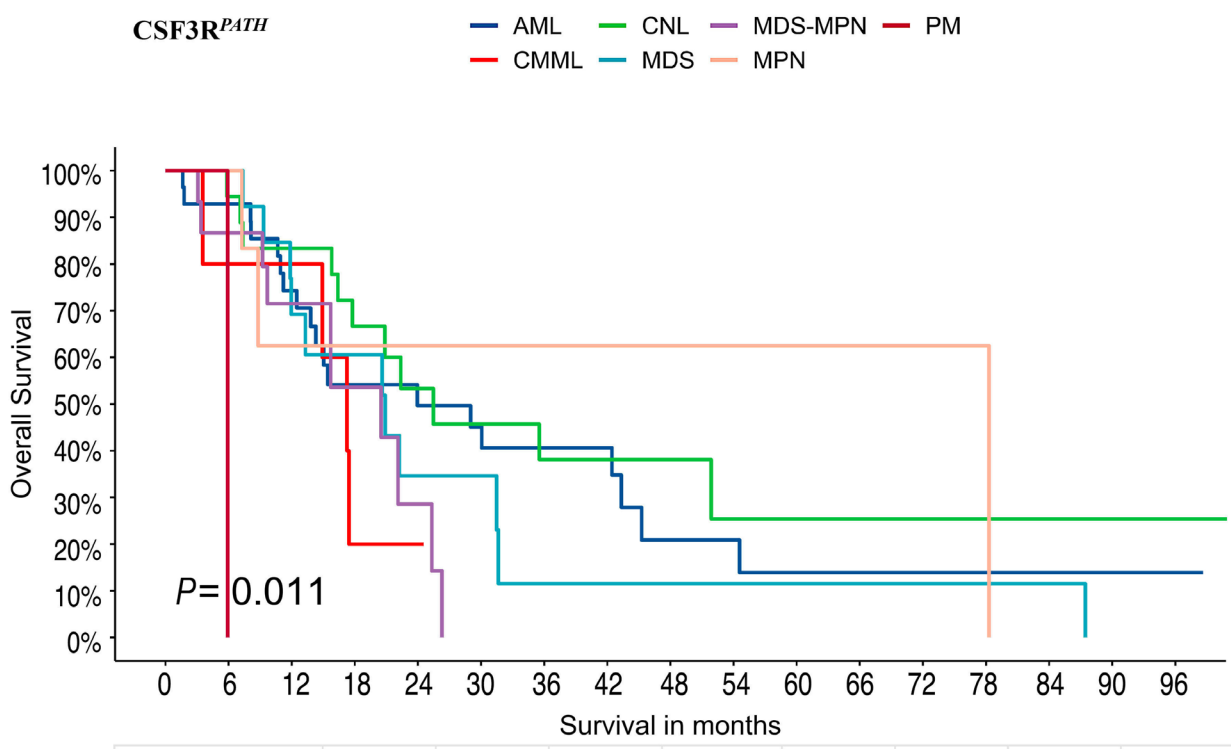
Table 1. Distribution of co-mutations across different disease groups.*

Gene	AML N=53 N (%)	CMML N=12 N (%)	CNL N=20 N (%)	MDS N=43 N (%)	MDS-MPN N=20 N (%)	MPN N=18 N (%)	PM N=15 N (%)	*CSF3R*^{VUS} N=91 N (%)	*CSF3R*^{PATH} N=91 N (%)	Total N=182 N (%)	*P*
ASXL1	17 (32)	6 (50)	12 (71)	15 (35)	6 (32)	5 (28)	1 (7)	16 (18)	46 (53)	62 (35)	0.02
TET2	5 (9)	4 (33)	0	9 (21)	6 (32)	2 (11)	4 (27)	15 (17)	15 (17)	30 (17)	0.07
SRSF2	9 (17)	2 (17)	8 (47)	3 (7)	2 (11)	2 (11)	1 (7)	9 (10)	18 (21)	27 (15)	0.02
DNMT3A	13 (25)	2 (17)	0	5 (12)	3 (16)	1 (6)	2 (13)	10 (11)	16 (18)	26 (15)	0.3
RUNX1	14 (26)	1 (8)	1 (6)	6 (14)	2 (11)	0	0	8 (9)	16 (18)	24 (14)	0.05
SETBP1	4 (8)	4 (33)	7 (41)	4 (9)	4 (21)	0	0	4 (4)	19 (22)	23 (13)	< 0.001
U2AF1	1 (2)	2 (17)	0	8 (19)	5 (26)	2 (11)	2 (13)	8 (9)	12 (14)	20 (11)	0.05
NRAS	10 (19)	4 (33)	0	1 (2)	2 (11)	0	0	5 (6)	12 (14)	17 (10)	0.004
EZH2	8 (15)	0	2 (12)	2 (5)	3 (16)	0	0	4 (4)	11 (13)	15 (8)	0.2
BCOR	6 (11)	2 (17)	0	4 (9)	0	0	1 (7)	7 (8)	6 (7)	13 (7)	0.4
IDH2	9 (17)	1 (8)	1 (6)	0	0	0	1 (7)	3 (3)	9 (10)	12 (7)	0.04
TP53	4 (8)	0	0	7 (16)	1 (5)	0	0	10 (11)	2 (2)	12 (7)	0.1
CEBPA	8 (15)	0	0	1 (2)	2 (11)	0	0	2 (2)	9 (10)	11 (6)	0.07
SF3B1	3 (6)	0	1 (6)	2 (5)	3 (16)	1 (6)	0	6 (7)	4 (5)	10 (6)	0.6
KRAS	2 (4)	4 (33)	1 (6)	0	1 (5)	1 (5)	0	4 (4)	5 (6)	9 (5)	0.001
ZRSR2	0	0	1 (6)	5 (12)	0	2 (11)	1 (7)	5 (6)	4 (5)	9 (5)	0.2
FLT3	7 (13)	1 (8)	0	0	1 (5)	0	0	5 (6)	4 (5)	9 (5)	0.08
JAK2	2 (4)	0	0	0	0	7 (40)	0	8 (9)	1 (1)	9 (5)	< 0.001
IDH1	4 (8)	0	0	2 (5)	0	2 (11)	0	3 (3)	5 (6)	8 (5)	0.5
STAG2	1 (2)	1 (8)	1 (6)	2 (5)	2 (11)	0	0	2 (2)	5 (6)	7 (4)	0.7
GATA2	3 (6)	0	0	2 (5)	0	1 (6)	0	2 (2)	4 (5)	6 (3)	0.8
WT1	5 (9)	0	0	0	1 (5)	0	0	1 (1)	5 (6)	6 (3)	0.2
CBL	2 (4)	1 (8)	1 (6)	0	0	1 (6)	0	3 (3)	2 (2)	5 (3)	0.7

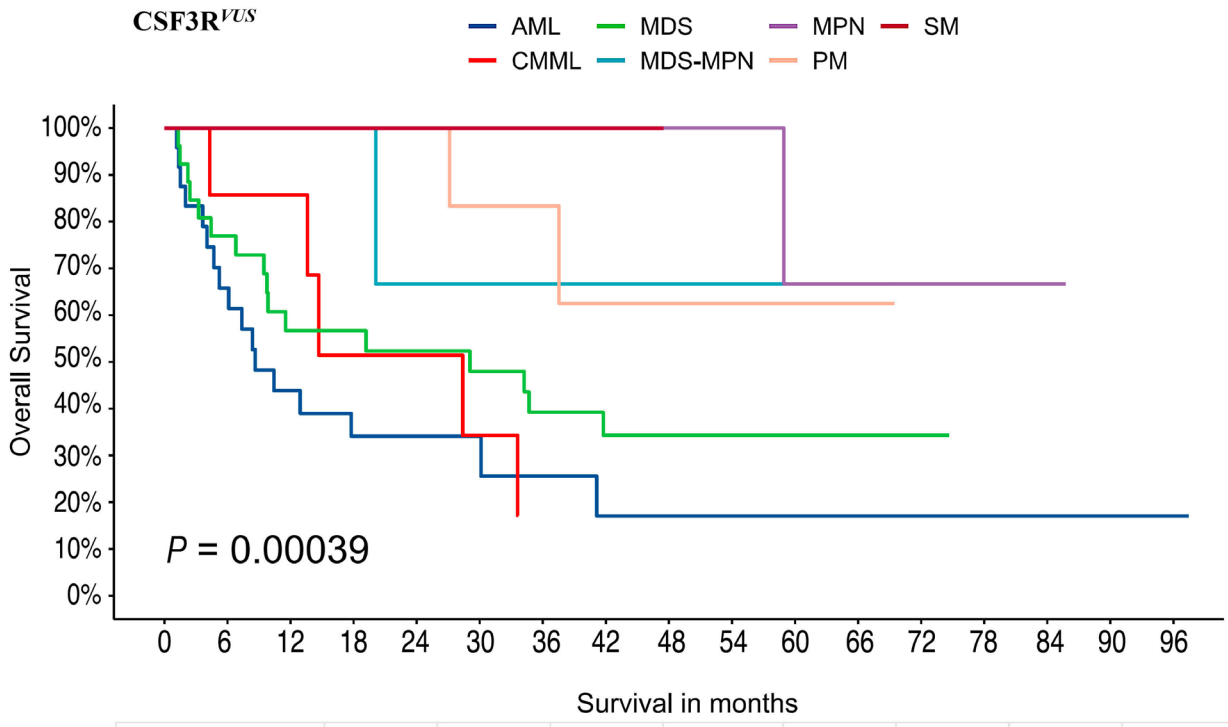
*This table includes only co-mutations with a prevalence of at least 2%. An all-inclusive table is shown in the *Online Supplementary Material*
AML: acute myeloid leukemia; CMML: chronic myelomonocytic leukemia; CNL: chronic neutrophilic leukemia; MDS: myelodysplastic syndrome; MDS-MPN: myelodysplastic/myeloproliferative neoplasm; MPN: myeloproliferative neoplasm; PM: premalignant condition; *CSF3R*^{VUS}: *CSF3R* variants of unknown significance; *CSF3R*^{PATH}: *CSF3R* pathogenic mutations.

lower VAF, with a median of 28% (IQR, 10-41) compared to *CSF3R*^{VUS}, with a median of 49% (IQR, 48-50; *P*<0.001). The latter observation suggested the possible inclusion of germline variants among patients with *CSF3R*^{VUS}. The most frequently identified *CSF3R*^{PATH} was T618I (N=53, 32%), observed in CNL (N=18, 34%), AML (N=15, 28%), and myelodysplastic/myeloproliferative neoplasm (N=8, 15%). *CSF3R*-E808K was the most frequent *CSF3R*^{VUS} (N=50, 30%) and was observed more commonly in AML (N=14, 8%), myelodysplastic syndrome (MDS) (N=17, 10%), and CMML (N=7, 4%). Figure 1 depicts details of *CSF3R* mutations/variants and associated myeloid neoplasms.

The median number of co-mutations was higher in patients with *CSF3R*^{PATH}, at three (range, 0-6), with 50 patients (58%) having more than three co-mutations, than in patients with *CSF3R*^{VUS}, who had a median of one (range, 0-5) co-mutation, with 25 patients (28%) having more than three co-mutations (*P*<0.001). Patients with AML



	AML (N=28)	CMML (N=5)	MDS (N=21)	MDS-MPN (N=15)	MPN (N=6)	PM (N=1)	CNL (N=20)	TOTAL (N=91)
Median Survival (months)	24	17	21	20	78	6	25	21



	AML (N=25)	CMML (N=7)	MDS (N=27)	MDS-MPN (N=5)	MPN (N=12)	PM (N=14)	SM (N=1)	TOTAL (N=91)
Median Survival (months)	9	28	29	NR	NR	NR	NR	37

Figure 2. Overall survival in patients with myeloid neoplasms with *CSF3R* mutations/variations. Overall survival of patients with myeloid neoplasms with *CSF3R* pathogenic mutations (*CSF3R*^{PATH}) (top) and variants of unknown significance (*CSF3R*^{VUS}) (bottom). AML: acute myeloid leukemia; CNL: chronic neutrophilic leukemia; MDS-MPN: myelodysplastic/myeloproliferative neoplasm; PM: premalignant conditions; CMML: chronic myelomonocytic leukemia; MDS: myelodysplastic syndrome; MPN: myeloproliferative neoplasms (myelofibrosis, polycythemia vera, chronic myeloid leukemia, and essential thrombocythemia); SM: systemic mastocytosis; NR: not reached.

and CMML displayed higher numbers of co-mutations. In patients with *CSF3R*^{PATH}, the most frequently observed co-mutations included *ASXL1* (N=46, 53%), *SETBP1* (N=19, 22%), and *SRSF2* (N=18, 20%). In patients with *CSF3R*^{VUS}, the most frequently observed co-mutations involved *ASXL1* (N=16, 18%), *TET2* (N=15, 17%), *DNMT3A* (N=10, 11%), and *TP53* (N=10, 11%). Table 1 and *Online Supplementary Table S1* show the distribution of co-mutations in different diseases.

In AML, the median age at diagnosis was 69 years (range, 17-90). *CSF3R*-T618I (N=15, 28%) was the most prevalent *CSF3R*^{PATH}, while *CSF3R*-E808K (N=14, 26%) was the most prevalent *CSF3R*^{VUS}. When compared to other myeloid neoplasms with *CSF3R*^{PATH}, AML patients with *CSF3R*^{PATH} were more likely to harbor co-mutations in *CEBPA, DNMT3A, IDH1, IDH2, NRAS, RUNX1*, and *WT1*. Among AML patients with *CSF3R*^{VUS}, *FLT3* and *KDM6A* were more common. In MDS patients, the median age at *CSF3R* mutation detection was 74 years (range, 37-93). The most common *CSF3R*^{PATH} were T618I (N=3, 19%) and Q749* (N=4, 25%). Other frequent mutation/variants included truncating *CSF3R* mutations/variants (Y752*, W791*, Q776*, Q770*, Q743*, and L751P-fs). The most common *CSF3R*^{VUS} in MDS was E808K (N=18, 42%), and it was more frequent in MDS than in other myeloid neoplasms (N=32, 26%; *P*=0.047). Among patients with *CSF3R*^{PATH}, co-mutations in *U2AF1* were more frequent in those with MDS (N=5, 31%) than in those with other myeloid neoplasms (N=6, 9%; *P*=0.01). *DDX41* mutations were more common in the *CSF3R*^{VUS} subcohort. Among the 20 patients with CNL, 18 (90%) harbored *CSF3R*-T618I and the median age at diagnosis was 70 years (range, 33-93). Compared to patients with other myeloid neoplasms with *CSF3R* mutations/variants, co-mutations were more frequent in CNL patients and involved *ASXL1* (71% *vs*. 34%; *P*<0.01), *SRSF2* (47% *vs*. 12%; *P*<0.01), and *SETBP1* (41% *vs*. 11%; *P*<0.01).

The median follow-up time for the entire cohort was 43 months (95% confidence interval [95% CI]: 34-49 months). Overall survival data across different myeloid neoplasms with *CSF3R* mutations/variants are shown in Figure 2. Patients with MPN (including myelofibrosis, polycythemia vera, chronic myeloid leukemia, and essential thrombocythemia) had a significantly longer median OS of 78 months (95% CI: 59 months-not reached) in patients with *CSF3R*^{PATH} and median OS not reached in those with *CSF3R*^{VUS}. In general, patients with *CSF3R*^{PATH} fared worse than those with *CSF3R*^{VUS}, in every disease group, with the distinct exception of AML, in which the opposite was true (Figure 2).

In AML patients, the median OS was 24 months (95% CI: 14-56 months) for those with *CSF3R*^{PATH} compared to 9 months (95% CI: 5 months-not reached) for those with *CSF3R*^{VUS} (*P*=0.1). Comparing the most common *CSF3R* mutations/variants in both cohorts, AML patients with the E808K variant had a worse OS of 5 months (95% CI: 4 months-not reached) compared to those with the T618I mutation who had a median OS of 30 months (95% CI: 12 months-not reached; hazard ratio [HR]=4, 95% CI: 1.7-10; *P*<0.01); significance was sustained after adjustment for European LeukemiaNet 2022 risk stratification, age, and transplant therapy (HR=4, 95% CI: 1.5-10.7; *P*<0.01). The median OS for MDS patients with *CSF3R*^{PATH} was 21 months (95% CI: 12 months-not reached) compared to 29 months (95% CI: 10 months-not reached) for MDS patients with *CSF3R*^{VUS} (*P*=0.6). Unlike the case with AML, *CSF3R*-E808K in MDS did not confer an inferior prognosis (OS 27 months *vs.* 21 months in patients with and without the specific variant, respectively; *P*=0.4). The median OS for patients with CNL was 25 months (95% CI: 18 months-not reached). The median time to blast transformation in 15 patients who transformed to AML from other myeloid neoplasms was 14 months, with a 5-year blast transformation-free survival of 52% (95% CI: 30-90%) for patients with *CSF3R*-^{PATH} compared to 89% (95% CI: 78-100%) for those with *CSF3R*^{VUS} (*P*<0.01).

The association between *CSF3R* mutations/variants and CNL has been well reported in the literature. The current study adds to this information in the context of a broader spectrum of *CSF3R* mutations/variants in myeloid neoplasms.[7-10] Our study is subject to limitations inherent to retrospective reviews, including the lack of functional studies, especially with regard to the distinction between germline variants and mutations. We were particularly intrigued by the frequent occurrence of the *CSF3R*-E808K variant and its prognostic relevance in AML.[11,12] Additional studies are needed to clarify the possible oncogenic/prognostic impact of *CSF3R*^{VUS} and recognize the fact that the characterizations regarding pathogenic *versus* VUS remain preliminary and are subject to change with future research.

Authors

Abiola Bolarinwa,[1] Natasha Szuber,[2] Rong He,[3] Animesh Pardanani,[1] Naseema Gangat[1] and Ayalew Tefferi[1]

[1]Division of Hematology, Mayo Clinic, Rochester, MN, USA; [2]University of Montreal, Montreal, Canada and [3]Division of Laboratory Medicine, Mayo Clinic, Rochester, MN, USA

Correspondence:
A. TEFFERI - tefferi.ayalew@mayo.edu

https://doi.org/10.3324/haematol.2025.288026

Received: April 10, 2025.
Accepted: June 23, 2025.
Early view: July 10, 2025.

Disclosures

NG has served on advisory boards for DISC Medicine and Agios.

Contributions

AB and AT designed the study, collected data, performed analyses, and wrote the paper. AT, AP and NG participated in patients' care. RH provided hematopathology and molecular laboratory expertise. All authors reviewed the final draft of the paper.

Data-sharing statement

Requests for original data should be made to the corresponding author.

References

1. Zhang H, Means S, Schultz AR, et al. Unpaired extracellular cysteine mutations of CSF3R mediate gain or loss of function. Cancer Res. 2017;77(16):4258-4267.
2. Fukunaga R, Ishizaka-Ikeda E, Seto Y, Nagata S. Expression cloning of a receptor for murine granulocyte colony-stimulating factor. Cell. 1990;61(2):341-350.
3. Tamada T, Honjo E, Maeda Y, et al. Homodimeric cross-over structure of the human granulocyte colony-stimulating factor (GCSF) receptor signaling complex. Proc Natl Acad Sci U S A. 2006;103(9):3135-3140.
4. Zhang H, Reister Schultz A, Luty S, et al. Characterization of the leukemogenic potential of distal cytoplasmic CSF3R truncation and missense mutations. Leukemia. 2017;31(12):2752-2760.
5. Pardanani A, Lasho TL, Laborde RR, et al. CSF3R T618I is a highly prevalent and specific mutation in chronic neutrophilic leukemia. Leukemia. 2013;27(9):1870-1873.
6. Szuber N, Finke CM, Lasho TL, et al. CSF3R-mutated chronic neutrophilic leukemia: long-term outcome in 19 consecutive patients and risk model for survival. Blood Cancer J. 2018;8(2):21.
7. Mohamed A, Gao J, Chen YH, et al. CSF3R mutated myeloid neoplasms: Beyond chronic neutrophilic leukemia. Hum Pathol. 2024;149:66-74.
8. Tefferi A, Thiele J, Vannucchi AM, Barbui T. An overview on CALR and CSF3R mutations and a proposal for revision of WHO diagnostic criteria for myeloproliferative neoplasms. Leukemia. 2014;28(7):1407-1413.
9. Maffei R, Paolini A, Conte B, et al. Distribution of different classes of CSF3R mutations and co-mutational pattern in 360 myeloid neoplasia. Ann Hematol. 2025;104(1):263-274.
10. Adema V, Hirsch CM, Przychodzen B, et al. Molecular spectrum of variants correlate with specific myeloid malignancies and secondary mutations. Blood. 2018;132(Supplement 1):4389.
11. Caron-L'Ecuyer J, Harnois M, Meunier MC, et al. Distinguishing germline from acquired pathogenic variants: key diagnostic features and clinical implications. Blood. 2024;144(Supplement 1):1783.
12. ClinVar. NM_000760.4(CSF3R):c.2422G>A (p.Glu808Lys). https://www.ncbi.nlm.nih.gov/clinvar/variation/434833/. Accessed March 31, 2025.