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*Published in:*
Clinical Genetics

*DOI:*
10.1111/cge.14083

*Publication date:*
2022

*Document version:*
Accepted manuscript

*Citation for pulished version (APA):*
Terkelsen, T., Brasch-Andersen, C., Illum, N., Busa, T., Missirian, C., Chandler, K., Holden, S. T., Jensen, U. B., & Fagerberg, C. R. (2022). Mono-allelic loss of YTHDF3 and neurodevelopmental disorder: clinical features of four individuals with 8q12.3 deletions. *Clinical Genetics*, *101*(2), 208-213. https://doi.org/10.1111/cge.14083

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**ORIGINAL ARTICLE**

# Mono-allelic loss of *YTHDF3* and neurodevelopmental disorder: clinical features of four individuals with 8q12.3 deletions

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**Conflicts of interest:** The authors declare no conflicts of interest.

Running head: YTHDF3 deletions

Tables/Illustrations: 2 (max 7)

This is the author manuscript accepted for publication and has undergone full peer review but has not been through the copyediting, typesetting, pagination and proofreading process, which may lead to differences between this version and the Version of Record. Please cite this article as doi: 10.1111/cge.14083

Word count (incl. title page, abstract, main text, legends, and references): 3,864 (max 6,000)

**ABSTRACT** (153 words; max 200)

The YTH domain family member 3 gene (*YTHDF3*) encodes a reader of the abundant N6-methyladenosine ($m^6A$) modification of eukaryotic mRNA, which plays an essential role in regulating mRNA stability and is necessary to achieve normal development of the central nervous system in animal models. *YTHDF3* has not previously been implicated in Mendelian disease despite a high probability of loss of function intolerance and statistical evidence of enrichment for gene-disruptive de novo variants in large-scale studies of individuals with intellectual disability and/or developmental delay. We report four individuals with deletion of 8q12.3, deletion size 1.38 – 2.60 Mb, encompassing YTHDF3, three of them were de novo, and in one case, the inheritance was unknown. Common features of the individuals (age range, 4 to 22 years) were developmental delay and/or intellectual disability. Two individuals underwent squint surgery. We suggest that haploinsufficiency of *YTHDF3* causes a neurodevelopmental disorder with developmental delay and intellectual disability of variable degree.

**Key Words**: Heterozygote; Chromosome deletion; YTHDF3 protein, human; Developmental delay; Intellectual disability

## INTRODUCTION

$N^6$-methyladenosine ($m^6A$) is an internal modification of eukaryotic mRNA [1], which plays an essential role in mRNA stability and regulation of gene expression [2]. This mechanism involves the interplay of so-called writers, erasers and readers of $m^6A$ [3]. Identified as readers of $m^6A$, the genes *YTHDF1* (MIM 616529), *YTHDF2* (MIM 610640), and *YTHDF3* (MIM 618669) encode three highly identical proteins that contain an $m^6A$-binding YTH domain and a prion-like low complexity domain [3]. The individual roles of the three proteins continue to be investigated. In some models, *YTHDF1* was proposed to facilitate the translation of $m^6A$-modfied mRNA [4], *YTHDF2* to accelerate the decay of $m^6A$-modified mRNA [5], and *YTHDF3* to recognize and buffer $m^6A$-modified mRNA in the cytosol [6 7]. More recently, other investigators have concluded that all three proteins act in the same way in promoting the decay of $m^6A$-modified mRNA [8 9].

A growing body of evidence implies an essential role for $m^6A$ during the normal development of the central nervous system [10], and YTH domain family members have been implicated in fertility, cancer development and immune responses [9]. At present it is uncertain if variants in the genes *YTHDF1*, *YTHDF2*, and *YTHDF3* have clinical implications. We report the phenotypes of four individuals heterozygous for deletions of 8q12.3 encompassing the *YTHDF3* gene.

## METHODS

Four individuals heterozygous for overlapping deletions of 8q12.3 including the *YTHDF3* gene were identified via the Database of Chromosomal Imbalance and Phenotype in Humans Using Ensembl Resources (DECIPHER)[11]. The deletions included *YTHDF3* and no other genes with a pLI score ≥ 0.90 (pLI = probability of Loss of function Intolerance), evaluated by search in gnomAD [12]. Larger deletions in DECIPHER that involved other genes with pLI scores > 0.9 were not included in this study. The deletions were identified by chromosomal microarray (CMA) in all individuals. The deletions (hg38) and protein-coding genes (GENCODE v32) were visualized in the UCSC Genome Browser (https://genome.ucsc.edu) [13]. Z-scores for growth measurements were

obtained from the WHO (for 0 to 2 years) and CDC charts (for 2 to 20 years) using PediTools (https://peditools.org) [14]. Z-scores for head circumference of older children were obtained from US reference data [15].

Written informed consent for publication was provided for all individuals. The study was registered with the ethics committee of the Region of Southern Denmark (S-20202000-88).

## RESULTS

We review the clinical and molecular genetic features of the four individuals heterozygous for deletions in 8q12.3 (Table 1).

### Clinical features

Individual P1 is a 6-year-old female with mild global developmental delay (DECIPHER #383076). She was born following a 38-week uncomplicated pregnancy. There were full Apgar scores. Birth weight was 3,150 g (-0.2 SD), length 50 cm (+0.5 SD), and head circumference 32 cm (-1.6 SD). Apart from a congenital umbilical hernia, for which she underwent surgery at age 20 months, she had no malformations or dysmorphic features. She had a tendency for childhood infections and serous otitis media. At the age of 3 years, she was referred to the pediatric department due to delayed development. She could sit independently at 8 months and crawled at 9-10 months, yet stopped again until 14 months, and walked independently at 24 months. A speech delay was evident. She spoke her first words after 24 months and spoke in sentences from around 4 years of age, although her impressive language was better according to the parents.

At the age of 6 years, weight was 23.6 kg (-0.4 SD), height 125 cm (-0.2 SD) and head circumference 50 cm (-0.7 SD). Neuropsychological evaluation showed mild intellectual disability based on the Wechsler Preschool and Primary Scale of Intelligence-Fourth Edition (WPPSI-IV) test: full scale intelligence quotient (IQ) = 63 (1$^{st}$ percentile), verbal comprehension score = 57 (0.2$^{nd}$ percentile), visuospatial score = 58 (0.3$^{rd}$ percentile), fluid reasoning score = 62 (1$^{st}$

percentile), working memory score = 96 (39th percentile), and processing speed score = 69 (2nd percentile).

Individual P2 is a 19-year-old male referred at age two years to the genetics department due to behavior anomalies (DECIPHER #259686). He was born following a 40-week uncomplicated pregnancy. Birth weight was 3900 g (+1.1 SD), length 51 cm (+0.6 SD), and head circumference 34 cm (-0.4 SD). His psychomotor development was normal (sitting 9 months, walking 15 months, no speech delay). He had scholarly difficulties and mild intellectual disability. He was able to read and write. He has no congenital malformations and no medical conditions. A cerebral MRI scan was normal. At the age of 14 years, weight was 56.5 kg (0.5 SD), height 167 cm (0.4 SD) and head circumference 56.5 cm (+0.7 to +1.3 SD). During adolescence, a diagnosis of pervasive developmental disorder was made.

Individual P3 is a 22-year-old male diagnosed with severe intellectual disability, autism and epilepsy (DECIPHER #277756). He was born following a pregnancy complicated by maternal substance abuse and was on the Special Care Baby Unit for 2 weeks with feeding difficulties and symptoms in keeping with neonatal abstinence. Birth weight was 3570g (+0.5 SD), whereas length and head circumference measurements were unavailable. He had no major dysmorphic features but was noted to have flat feet, a wide sandal gap and slender fingers. He was removed into the care of his grandmother at age 18 months. He had marked developmental delay: he did not walk until age 4 years and had very delayed speech and language with first words aged 3 years and understandable conversation from age 8 years. He started with febrile and afebrile tonic-clonic seizures at the age of 18 months. A cerebral MRI scan was normal. EEG showed spike and wave activity in the right centro-temporal region. He was started on anti-epileptic medication but it was difficult to get him to take it on a regular basis so it was discontinued. His seizure frequency decreased over time and are now mainly when he is unwell. He is partially sighted following on from an operation to correct a squint. He wears insoles for flat feet. He has a diagnosis of autism

and has a number of fixations and obsessions, as well as repetitive mannerisms. When last examined aged 15.5 years, he was overweight (+2.7 SD) and of tall stature (+1.3 to +2.1 SD). He eats a very limited diet. He cannot read or write and is unable to go out independently. He had a full time carer as he needs help with dressing and prompting to wash. He needs his food prepared for him. He enjoys fishing and goes with his carers three times a week. There is no ongoing contact with his birth parents but neither had any history of learning difficulties or epilepsy and his half-brother attended University.

Individual P4 is a 12-year-old male known with global development delay (DECIPHER #266517). He had surgery for bilateral strabismus at the age of 1.5 years, has no dysmorphic features and has normal general health. His early motor and language development were reported to be normal but at the age of 1.5 years his speech became repetitive and he lost the ability to use expressive language. Newborn measurements were unavailable. At age 4.5 years his growth parameters were within the normal range (height -0.7 SD, weight -0.7 to 0 SD, head circumference -1.3 SD). He initially attended a mainstream school but transferred to a school for children with complex needs. At age 12 years his growth parameters remain in the normal range (height and weight both -0.7 to 0 SD). He now has an extensive vocabulary but struggles to construct sentences and also with using tense. He is hypermobile and has dyspraxia. His weak core muscle and hand strength contribute to poor balance and coordination. He can write his name and simple sentences but uses an immature tripod grip which affects his writing ability. He is not able to use scissors. He is popular in class, imaginative and considerate and interacts with his friends and peers. While he can participate appropriately in group activities, he displays avoidant type behavior and prefers independent play. His motivation is inconsistent and has been noted to impact on his educational activities in class.

**Molecular genetic features**

The individuals were heterozygous for deletions in 8q12.3 that ranged in size from 1.38 Mb to 2.60 Mb (Figure 1).

Three deletions occurred *de novo*, whereas the parents of P3 were unavailable for testing. The deletion in P1 included no other genes known to be protein-coding than *YTHDF3*. The deletions in P2, P3 and P4 encompassed up to five other protein-coding genes than *YTHDF3*, none of which were predicted to exhibit haploinsufficiency, the pLI-scores being < 0.9. Only *CYP7B1* (MIM 603711; associated with hereditary spastic paraplegia; autosomal recessive inheritance) and *TTPA* (MIM 600415; associated with ataxia with isolated vitamin E deficiency; autosomal recessive inheritance) were known Mendelian disease-associated genes. These hereditary disorders were not considered relevant for the phenotype.

The CMA of P4 also identified two additional copy number variants (hg38) of uncertain significance. He was thus heterozygous for a *de novo* deletion of 741 kb in 8q13.1 (8:65,591,973-66,332,718) spanning eight genes, not known to be Mendelian disease-associated. But moreover, he was heterozygous for a *de novo* deletion of 2.59 Mb in 9q21.3 (9:78,571,738-81,153,868) that included a single protein-coding gene, *TLE4*, not known to be Mendelian disease associated, yet predicted to exhibit haploinsufficiency with a pLI-score of 1.0.

## DISCUSSION

We aimed to investigate the clinical impact of *YTHDF3* haploinsufficiency through the identification of individuals heterozygous for deletions of 8q12.3 encompassing *YTHDF3.* In three of the four individuals, the deletion occurred de novo, while inheritance was unknown in one. One individual (P4) also had two other de novo deletions making the impact of each deletion difficult to predict. The four individuals presented with developmental delay and/or intellectual disability, and absence of major dysmorphic features. While individuals P1 and P2 had rather non-specific mild intellectual disability, individual P3 was more severely affected with severe intellectual disability, epilepsy, and autism. The deletion in P1 encompassed the *YTHDF3* and no other genes; thus, the smallest deletion of overlap only encompasses this gene. *YTHDF3* is predicted to be intolerant for loss of

function (pLI-score 1.0), which makes haploinsufficiency of this gene a likely causative factor for the phenotype in the reported individuals. Previously, *YTHDF3* was identified as being significantly enriched for de novo likely gene-disruptive mutations among individuals with intellectual disability and/or developmental delay in large-scale studies [16]. The present study, however, gives the first detailed phenotypes of individuals with deletion of *YTHDF3*. Three out of four (75%) individuals suffered from a rather nonspecific phenotype of mild intellectual disability and/or developmental delay, whereas just one of the individuals had severe intellectual disability, epilepsy, and autism. As the phenotype of individual P3 might be influenced by other genetic or environmental factors, it is currently uncertain if haploinsufficiency of YTHDF3 per se is associated with non-specific mild intellectual disability or if it can also be associated with a more severe phenotype. However, given no management indications, whole exome sequencing was not initiated. It is also uncertain if deletions in 8q12.3 could be a risk factor for strabismus, which was observed in individuals P3 and P4, since squint is a frequent phenotype occurring in 5% of children [17]. The phenotyping of more individuals with loss of *YTHDF3* are needed to confirm the associated features.

It remains to be understood how the haploinsufficiency of *YTHDF3* can cause developmental delay and intellectual disability. There are other examples of neurodevelopmental disorders linked to genes involved in mRNA metabolism and transport, such as FMRP translational regulator 1 (*FMR1*; MIM 309550), associated with the fragile X syndrome and undergoing study as another reader of m$^6$A [18], UPF3B regulator of nonsense-mediated mRNA decay (*UPF3B*; MIM 300298) [19] and polyglutamine-binding protein 1 (*PQBP1*; MIM 300463) [20]. Interestingly, knockdown of *YTHDF3* with short interfering RNA oligos was previously demonstrated to reduce the translation efficiency and decay of m$^6$A-modified mRNA [6]. Transport of mRNA is essential for neurons, and all three YTH domain family members were reported in neuronal RNA granules [21]. Functional studies are needed to establish if *YTHDF3* impacts the stability of m$^6$A-modified mRNA independent from the other YTH domain family members in a context-dependent manner [9].

The study had certain limitations. We had used the pLI to classify genes as likely exhibiting haploinsufficiency; however, both dominant and recessive genes may have high pLI scores [22], and

a low pLI score is insufficient to exclude disease association [23]. The pLI score is affected by gene length and coverage, so that, for example, the pLI was <0.9 for *BHLHE22* deleted in individuals P2 and P3 despite no observations of LoF variants in this intron-less gene to date. This transcription factor-encoding gene is not known to be associated with Mendelian disease, but *BHLHE22* null mice exhibited abnormal motor and sensory cortex development in a study; a phenotype that was, however, not observed in heterozygous mice [24]. Moreover, a genome-wide association study linked the polymorphism rs16928927 within *NKAIN3* to developmental dyslexia at near genome wide significance [25]. In other words, we cannot rule out a potential neurodevelopmental impact of some of the other deleted genes despite the low pLI scores. Also, structural variants may have position effects on other distant genes outside the region [26]. Due to the lack of supplementary whole exome sequencing studies, contributions of additional genetic variants, including variants on the remaining allele of genes in the deleted region, cannot be ruled out. Alternative paternity could also be a potential source of false de novo variants in the study since identity testing was not performed. Lastly, developmental abnormalities are frequent in individuals submitted to DECIPHER, which could potentially have introduced a selection bias; however, the lack of *YTHDF3* deletions, except for small intronic deletions, observed in a large series of healthy individuals and compiled in the Database of Genomic Variants speaks against such bias. This first report on *YTHDF3* haploinsufficiency includes a limited number of known cases, and more cases are needed to verify the clinical disease spectrum. Despite of these limitations, this study adds to the emerging body of evidence implicating the m$^6$A machinery in neurodevelopmental disorders [10]. Future studies could address if other genes in the YTH-domain family are associated with human disease corresponding to observed roles in neural development [27 28].

In conclusion, we suggest that haploinsufficiency of *YTHDF3* causes a neurodevelopmental disorder characterized by intellectual disability and/or developmental delay of variable degree. The phenotyping of more individuals into later adulthood will be necessary to confirm the full range of features associated with mono-allelic loss of *YTHDF3*.

**ACKNOWLEDGEMENTS**

We wish to thank the patients and their families for taking part in the study.

This study makes use of data generated by the DECIPHER community [11]. A full list of centres who contributed to the generation of the data is available from http://decipher.sanger.ac.uk and via email from decipher@sanger.ac.uk. Funding for the Decipher project was provided by the Wellcome Trust.

**CONTRIBUTIONS**

Conceptualization: T.T., C.B.A., U.B.J., C.R.F. Investigation: T.T., C.B.A., N.I., T.B., C.M., K.C., S.H., C.R.F. Visualization: T.T., C.B.A. Writing-original draft: T.T. Writing-review and editing: C.B.A., U.B.J., C.R.F.

**FUNDING SOURCES**

This work was supported by The fund to support clinical research careers in the Region of Southern Denmark (C.R.F) and Aarhus University (T.T.). The funding sources had no involvement in the study.

**DATA AVAILABILITY STATEMENT**

All reported copy number variants have been submitted to DECIPHER as open-access (https://www.deciphergenomics.org).

**REFERENCES**

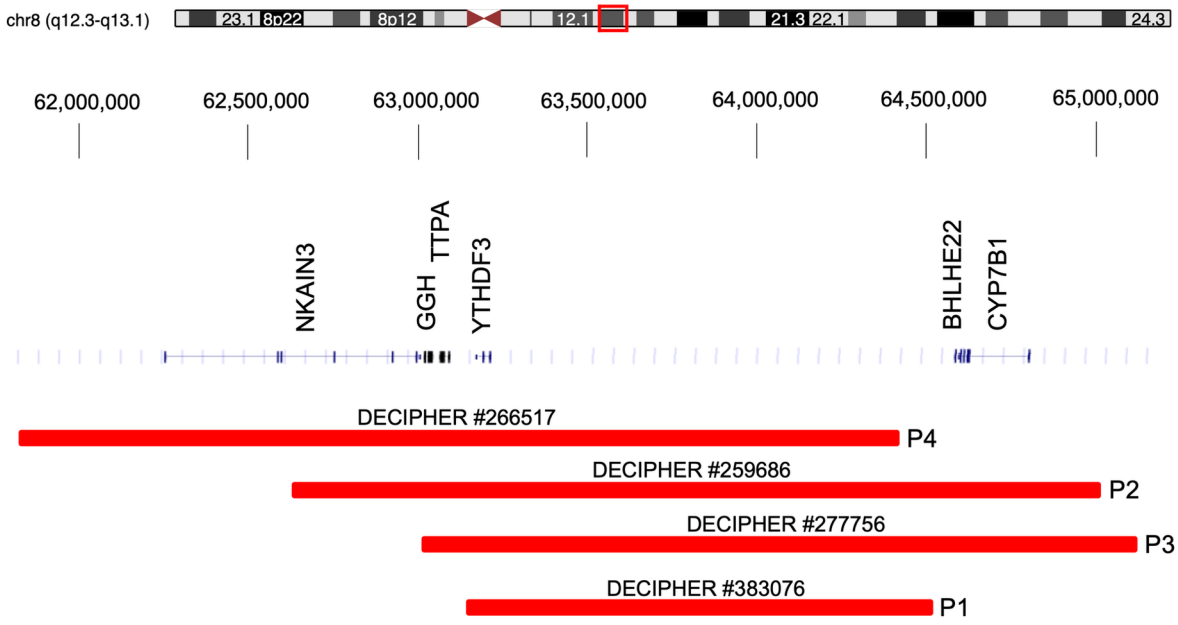
1. Desrosiers R, Friderici K, Rottman F. Identification of methylated nucleosides in messenger RNA from Novikoff hepatoma cells. *Proc Natl Acad Sci U S A* 1974;71(10):3971-5. doi: 10.1073/pnas.71.10.3971 [published Online First: 1974/10/01]
2. Wang X, He C. Reading RNA methylation codes through methyl-specific binding proteins. *RNA Biol* 2014;11(6):669-72. doi: 10.4161/rna.28829 [published Online First: 2014/05/16]

3. Patil DP, Pickering BF, Jaffrey SR. Reading m(6)A in the Transcriptome: m(6)A-Binding Proteins. *Trends Cell Biol* 2018;28(2):113-27. doi: 10.1016/j.tcb.2017.10.001 [published Online First: 2017/11/07]
4. Wang X, Zhao BS, Roundtree IA, et al. N(6)-methyladenosine Modulates Messenger RNA Translation Efficiency. *Cell* 2015;161(6):1388-99. doi: 10.1016/j.cell.2015.05.014 [published Online First: 2015/06/06]
5. Wang X, Lu Z, Gomez A, et al. N6-methyladenosine-dependent regulation of messenger RNA stability. *Nature* 2014;505(7481):117-20. doi: 10.1038/nature12730 [published Online First: 2013/11/29]
6. Shi H, Wang X, Lu Z, et al. YTHDF3 facilitates translation and decay of N(6)-methyladenosine-modified RNA. *Cell Res* 2017;27(3):315-28. doi: 10.1038/cr.2017.15 [published Online First: 2017/01/21]
7. Li A, Chen YS, Ping XL, et al. Cytoplasmic m(6)A reader YTHDF3 promotes mRNA translation. *Cell Res* 2017;27(3):444-47. doi: 10.1038/cr.2017.10 [published Online First: 2017/01/21]
8. Zaccara S, Jaffrey SR. A Unified Model for the Function of YTHDF Proteins in Regulating m(6)A-Modified mRNA. *Cell* 2020 doi: 10.1016/j.cell.2020.05.012 [published Online First: 2020/06/04]
9. Lasman L, Krupalnik V, Viukov S, et al. Context-dependent functional compensation between Ythdf m(6)A reader proteins. *Genes Dev* 2020;34(19-20):1373-91. doi: 10.1101/gad.340695.120 [published Online First: 2020/09/19]
10. Dermentzaki G, Lotti F. New Insights on the Role of N (6)-Methyladenosine RNA Methylation in the Physiology and Pathology of the Nervous System. *Front Mol Biosci* 2020;7:555372. doi: 10.3389/fmolb.2020.555372 [published Online First: 2020/09/29]
11. Firth HV, Richards SM, Bevan AP, et al. DECIPHER: Database of Chromosomal Imbalance and Phenotype in Humans Using Ensembl Resources. *Am J Hum Genet* 2009;84(4):524-33. doi: 10.1016/j.ajhg.2009.03.010 [published Online First: 2009/04/07]
12. Karczewski KJ, Francioli LC, Tiao G, et al. The mutational constraint spectrum quantified from variation in 141,456 humans. *Nature* 2020;581(7809):434-43. doi: 10.1038/s41586-020-2308-7 [published Online First: 2020/05/29]
13. Kent WJ, Sugnet CW, Furey TS, et al. The Human Genome Browser at UCSC. *Genome research* 2002;12(6):996-1006. doi: 10.1101/gr.229102
14. Chou JH, Roumiantsev S, Singh R. PediTools Electronic Growth Chart Calculators: Applications in Clinical Care, Research, and Quality Improvement. *J Med Internet Res* 2020;22(1):e16204. doi: 10.2196/16204 [published Online First: 2020/02/06]
15. Roche AF, Mukherjee D, Guo SM, et al. Head circumference reference data: birth to 18 years. *Pediatrics* 1987;79(5):706-12. [published Online First: 1987/05/01]
16. Coe BP, Stessman HAF, Sulovari A, et al. Neurodevelopmental disease genes implicated by de novo mutation and copy number variation morbidity. *Nat Genet* 2019;51(1):106-16. doi: 10.1038/s41588-018-0288-4 [published Online First: 2018/12/19]
17. Ye XC, Pegado V, Patel MS, et al. Strabismus genetics across a spectrum of eye misalignment disorders. *Clin Genet* 2014;86(2):103-11. doi: 10.1111/cge.12367 [published Online First: 2014/03/04]
18. Zhang F, Kang Y, Wang M, et al. Fragile X mental retardation protein modulates the stability of its m6A-marked messenger RNA targets. *Hum Mol Genet* 2018;27(22):3936-50. doi: 10.1093/hmg/ddy292 [published Online First: 2018/08/15]

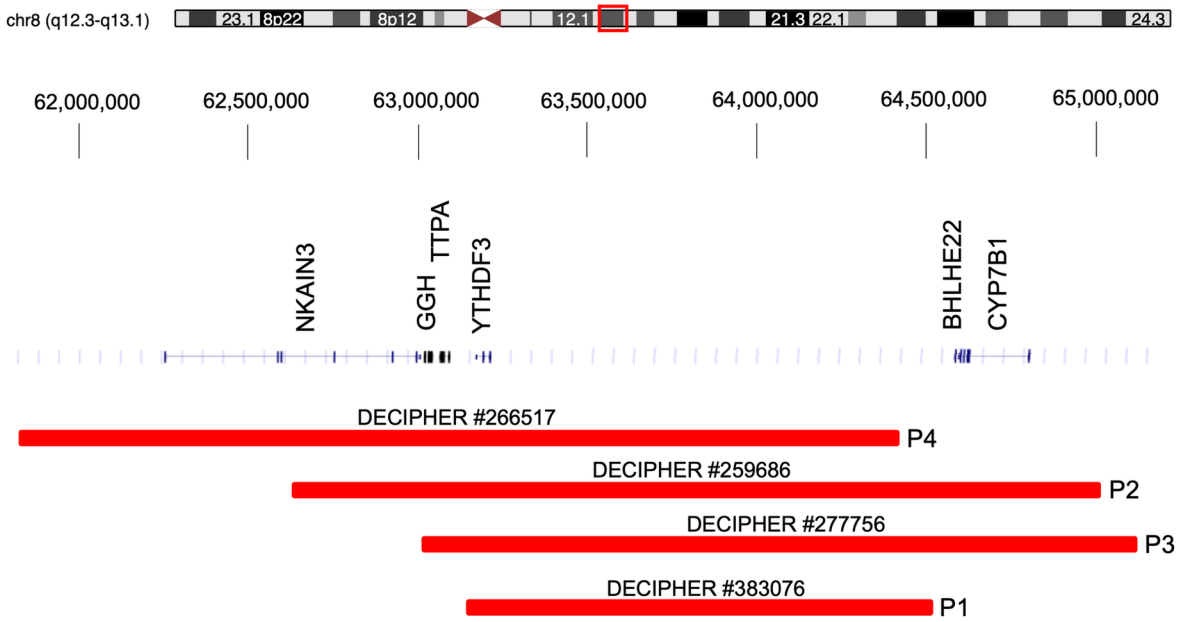
19. Tarpey PS, Raymond FL, Nguyen LS, et al. Mutations in UPF3B, a member of the nonsense-mediated mRNA decay complex, cause syndromic and nonsyndromic mental retardation. *Nat Genet* 2007;39(9):1127-33. doi: 10.1038/ng2100 [published Online First: 2007/08/21]
20. Wang Q, Moore MJ, Adelmant G, et al. PQBP1, a factor linked to intellectual disability, affects alternative splicing associated with neurite outgrowth. *Genes Dev* 2013;27(6):615-26. doi: 10.1101/gad.212308.112 [published Online First: 2013/03/21]
21. Fritzsche R, Karra D, Bennett KL, et al. Interactome of two diverse RNA granules links mRNA localization to translational repression in neurons. *Cell Rep* 2013;5(6):1749-62. doi: 10.1016/j.celrep.2013.11.023 [published Online First: 2013/12/24]
22. Fuller ZL, Berg JJ, Mostafavi H, et al. Measuring intolerance to mutation in human genetics. *Nat Genet* 2019;51(5):772-76. doi: 10.1038/s41588-019-0383-1 [published Online First: 2019/04/10]
23. Ziegler A, Colin E, Goudenège D, et al. A snapshot of some pLI score pitfalls. *Hum Mutat* 2019;40(7):839-41. doi: 10.1002/humu.23763 [published Online First: 2019/04/13]
24. Joshi PS, Molyneaux BJ, Feng L, et al. Bhlhb5 regulates the postmitotic acquisition of area identities in layers II-V of the developing neocortex. *Neuron* 2008;60(2):258-72. doi: 10.1016/j.neuron.2008.08.006 [published Online First: 2008/10/30]
25. Gialluisi A, Andlauer TFM, Mirza-Schreiber N, et al. Genome-wide association scan identifies new variants associated with a cognitive predictor of dyslexia. *Translational Psychiatry* 2019;9(1):77. doi: 10.1038/s41398-019-0402-0
26. Lupianez DG, Kraft K, Heinrich V, et al. Disruptions of topological chromatin domains cause pathogenic rewiring of gene-enhancer interactions. *Cell* 2015;161(5):1012-25. doi: 10.1016/j.cell.2015.04.004
27. Zhuang M, Li X, Zhu J, et al. The m6A reader YTHDF1 regulates axon guidance through translational control of Robo3.1 expression. *Nucleic Acids Res* 2019;47(9):4765-77. doi: 10.1093/nar/gkz157 [published Online First: 2019/03/08]
28. Li M, Zhao X, Wang W, et al. Ythdf2-mediated m(6)A mRNA clearance modulates neural development in mice. *Genome Biol* 2018;19(1):69. doi: 10.1186/s13059-018-1436-y [published Online First: 2018/06/02]

**FIGURE LEGENDS**

**Figure 1**. Overview of known protein-coding genes and the deletions in 8q12.3 (hg38) adapted from the UCSC Genome Browser (https://genome.ucsc.edu).




CGE_14083_FIGURE1.tiff




CGE_14083_GRAPHICAL_ABSTRACT.tiff

**Table 1**. Molecular genetic findings and clinical features of the four individuals.

| | P1 | P2 | P3 | P4 |
|---|---|---|---|---|
| Sex | F | M | M | M |
| Age at reporting | 6y | 19y | 22y | 12y |
| DECIPHER no. | 383076 | 259686 | 277756 | 266517 |
| Deletion size (Mb) | 1.38 | 2.39 | 2.11 | 2.60 |
| Coordinates (hg38) | 8:63,136,510-64,514,341 | 8:62,619,801-65,010,652 | 8:63,004,236-65,117,087 | 8:61,812,894-64,417,163 |
| Inheritance | Dn | Dn | NA | Dn |
| Birth weight | -0.2 SD | +1.1 SD | +0.5 SD | NA |
| Birth length | +0.5 SD | +0.6 SD | NA | NA |
| Birth head circumference | -1.6 SD | -0.4 SD | NA | NA |
| Weight (age) | -0.4 SD (6y) | +0.5 SD (14y) | +2.7 SD (15y) | -0.7 to 0 SD (12y) |
| Height (age) | -0.2 SD (6y) | +0.4 SD (14y) | +1.3 to +2.1 SD (15y) | -0.7 to 0 SD (12y) |
| Head circumference (age) | -0.7 SD (6y) | 0.7 to 1.3 SD (14y) | NA | -1.3 SD (4.5y) |
| Motor delay | Walked at age 2y | - | Walked at age 4y | Poor motor coordination |
| Speech delay | First words at age 2y | - | First words at age 3y | Repetitive words from age 1.5y, dyspraxia and difficulties in constructing sentences. |
| ID | Mild | Mild | Severe | ID not classified |
| Autism spectrum | - | PDD | Autism | - |
| Epilepsy | - | - | + | - |
| Cerebral MRI | NA | Normal | Normal | NA |
| Other | Umbilical hernia | - | Strabismus, flat feet, wide sandal gap, slender fingers | Strabismus; additional deletions of 8:65,591,973-66,332,718 (Dn) and 9:78,571,738-81,153,868 (Dn) |

Dn = de novo, ID = intellectual disability, m = months, NA = information not available, PDD = pervasive developmental disorder, SD = standard deviations, y = years.