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CASE REPORTS

# Prenatal Diagnosis for a Novel Missense Mutation in X-Linked Intellectual Disability Gene Followed by Favorable Pregnancy Outcome

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**Abstract** Intellectual disability (ID) is still unexplained in 60% of cases and prenatal diagnosis is very challenging for this condition. A second gravida presented to us at 6 weeks of gestation for counseling. Her previous child had been diagnosed with ID and autism. Detailed family history showed that her brother also had ID. Screening investigations were normal for the affected child. Exome sequencing report revealed variation of unknown significance (VOUS) on *SIN3A* gene and *UPF3B* gene. The variation in X-linked *UPF3B* gene was reclassified as novel pathogenic variation after segregation studies with parents and affected maternal uncle for both the genes variations. An amniocentesis was done at 18 weeks gestation for the novel mutation in the *UPF3B* gene and the fetus was found unaffected. The patient delivered a healthy male child who is doing well at two years of age. To conclude, we should not disregard VOUS on exome sequencing. Identification of VOUS requires careful genotype-phenotype correlation and segregation studies to counsel parents regarding the risk of having another affected child.

**Keywords** Intellectual disability · X-linked · Autism · *UPF3B* gene · Variation

## Background

Autism Spectrum Disorder (ASD) and Intellectual Disability (ID) are serious, coexisting, medical and social problems with a strong genetic components and heterogeneous etiology. Around 45–50% cases of ASD are eventually diagnosed with intellectual disability eventually [1]. X linked Intellectual disability accounts for 10% of cases of ID [2].

The discovery of the Fragile X locus in 1991has paved the way for molecular studies to detect the cause of X-linked intellectual disability (XLID) [3]. The advent of Next Generation Sequencing (NGS) has accelerated the discovery of more and more XLID genes. More than 141 XLID genes have been identified to date [4]. A large chunk of these genes causes non-syndromic XLID (NS-XLID) with ID as an isolated feature.

*UPF3B* gene on X chromosome encodes the regulator of nonsense transcripts 3B *(REN3B)* protein initially identified as a component of an exon–junction complex that promotes nonsense-mediated mRNA decay (NMD). Some authors have studied the functions of this gene and their implication in ID [5]. Tarpey et al. described the hemizygous variants in the *UPF3B* gene in affected males of 4 unrelated families for the first time [6].

Since then, many patients with ID have been studied by NGS and more variants have been reported in the UPF3B gene. according to the Human Gene Mutation Database (HGMD)1, 21 variants (17 pathogenic) have been identified in UPF3B to date. Six other pathogenic variants have been identified in ClinVar2, four of which overlap with the HGMD.

The aim of this report is to highlight the importance of genotype–phenotype correlation and segregation studies for a variation of uncertain significance on exome sequencing so that we can prevent the birth of another child with the same genetic disorder.

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## Case Report

Four generation pedigree of this Indian family is shown in Fig. 1.

A nonconsanguineous couple approached us at six weeks of pregnancy. Their previous male child had Intellectual disability and autistic features. He had an uneventful birth history with no neonatal complications. Development delay were noted since early infancy in all domains i.e. gross motor, fine motor, cognition and speech. Speech delay was profound. The child also had autistic features like lack of social reciprocation, poor interaction and self-isolation. At the time of genetic evaluation, he was 9.5 years old and was able to run, walk, hop and feed himself. Hearing and vision were normal. However, he did not follow commands, had no eye to eye contact and was able to speak only three word sentences. He was able to indicate bladder and bowel control and was attending a special school. On detailed family history, it was revealed that the proband's maternal uncle had almost similar features. The concerned uncle is 45 years old and unmarried and can do his routine activities with some help. He had never been genetically evaluated. On examination, the child had a long thin face, poor musculature, high nasal bridge, horizontal nystagmus and prognathia. There was no significant neurological or systemic involvement. Investigations including complete blood counts, congenital infection screening, detailed eye evaluation, brain stem evoked potential, magnetic resonance imaging brain, Tandem Mass spectrometry (TMS), Urine Gas Chromatography mass spectrometry were negative for any pathology. The intelligence quotient of the child was 55 (Borderline ID). Fragile X screen and microarray studies were normal. Whole exome sequencing revealed missense variation of unknown significance in two genes. There was a hemizygous variation, c.1306C > T in exon 11 of *UPF3B* gene and a heterozygous variation, c. 3602G > A in *SIN3A* gene. On genotype-phenotype correlation, other variations in the above mentioned genes are associated with craniofacial dysmorphism, intellectual disability and autism. *SIN3A* gene deletion has been reported with Witteveen-Kolk syndrome and 88.9% of missense changes are reported as benign as per the genomic database. This is more than the threshold of 51.0%.

In silico prediction tools, results were damaging for *UPF3B* and *SIN3A* variation. We carried out a segregation analysis of both parents and maternal uncle by Sanger sequencing. The maternal uncle was a hemizygous carrier of the same variation in *UPF3B* gene; the *SIN3A* variation was absent. The mother was an asymptomatic carrier of both gene variations and neither of the two variations was found in the father (Fig. 2). This variant was reclassified as a Class 2 variant (likely pathogenic), as per the guidelines laid down for interpretation of sequence variant by S Richards et al.in 2015. We concluded that the UPF3B gene was implicated for similar features in the child and the maternal uncle and counseled the parents for prenatal testing on this novel variant.

Amniocentesis was done at 18 weeks gestation for *UPF3B* variation only and the fetus did not harbor this novel mutation. The pregnancy continued with close surveillance and a 2.5 kg male baby was delivered at 35 weeks due to preterm labor pains. The baby is two years old and doing well.

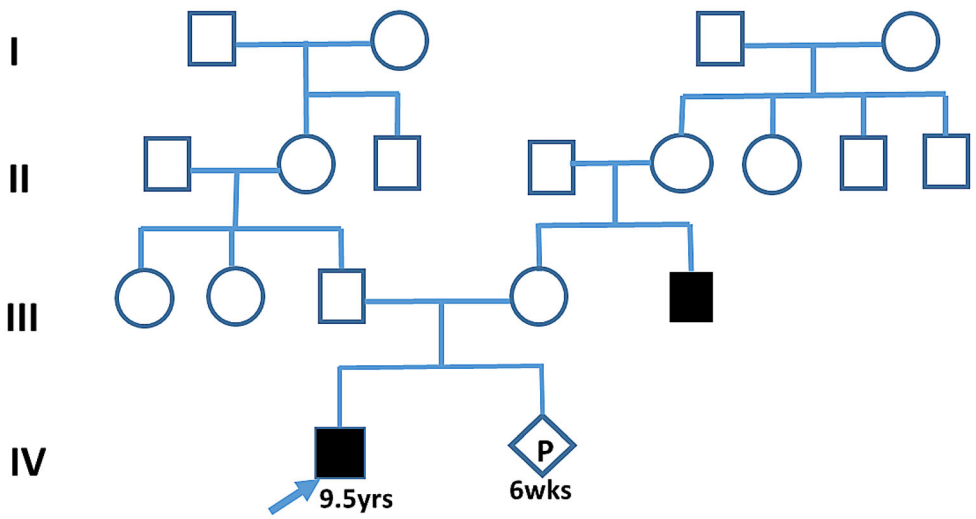


**Fig. 1** Four generation pedigree of the family. *Note* Shapes filled with black colour-Affected family members

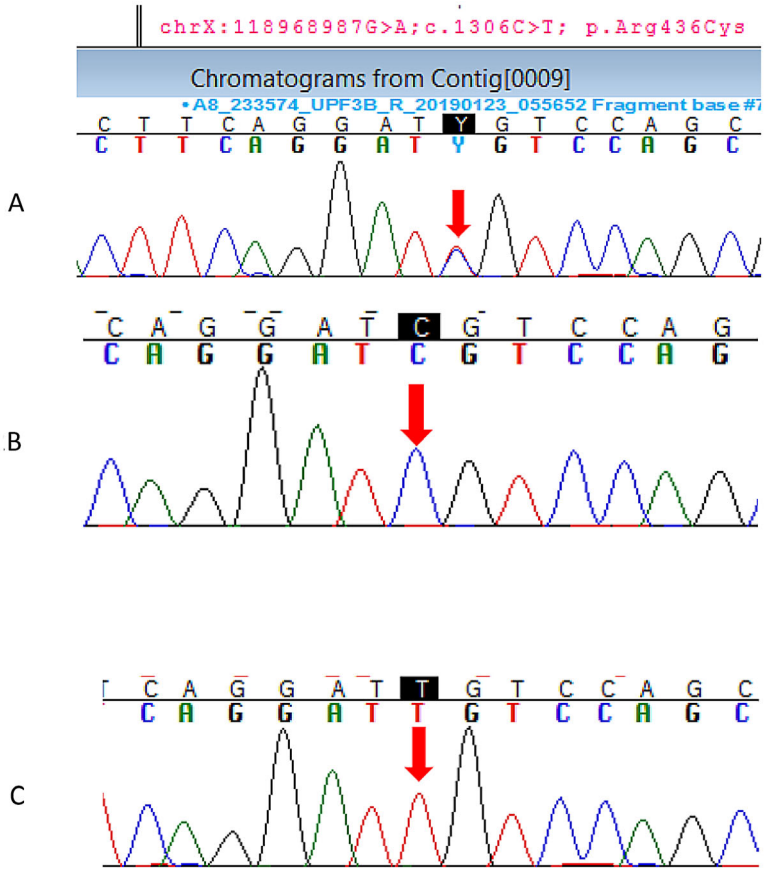


**Fig. 2** Fragment analysis of all family members. **a** The mother is a heterozygous carrier of c.1306C > T in *UPF3B* gene. **b** Proband's father not a carrier of c.1306C > T in *UPF3B* gene. **c** Proband's maternal uncle a carrier hemizygous c.1306C > T in *UPF3B* gene

## Discussion

To the best of our knowledge, this is the first case report where we present prenatal diagnosis of a novel missense variant c.1306C > T; p. (Arg436Cys) in the *UPF3B* gene. The sorting out of the definitive diagnosis out of inconclusive variations in two different genes appeared a dilemma for us especially when the family was very keen for prenatal diagnosis. *UPF3B* gene (OMIM 300,298) is located in Xq24 and encodes a protein involved in nonsense-mediated decay (NMD) of mRNA.

As per the recently curated database,71.4% of missense variants are reported as benign which is more than the threshold of 51.0% However, in our case, the variant was also present in the other living affected male and in one carrier female, and it was absent from the available unaffected male. Therefore, it completely co-segregated with the disease within the family and was reclassified as likely pathogenic (Class 2) as per ACMG classification of variants [7].

A study by Tahani Alrahbeni et al. also supported our diagnosis in which they concluded that *UPF3B* proteins with missense mutations found in patients with autism, schizophrenia and XLID are functionally impaired in NMD [8]. Large families with X-linked nonsyndromic intellectual disability have been detected with missense mutations by other studies [9, 10]. These mutations lead to the introduction of a premature termination codon and subsequent NMD of mutant *UPF3B* mRNA. *UPF3B* is widely expressed in neurons and also presents in dendritic spines, which are essential structures for proper neurotransmission and thus learning and memory processes [11].

## Implications for Clinical Practice

Meticulous analysis of the uncertain gene variations can prove helpful in preventing recurrence of the same genetic condition in future children especially when family approaches during an ongoing pregnancy.

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**Declarations**

**Conflict of interest** None.

## References


1. Tejada MI, Villate O, Ibarluzea N, de la Hoz AB, Martínez-Bouzas C, Beristain E, et al. Molecular and clinical characterization of a novel nonsense variant in exon 1 of the UPF3B gene found in a large spanish basque family (MRX82). Front Genet. 2019. https://doi.org/10.3389/fgene.2019.01074.
2. Tzschach A, Grasshoff U, Beck-Woedl S, Dufke C, Bauer C, Kehrer M, et al. Next-generation sequencing in X-linked intellectual disability. Eur J Hum Genet. 2015;23:1513–8. https://doi.org/10.1038/ejhg.2015.5.
3. Allen RC, Zoghbi HY, Moseley AB, Rosenblatt HM, Belmont JW. Methylation of HpaII and HhaI sites near the polymorphic CAG repeat in the human androgen-receptor gene correlates with X chromosome inactivation. Am J Hum Genet. 1992;51:1229–39.
4. Neri G, Schwartz CE, Lubs HA, Stevenson RE. X-linked intellectual disability update 2017. Am J Med Genet Part A. 2018;176:1375–88. https://doi.org/10.1002/ajmg.a.38710.
5. Kunz JB, Neu-Yilik G, Hentze MW, Kulozik AE, Gehring NH. Functions of *hUpf3a* and *hUpf3b* in nonsense-mediated mRNA decay and translation. RNA. 2006;12:1015–22. https://doi.org/10.1261/rna.12506.
6. Tarpey PS, Raymond FL, Nguyen LS, Rodriguez J, Hackett A, Vandeleur L, et al. Mutations in *UPF3B*, a member of the nonsense-mediated mRNA decay complex, cause syndromic and nonsyndromic mental retardation. Nat Genet. 2007;39:1127–33. https://doi.org/10.1038/ng2100.
7. Richards S, Aziz N, Bale S, Bick D, Das S, Gastier-Foster J, Grody WW, Hegde M, Lyon E, Spector E, Voelkerding K, Rehm HL. ACMG Laboratory Quality Assurance Committee. 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. 2015;17(5):405–24. https://doi.org/10.1038/gim.2015.30.
8. Alrahbeni T, Sartor F, Anderson J, Miedzybrodzka Z, McCaig C, Müller B. Full *UPF3B* function is critical for neuronal differentiation of neural stem cells. Mol Brain. 2015;8:33. https://doi.org/10.1186/s13041-015-0122-1.
9. Tarpey PS, Raymond FL, Nguyen LS, Rodriguez J, Hackett A, Vandeleur L, et al. Mutations in *UPF3B*, a member of the nonsense-mediated mRNA decay complex, cause syndromic and nonsyndromic mental retardation. Nat Genet. 2007;39(9):1127–33.
10. Xu X, Zhang L, Tong P, Xun G, Su W, Xiong Z, Zhu T, Zheng Y, Luo S, Pan Y, Xia K, Hu Z. Exome sequencing identifies UPF3B as the causative gene for a Chinese non-syndrome mental retardation pedigree. Clin Genet. 2013;83(6):560–4. https://doi.org/10.1111/cge.12014.
11. Laumonnier F, Shoubridge C, Antar C, Nguyen LS, Van Esch H, Kleefstra T, Briault S, Fryns JP, Hamel B, Chelly J, Ropers HH, Ronce N, Blesson S, Moraine C, Gécz J, Raynaud M. Mutations of the UPF3B gene, which encodes a protein widely expressed in neurons, are associated with nonspecific mental retardation with or without autism. Mol Psychiatry. 2010;15(7):767–76. https://doi.org/10.1038/mp.2009.14.