

Case Reports

Neonatal Seizure: A Rare Aetiology Easily Missed by Routine Metabolic Screening

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Abstract

We present the first Chinese case of D-bifunctional protein (DBP) deficiency, a single peroxisomal protein disorder. A non-dysmorphic, 2.53 kg hypotonic female infant developed seizure at fifty-six hours. After seizure onset, she lost her sucking and deep tendon reflex. On day 10, brain stem auditory evoked potential (BAEP) and visual evoked potential (VEP) studies showed marked impairment. Plasma amino acid, serum acylcarnitine pattern and urine organic acid analysis were normal. For persistent seizures, therapeutic trial of pyridoxine, biotin and folinic acid had no effect. Cerebrospinal fluid (CSF) glycine level and urine sulphite screen by Dipstick were normal. At eighth weeks of life, the markedly elevated level of very long chain fatty acid (VLCFA) level was identified. Subsequent biochemical tests revealed pattern consistent with peroxisomal DBP deficiency. Awareness of the suggestive clinical features and loopholes in the routine metabolic screening tests will facilitate early detection of this rare disorder.

Key words

Hypotonia; Neonate; Peroxisomal disorder; Seizure

Introduction

Seizures occur more often in the neonatal period than any other time of life. It may indicate the presence of a potentially treatable aetiology and should prompt an immediate evaluation to determine the cause and to institute specific therapy.

Peroxisomal disorders appear with a frequency of about 1:30,000¹ and are divided into two major categories: disorder of peroxisome biogenesis and disorder of single peroxisomal

protein. The prototype of the former disorder is Zellweger syndrome.² It is characterised by craniofacial dysmorphism and profound neurological abnormalities.² The single peroxisomal protein disorders are rare and features may mimic Zellweger syndrome, albeit absence of craniofacial dysmorphism. This report presents a baby girl with D-bifunctional protein (DBP) deficiency, a single peroxisomal protein disorder. Craniofacial dysmorphism was absent in this first Chinese case and her diagnosis was missed by routine metabolic screen but established subsequently by serum very long chain fatty acid (VLCFA) assay.

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Case Report

A 2.53 kg female infant was delivered at 38 week of gestation by elective cesarean section. Parents were non consanguineous and family history was noncontributory. Antenatal course was uneventful. She had normal liquor volume and fetal movements. Risk factors for birth asphyxia and perinatal infections were absent. Cord blood thyroid function was normal. Paediatrician assessment at twenty-

one hours of life found central hypotonia. Craniofacial dysmorphism and contractures were absent and all jerks were present. No myopathic face or tongue fasciculation was present. Gestational age as assessed by New Ballard Score corresponded to date. The baby remained well till fifty-six hours of life when she developed generalised tonic clonic seizure for fifteen seconds.

Initial laboratory findings revealed normal calcium, phosphate, magnesium, glucose, acid-base status, renal and liver functions. Her blood and cerebrospinal fluid (CSF) cultures were negative. Muscle enzymes, ammonia, lactate and pyruvate were not elevated. The CSF glucose was 4.0 mmol/L at the time when plasma glucose was 5.4 mmol/L. Computerised tomogram (CT) brain and electroencephalogram (EEG) study performed on day 3 showed no pathology and no epileptiform discharge respectively.

After seizure onset, her neurological status deteriorated. She lost her sucking reflex and examination revealed progressive loss of deep tendon reflexes (DTR). On day 10, brain stem auditory evoked potential (BAEP) revealed threshold at 80 db, compatible with moderate to severe impairment and visual evoked potential (VEP) showed markedly prolonged P100. Ophthalmologist's assessment on day 18 found normal cornea and fundi.

Investigations were then performed to look for white matter disorder in view of progressive loss of DTR. On day 12, nerve conduction velocity (NCV) and

electromyography (EMG) studies were unremarkable and somatosensory evoked potential (SSEP) study showed no identifiable waveforms.

Initially, the seizures were brief and ceased after phenobarbitone treatment. On day 13, seizure recurred and phenytoin was added. Metabolic screening at fasting and fed state was negative. Blood for carnitine and amino acids, and urine for reducing substances, amino and organic acid were normal. Very long chain fatty acid (VLCFA) assay was then requested for suspected peroxisomal disorder. The request was initially declined because of absence of craniofacial dysmorphism and presence of normal urine organic acid analysis and serum acylcarnitine pattern.

With deterioration in seizure control, the baby became ventilator dependent from day 14 onwards. Seizures persisted despite topiramate and high serum level of phenytoin. Trial of pyridoxine, biotin and folic acid had no effect. Magnetic resonance imaging study of the brain performed on day 35 detected no abnormalities. Further work up performed for uncontrolled seizures at seven weeks old revealed normal CSF glycine level and negative urine sulphite screen by Dipstick. Another request on VLCFA assay was made. VLCFA level was markedly elevated at eighth weeks of life. Further biochemical work-up in National Referral Laboratory in Women's & Children's Hospital in South Australia revealed a pattern consistent with peroxisomal D-bifunctional protein (DBP) deficiency (Table 1).

Table 1 Laboratory results indicating peroxisomal defect

	Very long chain fatty acids		Erythrocytes		Plasma
	C26:0/C22:0 ratio	C24:0/C22:0 ratio	Plasmalogens ¹		Bile acid intermediates ²
			C16:0	C18:0	C27 THCA
			ug/g haemoglobin		umol/L
Patient	0.427	2.094	276	376	2
Reference range	<0.035	0.550-1.150	125-215	220-405	<0.1

THCA, trihydroxycholestanic acid.

Comment:

1. Analysis of erythrocytes has shown that there are normal or slightly elevated levels of plasmalogens. Plasmalogen levels are routinely significantly decreased in infants affected by a peroxisomal biogenesis disorder. Therefore, these results strongly suggest that the baby is affected by an isolated disorder of peroxisomal beta-oxidation.
2. Investigation of the plasma bile acid intermediates revealed elevated levels of C27 THCA and varanic acid. However, the C29 dicarboxylic bile acid metabolite, which is increased in peroxisomal biogenesis disorders, was not elevated. This bile acid intermediate pattern is consistent with **peroxisomal D-bifunctional protein deficiency**.
3. Note that this diagnosis was also supported by the result of branched chain fatty acids analysis. This showed elevated levels of pristanic acid, in comparison with phytanic acid (results not shown).

The baby's seizure frequency and severity markedly decreased after elective tracheostomy performed at 4-5 months for ventilator dependency. After tracheostomy she was able to wean off mechanical ventilation. As part of the discharge plan, she was transferred to the general pediatric ward at 7 months. At 8 months old, she developed cardio-respiratory compromise and frequent seizure during an episode of parainfluenza type 3 infection. After discussion with parents, conservative approach was adopted as the parents fully understood the grave prognosis.

Discussion

Our patient presented with neonatal seizures and progressive worsening neurological condition that was not due to primary structural brain disease or infection. In approaching this encephalopathy, history and "routine metabolic screening" can help in ruling out common aetiologies like hypoxic ischaemia, hypoglycaemia, diabetic ketoacidosis, fluid and electrolytes disturbances, organ failure, intoxications and parainfectious encephalopathy. The panel of investigations performed for "routine metabolic screening" in our institution includes renal and liver functions, calcium, phosphate, magnesium, glucose, acid-base status, ammonia, lactate, pyruvate, paired CSF and plasma glucose, and urine for reducing substance and amino acids. Paired blood and urine for drug screening were done only if history or physical examination revealed features compatible with drug intoxication. Serum carnitine profile and amino acids and urine organic acid were performed only after discussion with biochemist for cases with suspicion of inborn error of metabolism (IEM). The results of the "routine metabolic screening" and IEM work up were normal in our case.

In our case, the progressive loss of DTR is an unexpected finding. While central hypotonia, seizure and lost of sucking reflex are features of gray matter pathology, loss of DTR is more a feature of white matter and neuromuscular pathology. BAEP, VEP, NCV, EMG and SSEP were performed to have a better delineation of the site of pathology. BAEP and VEP were markedly impaired. Ophthalmologic examination is an important evaluation of neurometabolic disorders.

Witnessing the progressively deteriorating course with unrevealing investigation findings, degenerative diseases were then considered. Though uncommon, certain degenerative disorders of the developing nervous system may manifest clinically in the neonatal period.² Apart from

distinctive dysmorphic craniofacial features, the description of Zellweger syndrome in renowned newborn neurology textbook totally matches with our patient.² Neurological syndrome in Zellweger syndrome includes severe visual, auditory impairment, marked hypotonia and weakness.² The latter two features are accompanied by areflexia and may be so severe as to raise the possibility of Werdnig-Hoffman disease.² Neonatal seizures are characteristic.² This reading prompts us to make a request for VLCFA assay.

While peroxisome deficiency disorder is a rare disorder, DBP deficiency is an even rarer disorder with estimated prevalence about 1:100,000.¹ Peroxisomal functions include both catabolic activities, e.g. beta-oxidation of VLCFA and anabolic activities, e.g. biosynthesis of plasmalogens and of bile acids.² DBP catalyzes the second and third step of peroxisomal beta-oxidation of fatty acids and fatty acid derivatives.¹ DBP has been shown to be indispensable for the breakdown of VLCFA such as C26:0, and alpha-methyl-branched-chain fatty acids such as pristanic acid and the bile acid intermediates dihydroxycholestanic acid (DHCA) and trihydroxycholestanic acid (THCA).¹

As DBP deficiency is so rare, little description can be found even in renowned neurology textbook. Distinctive phenotype might not be available in every case of DBP deficiency. The absence of classical dysmorphism illustrated in the present case is a good example.

In Ferdinandusse et al's report, extensive biochemical studies were performed in 126 DBP-deficient patients.¹ Virtually all children presented with neonatal hypotonia (98%) and seizures (93%) within the first month of life. External dysmorphism was present in only 53 among 79 patients (67%) for whom data were available and resembled those of patients with Zellweger syndrome.

For peroxisomal disorders, organic acid analysis may show dicarboxylic aciduria, which reflects impaired peroxisomal beta-oxidation.³ Also, the acylcarnitine pattern may be abnormal.⁴ However, diagnostic pitfall exists in that both findings can be normal, as in our case. Moreover, the decisive marker for peroxisomal diseases, with the exception of rhizomelic chondrodysplasia punctata, is the elevation of VLCFA in serum or cultured fibroblast.^{2,3}

Seizure is the most predominant feature in our case. A practical approach in management of neonatal seizure is to identify and treat the treatable causes.^{5,6} An algorithm depicting our approach is listed in Figure 1. Currently, no effective treatment is available for nonketotic hyerglycinaemia and sulfite oxidase deficiency. So, these disorders were omitted in the algorithm. Similarly no effective treatment is available for Zellweger syndrome.

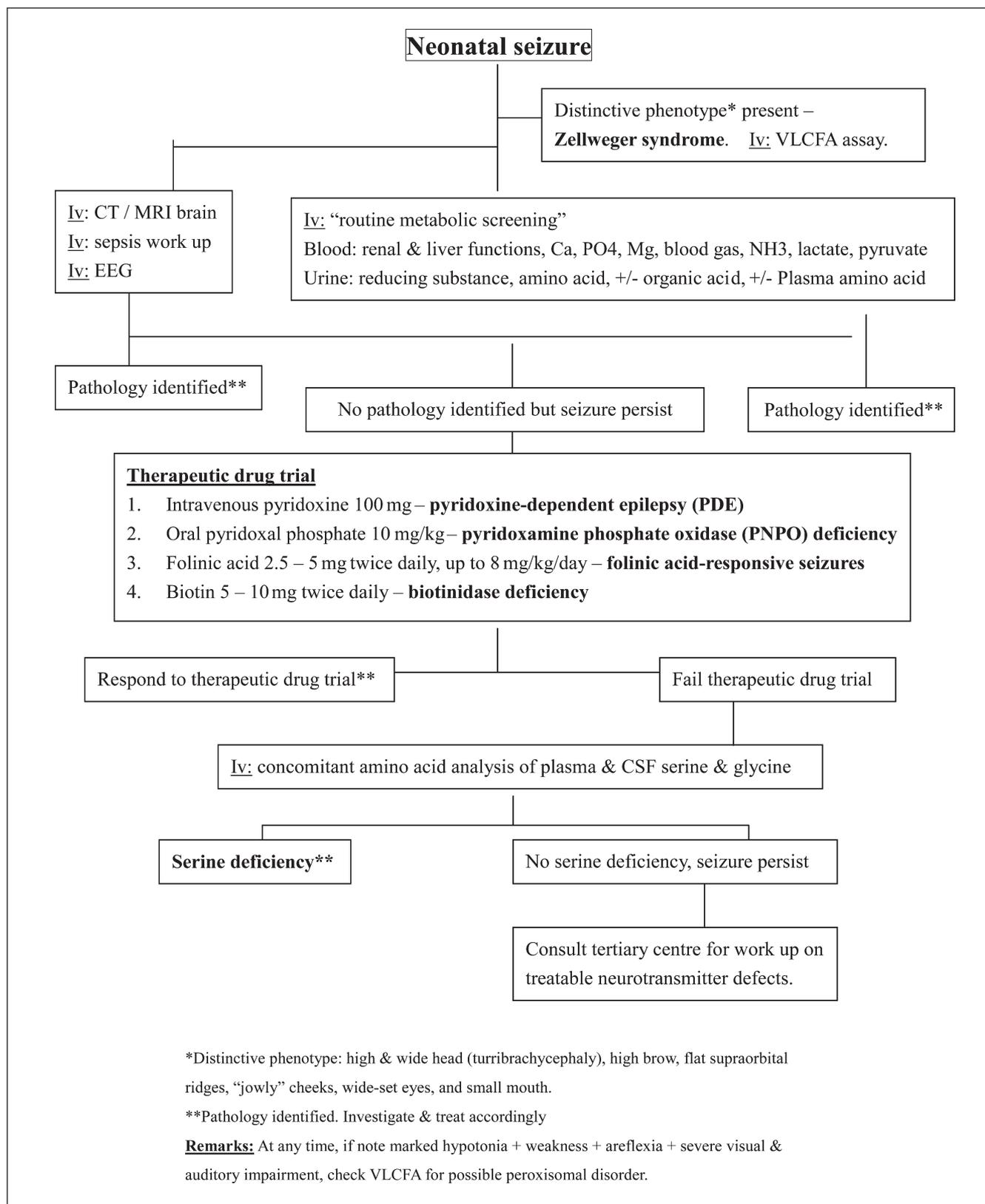


Figure 1 Approach to treatable neonatal seizures.

However, early diagnosis can reduce parental anxiety and aid genetic counseling.

Our view is that the request on VLCFA assay should be made on clinical grounds. As mentioned before, marked hypotonia, areflexia and weakness, together with severe visual and auditory impairment and seizures should prompt a request for VLCFA. While Zellweger syndrome is famous for its distinctive phenotype, other peroxisomal disorders may not have distinctive external dysmorphism. Early recognition relies on clinician's awareness of the constellation of suggestive features and loopholes in the "routine metabolic screening".

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