

Truncating Variants in *NAA15* Are Associated with Variable Levels of Intellectual Disability, Autism Spectrum Disorder, and Congenital Anomalies

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N-alpha-acetylation is a common co-translational protein modification that is essential for normal cell function in humans. We previously identified the genetic basis of an X-linked infantile lethal Mendelian disorder involving a c.109T>C (p.Ser37Pro) missense variant in *NAA10*, which encodes the catalytic subunit of the N-terminal acetyltransferase A (NatA) complex. The auxiliary subunit of the NatA complex, *NAA15*, is the dimeric binding partner for *NAA10*. Through a genotype-first approach with whole-exome or genome sequencing (WES/WGS) and targeted sequencing analysis, we identified and phenotypically characterized 38 individuals from 33 unrelated families with 25 different *de novo* or inherited, dominantly acting likely gene disrupting (LGD) variants in *NAA15*. Clinical features of affected individuals with LGD variants in *NAA15* include variable levels of intellectual disability, delayed speech and motor milestones, and autism spectrum disorder. Additionally, mild craniofacial dysmorphism, congenital cardiac anomalies, and seizures are present in some subjects. RNA analysis in cell lines from two individuals showed degradation of the transcripts with LGD variants, probably as a result of nonsense-mediated decay. Functional assays in yeast confirmed a deleterious effect for two of the LGD variants in *NAA15*. Further supporting a mechanism of haploinsufficiency, individuals with copy-number variant (CNV) deletions involving *NAA15* and surrounding genes can present with mild intellectual disability, mild dysmorphic features, motor delays, and decreased growth. We propose that defects in NatA-mediated N-terminal acetylation (NTA) lead to variable levels of neurodevelopmental disorders in humans, supporting the importance of the NatA complex in normal human development.

Advances in sequencing technologies such as whole-exome or genome sequencing (WES/WGS) have led to disease-gene association discoveries, functional annotation of the human genome, and improved diagnostic rates in individuals with suspected genetic disorders refractory to conventional diagnostic testing. An estimated diagnostic rate that often exceeds 25% can be achieved when WES/WGS is applied to otherwise undiagnosed complex cases.^{1–5} *NAA15* (N-alpha-acetyltransferase 15, MIM: 608000) was previously characterized as one of fifty-two risk genes for neurodevelopmental disorders by targeted sequencing of a large autism spectrum and intellectual disability (ASID) cohort.⁶ In another study of *de novo*

changes in severe congenital heart disease (CHD), likely gene disrupting (LGD) variants in *NAA15* were identified in two affected individuals in a cohort of 362 severe CHD cases; one of these individuals was known to have additional neurodevelopmental defects.⁷ In an effort to further characterize the clinical and molecular spectrum associated with genetic defects in *NAA15*, we ascertained, from 33 unrelated families, 38 individuals with truncating, presumably LGD (nonsense, frameshifting and splice) variants in *NAA15* via a collaborative world-wide effort among multiple institutions. As a result of comprehensive clinical evaluation and molecular analyses in all individuals, we propose that deleterious variants in *NAA15* are

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associated with variable levels of intellectual disability, developmental delay, autism spectrum disorder, dysmorphic features, and congenital cardiac anomalies.

This study was performed in accordance with protocols approved by the institutional review boards of the participating institutions (see [Supplemental Data](#)). Three affected individuals were recruited from the UK Deciphering Developmental Disorders (DDD) project (families 20, 27, and 30). Written informed consent was obtained from all study participants. The key clinical features of our cohorts are summarized in [Table 1](#). Detailed clinical summaries for each subject are provided in the [Supplemental Data](#). The use of GeneMatcher, a web-based tool for connecting researchers with an interest in the same gene,⁸ facilitated contact between international collaborators.

All subjects have variable degrees of neurodevelopmental disabilities, including impaired motor abilities (HP: 0001270), intellectual disability (HP: 0001249), impaired verbal abilities (HP: 0000750), and autism spectrum disorder (HP: 0000729) ([Table 1](#), [Table S1](#), and [Table S2](#)). Many subjects have impaired motor function, including fine-motor difficulties (n = 5, or 12%), mild ataxia (n = 1), abnormality of movement (n = 1), motor delay (n = 22, or 60%), and hypotonia (n = 5, or 14%). Various levels of intellectual disability are reported in almost all study subjects with available data; such disability includes mild,

moderate, or severe intellectual disability and learning difficulties with or without behavioral issues ([Table 1](#), [Table S1](#), and [Table S2](#)). The majority of affected individuals have verbal issues, including complete absence of speech, delayed language development, the need for sign language, or other speech difficulties. Most subjects also present with autism spectrum disorder (ASD) and/or other behavioral abnormalities. Individual 11 was noted to have marked hypersomnolence in early years, in apparent similarity to what was recently reported in a girl with a missense variant in *NAA10*.⁹ Minor facial dysmorphism was reported in some individuals ([Table 1](#)), but there were no consistent features noted nor a recognizable pattern of facial dysmorphism ([Figure 1](#) and [Figure S1](#)). The birth weight and length were low in some individuals; the most notable feature was a birth weight \leq 1st percentile in 7 out of 25 (28%) individuals with available information ([Table S3](#)). Some of the individuals remain small throughout life, whereas others are of normal stature and a few are above average height ([Table S3](#)).

Almost all individuals have normal or uncharacterized cardiac function ([Table 1](#)), with four exceptions. Individual 2 ([Figure 1](#)) has atrial ectopic (multifocal) tachycardia (HP: 0011701), treated with verapamil, and hypertension (HP: 0000822). Individual 3 had a ventricular septal defect (VSD), repaired surgically during infancy. Individual 17

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Table 1. Summary of Phenotypes

Phenotype	Number of individuals with phenotype	Number of individuals with relevant data	Percentage
Brain Structure and Function			
Intellectual disability (ID) ^a	23	23	100
ASD, ADHD, or behavioral issues	30	33	91
Abnormal brain MRI	2	11	18
Speech delay	32	33	97
Seizures	6	26	23
Motor Impairments			
Motor delay and related abnormalities	31	32	97
Muscle tone issues	7	18	39
Feeding difficulties	8	14	57
Cardiovascular			
Congenital cardiac defects	4	19	21
Major vessel anomalies	2	19	11
Arrhythmias	1	19	5
Hypertension	1	19	5
Other			
Mild dysmorphism	18	28	64
Skeletal or connective-tissue defects	8	20	40

^aIn individuals > 5 years of age, when IQ testing or other cognitive testing was performed.

has a Marfanoid habitus, with an aortic root at the upper limit of normal. By far the most severely affected, individual 19 has heterotaxy syndrome associated with a complex cardiac diagnosis of dextrocardia involving left superior and inferior venae cavae, total anomalous pulmonary venous return to the innominate vein, tricuspid atresia, hypoplastic right ventricle, double-outlet right ventricle, and transposed great arteries with severe pulmonary stenosis. The variant in this individual (c.1009_1012delGAAA) was previously reported in a cohort of 1,213 subjects with CHD and an increased prevalence of extracardiac congenital anomalies (CAs) and risk of neurodevelopmental disabilities (NDDs).^{7,10} Another LGD variant, c.2282C>A (p.Ser761*) in *NAA15*, was first reported in an individual with pulmonary stenosis, single left coronary artery, and tetralogy of Fallot (in the context of no reported neurodevelopmental disability), although we have been

unable to obtain additional information on this individual.⁷ A more recent analysis of this now expanded cohort of 2,871 CHD probands, including 2,645 parent-offspring trios, did not find any new variants in *NAA15*.¹¹ Given the low prevalence of CHD in our own cohort of 38 individuals, one caveat is that the expression of severe congenital heart disease could be due to variation at a second locus, a noncoding mutation outside of the exome, or some other additional variation undetected thus far.

A total of 25 presumably LGD variants contained in 12 of the 20 exons and two intron-exon boundaries of *NAA15* were identified from 33 unrelated families (Figure 2, Table S1, Table S2); these included nonsense variants (n = 13), canonical splice-site variants (n = 2), and frameshift variants (n = 10). The inheritance pattern of the variants was determined to be *de novo* for most subjects (22 families) through testing of parental samples. Familial inheritance was observed in three families (families 10, 22, and 28), and the corresponding *NAA15* LGD variant segregated with the neurocognitive phenotypes, including in one mildly affected parent in each family and in affected siblings in families 10 and 28. For Family 10, the read count data did not demonstrate any somatic mosaicism in the blood sample from the mother. Among the 25 variants identified, there were three recurrent variants, including c.228_232delCTTGA (p.Asp76Gluufs*20) (families 3 and 4, *de novo*), c.239_240delAT (p.His80Argufs*17) (families 6-11B, *de novo* in families 6 and 7, familial in family 10, and unknown inheritance in the rest of the families), and c.1009_1012delGAAA (p.Glu337Argufs*5) (families 19 and 20, *de novo*). We examined genomic context around the three recurrent loci to look for micro-homology that might increase the propensity for recurrent mutations and found that the most recurrent mutation, c.239_240delAT, occurs in the middle of one of 20 reported mutation hotspots, CATGT.¹² In addition, this recurrent variant, and another one, c.228_232delCTTGA, are close to each other in exon 3 in an area that is computationally predicted¹³ to form a quasipalindromic structure (lying distal to an even larger quasipalindromic structure), and the third recurrent mutation c.1009_1012delGAAA in exon 9 lies just distal to a quasipalindromic structure (Figure S2).¹⁴

Data from the Exome Aggregation Consortium (ExAC) study of 60,706 control individuals show that *NAA15* is likely intolerant to LGD variants (pLI = 1.00),¹⁵ that the residual variation intolerance score (RVIS) = -0.89 (among the 10.2% most LGD intolerant of human genes), and that LoF-FDR[ExAC] = 0.000224349.¹⁶ Excluding small cohorts (<100 probands, Table S4), we in total identified fourteen *de novo* variants in *NAA15* from six independent rare disease cohorts with a total sample size of ~36,731. Ten out of 14 cases are reported in detail here; the remaining four lack sufficient phenotype information. Our aggregate frequency of *de novo* LGD variants in affected individuals (~4.0 per 10,000) is significantly higher than the background rates estimated by Samocha et al.¹⁷ for LGD

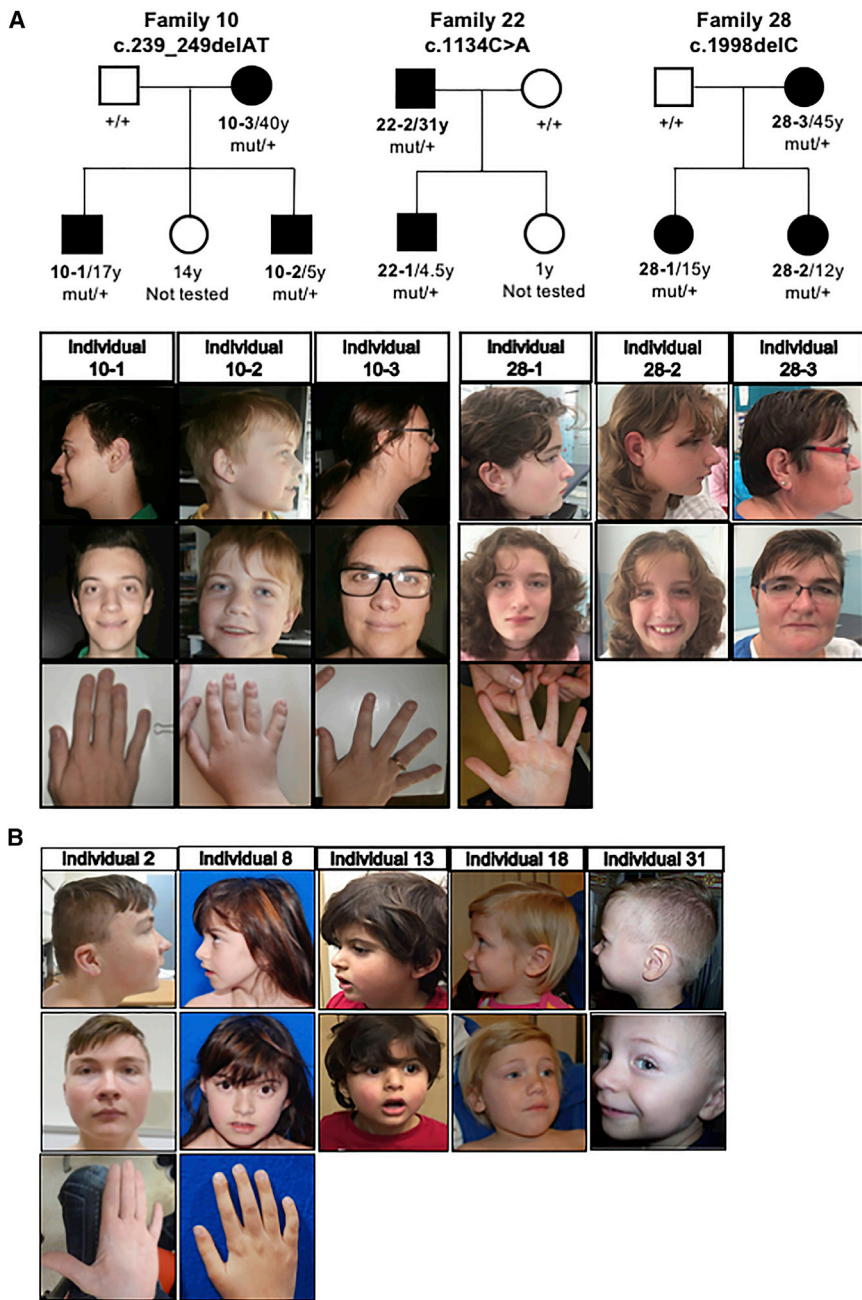


Figure 1. Pedigrees, Mild Facial Dysmorphology, and Hands of Individuals with Familial or *de novo* NAA15 LGD Variants

(A) Pedigrees are shown for the three families with inherited variants. **Family 10, Individual 10-1:** at age 17 years and 6 months, with prominent eyebrows, broad nose, and prominent chin. Hand appears normal. **Individual 10-2:** at 6 years and 6 months, with very well-developed philtral pillars. Hand appears normal. **Individual 10-3:** mother, with long mentum of the chin and relatively thick alae nasi. Hand appears normal. **Family 28, Individual 28-1:** at age 15 years, partial syndactyly in one hand, but otherwise not with particularly notable dysmorphology. **Individual 28-2:** sister, at age 12 years, who was not noted to have any obvious dysmorphology. **Individual 28-3:** Mother at age 45 years, with broad nose but otherwise not with notable dysmorphology.

(B) Minor facial dysmorphology was noted in some probands, but there were no reliably consistent features shared among them. **Individual 2:** at 17 years old, noted to have brachycephaly, appearance of ocular hypertelorism with short palpebral fissures, prominent nose tip with a longer columella of the nose, trapezoidal philtrum, and micrognathia without retrognathia. Also noted are small low-set, posteriorly rotated ears, with thickened and overfolded helix; hypoplastic distal phalanges on digits 2, 3, and 4; 5th finger with brachyclinodactyly; and persistence of fetal finger pads on the 3rd and 4th digit. **Individual 8:** at the age 8 years 9 months, noted to have thin philtrum, bulbous nasal tip, and 5th finger with brachyclinodactyly. **Individual 13:** at 4 years old, no facial dysmorphism noted. **Individual 18:** at 4 years and 3 months, with bulbous nose tip, thick alae nasi and anteverted nares, prominent cupid's bow and philtrum, long mentum of the chin, and simple ears. **Individual 31:** with epicanthus inversus, smooth philtrum, thin vermilion border of the upper lip, and sparse lateral eyebrows.

mutations (expected ~ 0.038 per 10,000; $p < 2.2 \times 10^{-16}$). If we further restrict the analysis to the three largest cohorts, each of which included more than 5,000 probands, the observed enrichment remains highly significant (nine *de novo* LGD variants among 33,831 total probands; $p = 2.48 \times 10^{-14}$). We acknowledge that there are limitations to comparing results from ExAC to a clinically ascertained cohort, particularly when one undertakes a genotype-first approach by actively searching for singleton cases with variants in *NAA15* by using different sequencing platforms and coverage levels.¹⁸ However, the average coverage for *NAA15* in ExAC and gnomAD databases is $47\times$ versus approximately $20\times$ coverage levels provided by clinically offered exome tests, suggesting that

the increased number of LGD variants in the current study is not due to higher exon coverage levels in clinical sequencing. Only six LGD variants in *NAA15* are reported in ExAC (Table S5), and 11 *NAA15* LGD variants are reported in the Genome Aggregation Database (gnomAD) (Table S6). Two of the variants that are recurrent and *de novo* in our research cohort (c.239_240delAT [p.His80Argfs*17] and c.228_232delCTTGA [p.Asp76Gluufs*20]) are present one time each in ExAC (and also duplicated in gnomAD, given that gnomAD includes many variants from ExAC). It should be noted that phenotypic information as well as the variant inheritance are not available on these individuals in ExAC or gnomAD. Given that the three parents in the inherited families (families 10, 22,

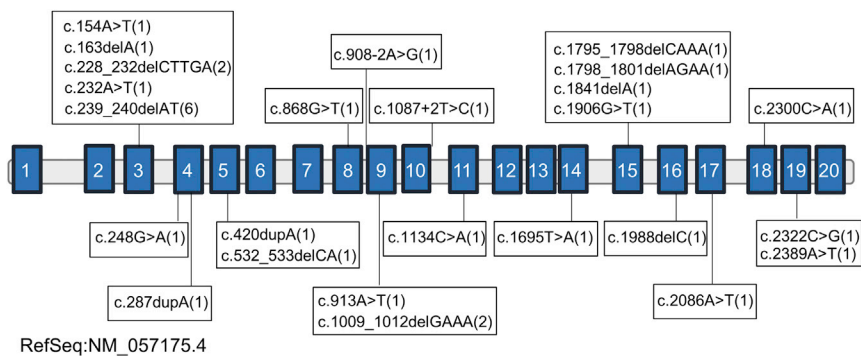


Figure 2. Exonic Localization of *NAA15* LGD Variants Identified in Subjects in This Study

Schematic representation of the genomic structure of human *NAA15*. Solid blue rectangles indicate exons, and the horizontal bars represent introns. *NAA15* variants with their relative positions in the gene are shown, and the number of affected individuals with the specific variants is shown in parentheses.

and 28) were only mildly affected, it is possible that such individuals could be found in cohorts such as ExAC or gnomAD. A recent study showed that ~2.8% of the ExAC population is associated with possible disease-associated genotypes,¹⁹ and it is well-known that genetic background can influence the expressivity of any given variant.

We sought to confirm whether any of the LGD variants might trigger nonsense-mediated decay (NMD) of the respective mutant RNA. For this, we made use of two research-subject-derived cell lines, including one lymphoblastoid cell line (LCL) from individual 10-1 (c.239_240delAT) (Figures 3A–3C and Supplemental Methods) and one induced pluripotent stem cell (iPS) line from individual 19 (c.1009_1012delGAAA) (Figures 3D–3F). Quantitative RT-PCR with primers 3' to the mutation demonstrated approximately 50% decreased total RNA in one cell passage from the LCLs from individual 10-1 in comparison to three control LCLs (Figure 3C, left panel), whereas the same assay (Figure 3F, right panel) and an additional Taqman assay (Figure S3) showed more variability in total RNA isolated from three different passages of the iPS line from individual 19 than from one control iPS line and a control human embryonic stem cell (hESC) line. Nonetheless, RT-PCR with primers spanning the mutation sites, followed by Sanger sequencing, did demonstrate substantially reduced mutant transcript in the LCL from individual 10-1 (Figure 3B) and almost complete absence of the mutant transcript in three different passages of the iPS line from individual 19 (a representative result from passage 16 is shown in Figure 3E). This reduction most likely occurs because the variant transcript is targeted for degradation via the nonsense-mediated decay (NMD) pathway.²⁰

We further explored the functional effects for two of the other LGDs in a yeast assay in which the human NatA complex can functionally replace yeast NatA, as shown by complementation of growth phenotypes^{21,22} and partial rescue of the NatA-specific Nt-acetylome.²³ Mutant *NAA15* (p.Thr55Hisfs*2 [c.163delA] from family 2 and p.Lys305* [c.913A>T] from family 18) failed to rescue the temperature-sensitive growth phenotype of yNatAΔ (Figure 4A and Tables S7, S8, and S9), suggesting that the two variants lead to reduced or abolished NatA activity, at least as assessed in this heterologous system. We further

verified human NatA expression in the yNatA deletion strain by immunoblotting (Figure 4B) against the HA epitope that was incorporated N-terminal to *NAA15*. In the context of overexpression from a plasmid, we detected both full-length HA-*NAA15* and HA-*NAA15* p.Lys305*, but not HA-*NAA15* p.Thr55Hisfs*2, suggesting that the mRNA for HA-*NAA15* p.Thr55Hisfs*2 is most likely undergoing complete NMD and/or that this truncated mini-protein is unstable, whereas truncated mini-protein HA-*NAA15* p.Lys305* can be expressed in this system but nonetheless does not provide functional rescue.

Distributed throughout the entire gene of *NAA15*, the 25 LGD variants we reported here are predicted to undergo NMD, leading to degradation of the mutant mRNA and thus loss of the aberrant protein product. Expression analysis from research-subject-derived lymphoblast cells or iPSCs confirmed under-representation of the mutant transcript in cDNA. In addition, the functional deficiency of human mutated *NAA15* was further supported by the growth rescue experiment in the yeast NatA-deficient strain, in which mutant human *NAA15* failed to restore the growth-deficiency phenotype. In light of these results, we propose haploinsufficiency of *NAA15* as the most likely mechanism for this newly recognized disease, although we readily acknowledge that some of the LGDs might not trigger complete NMD or might do so differentially in different tissues, leaving open the possibility for expression of a truncated *NAA15* protein, which could possibly act via a dominant-negative or gain-of-function mechanism in some individuals. *De novo* missense variants (c.1014G>T [p.Lys338Asn] and c.841G>C [p.Glu281Gln]) have been previously reported in two individuals with autism and intellectual disability, respectively;^{24,25} however, the deleterious effect of these missense variants has not been established and so will also require further functional studies, segregation in families, and/or proof of recurrence in multiple affected individuals. Further supporting our postulated mechanism of haploinsufficiency, when we searched the DECIPHER database²⁶ and our clinical cohorts for individuals with small microdeletions involving *NAA15*, the smallest deletion we could find is in a 31-year-old man carrying a *de novo* 2.73 Mb deletion, including *NAA15* and 17 other predicted genes. This man was noted as having mild intellectual disability, mild

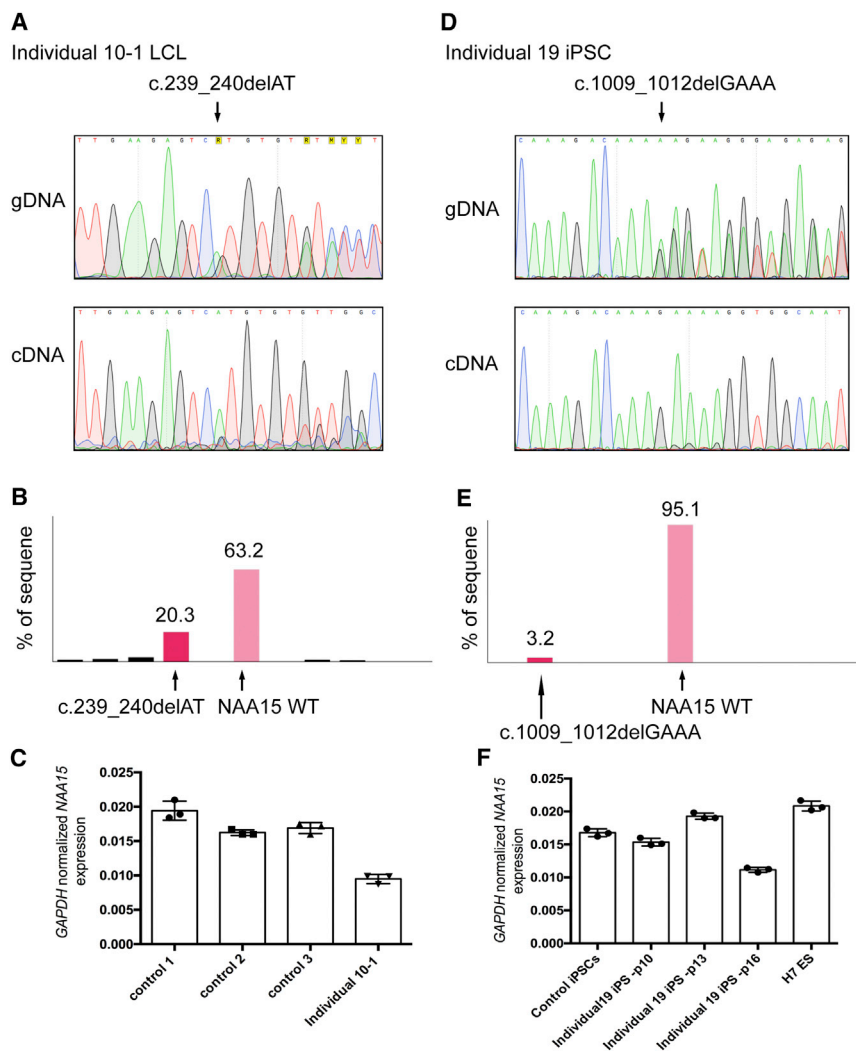


Figure 3. Expression Analysis of *NAA15* in Research-Subject-Derived Cell Lines

(A and D) Sanger sequencing of genomic DNA (top panel) and reverse-transcribed cDNA (bottom panel) isolated from a lymphoblastoid cell line (LCL) of individual 10-1 (c.239_240delAT) (A) and an induced pluripotent stem cell (iPS) line (passage 16) of individual 19 (c.1009_1012delGAAA) (D). (B and E) Quantification of different cDNA species from cDNA Sanger sequencing showing the relative ratio of WT *NAA15* versus c.239_240delAT (LCL line) (B) and (c.1009_1012delGAAA) (passage 16 iPS cell line) (E). (C and F) *NAA15* mRNA expression level analyzed by qPCR in research subject-derived cell lines (at passage numbers p10, p13, and p16), as compared to control cell lines (at passage 16). Error bars are standard deviation (SD), and the assay was performed three times per sample.

the auxiliary subunit binding with the catalytic subunit *NAA10* and localizes it to the ribosome, where this complex (named the NatA complex) serves as an N-terminal acetyltransferase (NAT).²⁹ This complex is evolutionarily conserved from yeast to vertebrates,²³ and the X-ray crystal structure of the 100 kDa holo-NatA complex from *Schizosaccharomyces pombe* shows that the NatA-*NAA15* auxiliary subunit contains 13 tetratricopeptide motifs and adopts a ring-like topology that wraps around the

dysmorphic features, motor delays in childhood, a low birth weight (−2SD), and adult height, weight, and head circumference all at the 10th centile (Figure S4). He has poor vision as a result of cortical visual impairment (CVI), which was not reported (but also not formally screened for) in any of the above reported individuals but which was found in some of the individuals with *NAA10* mutations.²⁷ It is also possible that his CVI could be due to some other missing gene in the CNV interval. There are currently 18 large heterozygous CNV deletions, including *NAA15* in the DECIPHER database;²⁶ these deletions range in size from 3.27 Mb to 24.30 Mb, and many are noted to be associated with global developmental delay or intellectual disability, supporting the case for haploinsufficiency of at least some of the genes in these CNV intervals. One individual with a *de novo* 5.2 Mb deletion died from a sudden cardiac event at the age of 35 (see Supplemental Case Reports).

Human *NAA15* encodes an 866 amino acid (~105 kDa) protein, *NAA15*, containing tetratricopeptide repeat domains and a putative bipartite nuclear localization signal.²⁸ Many studies have shown that *NAA15* acts as

NatA-*NAA10* subunit, an interaction that alters the *NAA10* active site for substrate-specific acetylation.³⁰ Mutation or loss of the NatA subunits in yeast (*Saccharomyces cerevisiae*) or human HeLa cells results in inhibited cell growth, cell apoptosis, and failure to enter the G₀ phase in the cell cycle.^{31,32} *Nat1* (ortholog of *NAA15*) knock-down flies have impaired locomotor activity and early adult lethality.⁶ *NAA10* and *NAA15* are both highly expressed in regions of cell division and migration during brain development and are downregulated as neurons differentiate in early postnatal development.^{33,34} *NAA15* has been shown to be expressed at low levels in most adult tissues (e.g., nervous system, heart, and reproductive system) (see GTEx Portal). However, RNA-seq data from human brain tissue suggests that upregulation of *NAA15* occurs in utero at eight weeks after conception and is developmentally downregulated thereafter, the highest expression being in the occipital neocortex and anterior cingulate (medial prefrontal) cortex (Figures S5A and S5B), supporting a role for *NAA15* in development of the nervous system. Similarly, in mice, upregulated expression of *NAA15* has been shown in regions of neuronal

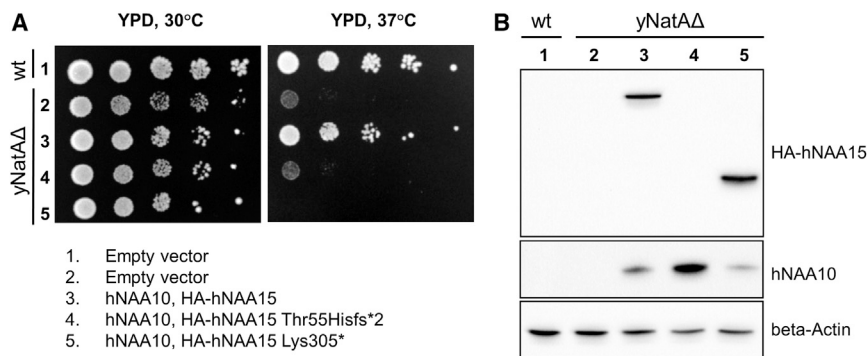


Figure 4. Truncation Mutations of Human NAA15 Impair NatA Function and Yeast Viability

(A) Serial dilution spot assay depicting the sensitivity of human NAA15 Thr55Hisfs*2 and Lys305* mutants to increased temperature in a *yna10Δ*, *yna15Δ* double-deletion background (*yNatAΔ*).

(B) Confirmation of human NatA expression by immunoblot analysis with anti-hNAA10 and anti-HA (for HA-hNAA15 detection) along with anti-beta Actin as a loading control.

migration, and proliferation in the neonatal mouse brain has been shown along with reduced expression as neurons differentiate during early postnatal development.^{33,34}

Genetic defects in *NAA10*, which is X-linked and encodes another member of the NatA complex, are associated with Ogden syndrome (MIM: 300855), Lenz microphthalmia (MIM: 309800), and intellectual disability (with variable cardiac involvement).^{27,35–39} In the case of Ogden syndrome, a total of eight boys from two families had a distinct combination of dysmorphology, hypotonia, global developmental delays, cardiac anomalies, cardiac arrhythmias and cardiomegaly, and the identical missense mutation segregated in multiple affected individuals in two unrelated families.⁴⁰ Different variants in *NAA10* have been reported, sometimes with only a mild intellectual-disability phenotype in heterozygous females, but also sometimes with hydrocephaly, supernumerary vertebrae, congenital heart defects, and arrhythmias, which are always more severe in the males.^{9,27,35–37} Although developmental delay and/or intellectual disability might be the only presenting feature, the additional cardiac, growth, dysmorphic features and other findings vary in type and severity. For the one family in which affected members had Lenz microphthalmia syndrome and a splice-site variant in *NAA10*, and in which proband-derived fibroblasts lacked expression of full-length NAA10 and displayed a cell-proliferation defect,⁴¹ it is not known why this family alone has such a dramatic ocular phenotype, although it is worth noting that 9/13 (69%) of the female subjects reported with missense variants in *NAA10* had some milder form of eye anomalies, including astigmatism, hyperopia and/or myopia.²⁷ Most studies have reported that missense mutations in *NAA10* decrease the enzymatic function of NAA10 and/or decrease its binding to NAA15.^{21,22,27,35,39,40}

In total, the presentations involving *NAA10* and *NAA15* appear to have phenotypic overlap but variability, and as such should be referred to more broadly as “*NAA10*-related syndrome” and “*NAA15*-related syndrome.” The extensive phenotypic variability is most likely related to genetic background differences and also to the spatial and temporal tissue-specific acetylation of a few N-terminal acetylation substrates by the NatA complex, although there are also suggested N-terminal acetylation (NTA)-

independent functions for *NAA10*.^{38,42} In the past few years, the first instance of NTA with relevance to cardiac function was reported and involved NTA of the cardiac voltage-gated sodium channel, Nav1.5, in tissues from individuals with end-stage heart failure.^{43,44} Indeed, protein quality control is of major relevance in heart failure.⁴⁵ Also, a 2015 study linked NTA and N-end-rule degradation to blood pressure regulation^{46,47} and demonstrated that N-terminal mutants of Rgs2, a key G-protein regulator, are differentially processed by NATs and the two branches of the N-end-rule pathway, leading to an imbalance in the signaling governing blood pressure. In regard to more common diseases and basic biology, there is emerging evidence that NTA of proteins are overexpressed or otherwise dysregulated in a variety of cancers, including lung, prostate, and liver cancers.^{48–54} NTA has been linked to neurodegenerative diseases such as Parkinson, Alzheimer, and Huntington disease, and NatA/NAA10 has been shown to contribute to the regulation of amyloid β -protein generation, to modulate the stabilization of Sup35 amyloid formation, and to prevent aggregation of Htt,^{55–60} supporting the importance of NTA in the progression of these diseases. Current findings link NTA to degradation of some proteins via Ac/N-degron-mediated recruitment of specific ubiquitin ligases.^{47,61–64} NTA might also influence protein-complex formation, as exemplified by the NEDD8 ligation enzymes,⁶⁵ along with prion formation.⁶⁰ Also, protein-specific targeting to membranes of the nucleus,⁶⁶ Golgi,^{67,68} and lysosomes⁶⁹ was shown to require NTA, but a general role in targeting is not supported.^{39,40,70}

In conclusion, we propose that disruption of NatA complex functionality can cause developmental disorders with variable expressivity. Future identification of additional affected individuals and studies in model organisms will be required if we are to continue to refine the clinical phenotype and determine the underlying mechanism whereby reduced expression or perturbed function of *NAA15* results in these phenotypes.

Supplemental Data

Supplemental Data include Supplemental Case reports, Figures S1–S4, a Supplemental Note, Supplemental Materials and Methods, Figures S1–S5, Tables S1–S9, Supplemental References,

and Supplemental Acknowledgments and can be found with this article online at <https://doi.org/10.1016/j.ajhg.2018.03.004>.

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Declaration of Interests

G.J.L. serves on advisory boards for GenePeaks and Seven Bridges Genomics. The Department of Molecular and Human Genetics at BCM derives revenue from molecular testing offered at Baylor Genetics Laboratories. J.R.L. has stock ownership in 23 and Me, is a paid consultant for Regeneron Pharmaceuticals, has stock options in Lasergen, and is a co-inventor on multiple United States and European patents related to molecular diagnostics for inherited neuropathies, eye diseases, and bacterial genomic fingerprinting. E.E.E. is on the scientific advisory board of DNAnexus. W.K.C. is on the scientific advisory board of the Regeneron Genetics Center. Richard Person and Rebecca Willaert are employees of GeneDx, a wholly owned subsidiary of OPKO Health.

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Web Resources

BrainSpan: Atlas of the Developing Brain, <http://www.brainspan.org/> (accessed 09/22/17)

ExAC, <http://exac.broadinstitute.org/>

gnomAD, <http://gnomad.broadinstitute.org/>

GTEx, <https://www.gtexportal.org/> (accessed 10/26/17)

OMIM, <http://www.omim.org>

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