Correspondence to: Vandana Puri
Department of Pathology, University College of Medical Sciences & GTB Hospital, Dilshad Garden, Delhi 110095, India
E-mail: drvandanapuri@gmail.com

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REFERENCES

1. Young JL Jr, Miller RW. Incidence of malignant tumors in U. S. children. J Pediatr 1975;86:254-8.
2. Pui CH, Relling MV, Downing JR. Acute lymphoblastic leukemia. N Engl J Med 2004;350:1535-48.
3. Jonsson OG, Sartain P, Ducore JM, Buchanan GR. Bone pain as an initial symptom of childhood acute lymphoblastic leukemia: association with nearly normal hematologic indexes. J Pediatr 1990;117:233-7.
4. McKay C, Furman WL. Hypercalcemia complicating childhood malignancies. Cancer 1993;72:256-60.
5. Hann IM, Lees PD, Palmer MK, Gupta S, Morris-Jones PH. Renal size as a prognostic factor in childhood acute lymphoblastic leukemia. Cancer 1981;48:207-9.
6. Bhatia NG, Sneha LM, Selvan SM, Scott JJ. Acute renal failure as an initial manifestation of acute lymphoblastic leukemia. Indian J Nephrol 2013;23:292-3.
7. Hibi S, Funaki H, Ochiai-Kanai R, et al. Hypercalcemia in children presenting with acute lymphoblastic leukemia. Int J Hematol 1997;66:353-7.
8. Rosol TJ, Capen CC. Mechanisms of cancer-induced hypercalcemia. Lab Invest 1992;67:680-702.
9. Shimonodan H, Nagayama J, Nagatoshi Y, et al. Acute lymphocytic leukemia in adolescence with multiple osteolytic lesions and hypercalcemia mediated by lymphoblast-producing parathyroid hormone-related peptide: a case report and review of the literature. Pediatr Blood Cancer 2005;45:333-9.
10. Lokadasan R, Prem S, Koshy SM, Jayasudha AV. Hypercalcaemia with disseminated osteolytic lesions: a rare presentation of childhood acute lymphoblastic leukaemia. Ecancermedicalscience 2015;9:542.
11. Turkmen K, Altintepe L, Guney I, et al. Uncontrolled hypertension secondary to leukemic cell infiltration of kidneys in a hemodialysis patient. Int J Nephrol Renovasc Dis 2010;3:65-8.
12. Larsen G, Loghman-Adham M. Acute renal failure with hyperuricemia as initial presentation of leukemia in children. J Pediatr Hematol Oncol 1996;18:191-4.

JAK2 exon 12 mutation-positive myeloproliferative neoplasm associated with recurrent thromboembolism

TO THE EDITOR: Detailed analysis of Philadelphia (Ph) chromosome-negative myeloproliferative neoplasms, essential thrombocythemia (ET), and polycythemia vera (PV) paved the way for the recognition of Janus kinase 2 (*JAK2*) as a potential genetic factor [1]. While most cases of these diseases are related to *JAK2* V617F, some are associated with *JAK2* exon 12 mutation. *JAK2* exon 12 mutation is a rare phenomenon that is not yet well studied. Although, the presence of *JAK2* exon 12 mutation is linked to PV and ET, its phenotypic features are not clear, especially in cases without ET and PV. The *JAK2* exon 12 G571S variant has also not been studied well, and its phenotypic features are unknown. We present a case of recurrent deep vein thrombosis (DVT) and pulmonary embolism that had a negative coagulation workup, but was positive for *JAK2* exon 12 (G571S) mutation. The presented case suggests a possible relationship between *JAK2* exon 12 mutation and recurrent thrombosis. While *JAK2* V617F is inherently related to thrombotic events, irrespective of the platelet level, *JAK2* exon 12 might have similar procoagulant effects. It is hypothesized that further molecular studies can reveal the underlying factors that cause this co-occurrence.

CASE

A 38-year-old Hispanic woman with a past medical history of pulmonary embolism, DVT, and asthma was seen for an evaluation of her recurrent episodes of thrombosis. She had experienced three episodes of DVT and one episode of pulmonary embolism. The first episode of DVT occurred 5 years ago, and the second episode happened 3 years ago. The pulmonary embolism occurred 1 year ago, following her third episode of DVT. Each DVT episode presented with lower leg edema and pain, and was confirmed by lower extremity ultrasound. The pulmonary embolism episodes presented as shortness of breath and low oxygen saturation.

She was given the following medications to take at home: Ipratropium-Albuterol (Duoneb Neb), 3 mL by inhalation every 4 hours as needed, and warfarin sodium (Coumadin), 12 mg orally once daily. However, she was non-adherent to the use of warfarin. The notable aspects of her family history were that her mother had diabetes mellitus and hypertension, while her father had colon cancer at age of 42. She was a current smoker and had been smoking 2 packs of cigarettes daily for 20 years. She denied alcohol and drug use. A review of systems was non-contributory.

A recent complete blood cell count revealed the following: white blood count 11.1×10^9/L, hemoglobin level 14.9 g/dL,

hematocrit 45.4%, mean corpuscular volume (MCV) 81.6 fL, mean corpuscular hemoglobin (MCH) level 30.7 pg, mean corpuscular hemoglobin concentration (MCHC) 32.8 g/dL, platelet count 555×10^9/L, neutrophil 46%, lymphocyte 41%, monocyte 4.6%, eosinophil 3.9%, and basophil 0.1%. A basic metabolic profile (BMP) showed the following levels: sodium 139 mmol/L, potassium 4.1 mmol/L, chloride 109 mmol/L, carbon dioxide 29 mmol/L, blood urea nitrogen (BUN) 6 mg/dL, creatinine 0.6 mg/dL, glucose 86 mg/dL, calcium 9.1 mg/dL, phosphorus 3.8 mg/dL, magnesium 2.2 mg/dL, total protein 7.8 g/dL, albumin 3.7 g/dL, aspartate amino-transferase (AST) 16 U/L, alanine amino-transferase (AST) 20 U/L, and alkaline phosphatase 119 U/L.

Her latest chest computed tomography showed a small incomplete filling defect in the right upper lobe, suggesting a chronic non-occluding pulmonary embolism. A coagulation workup, including lupus anticoagulant, protein C and S level/activity, antithrombin III level/activity, factor V level/activity, anti-cardiolipin IgG antibody, anti-cardiolipin IgM antibody, beta 2-GPI antibody (IgG,IgA, and IgM), Von Willebrand factor (VWF), factor VIII, methylenetetrahydrofolate reductase (*MTHFR*) gene mutation, homocysteine level, and prothrombin mutation analysis showed normal findings. For evaluation of high-normal platelet level, *JAK2* mutation was analyzed; no other cause of thrombocytosis had been identified in the patient's history or laboratory findings. The result showed G571S mutation in *JAK2* exon 12, and negative for *CALR* mutation. Total nucleic acid from plasma was used to sequence exons 12 and 13 using the polymerase chain reaction technique. The patient refused to undergo a bone marrow study.

DISCUSSION

The patient's platelet count has been in the higher range, from 350×10^9/L to 800×10^9/L, during the last 5 years. Some articles have mentioned that, in order to diagnose ET, it is necessary to have two platelet counts above 600×10^9/L that were measured 1 month apart. On the other hand, the recent World Health Organization definition reduces this threshold to 400×10^9/L, if that value is sustained. However, the laboratory findings for our patient did not satisfy either of the definitions of ET. The co-occurrence of the high platelet level and recurrent thrombosis, in the absence of other procoagulant abnormalities, highlights the possibility of a myeloproliferative disorder.

There is a proven correlation between PV and ET, and *JAK2* mutation [1]. Although most of the relevant cases are *JAK2* V617F, some are associated with *JAK2* exon 12 mutation. To date, there are more than 30 known *JAK2* exon 12 mutations. Nonetheless, *JAK2* exon 12 mutations are rare and have not been widely studied. In general, *JAK2* exon 12 mutations appear in younger individuals. Most of the reported cases of *JAK2* exon 12 mutation have been linked to PV or ET. Bone marrow analysis of *JAK2* exon 12 mutation cases show moderately hypercellular marrow with predominant erythroid hyperplasia. Patients with *JAK2* exon 12 mutation lack the prominent megakaryocyte clusters that can be seen in patients with *JAK2* V617F-positive PV. However, some patients with *JAK2* exon 12 mutation have elevated numbers of megakaryocytes [2].

JAK2 V617F has been associated with thrombotic events, such as Budd-Chiari syndrome. Interestingly, in *JAK2* V617F-positive patients, thrombotic events can occur even in the absence of increased red blood cells or platelets [3]. It has been shown that, in comparison with platelet quantity, platelet dysfunction is more important in thrombosis formation. To date, no association has been observed between *JAK2* exon 12-positive status (without erythrocytosis or essential thrombosis) and recurrent thrombus events. Limited number of studies regarding *JAK2* exon 12 have shown that mutations are more predominantly related to PV, with fewer cases of thrombosis [2].

ET is mainly caused by mutations in exon 9 of *CALR*, exon 10 of *MPL*, or *JAK2* V617F in more than 90% of cases. The remaining cases are known as "triple negative." In a recent study, Milosevic Feenstra *et al.* [4] performed whole-exome sequencing (WES) on samples from eight triple-negative patients. The authors were able to identify somatic mutations in three of the eight patients. For the three patients with clonal hematopoiesis, the analysis showed S204P and V285E mutations in *MPL*, and G571S mutation in *JAK2* exon12. However, Milosevic Feenstra *et al.* [4] did not describe any thrombotic event in the triple-negative patients (including the G571S mutation). Additionally, our case does not satisfy the criteria for ET, despite having high-normal platelet levels.

To date, the phenotypic features of the individual mutations of *JAK2* exon 12 are understudied and unknown. One study has described five cases of the abovementioned G571S variant [5]. Here, we have reported a case of recurrent DVT and pulmonary embolism in which the patient had a negative coagulation workup, but was positive for *JAK2* exon 12 (G571S) mutation. The presented case did not satisfy the criteria for PV and ET, even though our patient had a higher than normal platelet count over several years.

Vahid Tavakoli, Sundee Naing

Department of Internal Medicine, Richmond University Medical Center, Staten Island, NY, USA

Correspondence to: Vahid Tavakoli
Department of Internal Medicine, Richmond University Medical Center, 355 Bard Ave, Staten Island, NY 10310, USA
E-mail: v_tavakoli@yahoo.com

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REFERENCES

1. Baxter EJ, Scott LM, Campbell PJ, et al. Acquired mutation of the tyrosine kinase JAK2 in human myeloproliferative disorders. Lancet 2005;365:1054-61.
2. Scott LM. The JAK2 exon 12 mutations: a comprehensive review. Am J Hematol 2011;86:668-76.
3. Darwish Murad S, Plessier A, Hernandez-Guerra M, et al. Etiology, management, and outcome of the Budd-Chiari syndrome. Ann Intern Med 2009;151:167-75.
4. Milosevic Feenstra JD, Nivarthi H, Gisslinger H, et al. Whole-exome sequencing identifies novel MPL and JAK2 mutations in triple-negative myeloproliferative neoplasms. Blood 2016;127:325-32.
5. Alghasham N, Alnouri Y, Abalkhail H, Khalil S. Detection of mutations in JAK2 exons 12-15 by Sanger sequencing. Int J Lab Hematol 2016;38:34-41.

Is N-acetylcysteine infusion an effective treatment option in L-asparaginase associated hepatotoxicity?

TO THE EDITOR: L-asparaginase is an enzyme whose therapeutic effect has been well documented in children with acute lymphoblastic leukemia (ALL). The drug induces a relative asparagine deficiency that leads to the death of human lymphoblasts [1]. However, the mechanism of L-asparaginase-associated hepatotoxicity is not clear. There are few reports on the treatment of L-asparaginase-associated hepatotoxicity [2-4]. An experimental study conducted by Roesmann *et al.* [2] reported that when L-carnitine is administered in combination with L-asparaginase for steatosis, carnitine reduces the toxicity associated with L-asparaginase. Alshiekh-Nasany and Douer [3] reported that polyethylene glycol (PEG)-asparaginase related high-grade liver toxicity in adult patients could be rapidly and permanently reversed using the amino acid derivate L-carnitine. Al-Nawalki *et al.* [4] reported that histopathological macrovesicular steatosis was detected in 3 adult patients who developed L-asparaginase toxicity, and that the hepatotoxicity resolved after administration of carnitine and a vitamin B complex.

L-asparaginase is enzymatically active against glutamine, but with a significantly lower affinity against glutamine than L-asparagine [5]. L-glutaminase activity results in a reduction in the plasma glutamine level [6]. N-acetylcysteine (NAC) increases glutathione and antioxidant pools in hepatic cells, and thus increases the resistance of hepatic cell membranes [7]. Here, we describe a pediatric patient who developed severe hepatotoxicity accompanied by hyperamylasemia, hyperlipidemia, and elevated transaminase and bilirubin levels following the administration of 5 doses of L-asparaginase, and the parameters that were resolved following NAC infusion.

In the 2.5-year-old patient who was being followed-up after a diagnosis of pre-B ALL, induction therapy was initiated with a combination chemotherapy regimen comprising 60 mg/m^2 prednisolone (oral) (days 1–33), 1.5 mg/m^2 vincristine and 30 mg/m^2 daunorubicin (days 7, 15, 22, and 29), and 5,000 μ/m^2 L-asparaginase (*E. Coli*-Asp) (8 doses) (days 12, 15, 18, 21, 24, 27, 30, and 33). After the fifth L-asparaginase administration, the patient's laboratory results were as follows: alanine transaminase (ALT) 501 IU/L, aspartate transaminase (AST) 664 IU/L, gamma-glutamyl transferase (GGT) 1,297 IU/L, alkaline phosphatase (ALP) 246 IU/L, total/direct bilirubin 7/6.09 mg/dL, total cholesterol 272 mg/dL, triglyceride 571 mg/dL, and amylase 146 IU/L. The patient's clinical findings rapidly deteriorated, and abdominal distention and hepatomegaly were observed. Oral feeding was discontinued, and antibiotic (carbapenem and teicoplanin) and antifungal therapy (liposomal amphotericin B) was initiated. Tests for hepatitis A, B, and C, cytomegalovirus, Epstein Barr virus, parvovirus, and human immunodeficiency virus serology returned negative results. There was no growth in blood cultures. Abdominal computerized tomography revealed an increased liver size and density due to hepatosteatosis (Fig. 1). Drug-related toxic hepatitis was considered because of the progressively increased serum transaminase, bilirubin, amylase, and triglyceride levels. Liver biopsy could not be performed. The sixth dose of L-asparaginase was omitted. The NAC was started at a dose of 150 mg/kg for the first hour, 50 mg/kg for a subsequent 4 hours, and 100 mg/kg for the remaining 16 hours. Our patient received NAC infusion at a dose of 100 mg/kg/day over 4 days. The seventh administered dose of L-asparaginase was reduced by 50%. However, on day 2 of therapy, serum ALT was 689 IU/L, AST was 397 IU/L, and ALP was 302 IU/L. NAC therapy was re-started at an oral dose of 10 mg/kg daily and after the sixth day of NAC therapy, serum transaminase levels decreased to

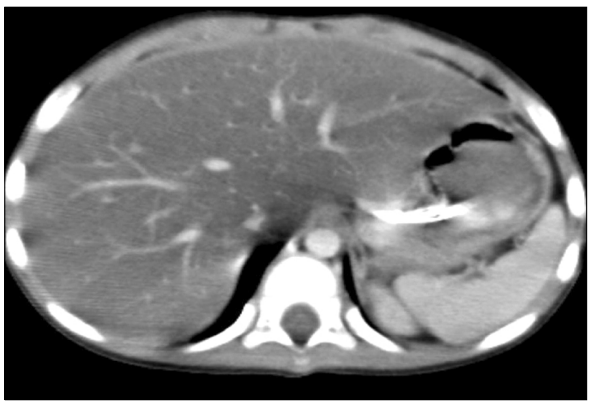

Fig. 1. Abdominal computed tomography showed a hepatomegaly and hepatosteatosis.