

## Clinical Genetics Short Report

### **CHEDDA syndrome is an underrecognized neurodevelopmental disorder with a highly restricted *ATNI* mutation spectrum**

#### **Running title: Expanded cohort of CHEDDA syndrome**

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#### **Conflict of Interest Statement**

The authors declare no conflict of interest. IMW is an employee of GeneDx, Inc..

#### **Data Availability Statement**

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## **Abstract**

We describe the clinical features of nine unrelated individuals with rare *de novo* missense or in-frame deletions/duplications within the 'HX motif' of exon 7 of *ATNI*. We previously proposed that individuals with such variants should be considered as being affected by the syndromic condition of Congenital Hypotonia, Epilepsy, Developmental delay, and Digital Anomalies (CHEDDA), distinct from Dentatorubral-Pallidoluysian Atrophy (DRPLA) secondary to expansion variants in exon 5 of *ATNI*.

We confirm that the universal phenotypic features of CHEDDA are distinctive facial features and global developmental delay. Infantile hypotonia and minor hand and feet differences are common and can present as arthrogryposis. Common comorbidities include severe feeding difficulties, often requiring gastrostomy support, as well as visual and hearing impairments. Epilepsy and congenital malformations of the brain, heart, and genitourinary systems are frequent but not universal.

Our study confirms the clinical entity of CHEDDA secondary to a mutational signature restricted to exon 7 of *ATNI*. We propose a clinical schedule for assessment upon diagnosis, surveillance, and early intervention including the potential of neuroimaging for prognostication.

## **Keywords**

Genetics, Rare diseases, Intellectual disability, Genomics, Arthrogryposis, Developmental delay, Neurodevelopmental disorder

## Main text

### Introduction

We recently reported eight unrelated individuals affected by a neurodevelopmental phenotype and multiple congenital anomalies with *de novo* missense and insertion variants within a 16-amino-acid "HX repeat" (where H stands for histidine and X is another amino acid) motif of *ATN1*<sup>1</sup>. We suggested the acronym CHEDDA for this syndrome (Congenital Hypotonia, Epilepsy, Developmental delay, Digital Anomalies) (MIM 618494) to distinguish this condition from the progressive autosomal dominant condition Dentatorubral-Pallidoluysian Atrophy (DRPLA: MIM 607462) caused by polyglutamine expansions in exon 5 of *ATN1* (MIM 125370).

In the original cohort, all eight individuals had a global developmental delay. Five had epilepsy (intractable to polytherapy in two), and six had a distinctive combination of neuroradiological abnormalities including cerebellar hypoplasia, thin corpus callosum, peri-Sylvian polymicrogyria and absent falx cerebri. Two individuals showed dramatic progression of parenchymal atrophy and had profound neurocognitive impairment. Six had visual impairment, mostly cortical. Extra-neurological congenital anomalies were common in this original cohort: including cardiac, renal and vertebral anomalies. Feeding difficulties were common: including dysphagia and gastroesophageal reflux with four out of the eight requiring orogastric tube/gastrostomy, or parenteral nutrition. All probands had a distinctive facial gestalt and many had overlapping toes and palmar crease anomalies<sup>1</sup>.

Herein we describe nine unrelated individuals with *de novo* missense/indel variants in the HX repeat motif, allowing us to confirm CHEDDA as a clinical entity and clarify its phenotype.

### METHODS

Through an international collaborative process, we collected de-identified clinical data from nine other unrelated individuals carrying *de novo* missense and indel/dup variants within a HX repeat motif of *ATN1*. In each participating center, written informed consent was obtained from the individual's legal guardians before genetic testing as approved by relevant local ethical committees. Clinical information was obtained by review of medical records and examination of affected individuals. Average faces were generated using asymmetry preservation and equal representation by individuals approaches previously described<sup>2</sup>.

### RESULTS

The molecular and clinical features of the additional nine individuals are summarised in Table 1.

Compared to the original cohort where all affected individuals had a significant degree of global developmental delay, we report a 2-year-old girl (individual four) who has a duplication of two amino acids, leucine and histidine, within the HX repeat motif of *ATN1* (where H stands for histidine and X stands for another amino acid) and only a mild neurodevelopmental phenotype. Unlike the rest of the expanded cohort, she did not present with congenital hypotonia. Instead, she presented at age 8 months due to concerns with her growth and development. At age 26 months, she can talk in short sentences and walk independently, and with early intervention her cognition was judged to be in the average range, with fine motor skills just below average. She does share the characteristic facial gestalt of CHEDDA and has other common functional features including severe dysphagia and feeding difficulties requiring gastrostomy feeds, and strabismus.

All other children with *de novo* variants in the HX domain in the cohort described herewith, as was the case for previously reported children with a diagnosis of CHEDDA, presented within the neonatal period and have global developmental delay. Four required NICU admission immediately after birth primarily due to significant respiratory complications: they had obstructive and/or central apnea, and three required respiratory support past the neonatal period. The respiratory support could be discontinued for one after jaw distraction ameliorated the micrognathia-related mechanical component and could be discontinued for the other individual after his central tone improved.

Feeding difficulties were significant issues in the children: apparently due to a combination of low tone, gastro-esophageal reflux, and oral phase dysphagia. Five have required gastrostomy feeds although a trial of oral feeding was able to commence in two.

All individuals had delayed gross motor milestones. Other than individual four, no other children are yet walking independently or can communicate verbally. Two children have an ongoing choreoathetoid movement disorder.

This expanded cohort has enabled us to clarify the core facial dysmorphic features of CHEDDA to be sparsity of lateral hairline, prominent ears, and thin upper lip. A composite face was generated highlighting the facial ‘gestalt’ of children with CHEDDA (**Figure 1**).

Those children with abnormalities of tone at birth (all but individual 4) were noted to have overlapping toes and three had single transverse palmar creases. One individual had arthrogryposis, likely reflective of unusual posturing and decreased movement *in utero*.

All nine individuals had visual concerns including cortical visual impairment in three individuals and strabismus requiring corrective surgery in one. Two have required prescription of glasses due to persistent abnormalities in visual acuity. Four individuals have hearing impairment of the conductive and sensorineural type, with one requiring surgery and another utilizing hearing aids.

Genitourinary anomalies in this cohort included one with renal hypoplasia, two with recurrent urinary tract infection and two with cryptorchidism - one bilateral, one unilateral. None had significant congenital cardiac abnormalities requiring surgery. Six individuals had major structural abnormalities on MRI brain (parenchymal atrophy, thin corpus callosum, pontine hypoplasia, cerebellar vermian hypoplasia, and Dandy-Walker spectrum). The Six individuals with abnormalities on MRI were the more severely impaired neurologically with profound global developmental delay in all and epilepsy and cortical visual impairment in four. In comparison, of the three individuals who did not have structural brain abnormalities identified, whilst two had epilepsy they were well controlled on monotherapy, none have cortical visual impairment, and developmental delay was more variable.

## DISCUSSION

This expanded cohort has allowed further delineation of the core phenotypic features characteristic of CHEDDA due to *de novo* variants in ATN1 disrupting the HX repeat motif: namely global developmental delay, feeding difficulties and distinctive facial features (**Table 1** and **Figure 1**). We recommend that common additional features require investigation to guide appropriate early intervention and prognostication (**Table 2**). Given that the absence of structural brain anomalies is suggestive of a better neurodevelopmental outcome, and that cervical stenosis has been identified in two affected individuals with CHEDDA, early neuroimaging may be considered whilst weighing up the potential risks of anesthesia and preferences of the family.

ATN1, a member of the atrophin protein family, is expressed in the brain and other organs, and is understood to act as a nuclear transcriptional regulator with important roles in the control of brain and other organ development<sup>3-7</sup>. All the variants in our original and expanded cohort occur within a HX repeat motif of *ATN1*. Of the genes that contain HX motifs, *REER* (MIM 214800), *AUTS2* (previously known as *KIAA0442*) (MIM615834), and more recently *FBRSL1* (MIM awaited) have also been reported to have rare variants associated with a human neurodevelopmental phenotype<sup>7-10</sup>. *De novo* missense or insertion variants in *REER* are associated with a range of neurodevelopmental phenotypes<sup>2,3</sup> (Neurodevelopmental disorder with or without anomalies of the brain, eye, or heart, MIM 616975), and strikingly the recurrent c.4313\_4318dupTCCACC, p.(Leu1438\_His1439dup), which lies in the HX repeat motif of *REER* is associated with a CHARGE-like multiple malformation syndrome<sup>9</sup>.

The fact that all individuals with a *de novo* variant in *ATN1* reported here and in our original paper<sup>1</sup> have the variant confined to the HX motif confirms the structural and functional significance of this region of the protein. Namely, it confirms the HX motif as a *bone fide* ‘hot spot’, in which *de novo* missense and indel variants which are absent from gnomAD should be considered as likely pathogenic as per ACMG guidelines (PM1)<sup>12</sup>. More studies are required to better understand the underlying pathophysiology of this group of HX repeat motif conditions.

Although the total number of reported individuals with CHEDDA due to variants in the HX motif are limited we note that the individual with the mildest phenotype (Individual 4 in this cohort) is the only affected individual to have an insertion of a histidine and another amino acid [p.Leu1063\_His1064dup]. All other individuals have either an amino acid substitution, or a deletion of one amino acid in this critical region, or insertion of two amino acids, neither of which are histidines. This may result in a less severe ‘dominant negative’ alteration to the 3D structure of the HX motif, as an explanation for her milder phenotype, as has been seen in other conditions due to variants in a restricted protein motif<sup>13</sup>. More clarity on genotypic-phenotypic correlations will be hopefully achieved as the number of identified individuals with CHEDDA expands. On the other hand, we note that our group was approached on several occasions by clinicians or laboratories reporting a *de novo* variant in exon 5 of *ATN1* on exome sequencing, however, on close examination all represented a variation in the CAG repeat region of *ATN1* which varies in the normal population between 6-35 repeats and could be confidently excluded as a causal mechanism in the proband<sup>14</sup>. Thus, care should be taken not to misinterpret normal variation between parents and child in the CAG repeat region in exon 5 of *ATN1* as pathogenic.

We are aware of three unrelated individuals with loss of function (frameshift or nonsense) variants in *ATN1* (personal communications). However, all three individuals lacked the distinctive features of CHEDDA and two had another confirmed pathogenic variant in another gene supportive of an alternative molecular diagnosis. This suggests that *de novo* loss of function variants in *ATN1* are unlikely to be causal of a phenotype, consistent with the presence of at least seven high quality ‘loss of function’ variants in *ATN1* in the database gnomAD (gnomAD v.2.1.1 non neuro) which should be depleted of individuals with a severe pediatric disease<sup>15</sup>.

In conclusion, our expanded cohort confirms a surprisingly restricted mutational spectrum for a gene that has the hallmarks of being intolerant to haploinsufficiency. *ATN1*-related CHEDDA syndrome is a clinically recognizable phenotype and this case series will hopefully contribute to the improved understanding of its natural history, which not only informs management decisions but also establishes a baseline against which future therapies can be evaluated.

## **Figure Legend**

Figure 1: Images of Children with CHEDDA Syndrome

(a) Composite image of faces of children with CHEDDA Syndrome. (b) Individual 1. (c) Individual 2. (d) Individual 3. (e) Individual 5. (f) Individual 6. (g) Individual 7. (h) Individual 8. (i) Individual 9.

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