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CASE REPORT

Management of paroxysmal nocturnal hemoglobinuria in CALR mutated post-essential thrombocythemia myelofibrosis: A case report

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Abstract

Paroxysmal nocturnal hemoglobinuria (PNH) results from the loss of erythrocyte surface proteins, leading to complement activation and its spectrum of effects. We explore this case of a 57-year-old man with post-essential thrombocythemia (ET) myelofibrosis (MF) who developed symptomatic anemia with evidence of hemolysis on lab work. While hemolysis was localized to be intramedullary based on workup, the exact diagnosis was undetermined, leading to a prolonged course of steroid therapy to control anemia. The hemolysis was eventually attributed to PNH diagnosed on flow cytometry and the patient was treated with complement inhibitors with eventual failure of therapy. He ultimately underwent a successful hematopoietic cell transplant (HCT) with post-transplantation flow cytometry showing complete resolution of PNH. While PNH has been identified as a progression of myelodysplastic syndromes, the mechanisms of its rare development in myeloproliferative neoplasms are poorly elucidated. Furthermore, its rarity and often vague symptoms make diagnosis and treatment a challenge. This is the second reported case of a JAK2-negative, CALR-positive post-ET MF and the first reported case to be treated with HCT. This case probes for further insight into the clinical significance between MF and PNH, its impact on management, and further consideration for HCT as curative therapy in such patients who fail complement inhibitor therapy.

KEYWORDS
CALR, case report, myelofibrosis, PNH, thrombocythemia

1 | INTRODUCTION

Paroxysmal nocturnal hemoglobinuria (PNH) is a rare disorder named for one of its uncommon symptoms. PNH can present with a collection of other non-specific symptoms including hemolysis, cytopenias, fatigue, and shortness of breath. A mutation of the PIGA gene results in a lack of CD55 and CD59 erythrocyte surface inhibitory proteins leading to uncontrolled complement activation [1]. Other downstream mutations causing PNH have also been identified. PNH is diagnosed through flow cytometry and treated with eculizumab, a monoclonal antibody that inhibits the terminal complement. However, bone marrow transplantation remains the only cure for PNH.

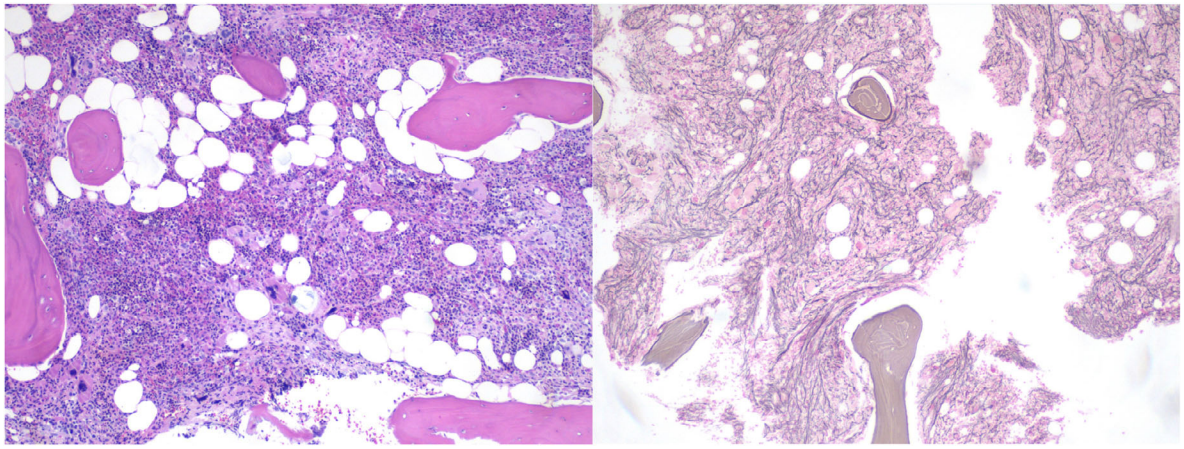

FIGURE 1 Left: Hypercellular bone marrow with increased trilineage hematopoiesis, megakaryocytic atypia, and clustering. Right: Reticulin Stain showing increased reticulin fibrosis consistent with myelofibrosis (MF)-2.

Although PNH clones commonly arise in myelodysplastic syndromes (MDS), they are uncommon in myeloproliferative neoplasms (MPNs) [2]. We present the second reported case of late-stage evolution of PNH in the setting of JAK2V617F-negative CALR-1 mutated post-essential thrombocythemia myelofibrosis (post-ET MF). Further, we examine the success of bone marrow transplantation as a curative therapy for PNH in bone marrow failure.

2 | CASE SUMMARY

A 57-year-old healthy man sought hematology evaluation due to nosebleeds and an elevated platelet count in the one-million range. Initial bone marrow biopsy showed megakaryocytosis with normal cytogenetics and JAK2 negative on molecular testing. CALR mutation testing was not performed at this time as the patient's diagnosis occurred years before the discovery of CALR mutation's association with MPNs in 2013. His ET was initially treated with anagrelide, hydroxyurea, and aspirin, but he experienced intolerable side effects. Eventually, treatment with interferon effectively maintained a stable platelet count (400–500 $\times$ 10^3/μL) but it was discontinued when he and his wife decided to plan for a pregnancy.

The patient opted to remain off therapy and maintained stable blood counts on routine monitoring (white blood cells [WBC] 6 $\times$ 10^3/μL; hemoglobin [Hgb] 12–13 g/dL; platelet count [Plt] 523 $\times$ 10^3/μL). Eventually, an upper respiratory infection led to a drop in hemoglobin (11.0 g/dL) without subsequent improvement, prompting a bone marrow biopsy. Biopsy revealed hypercellular marrow with increased grade 2 reticulin fibrosis, no increased blasts (< 1%), atypical localization of immature myeloid precursors, and clustering of large voluminous atypical megakaryocytes (Figure 1). DIPPS-plus score was 2, indicating the Intermediate-2 risk category for MF. He started ruxolitinib 10 mg daily leading to limited improvement in anemia prompting the addition of pegylated interferon alpha 45mcg weekly, but he remained with persistent anemia (Hgb 9.1 g/dL). As he became more anemic over months, he was started on darbepoetin alfa every 2–3 weeks without improvement. Due to worsening anemia requiring red blood cell (RBC) transfusions, the patient decided to stop both ruxolitinib and pegylated interferon. Repeat lab work showed WBC 5.9 $\times$ 10^3/μL, Hgb 7.3 g/dL, and Plt 269 $\times$ 10^3/μL with elevated erythropoietin at 1880 μU/L. Hemolysis labs significantly included low haptoglobin < 10 mg/dL, high LDH 1,046 U/L, total bilirubin 1.2 mg/dL, and reticulocyte count 7.5%. Direct coombs test was negative and cold hemagglutinin < 1:4, ruling out other causes of immune-mediated extramedullary hemolysis. Interval repeat molecular testing demonstrated a type 1 CALR mutation (52 bp del) at a variant allele frequency of 41% and was negative for JAK2 mutation. Re-introduction of prednisone improved hemoglobin levels, consistent with the immune basis for the patient's hemolysis and intramedullary hemolysis remained the working diagnosis as the underlying cause was not identified despite repeated workup and trials of prednisone. As a result, the patient remained controlled on prednisone for nearly 3.5 years.

Upon transferring care to a new clinician, new laboratory studies, including flow cytometry and bone marrow biopsy were performed. Flow cytometry revealed PNH clones with monocytes 83.79% and granulocytes 84.29% (Figure 2). Bone marrow biopsy was consistent with stage 2–3 MF (DIPPS-plus score of 3) without myelodysplasia; large hyper-lobulated hyperchromatic megakaryocytes are seen clustered. He started treatment with ravulizumab with concurrent prednisone dose reduction. Rivaroxaban was started for thrombosis prophylaxis though the patient did not experience any thrombotic complications. Hemoglobin improved to a peak of 9 g/dL on ravulizumab before the patient became refractory after 6 months of therapy and transitioned to pegcetacoplan which also failed after 6 months. He received supportive RBC transfusions as needed. Eventually, the patient made the decision to be evaluated for an allogeneic hematopoietic cell transplant (allo-HCT), which he underwent successfully. Post-transplant flow cytometry first showed a reduction in PNH clone size with complete clearance of PNH clone on follow-up flow cytometry.

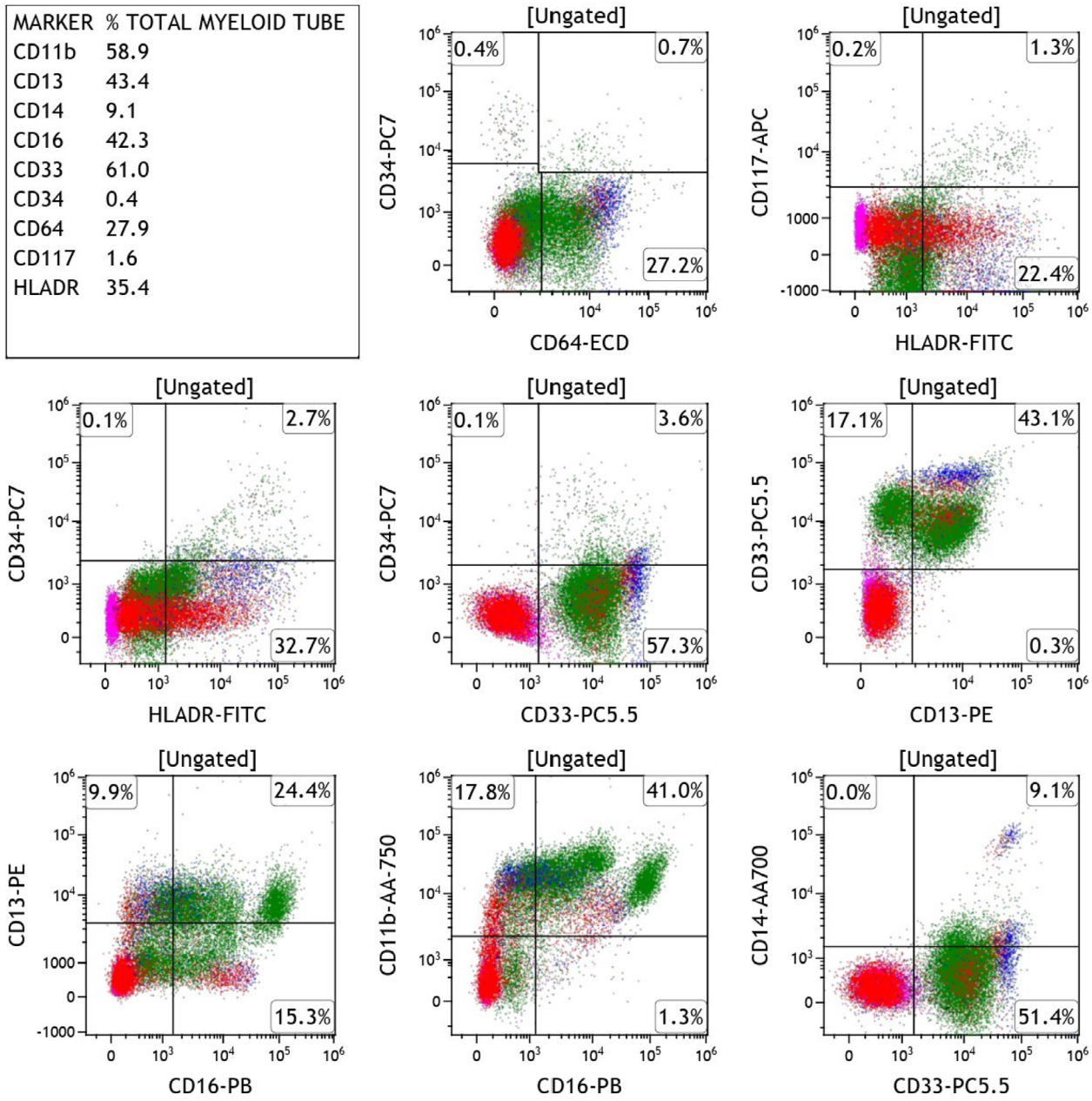

FIGURE 2 Flow cytometry indicating no increase in blasts.

3 | DISCUSSION

Our patient's case highlights the challenges and complexities involved in managing post-ET MF and the importance of considering PNH as a rare co-existing condition in patients who have persistent evidence of coombs negative hemolytic anemia. This case examines disease progression over a 23-year time frame, showing that seemingly minor changes in a patient's clinical state such as persistent anemia may reflect disease progression or new developments that should be worked up with a broad differential. It is easy to overlook slow longitudinal disease progression as a physician managing an established diagnosis for years.

Treatment for symptomatic PNH patients with hemolysis is complement inhibitors. Ravulizumab and eculizumab are among the first-line recommendations and act by binding C5 preventing activation. Pegcetacoplan is a second-like therapy for patients who fail C3 inhibitors and works by binding C3, leading to inhibition of both intravascular and extravascular hemolysis. For PNH patients with bone marrow failure due to MDS, HCT is recommended as curative therapy, though there are no clinical trials comparing response to allogenic HCT as opposed to complement inhibitor therapy. Small studies and cases showed curative outcomes in PNH patients with bone marrow failure due to severe MDS. Given PNH in the setting of severe MF, our patient ultimately underwent an HCT. This is the first reported case to

establish the success of HCT in patients with PNH in the setting of MPN with curative outcomes. HCT should be considered a treatment option for patients who do not respond to complement therapy or have bone marrow failure as a result of severe MF.

However, the limitation of these findings is that this is a case report focusing on one patient; the outcomes may not be generalizable to all patients in similar situations. We also do not have data to support any causative relationships. Further case studies and clinical trials are necessary to establish the repeated success of HCT in cases such as this, which may be difficult to carry out due to the rarity of this disease process.

Although rare, the development of PNH clones has been reported in prior case reports of patients with JAK2V617F and MPLW515L mutations driving a primary MPN, but only one other case reporting a CALR mutation was identified was identified [2]. Regardless of the mutation, PNH could arise in the context of any MPN as a consequence of a slow progression of disease leading to genetic changes over time in hematopoietic stem cells eventually leading to PNH. This supports the multiple-hit hypothesis. We posit that the amplification of PNH clones into clinically significant diseases may be correlated to the CALR mutation, which has been shown to cause excess cell proliferation [3]. This theory is supported by another case of late-stage PNH in CALR-positive ET [4].

The clinical significance of PNH in MPN is unclear, but one theory suggests the possible influence of PNH on the progression of MPN itself explaining the rise of clones in cases of late-stage disease [5]. A study examining the prevalence of PNH clones in various bone marrow diseases found PNH clones in a substantial number of cases [6]. As a result, routine screening for PNH may be beneficial in patients with bone marrow diseases, regardless of whether they have symptoms of hemolysis [6]. One downside of this study is the small sample size.

Another retrospective study suggests the opposite, purporting that symptoms or labs suggesting anemia in MPN are not suggestive of PNH [7]. While this study also had a small sample size, they recommend against standardized screening for PNH in patients with MPN specifically. Despite this, the proposal for PNH clone screening may remain valid for other intrinsic bone marrow diseases [6, 7]. One prospective study provides alternative causes of positive hemolysis markers in MPN by examining haptoglobin and LDH levels in MF patients without hemolytic anemia or liver cirrhosis. They found a correlation between these abnormal markers with high JAK-2 allele burden and inflammatory marker levels (C-reactive protein and albumin), but they did not look at other mutations such as CALR [8]. These conflicting findings and loose correlations highlight a gap in clinical understanding and practice for diagnosing and managing PNH in patients with MPN.

4 | CONCLUSION

This case highlights the challenges and complexities involved in managing MF and the importance of considering immune factors in intramedullary hemolysis, as well as the identification of rare coexisting conditions such as PNH that may influence patient care.

Our case of PNH in MF without any of the classical symptoms provides us insight into the genetic correlations between PNH and MF. Though there may not be a direct causative relationship, it probes further questions worth investigating regarding the clinical significance of PNH in such cases. This includes the factors that would lead one to consider PNH testing in patients with MF, the clinical significance of PNH in MF, and the impact on management, if any.

AUTHOR CONTRIBUTIONS

Mahija Cheekati contributed to the research, writing, editing, and submission of the manuscript. KarLeung Siu contributed to the patient's diagnosis and care, providing labs and work, and reviewing of manuscript. Rachel Ochs contributed to the patient's diagnosis and provided images of the patient's bone marrow biopsy and flow cytometry.

CONFLICT OF INTEREST STATEMENT

The authors declare no conflict of interest.

FUNDING INFORMATION

No outside funding was received for this project.

DATA AVAILABILITY STATEMENT

The data that support the discussion in this article is included in the references.

ETHICS STATEMENT

Not applicable. This did not include human subjects for research.

PATIENT CONSENT STATEMENT

The patient was informed of this case study.

CLINICAL TRIAL REGISTRATION

Not applicable. This is not a clinical trial.

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