Bleeding diathesis in a patient with a novel mutation in SERPINF2: A case study of alpha-2 antiplasmin deficiency

Marcela Torres1, Margaret Drummond-Borg2, Dorothy Adcock3, Elissa Dunlap4

1Department of Hematology/Oncology, Cook Children’s Medical Center, Fort Worth, TX; 2Department of Genetics, Cook Children’s Medical Center, Fort Worth, TX; 3Colorado Coagulation, Laboratory Corporation of America® Holdings, Englewood, CO; 4Texas College of Osteopathic Medicine UNT Health Science Center, Fort Worth, TX

BACKGROUND

Alpha-2 antiplasmin (Alpha2-AP) is a serine protease inhibitor that inactivates plasmin and prevents premature breakdown of fibrin clots. Deficiency of this enzyme can lead to spontaneous bleeding. Patients present with hemophilia-like symptoms, despite normal coagulation and platelet function studies. Less than 50 cases of congenital alpha-2 antiplasmin deficiency have been reported and most involve some form of consanguinity.

OBJECTIVE

To emphasize the importance of recognizing this disorder, to highlight the role of whole exome sequencing to identify pathogenic mutations, and to report a novel SERPINF2 gene mutation as a case of this disorder.

METHODS

To perform whole exome sequencing genomic DNA is fragmented by sonication and ligated to the Illumina multiplexing PE adaptors, then amplified. The DNA is then subjected to sequence analysis on Illumina HiSeq platforms. Data analysis and interpretation is done using Mercury. To assess Alpha2-AP levels diluted plasma is incubated with a precise excess of plasmin, resulting in rapid complex formation between plasmin and functional a 2-antiplasmin in the sample. Plasmin inhibition is directly proportional to antiplasmin in the sample. The residual plasmin hydrolyzes the chromogenic substrate and liberates the pNA chromophore which is read photometrically at 405 nm. Color intensity is inversely proportional to a 2-antiplasmin activity in the sample.

RESULTS

A 7-month-old patient presented with a hemarthrosis of the right knee that was assumed to be septic arthritis, despite negative cultures of the bloody synovial fluid. As she got older, she experienced significant bruising and spontaneous hematomas, necessitating a hematologic consultation. Complete blood count (CBC), Von Willebrand Factor (VWF) activity and antigen, prothrombin time (PT), partial thromboplastin time (PTT), thrombin time, platelet function analysis, and fibrinogen levels were all normal. At 3 years of age, she had a second suspected hemarthrosis of the right knee, prompting the following tests: plasminogen activator inhibitor-1 levels, euglobulin lysis time, factor XIII activity and platelet aggregation studies. All tests showed normal results. In addition, she had two episodes of hemorrhage after a tooth extraction and after losing a tooth, necessitating red blood cell and fresh frozen plasma transfusions. Aminocaproic acid was also used to stop the bleeds. Finally, whole exome sequencing revealed a novel homozygous mutation in the SERPINF2 gene. Alpha-2 antiplasmin activity was then measured at <21%. The parents were also sequenced and found to be heterozygous for the same mutation. The causative mutation, a single amino acid substitution (p.R403H; c.1208G>A), has not been previously reported in patients with alpha-2 antiplasmin deficiency. Family history was positive for consanguinity (patient’s great grandfathers were first cousins). These findings suggest that this mutation has likely been present in several generations of this patient’s family and follows the pattern of an autosomal recessive disorder. This case highlights the difficulties in diagnosing bleeding disorders that involve the fibrinolytic pathway, and it illustrates how whole exome gene mutation analysis can aid in the diagnosis of these rare disorders.

REFERENCES


CONCLUSION

This case highlights the difficulties in diagnosing bleeding disorders that involve the fibrinolytic pathway, and it illustrates how whole exome gene mutation analysis can aid in the diagnosis of these rare disorders. Another aspect of identifying a previously non-reported pathogenic mutation of the SERPINF2 gene is its potential value in providing additional insight into the molecular mechanisms of alpha-2 antiplasmin deficiency.