

Editorial

Implications of *CSF3R* and other Novel Mutations in Chronic Neutrophilic Leukemia

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Editorial

Chronic Neutrophilic Leukemia (CNL) is a rare but potentially aggressive type of Myeloproliferative Neoplasm (MPN) [1]. According to the current 2008 World Health Organization (WHO) diagnostic criteria, CNL is characterized by peripheral blood leukocytosis of $\geq 25 \times 10^9/L$ (80% of which are mature neutrophils and less than 10% are immature granulocytes); hepatosplenomegaly; a hypercellular bone marrow with myeloid hyperplasia, normal myeloid maturation and <5% blasts (less than 1% in the peripheral blood) [1]. Molecular and cytogenetic analysis must be performed to confirm the absence of Philadelphia chromosome with translocation t(9;22) (*BCR-ABL1*), and rearrangements in genes encoding platelet derived growth factor receptors alpha and beta (*PDGFRA/B*) and fibroblast growth factor receptor 1 (*FGFR1*) [1]. CNL is essentially a diagnosis of exclusion among the MPNs. Additional molecular analyses in the pathogenesis of CNL have been recently studied to better specify the diagnostic criteria of CNL and propose potential therapeutic targets.

Recent studies have determined that a particular mutation in the colony stimulating factor 3 receptor (*CSF3R*) may play a role in the development of CNL [2]. Germline *CSF3R* mutations have been previously reported in more than 30% of patients with Severe Congenital Neutropenia (SCN) [3]. The *CSF3R* gene is located on the short arm of chromosome 1. The gene encodes the transmembrane receptor for granulocyte colony-stimulating factor (G-CSF), which is involved in the proliferation, differentiation and function of granulocytes [2,3]. *CSF3R* signals through a number of pathways including the JAK-STAT pathway and the SRC tyrosine kinase pathway. Mutations that alter the receptor structurally and functionally disrupt its ability to regulate granulocytic differentiation and increase granulocytic proliferative capacity [2].

In a seminal study, Maxson et al. reported the usefulness of *CSF3R* mutation for the diagnosis of CNL [2]. In 27 patients with CNL and aCML, the *CSFR3* mutation was identified in 16 patients (59%). 90% of the patients with CNL contained a *CSF3R* mutation, whereas only 45% of patients with aCML contained a mutation. The most common mutations were detected in the membrane proximal region and include T618I (consists of a single base substitution of C-to-T at nucleotide 1853 on exon 14) and T615A. Other mutations that were

noted include frame shift and nonsense mutations in the cytoplasmic tail that lead to truncation. Through drug sensitivity profiling, it was observed that mutations that lead to truncation of the cytoplasmic tail lead to dysregulation in the SRC family-TNK2 kinases, whereas mutations in membrane proximal lead to dysregulation in JAK family kinases. An additional finding *via in vitro* drug sensitivity assays was the overall clinical improvement noted in patients with CNL that had *CSF3R* T681I who were treated with ruxolitinib, a JAK1/2 inhibitor, whereas those who had truncated mutations responded selectively to dasatinib, a SRC kinase inhibitor. This observation supported the theory that patients with CNL may show differences in sensitivity to different pathway inhibitors based on the type of *CSF3R* mutation they harbored.

Pardanani et al. further validated the previous findings by sequencing exons 14-17 of *CSF3R* in clinical cases suggestive of CNL or aCML [4]. Out of 54 total cases, 12 cases met WHO-defined criteria for CNL and 9 cases were WHO-defined aCML [3]. An overall mutational frequency of 100% was observed in WHO-defined CNL cases, in contrast to 0% seen in WHO-defined aCML. The primary mutation encountered was *CSF3R* T618I (n=10). They also reported *SETBP1* mutations in 33% patients. Another update of this series has reported *SETBP1* mutation in 40% in *CSF3R* mutated CNL [5]. In another study, a group of 14 *CSF3R* mutated CNL patients were screened for additional mutations [6]. The most frequent co-mutation was *ASXL1* (8 patients; 57%). Five patients' harbored *SETBP1* mutations (38%), 2 patients had both mutations. Two patients with *SETBP1* (not *ASXL1*) mutations developed blastic transformation. Two patients evolved into Chronic Myelomonocytic Leukemia (CMML); both these patient had *ASXL1* mutations but wild type *SETBP1* gene. On a multivariate analysis, *ASXL1* mutation and thrombocytopenia were reported to be independent risk factors for survival. Other rarely reported mutations in CNL include *JAK2* V617F, and *CALR*.

Since the discovery of *CSF3R* mutations in CNL, several cases of *CSF3R* mutated CNL and aCML have reported that received Ruxolitinib. A patient carrying *CSF3R* T618I was reported to have dose dependent reduction in neutrophil count and resolution of thrombocytopenia [7]. At present, ruxolitinib remains a second line treatment in patients who fail hydroxyurea [8].

Two cases of *CSF3R* and *SETBP1* mutated CNL have been reported so far that failed to respond to ruxolitinib (JAK1/2 inhibitor). This raises the possibility of ineffectiveness of JAK inhibitors in *CSF3R* mutated CNL cases harboring *SETBP1* mutation [5].

These findings, as well as other gene mutations recently found to be implicated in CNL, such as mutations in set binding protein 1 (*SETBP1*) and *JAK2*, have culminated in a revision of the current WHO diagnostic classification of CNL to include the molecular

criterion [9]. Presence of *CSF3R* T618I or other activating mutations is one of the diagnostic criteria in the current updated WHO 2016 classification of hematopoietic neoplasms. In the absence of *CSF3R* mutation, persistent neutrophilia, splenomegaly and no identifiable cause of reactive neutrophilia including plasma cell neoplasm, should be demonstrated. In presence of any these confounding factors, clonality of myeloid cells must be demonstrated by cytogenetic or molecular studies [9].

With the recent discovery of *CSF3R* mutations, CNL no longer remains a diagnosis of exclusion. Presence of *CSF3R* and /or other recurrent mutations makes its distinction easier from reactive leukocytosis. This has also led to better characterization of the disease and easier differentiation from its close mimicker, atypical chronic myeloid leukemia. The *CSF3R* mutations have proven potential to not only the diagnosis of CNL but to the development of possible target therapies also. *SETBP1* and *ASXL1* mutations appear to be of prognostic significance and correlate with disease progression.

References

1. Bain BJ, Brunning RD, Vardiman JW, Thiele J, Swerdlow SH, Campo E, et al. Chronic neutrophilic leukaemia WHO Classification of Tumors of Haematopoietic and Lymphoid Tissues. Lyon, France: IARC Press. 2008: 38-39.
2. Maxson JE, Gotlib J, Pollyea DA. Oncogenic *CSF3R* mutations in chronic neutrophilic leukemia and atypical CML. *N Engl J Med*. 2013; 19: 1781-1790.
3. Klimiankou M, Mellor-Heineke S, Zeidler C. Role of *CSF3R* mutations in the pathomechanism of congenital neutropenia and secondary acute myeloid leukemia. *Ann NY Acad Sci*. 2016; 1: 119-125.
4. Pardanani A, Lasho TL, Laborde RR. *CSF3R* T618I is a highly prevalent and specific mutation in chronic neutrophilic leukemia. *Leukemia*. 2013; 27: 1870-1873.
5. Lasho TL, Mims A, Elliott MA. Chronic neutrophilic leukemia with concurrent *CSF3R* and *SETBP1* mutations: Single colony clonality studies, *in vitro* sensitivity to JAK inhibitors and lack of treatment response to ruxolitinib. *Leukemia*. 2014; 28: 1363-1365.
6. Elliot MA, Pardanani A, Hanson LA. *ASXL1* mutations are frequent and prognostically detrimental in *CSF3R*-mutated chronic neutrophilic leukemia. *Am J Hematol*. 2015; 90: 653-656.
7. Stahl M, Xu ML, Steensma DP. Clinical response to ruxolitinib in *CSF3R* T618-mutated chronic neutrophilic leukemia. *Ann Hematol*. 2016; 95: 1197.
8. Elliott MA, Tefferi A. Chronic neutrophilic leukemia 2016: Update on diagnosis, molecular genetics, prognosis and management. *Am J Hematol*. 2016; 913: 341-349.
9. Arber D, Orazi A, Hasserjian R. The 2016 revision to the World Health Organization (WHO) classification of myeloid neoplasms and acute leukemia. *Blood*. 2016.