UC San Diego
UC San Diego Previously Published Works

Title
Novel Factor XIII variant identified through whole-genome sequencing in a child with intracranial hemorrhage

Permalink
https://escholarship.org/uc/item/4x50v203

Journal
Molecular Case Studies, 4(6)

ISSN
2373-2865

Authors
Briggs, Benjamin
James, Kiely N
Chowdhury, Shimul
et al.

Publication Date
2018-12-01

DOI
10.1101/mcs.a003525

Peer reviewed

Novel Factor XIII variant identified through whole-genome sequencing in a child with intracranial hemorrhage

Benjamin Briggs,[1,2] Kiely N. James,[1] Shimul Chowdhury,[1] Courtney Thornburg,[2] Lauge Farnaes,[1] David Dimmock,[1] and Stephen F. Kingsmore[1] on behalf of the RCIGM Investigators[1]

[1]Rady Children's Institute of Genomic Medicine, University of California, San Diego, California 92123, USA; [2]Department of Pediatrics, Division of Hematology and Oncology, University of California, San Diego, California 92123, USA

Abstract Pediatric stroke can be either hemorrhagic or ischemic, with ∼5% of hemorrhagic strokes being caused by genetic coagulopathies. We report an 8 mo old presenting with a hemorrhagic stroke caused by severe Factor XIII deficiency (OMIM # 613225) in whom rapid whole-genome sequencing identified a novel variant in the *F13A1* gene c.1352_1353delAT (p.His451ArgfsTer29).

[Supplemental material is available for this article.]

CASE PRESENTATION

An 8-mo-old male presented to his pediatrician's office for right arm paralysis and was brought to our emergency department by air ambulance. He had fallen <1 ft in height from a push car 1 wk prior. He did not have loss of consciousness but had progressive altered mental status starting the day after the fall. He was born at term by cesarean section and had no bleeding complications at birth, such as cephalohematoma or prolonged bleeding from heel sticks. The parents noted minimal bleeding at the time of umbilical cord detachment, and the patient never underwent circumcision. He had no hematoma formation with his immunizations. There was no family history of bleeding complications or recurrent miscarriage and no known consanguinity. A computed tomography (CT) angiogram of the head with and without contrast revealed a left frontal parietal, intra-axial mass, measuring ∼4.3 × 5 × 4.1 cm, which was concerning for hemorrhagic neoplasm. The patient's activated partial thromboplastin time (aPTT) was 35 sec (reference range of 24–38 sec) and prothrombin time (PT) was 13.5 sec with an international normalized ratio of 1.1 (reference range of 11.4–14.0 sec and 0.9–1.2, respectively). The fibrinogen was 224 mg/dl (reference range of 160–425 mg/dl) and platelet count was 375 × 10^3/µl (reference range of 140–440 × 10^3/µl). Magnetic resonance imaging (MRI) of the brain with and without contrast showed a 5 × 5.4 × 4.6-cm heterogeneous intra-axial left frontal lobe lesion with a fluid level with sheet-like contrast enhancement. The margins of the lesions were well-defined and revealed chronic hemosiderin deposition leading to the suspicion that this was a large intraparenchymal hematoma from a recent cavernoma bleeding event. The patient was taken to the operating room on his fifth hospital day and the lesion was removed. The patient received no hemostatic prophylaxis or treatment prior to surgery and had no postoperative bleeding complications.

Corresponding author:
bbriggs@rchsd.org

Ontology terms: intracranial hemorrhage; stroke-like episodes

Histopathology revealed the lesion to be hematoma without evidence of vascular malformation. Given the diagnostic challenge, a cerebral cavernous malformation (CCM) genetic panel (analysis of *CCM2*, *KRIT1*, and *PDCD10*) was sent, in addition to rapid, trio, whole-genome sequencing (WGS) as part of an IRB-approved clinical trial (clinicaltrials.gov # NCT02917460).

TECHNICAL ANALYSIS AND METHODS

Blood was drawn following consent for trio rapid WGS. DNA was subsequently extracted using EZ1 DSP DNA Blood Kit and sequenced on a NovaSeq 6000 (Illumina). Rapid alignment and nucleotide variant calling was performed using the Dragen (Edico Genome) hardware and software (Miller et al. 2015). Yield was 153.9 Gb, 136.7 Gb, and 162.9 Gb for the proband, mother, and father, respectively. This resulted in 4,694,894, 4,717,798, and 4,715,319 distinct variant calls, respectively (Supplemental Data 1). Variants were annotated and analyzed in Opal (Omicia) (Coonrod et al. 2013). Initially, variants were filtered to retain those with allele frequencies of <1% in the Exome Variant Server, 1000 Genomes Samples, and Exome Aggregation Consortium database (Karczewski et al. 2016; http://evs.gs.washington.edu/EVS/2016). A gene panel was built in Phenolyzer (Yang et al. 2015) using Human Phenotype Ontology (HPO) (Köhler et al. 2017). See Table 1 for phenotype features. This panel included 367 genes related to the following HPO terms: Cavernous hemangioma (HP: 0001048) and Hemiparesis (HP: 0001269). This method identified homozygosity for *F13A1* c.1352_1353delAT (p.His451ArgfsTer29) (see Table 2), which was classified as likely pathogenic based on the guidelines established by the American College of Medical Genetics and Genomics and the Association for Molecular Pathology (Richards et al. 2015).

VARIANT INTERPRETATION

Our patient had a novel variant c.1352_1353delAT (p.His451ArgfsTer29) in the *F13A1* gene detected in a homozygous state that encodes the A subunit of Factor XIII (FXIII). The majority of patients with FXIII deficiency have mutations in the A subunit with mutation in the B subunit comprising <5% of cases (Ivaškevičius et al. 2007). The variant was confirmed by Sanger

Table 1. Phenotypic features

Factor XIII deficiency	Proband (II-1)	Relevance/alternate explanation
Epistaxis	No	
Gum bleeding	No	
Hemarthrosis	No	
Umbilical bleeding after birth	Yes	Based on parental report, did not require medical intervention
Ecchymosis	No	
Hematomas	No	
Subcutaneous bleeding	No	
Intracranial bleeding	Yes	
Bleeding tendency	No	Did not have bleeding with neurosurgical intervention
Deficiency of Factor XIII	Yes	Factor XIII assay revealed activity of <4%
Impaired wound healing	No	Craniotomy incision healed without complication
Miscarriage	N/A	

The list of clinical features is based on the OMIM clinical synopsis related to *F13A1* gene (#613225; Factor XIII subunit A deficiency).

Table 2. Genomic findings

Gene	Genomic location	HGVS cDNA	HGVS protein	Zygosity	Parent of origin	Variant interpretation
F13A1	Chr 6: 6182326	ENST00000264870: c.1352_1353delAT	p.His451ArgfsTer29	Homozygous	Maternal and paternal	Likely pathogenic

sequencing and analysis of the parents confirmed their carrier status. This frameshifting variant in exon 11 of 15 introduces a premature stop codon and is therefore predicted to result in loss of normal protein function. This variant has not been previously reported or functionally characterized in the literature, but multiple frameshifting variants downstream from this variant have been reported in the Human Gene Mutation Database (Ivaškevičius et al. 2017). It is absent from ExAC and gnomAD population databases. Based on these criteria, the variant was classified as likely pathogenic (Supplemental Data 2). The variant was found within an 11-Mb region of homozygosity. An elevated homozygous/heterozygous variant ratio and multiple regions of homozygosity of >5 Mb, which are typical of individuals whose parents have moderate shared ancestry, were found in this individual (Supplemental Data 1). This matched the patient's demographics, as both of his parents were from the same small rural community in Mexico. This family history and its genomic signature predispose individuals for a heightened risk of autosomal recessive conditions. Of note, the patient's CCM genetic panel also returned negative.

Hematology was immediately consulted and urgent outpatient evaluation was scheduled. A clot lysis screen was abnormal with the clot dissolving in urea but stable in saline. A FXIII assay revealed an activity of <4% confirming the diagnosis of severe FXIII deficiency. The patient was then started on prophylaxis with coagulation FXIII A-subunit (Recombinant) (Tretten, Novo Nordisk) 35 international units per kilogram of body weight every 4 wk and has had no further bleeding complications. FXIII levels were not measured in the parents, given their lack of bleeding history and their heterozygous state. FXIII levels were recommended for two older brothers with genetic testing at the time of family planning to determine if they were carriers of the FXIII variant.

Pediatric stroke is an acute neurologic deficit persisting for at least 24 h in patients 28 d to 18 yr resulting from a disturbance in cerebral blood flow (Gumer et al. 2014). Stroke is typically defined as either ischemic or hemorrhagic, with hemorrhagic stroke accounting for ~39%–54% of pediatric strokes and intracerebral hemorrhage (ICH) accounting for ~23% (Lo 2011). Vascular malformations, followed by brain tumors, are the most common underlying cause for hemorrhagic stroke and in ~5% of the described cases a genetic coagulopathy is found to be the cause, but unfortunately the studies did not further clarify the type of coagulopathy (Lo 2011). FXIII deficiency is a rare autosomal recessive bleeding disorder in which 17% of affected individuals present with ICH as their sentinel bleed (Naderi et al. 2014a). In a study of patients with central nervous system bleeding (CNSB) and rare bleeding disorders 7 out of 15 patients with FXIII deficiency had CNSB as their primary bleeding episode (Siboni et al. 2012). FXIII deficiency is the congenital bleeding disorder that is most commonly complicated by ICH, with around one-third of patients experiencing this complication (Dorgalaleh et al. 2016). CNSB can occur spontaneously or after mild trauma bleed (Naderi et al. 2014b), with spontaneous being more common (Siboni et al. 2012). More than 90% of ICH in congenital FXIII deficiency are intraparenchymal, and about one-third of patients experienced them before the age of two (Dorgalaleh et al. 2016). ICH can also occur in patients with severe hemophilia A and B, FVII deficiency, FX deficiency, FV deficiency, FII deficiency, and afibrinogenemia (Tabibian et al. 2018). In addition to ICH, FXIII

deficiency is also associated with umbilical cord bleeding, soft tissue bruising, mucosal bleeding, poor wound healing, and recurrent miscarriage (Naderi et al. 2014a).

Our patient presented with ICH as his sentinel bleed. The cause of the bleed was only identified after WGS identified a novel variant in the *F13A1* and functional testing confirmed the diagnosis of severe FXIII deficiency, which is a very unusual way for FXIII deficiency to be identified. A likely reason for the initial diagnostic confusion was the appearance of the bleed on CT and MRI, which was likely a result of our patient rebleeding into a site of an old hemorrhage, which may not be unexpected as one-third of patients with FXIII deficiency and CNSB have had a recurrence (Siboni et al. 2012).

SUMMARY

We report an 8 mo old presenting with a hemorrhagic stroke found to be caused by severe FXIII deficiency after rapid WGS identified a novel variant in the *F13A1* gene c.1352_1353delAT (p.His451ArgfsTer29). This case highlights the importance of having a broad differential diagnosis for ICH. Although only 5% of hemorrhagic stroke is secondary to a bleeding disorder (Lo 2011), all children presenting with ICH should have laboratory evaluation and consideration of hematology consultation. Additionally, if an inherited bleeding disorder is suspected in the setting of normal aPTT, PT, fibrinogen, and platelet count, then FXIII testing should be strongly considered. It is also important to recognize how critical WGS was in achieving the correct diagnosis for our patient, allowing for the timely factor replacement and mitigation of further bleeding complications. In particular, identification of the FXIII subunit A variant allowed for initiation of subunit A-specific therapy with recombinant FXIII subunit A. Without this information, a plasma-derived FXIII that contains both subunits must be used, which has a potential infection risk from human blood.

ADDITIONAL INFORMATION

Data Deposition and Access

The ClinVar (https://www.ncbi.nlm.nih.gov/clinvar/) accession number is SCV000804842.

Ethics Statement

Author Contributions

REFERENCES

Coonrod EM, Margraf RL, Russel A, Voelkerding KV, Reese MG. 2013. Clinical analysis of genome next-generation sequencing data using the Omicia platform. *Expert Rev Mol Diagn* **13:** 529–540.

Dorgalaleh A, Naderi M, Shamsizadeh M. 2016. Morbidity and mortality in a large number of Iranian patients with severe congenital factor XIII deficiency. *Ann Hematol* **95:** 451–455.

Gumer LB, Del Vecchio M, Aronoff S. 2014. Strokes in children: a systematic review. *Pediatr Emerg Care* **30:** 660–664.

Ivaškevičius V, Seitz R, Kohler HP, Schroeder V, Muszbek L, Ariens RA, Seifried E, Oldenburg J; Study Group. 2007. International registry on factor XIII deficiency: a basis formed mostly on European data. *Thromb Haemost* **97:** 914–921.

Ivaškevičius V, Biswas A, Garly ML, Oldenburg J. 2017. Comparison of *F13A1* gene mutations in 73 patients treated with recombinant FXIII-A$_2$. *Haemophilia* **23:** e194–e203.

Karczewski KJ, Weisburd B, Thomas B, Solomonson M, Ruderfer DM, Kavanagh D, Hamamsy T, Lek M, Samocha KE, Cummings BB, et al. 2016. The ExAC browser: display reference data information from over 60 000 exomes. *Nucleic Acids Res* **45:** D840–D845.

Köhler S, Vasilevsky NA, Engelstad M, Foster E, McMurry J, Aymé S, Baynam G, Bello SM, Boerkoel CF, Boycott KM, et al. 2017. The Human Phenotype Ontology in 2017. *Nucleic Acids Res* **45:** D865–D876.

Lo WD. 2011. Childhood hemorrhagic stroke: an important but understudied problem. *J Child Neurol* **26:** 1174–1185.

Miller NA, Farrow EG, Gibson M, Willig LK, Twist G, Yoo B, Marrs T, Corder S, Krivohlavek L, Walter A, et al. 2015. A 26-hour system of highly sensitive whole genome sequencing for emergency management of genetic diseases. *Genome Med* **7:** 100.

Naderi M, Dorgalaleh A, Alizadeh S, Tabibian S, Hosseini S, Shamsizadeh M, Bamedi T. 2014a. Clinical manifestations and management of life-threatening bleeding in the largest group of patients with severe factor XIII deficiency. *Int J Hematol* **100:** 443–449.

Naderi M, Zarei T, Haghpanah S, Eshghi P, Miri-Moghaddam E, Karimi M. 2014b. Intracranial hemorrhage pattern in the patients with factor XIII deficiency. *Ann Hematol* **93:** 693–697.

Richards S, Aziz N, Bale S, Bick D, Das S, Gastier-Foster J, Grody WW, Hegde M, Lyon E, Spector E, et al. 2015. Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology. *Genet Med* **17:** 405–424.

Siboni SM, Zanon E, Sottilotta G, Consonni D, Castaman G, Mikovic D, Biondo F, Tagliaferri A, Iorio A, Mannucci PM, et al. 2012. Central nervous system bleeding in patients with rare bleeding disorders. *Haemophilia* **18:** 34–38.

Tabibian S, Motlagh H, Naderi M, Dorgalaleh A. 2018. Intracranial hemorrhage in congenital bleeding disorders. *Blood Coagul Fibrinolysis* **29:** 1–11.

Yang H, Robinson PN, Wang K. 2015. Phenolyzer: phenotype-based prioritization of candidate genes for human diseases. *Nat Methods* **12:** 841–843.