


Congenital Myasthenic Syndrome Due to Choline Acetyltransferase Mutations in Infants: Clinical Suspicion and Comprehensive Electrophysiological Assessment Are Important for Early Diagnosis

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Abstract

Congenital myasthenic syndromes are inherited disorders caused by various defects in neuromuscular transmission. Although the typical presentation is fatigable weakness with prominent cranial involvement, neonates can lack these hallmark manifestations, and in those with choline acetyltransferase gene mutations, basal electrophysiological testing can yield negative findings. The authors report the case of a male infant presenting at birth with oculomotor and bulbofacial weakness, hypotonia, clubfoot, and severe respiratory insufficiency. Electromyography showed myogenic signs, and basal repetitive nerve stimulation yielded negative findings. Since age 6 months, the infant had progressively improved, acquiring autonomous respiration. Prolonged subtetanic repetitive nerve stimulation disclosed a marked decremental response compatible with suspected congenital myasthenic syndrome with episodic apnea. Genetic testing identified 2 novel choline acetyltransferase mutations (R470X, F580C). Keeping a high clinical suspicion of this rare condition and undertaking early comprehensive electrophysiological assessments including prolonged repetitive nerve stimulation (10 Hz for 5 minutes) can expedite the diagnosis.

Keywords

congenital myasthenic syndrome with episodic apnea, choline acetyltransferase mutations, electrodiagnosis, prolonged subtetanic repetitive nerve stimulation, hypotonia, neonatal respiratory paralysis

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Congenital myasthenic syndromes are inherited disorders caused by heterogeneous alterations in neuromuscular transmission, classified according to the affected protein's localization in presynaptic, synaptic, and postsynaptic subtypes.¹ The major presynaptic subtype is congenital myasthenic syndrome with episodic apnea (MIM254210), caused by recessive choline acetyltransferase gene mutations, resulting in impaired acetylcholine synthesis.^{2,3}

Although the various congenital myasthenic syndromes share a common clinical presentation, characterized by fatigable weakness with prominent ocular and bulbofacial involvement, a distinctive feature in congenital myasthenic syndrome with episodic apnea is the presence of episodic apneas triggered by fever, infection, or vomiting. Some infants present at birth with bulbar and respiratory paralysis, often

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Figure 1. Photographs of the baby showing ocular and facial paresis at the age of 1 day (A), 3 months (B), 9 months (C), and 3 years (D).

improving within several months. Because typical myasthenic signs are often difficult to recognize in neonates, when examining a floppy infant showing some of these signs, a high index of clinical suspicion is required.⁴ This case report describing an infant who had congenital myasthenic syndrome with episodic apnea confirmed by molecular genetic testing provides a useful reminder that when a neonate presents with clues suggesting this syndrome, physicians should undertake early complete electrophysiological assessment.

Case Report

A male infant, born at the 39th week of gestation, presented at birth with hypotonia, drooping eyelids, clubfoot, and respiratory failure needing mechanical ventilation (Figure 1A). The Apgar score at birth was low: 2 of 10 at 1 minute and 4 of 10 at 5 and 10 minutes. Ultrasound studies during pregnancy showed polyhydramnios and cerebellar hypoplasia confirmed by fetal and postnatal magnetic resonance imaging. The family medical history reported that the proband's father since infancy had successfully carried a ventriculoperitoneal shunt for Dandy-Walker syndrome. After birth, the proband was admitted to a city hospital neonatal intensive care unit, where he underwent nasogastric tube feeding and continuous controlled mechanical ventilation for persistent respiratory insufficiency. Extended investigations including creatine kinase, metabolic studies, and genetic analyses for congenital myotonic dystrophy, Prader-Willi syndrome, and spinal muscular

atrophy yielded negative findings. At 3 months of age (Figure 1B), the patient was transferred to the authors' pediatric intensive care unit. The neurological examination at admission showed poor behavioral contact, ptosis, ophthalmoparesis, expressionless face, reduced sucking, hypotonia, and poor general movements. The infant still required nasogastric tube feeding and persistent mechanical ventilation after weaning failed. Transdiaphragmatic pressures measured through intensive care instrumentation (esophageal/gastric catheter and pneumotachometer, a device for measuring airflow) suggested diaphragmatic paralysis. An electroencephalogram disclosed normal sleep-wake cycles. Multimodal evoked potentials were normal. Nerve conduction studies and baseline low-frequency repetitive stimulation (10 stimuli at 2 Hz) of median and ulnar nerves yielded normal findings, and needle electromyography showed slight myogenic signs. A muscle biopsy specimen contained a prevalence of type 2 fibers. Because the infant's ocular, facial, and limb strength continued to fluctuate day by day, we started a therapeutic trial with the cholinesterase inhibitor pyridostigmine orally up to 6 mg/kg/d divided into 5 doses, which slightly improved ptosis, movements, and ventilation. Encouraged by this improvement, the authors reconsidered a possible neuromuscular transmission defect. To investigate this possibility, they undertook comprehensive electrophysiological testing (Table 1).⁵ A single supramaximal stimulus applied to the ulnar and median nerves failed to elicit repetitive compound motor action potentials, thus excluding acetylcholinesterase deficiency and slow-channel syndromes.

Table 1. Electrophysiological Testing Undertaken in the Infant With Congenital Myasthenic Syndrome.

Electrophysiological test	Result
Motor and sensory median and ulnar nerve conduction studies	Normal
Needle electromyography in cranial and limb muscles	Slightly myogenic signs
Single supramaximal stimulus to detect a possible repetitive CMAP in 2 distal muscles	No repetitive CMAP
Baseline low-frequency RNS (train of 10 supramaximal stimuli at 2 Hz) in 2 distal muscles	First assessment: normal Second assessment: 20% CMAP decrement (borderline value for small infants)
Tetanzing stimulation at 50 Hz for 10 seconds, followed by baseline RNS every minute for 5 minutes	During stimulation: 55% CMAP increment (slightly abnormal) RNS after tetanzing stimulation: no significant CMAP changes
PRNS followed by baseline RNS every minute for 5 minutes	During PRNS: 90% CMAP decrement (abnormal) RNS 1 minute after PRNS: 60% CMAP decrement (abnormal) RNS 5 minutes after PRNS: CMAP recovery to baseline
Frontalis muscle SSFEMG	Average jitter of 88 microseconds (abnormal)

Abbreviations: CMAP, compound muscle action potential; RNS, repetitive nerve stimulation; PRNS, prolonged subtetanic repetitive nerve stimulation at 10 Hz for 5 minutes; SSFEMG, stimulated single-fiber electromyography.

Baseline low-frequency repetitive ulnar nerve stimulation (10 stimuli at 2 Hz) induced a mild decremental response (20%) (borderline value for small infants). High-frequency nerve stimulation (50 Hz for 10 seconds) elicited a 55% increment (slightly elevated, but not indicative of Lambert-Eaton-like congenital myasthenic syndrome), and the following low-frequency stimulation tests did not induce a significant decrement. Conversely, prolonged subtetanic repetitive ulnar nerve stimulation (10 Hz for 5 minutes) induced a marked decremental response (90%) after 2 minutes. One minute after this prolonged stimulation ended, standard low-frequency stimulation (10 stimuli at 2 Hz) still induced a 60% decrement, but within 5 minutes, values returned to baseline. Finally, frontal muscle-stimulated single-fiber electromyography showed increased jitter (average jitter, 88 microseconds).

Antibodies against acetylcholine receptor and muscle-specific tyrosine kinase tested negative. The authors informed the infant's parents about the diagnosis of congenital myasthenic syndrome, and they agreed to tracheostomy and percutaneous gastrostomy. From the age of 6 to 9 months, facial, bulbar, axial, and limb strength and breathing progressively improved, but the improvement had no direct relationship to the pyridostigmine dose. The infant was discharged from the hospital at the age of 9 months and was able to breathe spontaneously (Figure 1C). Oral pyridostigmine was progressively increased to 9 mg/kg/d (the current dose) divided into 6 doses, and at the age of 1.5 years, 3,4-diaminopyridine was added at up to 0.8 mg/kg/d divided into 4 doses, with both medications inducing further overall improvement. He has been able to walk since the age of 2.5 years. At age 3 years, the proband shows moderate ocular and facial weakness and slight-to-moderate motor and language delay, whereas his overall psychological development appears normal (Figure 1D). Feeding is entirely oral. The child uses apnea monitors during the night, although he breathes autonomously and needs ventilatory support only rarely during infections. The overall "electroclinical" picture prompted the authors to test for choline acetyltransferase

and rapsyn mutations. Genetic diagnosis performed at the Medizinisch Genetisches Zentrum in Munich, Germany, by 1 of the coauthors (AA) identified 2 compound heterozygous mutations in the choline acetyltransferase gene: 1408C>T (R470X) in exon 11 and 1739T>G (F580C) in exon 13. The first mutation has not been described previously and leads to a premature stop codon, predicting a loss of protein function, most probably through nonsense-mediated messenger ribonucleic acid (RNA) decay. The second mutation is unknown as a polymorphism or pathogenic mutation from the literature or mutation databases, but according to a software prediction program (PolyPhen), this variant is probably damaging (score, 0.996). The mother carries the first mutation, and the father carries the second.

Discussion

The authors consider this rare case of an infant with congenital myasthenic syndrome due to novel choline acetyltransferase gene mutations^{2,3} clinically and neurophysiologically interesting and instructive. First, it illustrates how the clinical features in newborns and infants can mislead the diagnosis of congenital myasthenic syndrome and lead clinicians to suspect other central or peripheral nervous system diseases. Second, it underlines how comprehensive electrodiagnostic testing (Table 1) helps to avoid false negatives from basal stimulation testing alone and distinguish the specific congenital myasthenic syndrome subtype for targeted genetic testing. This case also underlines that in the choline acetyltransferase deficiency subtype, prolonged subtetanic repetitive nerve stimulation, still poorly used in infants in many laboratories,⁶ is extremely useful.

The first diagnostic pitfall the physicians encountered in this case arose because the proband's cerebellar malformation and father's Dandy-Walker syndrome pointed to a central nervous system disease rather than a peripheral disorder, and the initial diagnostic work-up underestimated the congenital myasthenic syndromes, rarely reported in case series studies describing the causes of neonatal hypotonia.⁷⁻⁹ In the differential diagnosis of

the infant, the authors had to consider central nervous system diseases, neuropathy, spinal muscular atrophy, myopathy, and neuromuscular transmission disorders. As well as hypotonia and weakness, in our patient, the authors also recognized prominent facial, ocular, and bulbar weakness that fluctuated in intensity, thus corroborating the suspicion of a neuromuscular transmission disorder according to expert recommendations.^{10,11}

The second diagnostic pitfall the authors encountered was the initial misleading electrodiagnosis limited to basal electrophysiological testing. In suspecting congenital myasthenic syndrome, the operator responsible for electrodiagnosis must undertake an extensive electrophysiological assessment.⁵ Clinical neurophysiologists should be able to manage technical difficulties related to the infant's small size and poor collaboration.¹² They need to be familiar with the various neurophysiological changes taking place during development. The authors' experience confirms that repetitive nerve stimulation testing at low frequency (10 stimuli at 2-3 Hz) is insufficient to rule out the diagnosis of congenital myasthenic syndrome with episodic apnea.¹⁻³ Conversely, when the authors undertook comprehensive electrophysiological testing, prolonged subtetanic stimulation (10 Hz for 5 minutes) elicited a marked decremental response with slow recovery. This abnormality was compatible with congenital myasthenic syndrome with episodic apnea, prompting the authors to test the choline acetyltransferase gene.¹⁻³ Prolonged subtetanic nerve stimulation is a simple technique but scarcely used in infants, as highlighted by its absence in a recent multicenter follow-up study of patients with choline acetyltransferase mutations.⁶ It is especially useful in infants who cannot maintain the sustained voluntary muscle activity used in collaborating patients to induce postexercise exhaustion. No published studies have measured the diagnostic sensitivity and specificity of prolonged subtetanic stimulation in infants with choline acetyltransferase deficiency, but clinical and laboratory data suggest that they are considerable.¹⁻³ In vitro studies show that 10-Hz stimulation for 5 minutes of affected small muscle bundles totally abolished the amplitude of the extracellularly recorded compound muscle action potential and progressively decreased the amplitude of the intracellularly recorded endplate potential by approximately 90% with recovery over 7 minutes.³ These events are attributed to slow acetylcholine resynthesis.

If the extensive repetitive nerve stimulation protocol shows no signs indicating a neuromuscular transmission disorder, the infant should therefore undergo stimulated single-fiber electromyography.⁵ Some investigators have proposed this neurophysiological technique as the initial electrophysiological step in children with a suspected neuromuscular defect.¹³ Whereas single-fiber electromyography, a technique that requires specific expertise, is highly sensitive in detecting neuromuscular junction disorders, an extensive nerve stimulation protocol is easy for all pediatric neurophysiologists to undertake and provides essential clues in differentiating the specific congenital myasthenic syndrome subtypes for targeted genetic testing.^{1,5}

Finally, the patient's myasthenic symptoms responded to a therapeutic trial with pyridostigmine, thus helping the diagnosis.

Caution is nevertheless needed, remembering that pyridostigmine is ineffective or even detrimental in some congenital myasthenic syndrome subtypes, such as acetylcholinesterase deficiency, slow-channel syndromes, or downstream of kinase-7 (DOK7) mutations that benefit from ephedrine or open-channel blockers.¹

In conclusion, a rational clinical, electrophysiological, and genetic approach to infants with symptoms suggesting congenital myasthenic syndrome allows clinicians to provide early appropriate treatment and genetic counseling to families, saving time and resources. Sending deoxyribonucleic acid (DNA) samples to an expert and updated genetic laboratory with experience in all congenital myasthenic syndrome subtypes allows future research in patients in whom the initial genetic analysis results proved negative.¹⁴

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Author Contributions

RD, the neurologist who principally followed up with the patient, made the major contribution in drafting the article. AA performed the genetic analysis and interpreted the genetic data. RD and PS contributed by performing and interpreting the neurophysiological studies. GPC and ADF contributed to neurological management and helped in drafting the article. GC performed the respiratory physiological study and drafted the relative part of the history. FN conducted family genetic counseling and helped in drafting the article. HL gave an essential expert opinion on targeted genetic analysis and made a substantial contribution in revising the article. SB and HL were both mentors in this work.

Declaration of Conflicting Interests

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Ethical Approval

The authors received informed patient consent forms from the baby's parents.

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