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Congenital myasthenic syndrome caused by a frameshift insertion mutation in *GFPT1*

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Abstract

Objective

Description of a new variant of the glutamine-fructose-6-phosphate transaminase 1 (*GFPT1*) gene causing congenital myasthenic syndrome (CMS) in 3 children from 2 unrelated families.

Methods

Muscle biopsies, EMG, and whole-exome sequencing were performed.

Results

All 3 patients presented with congenital hypotonia, muscle weakness, respiratory insufficiency, head lag, areflexia, and gastrointestinal dysfunction. Genetic analysis identified a homozygous frameshift insertion in the *GFPT1* gene (NM_001244710.1: c.686dupC; p.Arg230Ter) that was shared by all 3 patients. In one of the patients, inheritance of the variant was through uniparental disomy (UPD) with maternal origin. Repetitive nerve stimulation and single-fiber EMG was consistent with the clinical diagnosis of CMS with a postjunctional defect. Ultrastructural evaluation of the muscle biopsy from one of the patients showed extremely attenuated postsynaptic folds at neuromuscular junctions and extensive autophagic vacuolar pathology.

Conclusions

These results expand on the spectrum of known loss-of-function *GFPT1* mutations in CMS12 and in one family demonstrate a novel mode of inheritance due to UPD.

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UCLA Clinical Genomics Center coinvestigators are listed in the appendix 2 at the end of the article.

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Glossary

C4RCD = Center for Rare Childhood Disorders; **CMS** = congenital myasthenic syndrome; **GFPT1** = glutamine-fructose-6-phosphate transaminase 1; **LCRH** = long contiguous regions of homozygosity; **NEB** = nebulin; **NMJ** = neuromuscular junction; **RNS** = repetitive nerve stimulation; **SMA** = spinal muscular atrophy; **TA** = tubular aggregate; **UPD** = uniparental disomy; **WES** = whole-exome sequencing.

Congenital myasthenic syndrome (CMS) is a genetically and clinically heterogeneous group of disorders with compromised neuromuscular transmission. Common underlying symptoms are fatigable skeletal muscle weakness confined to the ocular, bulbar, or limb-girdle muscles.¹ Classification of CMS is based on inheritance pattern and localization of defects at the neuromuscular junction (NMJ).^{1,2} To date, over 30 genes have been identified to cause CMS, which are functionally critical for the development and maintenance of NMJ.^{3,4} Mutations in genes involved in the N-linked glycosylation pathway have also been identified to cause CMS. Genetic linkage analysis and whole-exome sequencing (WES) experiments have identified biallelic mutations in the gene encoding glutamine-fructose-6-phosphate transaminase 1 (*GFPT1*, Online Mendelian Inheritance in Man [OMIM]: 138292) as the cause of CMS type 12 (OMIM:610524). *GFPT1* enzyme converts fructose-6-phosphate to glucosamine-6-phosphate through a rate-limiting transaminase reaction that regulates the biosynthesis of N-acetylglucosamine and protein glycosylation.^{2,5} Most of the biallelic *GFPT1* gene mutations that cause CMS12 are missense, nonsense, or frameshift with protein truncation, and also a recurrent 3'-untranslated region mutation has also been reported.^{2,5,6} Patients with *GFPT1* mutation display characteristic symptoms such as limb-girdle weakness and tubular aggregates (TAs) in skeletal muscle fibers.⁵ Patients also show evidence of decremental response to repetitive nerve stimulation (RNS), indicating NMJ dysfunction.⁷ In this study, we present clinical, neurophysiologic, histopathologic, and genetic findings in 2 unrelated families with 3 affected children diagnosed with CMS12 who share the same loss-of-function *GFPT1* variant. This report expands on current knowledge of the effects of pathogenic *GFPT1* variants.

Methods

Standard protocol approvals, registrations, and patient consents

Family A was enrolled in the research protocol at the Center for Rare Childhood Disorders (C4RCD). The protocol was reviewed and approved by the Western Institutional Review Board (Protocol #20120789). Informed consent for sequencing and data usage was obtained from each participant or their guardian for children younger than the age of 18. Patient A1 had clinical evaluation at C4RCD. Patients from family B were evaluated at the Department of Neurology, The University of California, Los Angeles (patient B1), and Providence Medical Group (patient B2).

Whole-exome sequencing

Patient A1 had trio WES and analysis at TGen. Sequence data were analyzed using in-house-developed pipeline that included alignment, quality filtering, single nucleotide variant and indel calling, copy number analysis, variant annotation, and filtering.⁸ The annotated variant list was filtered using genetic models including de novo, homozygous recessive, compound heterozygous, and X-linked. Candidate variants were prioritized based on population frequency, in-silico predictions, and clinical phenotype. Candidate variants were validated by Sanger sequencing.

Patient B1 had clinical WES at The University of California, Los Angeles Clinical Genomics Center as a singleton.⁹ Patient B2 underwent targeted *GFPT1* gene sequencing by Pacific Biosciences.

Neurophysiologic studies

Nerve conduction, EMG, and repetitive stimulation studies were performed on patient A1. Patient B1 had single fiber EMG on the left extensor digitorum communis.

Data availability

Variant level data will be submitted and made available in ClinVar.

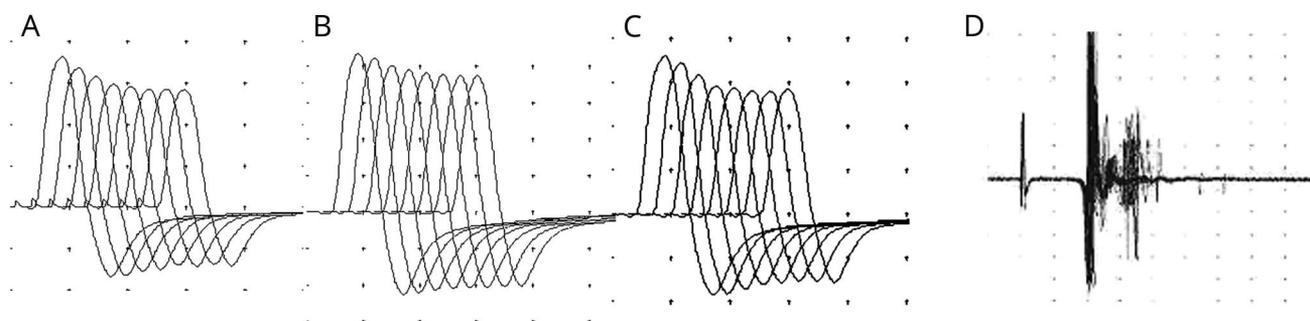
Results

Clinical history

Family A, patient A1

This is a 11-year-old Hispanic girl with severe muscle weakness since birth. She was born at term by emergency C-section because of fetal heart rate irregularities. She required bag-mask ventilation, was electively intubated, and required mechanical ventilation. A gastrostomy tube was placed at 1 month of age. Physical examination revealed severe hypotonia, weakness, decreased spontaneous movement, and areflexia. At 5 months of age, she demonstrated severe head lag with hypotonia. Fluctuating responses were seen with deep tendon reflexes. An examination at 2.5 years revealed significant diffuse hypotonia, severe muscle weakness, facial diplegia, minimal to no movement of her shoulders and proximal legs, flabby muscles, severe head lag, and hyporeflexia. She had severe scoliosis, requiring the use of a neck collar and thoracic brace. Based on her EMG findings (figure 1), she was treated with pyridostigmine and had a partial response. At 3 years of age, she underwent tracheostomy and was treated with 3,4-diaminopyridine together with pyridostigmine. The addition of 3,4-diaminopyridine did not result in significant clinical

Figure 1 Clinical neurophysiology



Decremental response on 3 Hz repetitive nerve stimulation of hand muscle of patient A showing a postjunctional neuromuscular deficit by postactivation facilitation and then exhaustion; decrement of 13% at rest (A), 7% immediately after 30 seconds exercise (B), and 15% at 360 seconds after the exercise (C). Stimulated (15 c/s) single-fiber EMG study on extensor digitorum communis muscle on patient B1 demonstrating a large jitter on the last unit (D).

improvement and was discontinued because of changes in her ECG. There was no family history of muscle disorders, and both parents were unaffected. Previous molecular testing included a chromosomal microarray, which revealed 2 long contiguous regions of homozygosity (LCRH) of 57.6, and 89.8 Mbases on chromosome 2p21q11.2 and 2q22.1q36.3, respectively. Genetic testing was negative for spinal muscular atrophy (SMA), Prader-Willi syndrome, congenital myotonic dystrophy, and the genes linked to CMS known before year 2010 (table 1). Targeted testing for *SURF1*, *SCO1*, *SCO2*, and *COX10* mutations was negative.

Family B, patient B1

This is an 8-year-old boy of Mexican ancestry with congenital hypotonia, low muscle bulk, dysphagia requiring gastrostomy

tube feeding, recurrent respiratory infections, and with a tracheostomy. He was born at 39 weeks gestation to a 34-year-old G9 P7 mother via vaginal delivery after an uncomplicated pregnancy. At initial evaluation, he was noted to be floppy, with a sluggishly reactive left pupil. He eventually underwent gastrostomy tube placement and tracheostomy and needed mechanical ventilation. At 1 month, he was found to have bilateral cryptorchidism, micrognathia, a high arched palate without evidence of cleft, head lag, marked decrease in proximal tone, and absent deep tendon reflexes. At 10 months of age, he weighed 3.6 kg and was bright and alert. There was minimal spontaneous movement, minimal movement of the arms, and no movement of the legs at all. He slipped through at the shoulders on vertical suspension and had a severe head lag. Tone was slightly better in

Table 1 Variants associated with CMS and ruled out

Patient	GT	HGNC	HGVS.c	HGVS.p	SIFT	Polyphen	Consequence	gnomAD MAF (%)	Note
B1	0/1	<i>NEB</i>	c.18660G>C	p.Leu6220Phe	Tolerated	Benign	Missense	0.02925	Comp het
B1	0/1	<i>NEB</i>	c.5696C>T	p.Thr1899Ile	Tolerated	Probably damaging	Missense	0.04445	Comp het
B1	0/1	<i>CHRND</i>	c.117C>G	p.Asn39Lys	Tolerated	Benign	Missense	0.5509	High population frequency
B1	0/1	<i>LAMB2</i>	c.4751A>G	p.Glu1584Gly	Tolerated	Benign	Missense	0.00041	Het for autosomal recessive disorder
A1	0/1	<i>MUSK</i>	c.2553T>A	p.Ser851Arg	Deleterious	Possibly damaging	Missense	—	—
A1	0/1	<i>CHAT</i>	c.1682G>A	p.Arg561Gln	Tolerated	Probably damaging	Missense	0.75	—
A1	0/1	<i>RAPSN</i>	c.614G>A	p.Arg205Gln	Tolerated	Benign	Missense	1.585	High population frequency
A1	0/1	<i>SCN4A</i>	c.404T>C	p.Met135Thr	Tolerated	Benign	Missense	0.15	—

Abbreviations: CMS = congenital myasthenic syndrome; comp het = compound heterozygous; GT = genotype; HGNC = HUGO Gene Nomenclature Committee; HGVS.c = Human Genome Variation Society coding DNA reference sequence position; HGVS.p = HGVS protein reference sequence position; MAF = minor allele frequency; *NEB* = nebulin; SIFT = Sorting Intolerant from Tolerant.

the legs than the arms. At the age of 2 years, he was able to breathe on his own, requiring intermittent mechanical ventilation during hospitalizations for respiratory infections. At the age of 8 years, he wore a neck brace and thoracolumbosacral orthosis. He had a tracheostomy tube, ate pureed food by mouth, and received supplemental nutrition by G-tube. The patient had normal speech and intelligence and attended 2nd grade in a special needs school. His medications included budesonide, cholecalciferol, guar gum, levalbuterol, and docusate. He started 3,4-diaminopyridine at the age of 8 years, without benefit. Previous molecular testing included a normal karyotype, negative testing for Prader-Willi, SMA, and congenital myotonic dystrophy. Enzymatic and metabolic assays for creatine kinase, thyroid function, serum cortisol, plasma ammonia, quantitative amino acid profile, very long chain fatty acids, pyruvate, lactate, and acylcarnitine profile were all normal.

Family B, patient B2

This boy is the younger sibling of patient B1, and he died at the age of 2 years. He was born at 37-1/2 weeks of gestation after a pregnancy complicated by gestational diabetes and hypertension. He was intubated immediately after delivery and required resuscitation with chest compression. In the neonatal intensive care unit, he was started on a therapeutic hypothermia protocol for presumed hypoxic-ischemic encephalopathy. On initial evaluation, EEG was abnormal (presumably because of hypothermia) but showed no evidence of seizures and improved after rewarming. Echocardiogram and renal ultrasound were normal. Head ultrasound revealed grade 1 germinal matrix hemorrhage, and brain MRI showed left germinal matrix and intraventricular hemorrhage. He had a broad nasal bridge, ptosis, micrognathia, a tented upper lip, high and narrow palate, slightly wide set nipples, and descended testes. He also had loose skin on his neck and around his thighs. His torso appeared long, and there was a limitation of elbow extension to approximately 150°, and he had diffusely decreased muscle tone and absent tendon reflexes. Some benefit from pyridostigmine treatment was observed.

Neurophysiologic findings

Clinical neurophysiology

Patient A1

Nerve conduction studies at the age of 2 years revealed low compound muscle action potentials, especially in the trapezius and facial muscles. EMG showed rapid recruitment of motor units suggestive of a myopathy. There was no spontaneous activity in limb muscles. A 13% decremental response to 3 Hz RNS was seen only in the hand muscles and not in the trapezius muscle. At the age of 6 years, the RNS revealed postactivation facilitation and postexercise exhaustion suggestive of a postsynaptic NMJ defect (figure 1, A–C).

Patient B1

At 10 months, a reproducible decrement of 18% was seen on RNS of the left abductor pollicis brevis muscle. At the age of 6 years, RNS demonstrated a 14% decremental response of

abductor pollicis minimi and a 39% decremental response on the trapezius. Radial nerve-stimulated single-fiber EMG of the left extensor digitorum communis muscle demonstrated a dramatically increased variability of latency (jitter) in 11 pairs of the 14 pairs tested, with an average mean consecutive difference value of 165.5 microseconds (normal <36.6) and blocking seen in 8 pairs of 14 total tested, suggesting a disorder of neuromuscular transmission (figure 1D).

Muscle biopsy

Patient A1

A quadriceps muscle biopsy performed at 1 month of age was nondiagnostic. No residual tissue was available for further evaluation including electron microscopy after the genetic diagnosis.

Patient B1

A left quadriceps muscle biopsy at the age of 1 month showed a population of abnormally small muscle fibers that was suggestive of a myopathy. No residual tissue was available for further evaluation after genetic diagnosis.

Patient B2

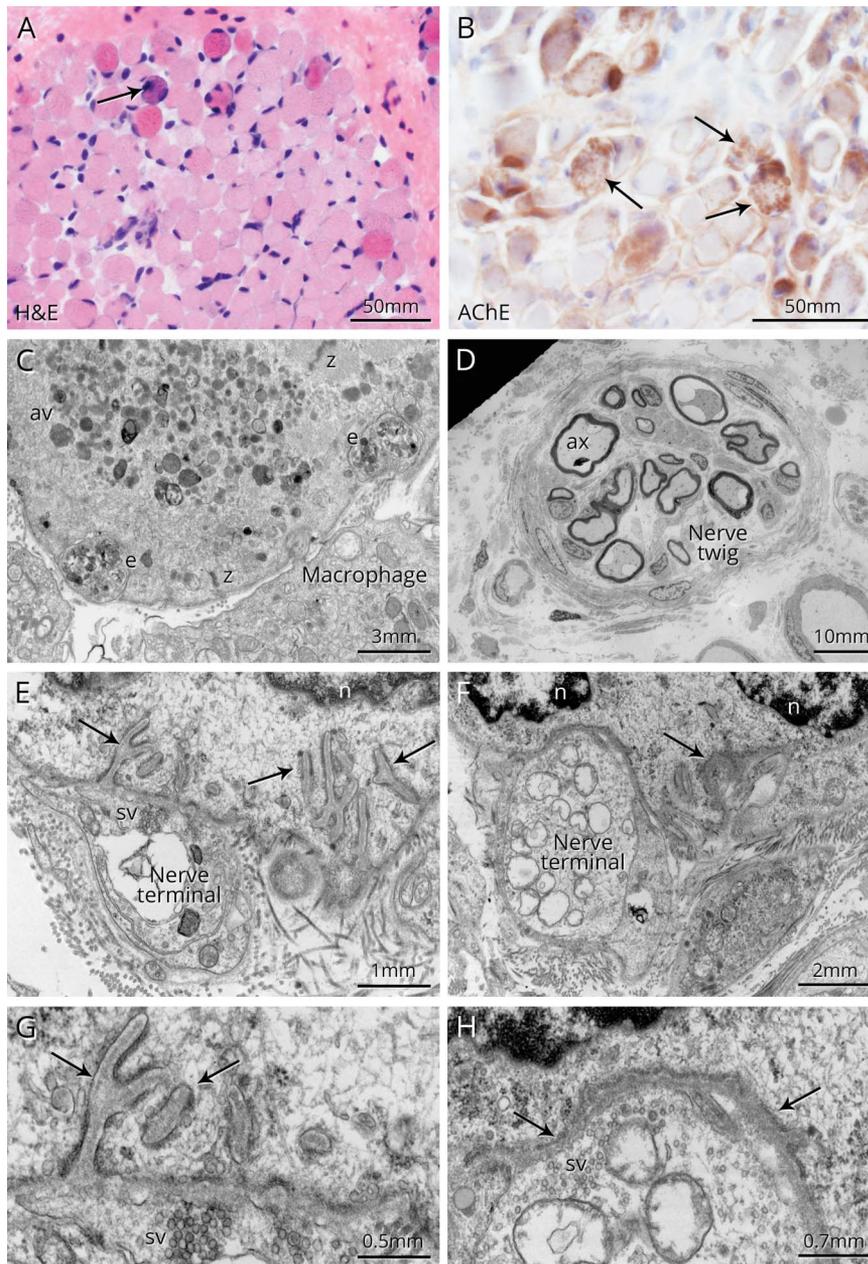
Light microscopy and transmission electron microscopy revealed a mild necrotizing myopathy with extensive autophagic vacuolar pathology (figure 2A). Autophagic vacuoles had sarcolemmal features in that acetylcholinesterase was localized at the surface of cytoplasmic vacuoles (figure 2B). Extrusion of autophagic vacuoles from affected muscle fibers was noted (figure 2C). Intramuscular nerve twigs seemed normal in cryosections (not shown) and in electron micrographs (figure 2D). By contrast, NMJs had varying degrees of postsynaptic fold attenuation (figure 2, E–H), whereas synaptic vesicles were numerous within nerve terminals. Complete absence of NMJ end plate folds was noted in association with some nerve terminals (figure 2H). No TAs were observed.

Genetic findings

Patient A1

An average of 61X coverage was obtained in the trio exomes. Previous chromosomal microarray data suggested that uniparental disomy (UPD) of chromosome 2 and that pathogenic homozygous variants on chromosome 2 may contribute to the patient's symptoms. Copy number and homozygosity analysis of the exome data confirmed the copy neutral long contiguous stretches of homozygosity reported by array comparative genomic hybridization (A 3). A homozygous, frameshift insertion was discovered in *GFPT1* at 2p13.3 (chr2:69581443insG; NM_001244710.1:c.686dupC; p.Arg230Ter) (figure 3). This variant is at the 5' end of exon 9, adjacent to the splice acceptor site of the muscle/heart specific transcript (NM_001244710), and results in the introduction of a termination codon. The variant was observed in the mother as a heterozygous change, whereas the father was a homozygous reference, suggesting UPD. These

Figure 2 Muscle biopsy pathology of patient B2



The cryosection H&E image (A) shows increased variation in fiber size and a mild degree of regeneration (arrow), suggesting that the patient has a mild, necrotizing myopathy. AChE enzyme histochemistry (B) highlights numerous cytoplasmic autophagic vacuoles with sarcolemmal features (arrows). Extensive autophagy is confirmed by electron microscopy (C). av occupy most of the intracellular space of the muscle fiber in the upper portion of the micrograph. Only rare sarcomeres are present, as noted by Z-lines (z) in this image. Extrusion of autophagic vacuoles (E) is evident at the surface of this muscle fiber. Intramuscular nerve twigs appear normal (D). Two of numerous NMJs observed in the biopsy are illustrated in panels (E-H). There are varying degrees of postsynaptic fold attenuation (arrows), milder in panels E and G and complete loss of NMJ endplate folds in panels F and H. Nerve terminals contain numerous sv. Portions of muscle fiber nuclei are present along the top of panels E, F, and H (n). AChE = acetylcholinesterase; av = autophagic vacuoles; H&E = hematoxylin & eosin; NMJ = neuromuscular junction; sv = synaptic vesicles.

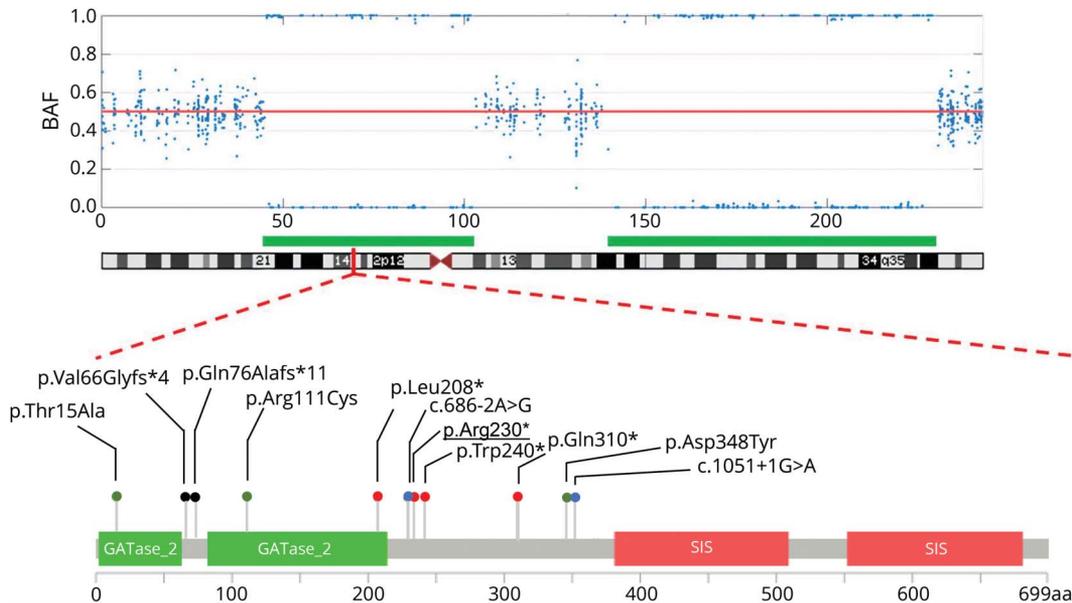
findings were independently confirmed by Sanger sequencing and confirmed loss of the paternal chromosome 2 segment (figure 4, A and B). This variant is observed in a single individual (1/219894) as a heterozygous change in the gnomAD v2.1.1 database.¹⁰ In-silico predictions suggested that *GFPT1* is intolerant to loss-of function variation (Exome Aggregation Consortium probability of being loss-of-function intolerant = 0.97). There are 10 *GFPT1* variants in ClinVar, in patients with CMS12, classified as pathogenic or likely pathogenic, which include missense, stop gained, and frameshift indels (table 2). Evaluation of the known genes associated with CMS revealed that Patient A1 was heterozygous for variants in *CHAT*, *MUSK*,

RAPSN, and *SCN4A* that were ruled out because of insufficient evidence for pathogenicity using American College of Medical Genetics criteria.¹¹ There were no additional candidates outside the region of LCRH found to correlate with the patient's symptoms.

Patient B1

An average of 154X coverage was obtained for this singleton WES. Variants of uncertain clinical significance were identified in nebulin (*NEB*), *SH3T2C*, *POLG*, and *GFPT1* genes. After clinical correlation, potential compound heterozygous variants in the gene (*NEB*, OMIM:161650) were reported.⁹

Figure 3 *GFPT1* gene and previously reported, ClinVar pathogenic and likely pathogenic variants in CMS12



BAF plot from WES of patient A1 (top) demonstrates long continuous regions of homozygosity (blue dots with BAF ~1 or BAF ~0) that overlaps with previous array CGH data (green horizontal bars). Red, vertical bar at 2p13.3 on the cytoband indicates the position of *GFPT1* gene within the LCRH. Below the cytoband, domain structure of *GFPT1* and ClinVar pathogenic and likely pathogenic mutations in relation to *GFPT1* domains of NM_001244710 and to p.Arg230* (underlined) identified in this study. Variants are colored based on their functional impact (green = missense, black = frameshift, red = nonsense, and blue = splice). BAF = B allele frequency; CGH = comparative genomic hybridization; *GFPT1* = glutamine-fructose-6-phosphate transaminase 1; LCRH = long contiguous regions of homozygosity; SIS = sugar isomerase domain; WES = whole-exome sequencing.

NEB is associated with autosomal recessive nemaline myopathy type 2 (OMIM: 256030) which carries similar features to CMS but is also associated with nemaline rods.¹² Parents were not available to determine phase of the 2 *NEB* variants. Reanalysis of exome data was requested, and the *GFPT1* p. Arg230Ter variant was reclassified as likely pathogenic. Other CMS-related variants were also identified in *CHRNA2* and *LAMB2* but were ruled out per American College of Medical Genetics guidelines.

Patient B2

Sequencing in the patient and his parents using congenital myopathy gene panel testing (Pacific Biosciences, Menlo Park, CA) identified the same p.Arg230Ter homozygous variant in B2 and demonstrated that both parents were heterozygous.

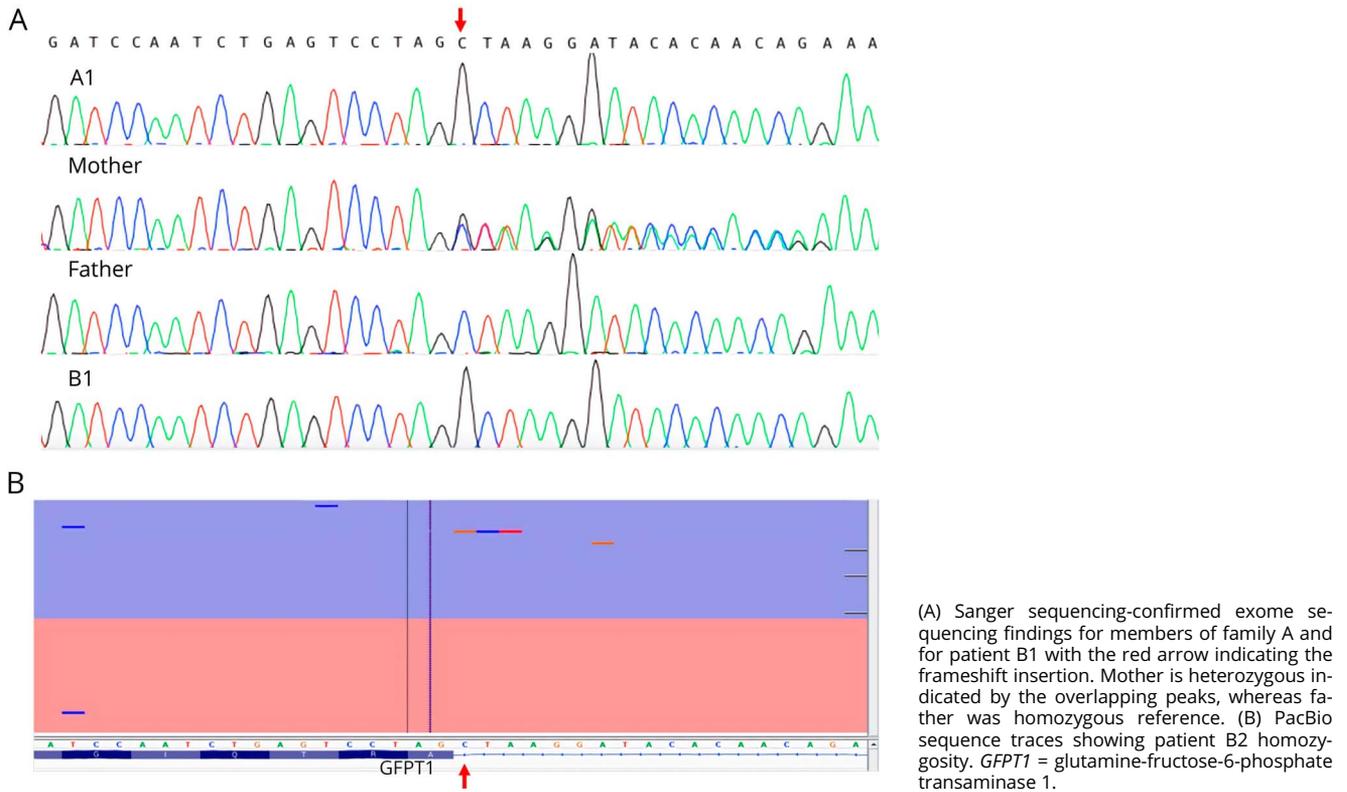
Discussion

We report the first case of maternal UPD of chromosome 2 in CMS and with a novel loss-of-function variant in *GFPT1* and provide support for pathogenicity of this variant by describing another family with CMS sharing the same variant. These 3 patients, from 2 unrelated families, with similar clinical phenotypes further expand the clinical spectrum of CMS12. Other CMS-related genetic variants were excluded in these families (table 1). From a clinical perspective, all patients presented with the classic clinical features of CMS12,

including hypotonia, general muscle weakness, little to no movement of shoulders and proximal legs, feeding difficulty, and EMG decrement in 2 of the 3 patients (table 3). However, they also share symptoms not commonly observed in CMS12. All of them manifested with respiratory insufficiency that required ventilator support at one point which is mostly associated with *DOK7* or *RAPSN* deficiency.¹³ Although CMS12 usually manifests within the first decade of life, only a few cases have been reported with congenital onset.⁵ Here, all 3 patients manifested with clinical symptoms at birth. In fact, 2 patients previously reported with early onset of disease carried splice altering variants at the muscle specific exon 9 junction and predicted biallelic loss of *GFPT1*, suggesting that loss-of-function variants confined to the muscle and heart-specific transcript adversely affects muscle fiber integrity and may result in earlier onset and a more severe clinical course.^{14,15}

Although the addition of 3,4-diaminopyridine to pyridostigmine treatment resulted in a positive response in one of the patients with *GFPT1* mutation (B2), it did not improve symptoms in Patient A1 or B1, indicating heterogeneity in response to the drug treatment. Selcen et al.¹⁴, reported a female patient with nonambulatory status, requiring respiratory support and a G-tube, harboring a nonsense and a splice-altering variant; her muscle biopsy demonstrated autophagic vacuolar pathology and attenuation of NMJ end plate folds, similar to the findings in the most severely affected (patient B2) of our children and absence of a ~70 kd glycoprotein. Muscle biopsies from many of the reported CMS12 cases have shown

Figure 4 Sequence traces of p.Arg230*



TAs, leading to the term congenital myasthenic syndrome with tubular aggregates⁵ None of our patients demonstrated TAs on histopathologic studies.

The p.Arg230Ter variant has not been described before in patients with CMS12 and is predicted to result in loss of GFPT1 protein in muscle and heart cells.¹⁶ Although this

Table 2 Clinvar pathogenic GFPT1 variants in CMS12 based on ClinVar release date April 29, 2020

HGVS.c	HGVS.p	Pubmed ID	Clinvar variant ID
NM_002056.4:c.1051+1G>A	^a	23794683	540351
NM_002056.4:c.1042G>T	p.Asp348Tyr	21310273	29738
NM_002056.4:c.928C>T	p.Gln310Ter	28492532	570975
NM_001244710.2:c.964C>T	p.Arg322Ter	23794683	859066
NM_001244710.1:c.719G>A	p.Trp240Ter	21310273	29737
NM_002056.4:c.621del	p.Leu207_Leu208insTer	21310273	29740
NM_002056.4:c.452C>A	p.Thr151Lys	30635494	631478
NM_001244710.1:c.331C>T	p.Arg111Cys	21310273 8664562	29735
NM_001244710.1:c.223dupA	p.Gln76Alafs11 ^a	21310273	29736
NM_002056.3:c.197_201delTTAAG	p.Val66Glyfs4 ^a	28492532	583230
NM_001244710.1:c.43A>G	p.Thr15Ala	12467753 12467753	29739
NM_002056.4:c.41G>T	p.Arg14Leu	30635494	631477
NM_001244710.1:c.686-2A>G	^a	28464723 23794683	540353

Abbreviations: CMS = congenital myasthenic syndrome; *GFPT1* = glutamine-fructose-6-phosphate transaminase 1.
^a Splice mutation.

Table 3 Clinical summary of patients with GFPT1 mutations

Patient ID or publication source	A1	B1	B2	(Ref 15)	(Ref 14)
Sex	Female	Male	Male	Male	Female
Mutation (HGVS.c)	c.686dupC ^a	c.686dupC/ c.686dupC	c.686dupC/ c.686dupC	c.686-2A>G/c.686- 2A>G	c.686-2A>G/ c.910C>T
APGAR	5,6,6	1,6,7	1,2,5		
Generalized muscle weakness	b	b	b		
Ambulation	a	a		b	a
Hypotonia	b	b	b	a	b
Head lag/muscle weakness	b	b	b	b	
Areflexia	b	b	b		
Prominent neck flexor weakness	b	b		b	b
Scoliosis	Severe ^b	a	a	Mild ^b	
Muscle atrophy	b	b		b	
Pes cavus deformity				a	
Ocular/facial/bulbar weakness	Facial diplegia	b	b	a	b
Myopathy	b	b	b	b	b
Muscular dystrophy				b	b
Respiratory insufficiency	b	b	Transient	a	
Pneumonia	b	b	.		
Mechanical ventilation	b	b	b	a	b
Gastrointestinal dysfunction	G-tube ^b	G-tube ^b	G-tube ^b	a	G-tube ^b
Cardiovascular irregularities	b			a	

Abbreviations: APGAR = appearance, pulse, grimace, activity, and respiration; HGVS.c = Human Genome Variation Society coding DNA reference sequence position.

^a Not observed. Empty cells indicate no information.

^b Observed.

genetic variant is shared among all 3 of the described children, the variation in severity and spectrum of symptoms suggest pathophysiologic complexity. To the best of our knowledge, patient A1 is the first instance of maternal UPD of chromosome 2, homozygosity for the *GFPT1* nonsense mutation located within the UPD region, inherited from a mother who was heterozygous for the pathogenic *GFPT1* variant. Glycosylation deficiency is one of the causes of CMS, and mutations have been identified in several glycosylation pathway genes including *DPAGT1*, *ALG2*, *ALG14*, and *GFPT1*.^{5,17,18} The exact mechanism by which glycosylation deficiency causes the NMJ defect is still unclear. Loss of *GFPT1* protein can result in abnormal glycosylation in muscle cells, reduced acetylcholine receptor cell surface expression, and abnormal sialylation of transmembrane muscle proteins.^{14,19,20} Electrophysiologic studies in one of our children who had UPD and predicted loss of function of the *GFPT1*, suggesting a postsynaptic NMJ defect. Reduction in *GFPT1* induces structural changes in the end plate characterized by loss of junctional folds and denervation-reinnervation process affecting NMJ.¹⁹ However,

the molecular consequences of absence of the ubiquitously expressed *GFPT1* protein is not fully understood. This report expands the clinical, genetic, histologic and electrophysiologic spectrum of *GFPT1* gene mutations related CMS12.

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Disclosure

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Appendix 1 (continued)

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Continued

Appendix 2 (continued)

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