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Extending the Mutation Spectrum for Galloway–Mowat Syndrome to Include Homozygous Missense Mutations in the *WDR73* Gene

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Abstract

Galloway–Mowat syndrome is a rare autosomal-recessive disorder classically described as the combination of microcephaly and nephrotic syndrome. Recently, homozygous truncating mutations in *WDR73* (*WD repeat domain 73*) were described in two of 31 unrelated families with Galloway–Mowat syndrome which was followed by a report of two sibs in an Egyptian consanguineous family. In this report, seven affecteds from four families showing biallelic missense mutations in *WDR73* were identified by exome sequencing and confirmed to follow a recessive model of inheritance. Three-dimensional modeling predicted conformational alterations as a result of the mutation, supporting pathogenicity. An additional 13 families with microcephaly and renal phenotype were negative for *WDR73* mutations. Missense mutations in the *WDR73* gene are reported for the first time in Galloway–Mowat syndrome. A detailed phenotypic comparison of all reported *WDR73*-linked Galloway–Mowat syndrome patients with *WDR73* negative patients showed that *WDR73* mutations are limited to those with classical Galloway–Mowat syndrome features, in addition to cerebellar atrophy, thin corpus callosum, brain stem

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SUPPORTING INFORMATION

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hypoplasia, occasional coarse face, late-onset and mostly slow progressive nephrotic syndrome, and frequent epilepsy.

Keywords

Galloway–Mowat syndrome; cerebellar atrophy; coarse face; *WDR73*; nephrotic syndrome

INTRODUCTION

Galloway–Mowat syndrome (GMS; OMIM 251300) was first described in 1968 in two siblings presenting with nephrotic syndrome, microcephaly, and hiatal hernia [Galloway and Mowat, 1968]. Subsequently, more than 60 cases with similar clinical features have been described, significantly expanding the clinical phenotype to include central nervous system anomalies, epilepsy, optic nerve hypoplasia, low birth weight, intrauterine growth retardation, and hypothyroidism, mostly with an autosomal recessive inheritance pattern.

Previous reports have suggested three distinct clinical presentations of patients with microcephaly and nephrotic syndrome, based upon the presence of cerebral cortical gyral abnormalities, age of onset, progression, steroid-responsiveness of nephrosis, developmental delay, and the presence of skeletal dysplasias—the first is a classical presentation of Galloway–Mowat syndrome, a second syndrome of microcephaly, nephrotic syndrome, and developmental delay (MNSDD), and a third syndrome of microcephaly, developmental delay, and spondylorhizomelic short stature [Meyers et al., 1999]. Recent reports have recognized that GMS is clinically heterogeneous, and should be considered in children even in the absence of renal involvement at presentation [Sano et al., 1995; Meyers et al., 1999; Colin et al., 2014].

Renal abnormalities in GMS range from isolated proteinuria to steroid resistant nephrosis, which may also progress to end-stage kidney disease, multiple organ failures, and contribute to mortality. Although the early onset form of nephrotic syndrome is more common, late-onset has also been reported [Steiss et al., 2005]. Renal biopsies have revealed various histopathology including diffuse mesangial sclerosis, focal segmental glomerulosclerosis, mesangial proliferation, increased mesangial matrix, and minimal change disease [Meyers et al., 1999; Steiss et al., 2005; Sartelet et al., 2008].

Microcephaly is reported at the time of birth or noted later in life, frequently accompanied by cerebellar and corpus callosal hypoplasia, neural migration defects, and other brain malformations. Epilepsy and intellectual disability are also common [Meyers et al., 1999; Pezzella et al., 2010; Ekstrand et al., 2012]. Prenatal and perinatal findings include low birth weight, intrauterine growth retardation, oligohydramnios, mild facial dysmorphisms that can largely be attributed to the decrease in head circumference, hiatal hernia, joint contractures, and eye malformations such as microphthalmia, optic atrophy, and corneal opacities [Hou and Wang, 1995; Meyers et al., 1999; Chen et al., 2011].

Recently, homozygous protein truncating mutations in *WDR73* were identified in two of 31 families meeting criteria for GMS. *WDR73* maps to chromosome 15q25.2 and encodes a

378 amino acid protein with six WD40 repeats that form a six-bladed propeller structure. Proteins that contain WD40 repeat domains are predicted to function as scaffolds for the assembly of protein complexes. *WDR73* is expressed in brain and kidney, localizing to spindle poles during mitosis, suggesting a critical role in the maintenance of cell architecture and cell survival. Among the non-cortical regions, the highest expression was observed in the cerebellar cortex. Moreover, morpholino injected zebrafish embryos showed signs of developmental delay with a curved tail region, reduced head size, and brain morphology defects such as poorly expanded and differentiated midbrain and cerebellum. Moreover, fewer proliferating progenitor cells were observed in morphant embryos. The data points to a role for zebrafish *wdr73* in regulating either neural progenitor cell cycle length, or promoting cell cycle exit, and neuronal differentiation [Colin et al., 2014; Ben-Omran et al., 2015]. This study examines the prevalence of *WDR73* mutations in a cohort of patients with neurodevelopmental disorders, and refines the clinical spectrum of *WDR73*-related GMS.

CLINICAL DATA

Family 1486 was of Palestinian origin and had two similarly affected children from a first-cousin marriage (Fig. 1A). Patient II-1 was a female born at term by NSD (normal spontaneous delivery) after an uneventful pregnancy, with normal birth weight and OFC (occipito-frontal circumference) by report. She was referred for evaluation at 6 months of age due to weakness and poor feeding. Physical exam demonstrated severe hypotonia with poor head control. Seizures began at age 2 years with arm flexing and crying. She received multiple trials of antiepileptic medications including phenobarbital and valproate, which did not alter the frequency or intensity. MRI revealed a thin corpus callosum and severe cerebellar atrophy.

The family immigrated to the United States at age 5 years. Evaluation demonstrated severe microcephaly with an OFC of 44.5 cm (−7/−6 SD). She did not visually track, sit, reach, speak, or make purposeful movements. She had diffuse hypertonia and 3+ brisk deep tendon reflexes, with contractures of elbows and ankles bilaterally.

MRI (Magnetic resonance imaging) revealed a thin corpus callosum, mildly enlarged ventricles, and significant cerebellar atrophy (Fig. 1C). Workup included karyotype of 46,XX, and normal acylcarnitines, CPK (creatine phosphokinase), and mass spectrometric analysis of transferrin. Other laboratory findings revealed mild anemia, hypothyroidism and borderline lactate, pyruvate, and creatine levels with the presence of proteinuria. She was diagnosed with nephrosis at about age 4, which progressed in spite of steroid treatment. Renal ultrasound demonstrated bilateral enlarged kidneys (12 cm diameter) and increased echogenicity of the cortex, without evidence of hydronephrosis. Serum creatinine level was 1.9 mg/dl and BUN 80 mg/dl, suggesting impaired clearance. Renal biopsy revealed collapsing variant of focal segmental glomerular sclerosis (Fig. 1D). ANA and ANCA were negative. She expired at 2–5 months of age due to cardiac and respiratory arrest, secondary to enterocolitis, bowel necrosis and renal failure.

Her sister (II-3) was born at 39 weeks gestation by NSD, with normal growth parameters. At 2 months the family noted hypotonia and lack of visual tracking. At 6 months OFC was 42

cm (-1 SD). MRI demonstrated diffuse white matter volume loss with vermis hypoplasia and thinning of the corpus callosum (Fig. 1C). At 15 months OFC was 44.5 cm ($-3/-2$ SD) and weight of 9.3 kg ($-2/-1$ SD). She had poor head control, axial hypotonia, and could not sit without support. She had exotropia and did not visually track. She had no words and minimal vocalization. Neurological exam was significant for appendicular hypertonia and 3+ deep tendon reflexes in the upper and lower extremities with persistent Moro response. Eye exam revealed intermittent alternating exotropia, partial optic atrophy, cortical visual impairment, and temporal optic disc pallor. Renal ultrasound and metabolic screening have remained normal until age 1.5 years.

Family 2027 from Egypt had two similarly affected siblings from a first-cousin marriage. The family history was otherwise unremarkable, with one healthy older sibling (Fig. 1A). Patient II-2 was born at term by NSD after an uneventful pregnancy. She came to attention at 9 months of age due to unsteadiness in sitting. Her physical exam demonstrated hypotonia, truncal ataxia, and oculomotor apraxia. She had a coarsened facies with full lips and cheeks, broad nose with fleshy tip, prominent forehead, and frontal hirsutism (Fig. 1B). MRI demonstrated cerebellar atrophy and deep white matter changes, especially in the occipital region (Fig. 1C). Re-examination at 3 years revealed microcephaly (OFC: 43.5 cm, $-6/-5$ SD) with moderate developmental delay, absent speech, and significant truncal ataxia. She was without detectable proteinuria.

Her brother (II-3) was born NSD at term, with noted hypotonia. Examination at 2 years showed an OFC of 46 cm ($-4/-3$ SD), without seizures, but with lack of independent walking and with truncal ataxia. Facial features were similarly coarsened (Fig. 1B), and there was no speech. MRI revealed cerebellar and frontotemporal cerebral atrophy (Fig. 1C). Abdominal ultrasound, and renal chemistries, and urine protein testing were normal.

The patient from family 2286 was born at 37 GW by NSD following treatment for premature labor to double first-cousin parents. Family history was unremarkable except for a healthy sibling who died of SIDS (sudden infant death syndrome) at 1 year of age, and another sibling with developmental delay (but without microcephaly) who died of acute infection at 2 years of age (Fig. 1A). The child was evaluated at 9 months due to night time seizure episodes and increasing muscle tone. He had global developmental delay, poor head control, and was unable to sit. Physical exam revealed full lips and cheeks, a broad nasal root, hirsutism, and a broad forehead (Fig. 1B). Motor examination demonstrated hypotonia with brisk reflexes, cranial MRI showed cerebellar atrophy and thin corpus callosum (Fig. 1C). Metabolic and biochemical investigations revealed proteinuria. EEG at 4 years documented generalized epileptiform activity, treated with carbamazepine therapy.

At 11 years, he had a height of 117 cm ($-4/-3$ SD), weight 27 kg (-2 SD), and OFC 47 cm (-5 SD). He was able to sit without support but was unable to independently ambulate or stand. Speech comprehension and production was lacking. Proteinuria progressed, resulting in edema of the face and extremities. Renal biopsy demonstrated diffuse mesangial sclerosis and an abdominal ultrasonography documented smaller than average kidneys. Additional studies revealed normal hearing and vision. Chromosome analysis yielded a normal male karyotype (46,XY).

Family 2889, also from Egypt, presented two affected siblings from a first cousin marriage, with unremarkable family history (Fig. 1A). Patient II-3 was born by NSD at term, with unknown growth parameters. Evaluation at 8 years showed severe global developmental delay and microcephaly (OFC: 45.5 cm, $-7/-6$ SD). She could not walk, could sit only with support and had not developed speech. There was central hypotonia with spasticity and brisk peripheral reflexes, coarsened facial features with full lips and cheeks, hirsutism and a very broad nasal root with a fleshy tip. She also had bushy eyebrows, bi-temporal narrowing, and a long and smooth philtrum (Fig. 1B). Cranial MRI revealed severe cerebellar atrophy, mild brainstem hypoplasia, deep white matter changes especially pronounced in the occipital area, and general cerebral atrophy (Fig. 1C). A 24-hr urine collection showed elevated protein levels.

Her sister (II-4) was also born at term by NSD. At age 2 she had OFC 42 cm ($-6/-5$ SD), was unable to sit independently and had not developed speech or communication. Her facial features, although similar to her older sister, were less coarsened (Fig. 1B). She had central hypotonia with spasticity in her extremities and brisk deep tendon reflexes. MRI demonstrated cerebellar atrophy with brain stem hypoplasia and a thin corpus callosum (Fig. 1C). Renal evaluation demonstrated elevated protein levels in the urine.

MATERIALS AND METHODS

Patient Ascertainment

Patients presenting with features of neurodevelopmental delay or regression, ataxia, intellectual disability, autism, epilepsy, or structural brain malformations, were recruited from child neurology clinics throughout the Middle East, North Africa, and Central Asia between the years of 2004 and 2014. Recruitment was focused in the major population centers of the Middle East including Morocco, Libya, Egypt, Saudi Arabia, Kuwait, UAE, Oman, Jordan, Pakistan, Turkey, and Iran with consanguinity rates (i.e., rate of marriage between first or second cousins) of approximately 50% compared with $<1\%$ in US and Western Europe. Among the recruited cohort, consanguinity was present in 63% of parents, suggesting some bias in sampling towards those with affected children due to recessive disease. Consenting and sampling was performed on both parents and all available genetically informative siblings to include affected and unaffected members, as well as extended family members if appropriate, consistent with IRB guidelines approved by the ethical committee and according to the Declaration of Helsinki. General and neurological examination, clinical records, radiographs, photographs, videos documenting movement, and past history were reviewed and patients were examined by one or more of the authors. Blood and/or saliva was collected on all consenting potentially informative family members. DNA was extracted with the Qiagen AutoPure instrument, and was subjected to quality control measures for concentration/purity and to confirm inheritance for subsequent genetic investigation.

Whole Exome Sequencing

WES (whole exome sequencing) was performed on two affected members per family when available, or both parents and affected member from singleton cases. Genomic DNA was

subject to Agilent Human All Exon 50 Mb kit library preparation, then paired-end sequencing (2×150 bp) on Illumina HiSeq 2000 instrument. For each patient sample, >90% of the exome was covered at >30 $\times$. Sequences were aligned to hg19 and variants identified through the GATK pipeline [Van der Auwera et al., 2013]. Variations were annotated with in-house software and the SeattleSeq server [Dixon-Salazar et al., 2012]. All variants were prioritized by allele frequency, conservation, and predicted effect on protein function. Variants in *WDR73* were analyzed for segregation with disease within family members by Sanger sequencing of all informative available members.

Sanger Sequencing

Primers were designed using the Primer3 program and tested for specificity using NIH BLAST software. PCR (polymerase chain reaction) products were treated with Exonuclease I (Fermentas, Vilnius, Lithuania) and Shrimp Alkaline Phosphatase (USB Corporation, Cleveland, OH) and sequenced using BigDye terminator cycle sequencing Kit v.3.1 on an ABI 3100 DNA analyzer (Applied Biosystems, Carlsbad, CA). Sequence data were analyzed by Sequencher 4.9 (Gene Codes Corporation, Ann Arbor, MI) to test segregation of the mutation with the disorder under a recessive mode of inheritance, taking advantage of all informative meioses in each family.

Modeling

Three-dimensional models were predicted using the Iterative threading assembly refinement (I-TASSER) server that utilizes a combination of homology modeling and threading algorithms. The structures were evaluated with Protein structure analysis (ProSA) for global and local model evaluations, visualized in PyMol. Electrostatic surfaces were generated with the APBS plugin for PyMol.

RESULTS

Exome sequencing revealed homozygous missense mutation (c.287 G>A, p.Arg96Lys) in the affected from families 1486 and 2286; and another homozygous missense mutation (c.602 G>A, p.Gly201-Glu) in affected from families 2027 and 2889. All mutations were confirmed by Sanger sequencing of all samples from each family to both validate the variant, as well as and confirm that the mutation segregated according to a recessive model with full penetrance. Tested parents were confirmed to be heterozygous carriers, and unaffected siblings in the families were confirmed as either carriers or wild-type (Suppl. Fig. S1). These variants were not seen in our in-house exome database containing more than 1,800 individuals for whom whole-exome sequencing is available (of these 1,000 are of matching Arab/Middle East descent), nor was it evident in the NHLBI Exome Variant server or in any public SNP database.

WDR73 has a β -propeller topology, containing 25- β sheets and 1- α helix at the N-terminus as predicted by both secondary structure analysis programs (SCRATCH, PsiPred, JPred) as well as predicted by tertiary structure. Visualization in PyMol revealed the location of the altered residue. Alignment of the predicted structures revealed conformational differences between the wild-type protein, the p.Arg96Lys protein, and the p.Gly201Glu protein (Fig.

2), including discordance between the beginning and end of structural motifs as well as the rotation of bonds of the amino acids. Further analysis of the electrostatic surfaces of the proteins revealed shifts of electrostatic potential on the outside of the structure.

In the p.Gly201Glu model, the residue occurs inside of a β sheet located on the outside of the molecule where most of the protein–protein specificity probably occurs. Within the β sheet, there was a predicted rotation of the bonds where the sheet became less twisted without the increased rotational mobility of the glycine. Furthermore, there was a large difference of electrostatic potential on the outside of the molecule due to the introduction of the acidic side chains of the new residue.

In the p.Arg96Lys model, the lysine did not fall within a β sheet, but instead on a coil. Although in the wild-type protein residue 96 was also a basic residue, the conformation of the mutant protein presented a positive potential closer to the central axis—the presumed active site of the protein. Analysis of electrostatic potential revealed alterations of charge on the outside of the propeller, suggesting disruptions between specific protein–protein interaction domains of the molecule.

In our cohort were 13 additional consanguineous families with one or more affected individual with microcephaly and a renal phenotype, ranging from isolated proteinuria to nephrotic syndrome. No mutations in *WDR73* that would account for their symptoms were encountered, and all had clinical features that would distinguish them from the patients with *WDR73* mutations (i.e., microcephaly only -2 SD by mid-childhood, absence of severe intellectual disability, presence of renal symptoms earlier or more severe or of different type). The average onset of nephrotic syndrome in this cohort was 1.8 years that was significantly earlier than the *WDR73* positive cases. Furthermore, among 467 individuals with microcephaly without renal features, on whom exome sequencing was performed, no mutations in *WDR73* that would account for their symptoms were encountered, suggesting that the clinical features are both sensitive and specific to *WDR73* mutations.

DISCUSSION

We report the confirmation of homozygous mutations in *WDR73* in seven patients that are consistent with a diagnosis of GMS, and clarify the phenotypic spectrum (Table I). Our approach was different from traditional position cloning approaches, in that we recruited a large and clinically diverse cohort with neurodevelopmental presentations. Taking a “sequence first” approach, we screened this cohort with exome sequencing, and identified patients with homozygous deleterious *WDR73* mutations. This approach may be more likely to identify diverse and partially penetrant phenotypic features, because recruitment was not limited only to those presenting with specifically defined GMS syndromic features. Indeed, several of the patients with *WDR73* mutations did not show evidence of proteinuria until the current time, and were not diagnosed with GMS at the time of sequencing, but all patients with mutations over the age of 5 years showed renal involvement.

Among *WDR73*-mutated individuals was the uniform presence of progressive microcephaly, often with normal OFC at birth, then ranging from -3 to -7 SD in the first decade of life,

indicating slowed postnatal growth. Minimal differences in OFC were observed in siblings, however, differences in microcephaly percentiles were evident between families indicating that environmental and nutritional factors, as well as familial/population-level growth potentials, may influence the underlying defect.

Cerebellar atrophy seen in MR imaging is also consistent within our series, and correlated with symptoms of truncal ataxia, hypotonia, and dysmetria in some individuals. Epilepsy was variable, as was optic atrophy. In previous reports of GMS, cerebellar atrophy was an infrequent finding [Steiss et al., 2005]. Therefore the presence of cerebellar atrophy in all seven of our patients, and thus all 12 patients reported so far with *WDR73* positive GMS is a notable predictive feature.

Other common pathologies observed in cranial MRI were thinning of corpus callosum in half of the patients and brain stem hypoplasia in 80%. These findings were reported only rarely in GMS patients [Pezzella et al., 2010]. These frequent additional findings may also be considered features of *WDR73*-mutated patient when GMS is in the differential diagnosis.

Epilepsy was encountered in seven of the twelve patients with *WDR73* mutations reported to date, making it a common clinical feature of this group. However, the onset, type, and frequency of the seizures as their response to therapy varied greatly between the patients as well. There was also intrafamilial variability as one sibling in the family suffered from epilepsy while the other was seizure-free. Epilepsy was also observed previously in some patients with GMS, prior to gene discovery [Pezzella et al., 2010].

There were no hiatal hernia cases in the *WDR73*-linked patients reported so far, which was one of the features observed in the original siblings reported by Galloway and Mowat [1968]. Although the number of patients with *WDR73* is not large enough to exclude mutations in patients with hiatal hernia, the total absence of hiatal hernia so far should be taken into consideration while making diagnostic decisions.

Another interesting observation was the presence of coarse facial features in five patients from three families (2027, 2286, and 2889) in our cohort. These features were not dissimilar to the previously described typical microcephalic faces encompassing sloping forehead, pinched nose, micrognathia, and large ears. Facial dysmorphisms were not reported in the first published *WDR73*-mutated patients [Colin et al., 2014], but we communicated with the authors and confirmed coarsened facies, full lips and cheek, bushy eyebrows, wide nasal root, and bulging forehead in siblings of family A. Also, the other two Egyptian siblings had coarse facial features as well, [Ben-Omran et al., 2015] suggesting that nine out of twelve patients had these particular facies.

All *WDR73*-mutated patients displayed severe intellectual disability in contrast to variable intellectual disability of GMS patients reported prior to gene discovery [Pezzella et al., 2010]. The difference in severity may be attributed to the effects of overlapping epilepsy, additional neurodevelopmental brain anomalies, and optic atrophy in this subgroup. We hypothesize that the more pronounced structural brain defects seen in *WDR73*-mutant GMS patients correlates with the degree of severity of intellectual disability.

Microcephaly with nephrotic syndrome has historically been the major GMS diagnostic criteria, yet in the *WDR73*-mutation positive group, eight out of twelve patients had proteinuria with onsets varying from 2 years to 8 years of age (median age 5), later on average compared with the total of >60 GMS cases reported in the literature [Sano et al., 1995]. Four of these patients had renal biopsies, two with collapsing type focal segmental glomerulosclerosis (FSGS), one with non-collapsing FSGS, and one diffuse mesangial sclerosis pathology. Two patients have died due to nephrotic syndrome related complications, both of which were rapidly progressive cases after the initial onset. The rest of the patients are still being followed for proteinuria and renal insufficiency keeping in mind that symptoms can appear later in life and progress after onset.

As a summary, the limited number of patients within *WDR73* positive GMS is characterized by microcephaly, cerebellar atrophy, severe intellectual disability, and frequent abnormalities such as thin corpus callosum, brain stem hypoplasia, epilepsy, occasional coarse face, and a late-onset and mostly slow progressing nephrotic syndrome. These features can help distinguish *WDR73*-positive patients from the general GMS patient group, thereby facilitating clinical and molecular diagnoses in the outpatient clinics.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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FIG. 1.

(A) Pedigrees. Pedigrees showing autosomal recessive inheritance pattern in families. Individual 2286-II-7 shown with gray shading is not affected similarly. (B) Gestalt of the patients. Facial pictures of the affecteds showing coarse facial features such as full lips and cheeks, broad forehead, bushy eyebrows, flat nasal root with fleshy nasal tip. (C) Brain imaging studies. MR findings of affected individuals showing cerebellar atrophy, and thin corpus callosum [arrows] and brain stem hypoplasia [arrowheads]. (D) Histopathological studies. Histological slide from the renal biopsy of 1486-II-3 showing glomerular changes illustrating the collapsing variant of focal segmental glomerular sclerosis. (200× magnification, Jones silver stain-top, PAS stains-middle and bottom). [Color figure can be seen in the online version of this article, available at <http://wileyonlinelibrary.com/journal/ajmga>].

FIG. 2.

Protein modeling. (A) Comparison of the wild-type protein and the predictions of the outcome of the missense mutations. (B) Comparison of the analyses of electrostatic potential of the wild-type and the mutated proteins. [Color figure can be seen in the online version of this article, available at <http://wileyonlinelibrary.com/journal/ajmga>].

TABLE I

Comparison of the Clinical Manifestations With Patients Reported With WDR73 Mutations (A-II-3, A-II-4, and B-II-3) from Colin et al. [2014] and (II-4 and II-6) Ben-Omran et al. [2015]

	A-II-3	A-II-4	B-II-3	II-4	II-6	1486-2-1	1486-2-3	2027-2-2	2027-2-3	2286-2-5	2889-2-3	2889-2-4
Gender	M	M	M	F	F	F	F	F	M	M	F	F
Origin	Morocco	Morocco	Turkish	Egypt	Egypt	Palestinian	Palestinian	Egypt	Egypt	Syria	Egypt	Egypt
Consanguinity	-	-	+	+	+	+	+	+	+	+	+	+
Mutation type	Nonsense	Nonsense	Nonsense	Nonsense	Nonsense	Missense	Missense	Missense	Missense	Missense	Missense	Missense
Age	5 years	7 years	13 years	11 years	4 years	5 years	1.5 years	3 years	2 years	11 years	8 years	2 years
Microcephaly	+ (-3 SD)	+ (-2,5 SD)	+ (-3 SD)	+ (-1,8 SD)	+ (-1,15 SD)	+ (-7/-6 SD)	+ (-6/-5 SD)	+ (-6/-5 SD)	+ (-4/-3 SD)	+ (-5 SD)	+ (-7/-6 SD)	+ (-6/-5 SD)
Epilepsy	+	+	-	+	+	+	-	-	-	+	+	-
Cerebellar atrophy	+	+	+	+	+	+	+	+	+	+	+	+
Thin corpus callosum	+	-	+	+	+	(w/pons) +	+	-	-	-	+	-
Brain stem hypoplasia	+	-	+	+	+	+	+	-	+	+	+	+
Coarse facies	-	-	-	+	+	-	+	-	+	+	+	+
Optic atrophy	+	+	+	+	+	+	-	-	-	-	-	-
Hiatal hernia	-	-	-	-	-	-	-	-	-	-	-	-
Intellectual disability	+ (Severe)	+ (Severe)	+ (Severe)	+ (Severe)	+ (Severe)	+ (Severe)	+ (Severe)	+ (Severe)	+ (Severe)	+ (Severe)	+ (Severe)	+ (Severe)
Proteinuria	+	-	±	+	+	+	-	-	-	+	+	+
Histology	FSGS (collapsing)	-	FSGS	N/A	N/A	FSGS (collapsing)	N/A	N/A	N/A	Diffuse mesangial sclerosis	N/A	N/A

F, female; FSGS, focal segmental glomerulosclerosis; M, Male, N/A, not available; SD, standard deviation; w/, with.