Official Journal of the European Paediatric Neurology Society

Original article

Are diagnostic magnetic resonance patterns life-saving in children with biotin-thiamine-responsive basal ganglia disease?

Tülay Kamaşak [a,*], Cengiz Havalı [b], Hülya İnce [c], İlker Eyüboğlu [d], Alper Han Çebi [e], Sevim Sahin [a], Ali Cansu [a], Kursad Aydin [f]

[a] Karadeniz Technical University, Department of Pediatric Neurology, Trabzon, Turkey
[b] University of Health Sciences Bursa Training and Research Hospital, Bursa, Turkey
[c] Bahcesehir University Medical Faculty Hospital, Samsun, Turkey
[d] Karadeniz Technical University, Department of Radiology, Trabzon, Turkey
[e] Karadeniz Technical University, Department of Genetic, Trabzon, Turkey
[f] Medipol University of Hospital, İstanbul, Turkey

ARTICLE INFO

Article history:
Received 15 May 2018
Received in revised form
14 June 2018
Accepted 25 June 2018

Keywords:
Biotin-thiamine responsive basal ganglia disease
BTBGD
SLC19A3 mutations
Encephalopathy

ABSTRACT

Background: Biotin-thiamine responsive basal ganglia disease (BTBGD) is an autosomal recessive disorder caused by mutations in the *SLC19A3* gene and characterized by recurrent sub-acute episodes of encephalopathy that typically starts in early childhood. This study describes characteristic clinical and magnetic resonance imaging (MRI) findings of six cases of BTBGD diagnosed with newly identified mutations and genetically confirmed, with very early and different presentations compared to cases in the previous literature.

Methods: Six patients referred from different centers with similar clinical findings were diagnosed with BTBGD with newly identified mutations in the *SLC19A3* gene. Two novel mutations in the *SLC19A3* gene were identified in two patients at whole exome sequencing analysis. The clinical characteristics, responses to treatment, and electroencephalography (EEG) and MRI findings of these patients were examined. The other four patients presented with similar clinical and cranial MRI findings. These patients were therefore started on high-dose biotin and thiamine therapy, and mutation analysis concerning the *SLC19A3* gene was performed. Responses to treatment, clinical courses, EEG findings and follow-up MRI were recorded for all these patients.

Results: Age at onset of symptoms ranged from 1 to 3 months. The first symptoms were generally persistent crying and restlessness. Seizures occurred in five of the six patients. Cranial magnetic resonance imaging revealed involvement in the basal ganglia, brain stem, and the parietal and frontal regions in general. The first two patients were siblings, and both exhibited a novel mutation of the *SLC19A3* gene. The third and fourth patients were also siblings and also exhibited a similar novel mutation of the *SLC19A3* gene. The fifth and

* *Corresponding author*. Fax: +90 462 325 32 05.
E-mail addresses: tkamasak@hotmail.com (T. Kamaşak), cengizhavali@gmail.com (C. Havalı), drhulyaince@yahoo.com (H. İnce), ilkereyuboglu@gmail.com (İ. Eyüboğlu), dralphancebi@yahoo.com (A.H. Çebi), sevimsahin1@yahoo.com (S. Sahin), acansu2011@hotmail.com (A. Cansu), kursadaydin@hotmail.com (K. Aydin).
https://doi.org/10.1016/j.ejpn.2018.06.009
1090-3798/© 2018 European Paediatric Neurology Society. Published by Elsevier Ltd. All rights reserved.

sixth patients were not related, and a newly identified mutation was detected in both these subjects. Three novel mutations were thus detected in six patients.

Conclusion: BTBGD is a progressive disease that can lead to severe disability and death. Early diagnosis of treatable diseases such as BTBGD is important in order to prevent long-term complications and disability.

© 2018 European Paediatric Neurology Society. Published by Elsevier Ltd. All rights reserved.

1. Introduction

Biotin-thiamine responsive basal ganglia disease (OMIM: 607483) is an autosomal recessive disorder caused by *SLC19A3* gene mutations.[1] It is characterized by recurrent subacute encephalopathy manifesting as confusion, seizures, ataxia, dystonia, supranuclear facial palsy, external ophthalmoplegia and dysphagia. Hemiparesis or quadriparesis may be observed. Episodes are often triggered by febrile illness or mild stress. More rarely, BTBGD presents as chronic or slowly progressive dystonia, seizures and psychomotor delay.[2] The aim of this study was to investigate the clinical and radiological features of six patients diagnosed with BTBGD with novel mutations of the *SLC19A3* gene, with symptoms such as restlessness, persistent crying and seizures. The *SLC19A3* gene encodes the transporter protein hTHTR2, which transfers thiamine into the cell. Mutations in this pathway can lead to consequences such as poor transport or reduced levels of thiamine. Increased *SLC19A3* expression in the case of hypoxia and stress cannot be achieved in individuals with this mutation. High doses of thiamine may make some contribution to cellular intake. Biotin is essential for normal expression of *SLC19A3* and is administered in order to provide optimal expression.[1–3]

The six patients in this study were diagnosed with novel mutations in the *SLC19A3* gene. The purpose of this study was to investigate the typical presentation findings, MRI findings, clinical courses, and responses to treatment of our cases with more mortal courses compared to cases in the previous literature.

2. Materials and methods

Two siblings were admitted to the Karadeniz Technical University Medical Faculty Hospital with restlessness and seizures. Mitochondrial disease was suspected following MRI. Two cases admitted to the University of Health Sciences Bursa Training and Research Hospital due to downward deviation of the eyes, restlessness, recurrent anxiety and tonic contractions were considered as BTBGD at cranial MRI. The sister of one of these two patients was diagnosed BTBGD with the same mutation. Another patient admitted to the Bahcesehir University Medical Faculty Hospital had a history of seizure onset and similar cranial MR findings and was also diagnosed with BTBGD. These six subjects were investigated for the purpose of performing genetic analyses, examining clinical and MRI findings, and monitoring treatment responses. All of the patients were examined the time of presentation to hospital to the final visit. Neurological examination results and treatment responses were recorded before and after treatment. We interviewed the parents and recorded seizure frequencies. The EEG records were also examined. Whole Genome Sequencing of Human mtDNA analyzes were normal. Whole exome sequencing analysis were planned for the first two patients of Karadeniz Technical University Medical Faculty Hospital. For other patients *SLC19A3* gene sequence analysis were performed after cranial MR.

3. Results

Six patients were admitted to outpatient clinics due to restlessness and seizures. Mitochondrial disease and BTBGD were suspected based on cranial MRI findings. Causes of admission, cranial MRI findings, initial diagnoses and treatments are summarized in Table 1.

3.1. *Patients 1 and 2, c.623_624insA mutation*

These two patients were siblings, a boy of 4 and a girl of 1.5 years (Fig. 1). Both presented with restlessness, persistent crying and contractions. The first patient initially presented to hospital at the age of 2 months due to excessive crying and restlessness. Leigh syndrome was suspected at cranial MRI at this time, and the patient was monitored accordingly. Motor development milestone delay was subsequently observed, and hypotonicity and seizures were added to the manifestation. At the age of 2.5 years, the patient experienced infection-triggered encephalopathy attack. During this time, his one-month old sister presented to our hospital due to excessive crying and restlessness. Whole Genome Sequencing of Human mtDNA analyzes was normal, and whole exome sequencing analysis was planned for the two patients. Hyperintense lesions were observed on T2-weighted images and diffusion-weighted images in the bilateral basal ganglia and anterior segment of both parts of the thalamus at cranial MRI in Patient 1 (Fig. 2a–c). MR spectroscopy revealed a pathological lactate peak in white matter areas, an increased Cho/Cr ratio and a mild decrease in the NAA/Cr ratio, and findings compatible with an active demyelinating process and neuroaxonal damage (Fig. 3). Hyperintense lesions on T2-weighted images in the bilateral thalamus, basal ganglia, brainstem, and the frontal and parietal lobe were seen at cranial MR in Patient 2 (Fig. 4a). No significant response was

Table 1 – Clinical summary of patients.

	Age[a] (day)/ gender	Clinical findings	First diagnosis	MRI	Gene mutation	Treatment and time	Outcome
Patient 1	60/M	Excessive crying, irritability, seizures, mental motor regression	Mitochondrial disorder	bilateral globus pallidus, putamina, thalamus, pons, both frontal lobes	c.623_624insA	(First hospitalization)- Coenzim Q, Tiamin, Carnitine, Riboflavin, Antiepileptics (more than 2)	Decrease in seizures, Spontaneous respiration was restored without mechanical ventilator support
Patient 2	30/F	Excessive crying, irritability, seizures	Mitochondrial disorder	pons, medial thalami, putamina, frontal and, parietal lobes	c.623_624insA	(First hospitalization)- Coenzim Q, Tiamin, Carnitine, Riboflavin, Antiepileptics (more than 2), ACTH	Decrease in seizures,
Patient 3	30/F	Excessive crying, irritability, downward deviation of the eyes, seizures	BTBGD	Pons, putamina, globus pallidi and thalami, cortical and sub-cortical cerebral hemispheres	c.620delinsAA	2 month/Biotin and Thiamine	Neuromotor developmental retardation, seizure continues
Patient 4	30/F	Excessive crying, irritability	BTBGD	bilateral perirolandic area, lateral thalami and posterior putamina	c.620delinsAA	1 month/Biotin and Thiamine	No significant change yet
Patient 5	37/M	Excessive crying, irritability, spasms, Respiratory insufficiency	BTBGD	Pons, bilaterally globus pallidus, putamina and thalami	c.894T > G (p.Y298[a])	1 month/Biotin and Thiamine	The neuromotor development retardation. Seizure not seen
Patient 6	23/F	Seizure, respiratory insufficiency	Mitochondrial disorder	putamina and bilaterally frontal and parietal lobe	c.894T > G (p.Tyr298)	1 month/Biotin and Thiamine	Exitus

[a] The age of the patients at the time of admission.

achieved to 100 mg thiamine therapy in either case, and mitochondrial disease was suspected after cranial imaging. Follow-up MRI showed atrophy, especially in the thalamus and basal ganglia (Fig. 2d). However there was a significant decline in progression of disease after treatment with high doses of thiamine and biotin (300 mg and 10 mg/kg/day, respectively) after genetic analysis. The number of seizures decreased from one seizure per day to one or two seizure per month. The patients' interest in their surroundings increased. Their mother told that they responding with reaction when they see her.

3.2. *Patients 3 and 4, c.620delinsAA mutation*

The third patient is now 2.5 years old (Fig. 5). She was a one-month-old girl when she applied to the hospital. Her eyes

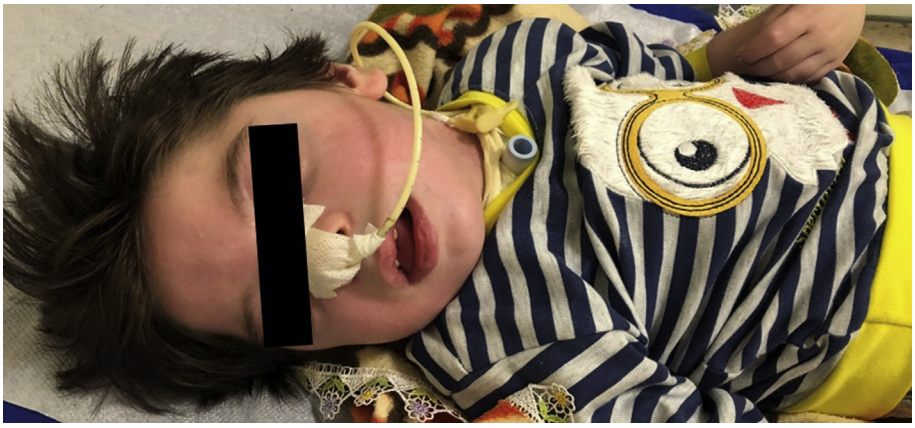

Fig. 1 – Patients 1, after treatment.

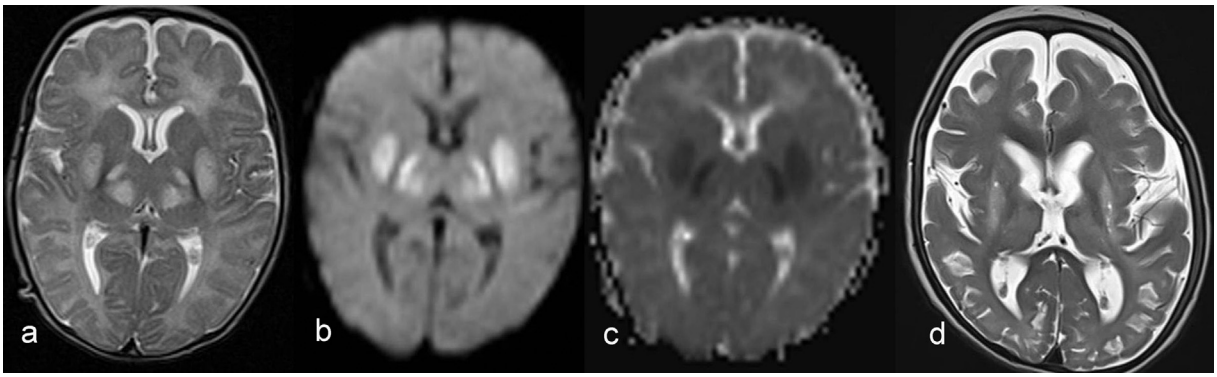

Fig. 2 – Axial T2-weighted MR image (a) shows symmetric hyperintense lesions in putamina and medial thalami. Diffusion-weighted image (b), and ADC map (c) findings are consistent with restricted diffusion. Following MR image (d) reveals diffuse cerebral atrophy and focal necrotic areas in basal ganglia.

deviated downward for approximately 5–10 s and then resumed their normal position. She was restlessness, with accompanying seizure-like eye movements and subsequent nutritional problems, spasticity and seizures. High signal areas were observed in the cortical and sub-cortical cerebral hemispheres, basal ganglia, brainstem and thalami at MRI (Fig. 6a–c). Since her cranial MRI findings closely resembled those of previous patients, she was diagnosed with BTBGD and started on high-dose thiamine and biotin therapies. The number of seizures decreased from one or two seizure per week to one or two seizure per month. The decline in the stages of motor development stopped, but she still could not sit alone. At follow-up MRI two months after treatment, regression was detected in the lesions in the pons, basal ganglia and thalami (Fig. 6d,e). In addition, cystic encephalomalacia was observed in the cerebral hemispheres (Fig. 6f).

The fourth patient, a one-month-old girl, was the sister of the third patient (Fig. 7). When she was admitted to hospital due to persistent cry and restlessness, cranial MR was evaluated, and genetic mutation was investigated due to the findings observed in her sister. High-dose thiamine and biotin therapy was initiated. Increased signal intensity was observed in bilateral perirolandic area, lateral thalami and posterior putamina (Fig. 8). The same genetic mutation as in the sister was identified.

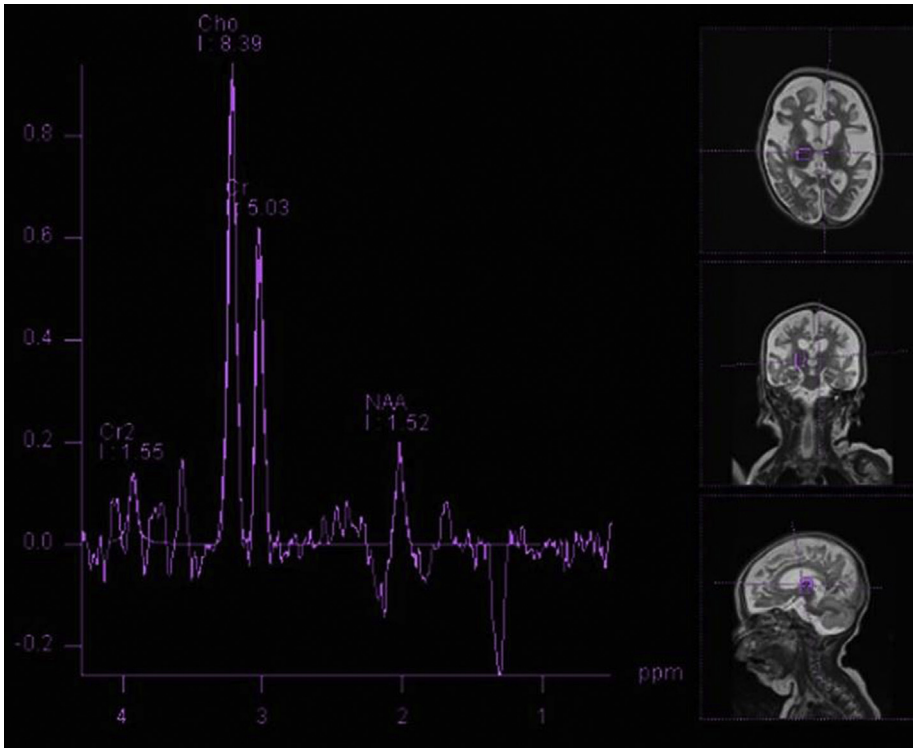

Fig. 3 – Decreased NAA and lactate peak (1.3 ppm), on TE 135 single voxel MR spectroscopy image, are consistent with neuronal loss in the right thalamus.

3.3. *Patients 5 and 6, c.894T > G (p.Y298*)*

The fifth patient was a 37-day-old boy admitted to hospital due to persistent crying and restlessness (he is one years old now). Respiratory insufficiency was determined, and spasticity and contractions were observed during follow-up. The patient was admitted to intensive care with mechanical ventilator support. Cortical and sub-cortical involvement was observed in the bilateral frontal and parietal lobes at MRI. Increased signal intensity was seen in the thalami, basal ganglia and brainstem on T2-weighted images (Fig. 9a,b). BTBGD was suspected, and the patient was started on high-dose biotin and thiamine therapies (10 mg/kg biotin, 300 mg thiamine). Genetic analysis was performed for *SLC19A3* mutation. Four weeks after treatment, regression was detected in the lesions at follow-up MRI (Fig. 9c,d). No seizures occurred in this patient after high-dose thiamin and biotin therapies, which were started immediately after receipt of the genetic analysis results. Despite rapid treatment and an appropriate dosage being applied, the patient's mental and motor development was significantly delayed for his age.

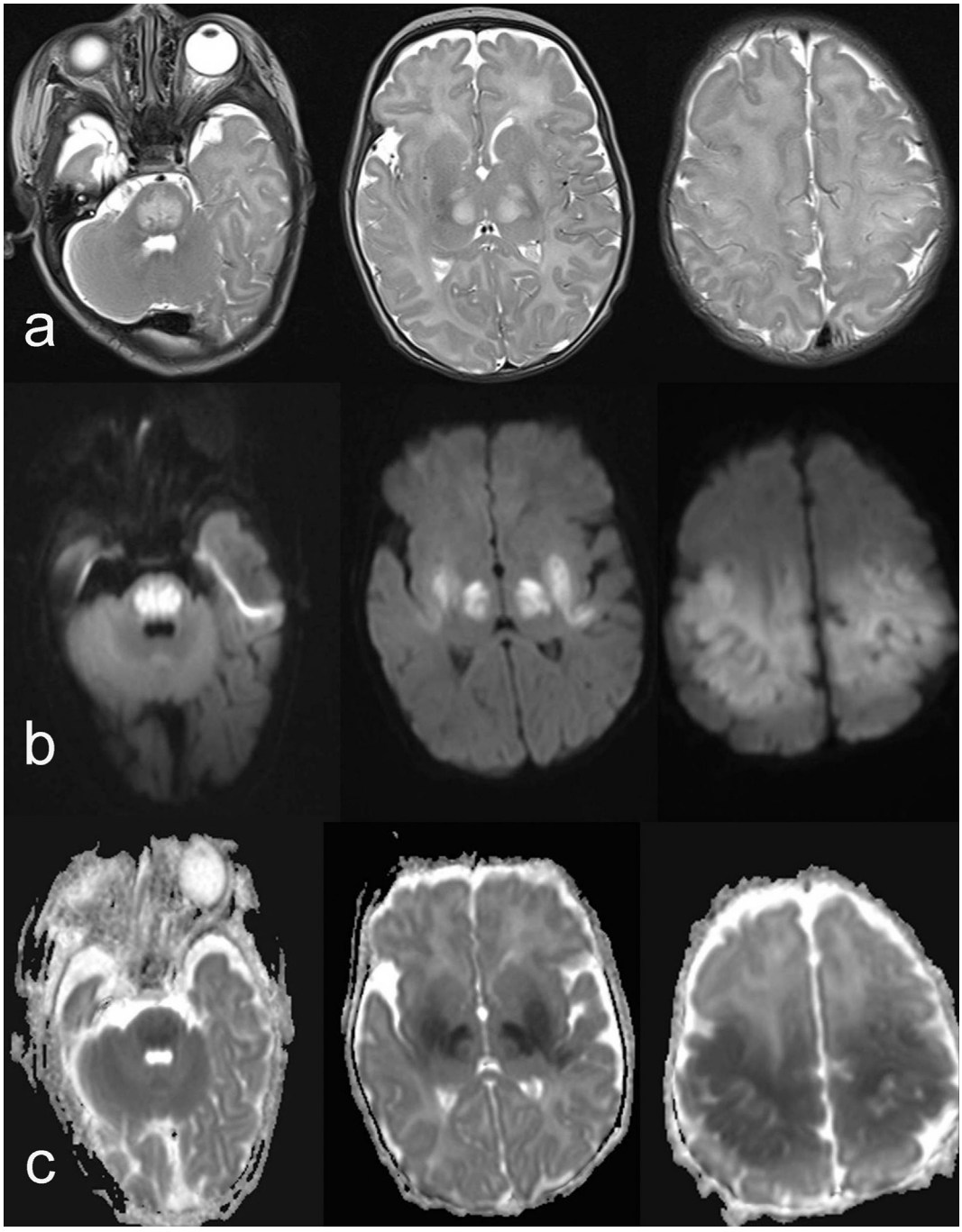

Fig. 4 – Axial T2-weighted MR images (a) show increased signal intensity in pons, medial thalami, putamina, frontal and, parietal lobes. There is restricted diffusion on DWI (b) and ADC map (c).

Fig. 5 – Patients 3, after treatment.

The sixth patient was a 23-day-old girl. She was admitted to the emergency department with status epilepticus, and was then transferred to the intensive care unit under anesthesia and attached to a mechanical ventilator due to refractory seizure. While in the emergency department due to seizures we learned that she had undergone clonic seizures for two days. A similar history was reported in two brothers who had died at the same age. Restlessness was the most common clinical finding in all patients. Increased signal was observed in the cerebral hemispheres, including the perirolandic cortex of the frontal and parietal lobes, basal ganglia, thalami and brain stem on T2-weighted MR images. Diffusion-weighted MRI showed restricted diffusion in the same regions (Fig. 10a,c). This case was diagnosed as status epilepticus.

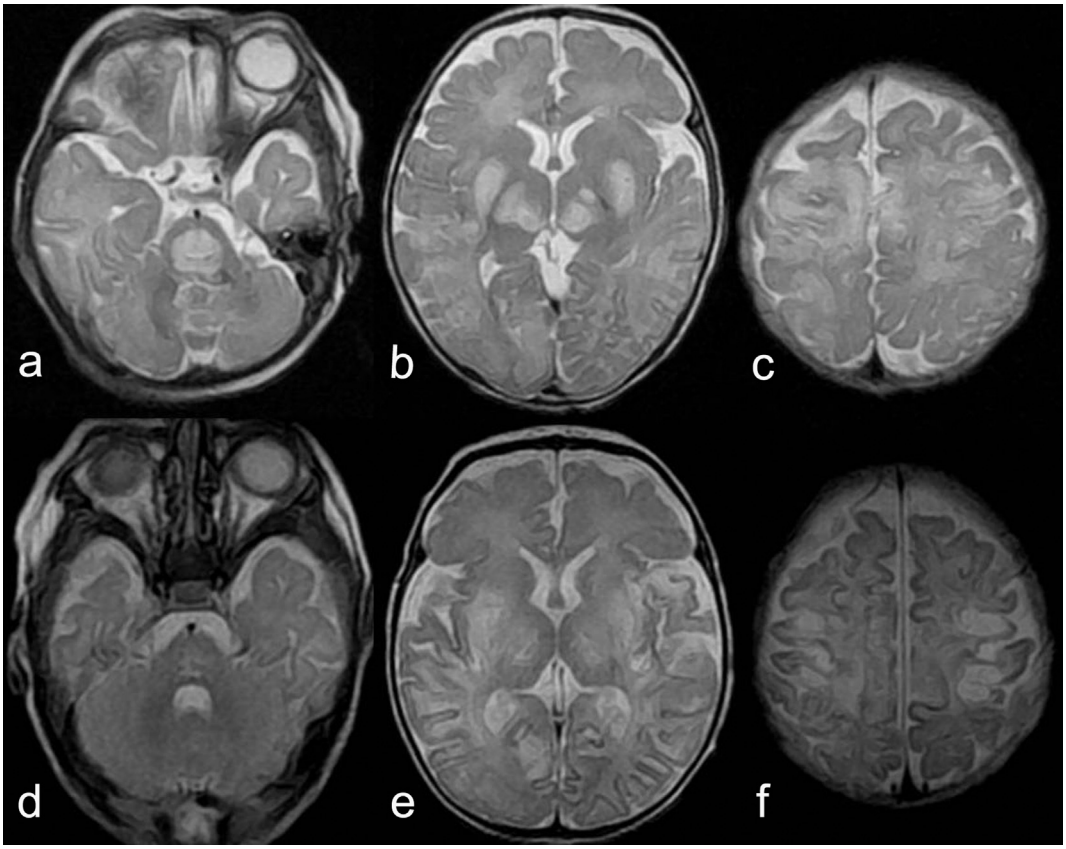

Fig. 6 – Axial T2-weighted MR images before (a–c) and 2 months after (d–f) treatment with thiamin and biotin. Increased signal intensity in the pons (a). Abnormal high signal and swelling of the putamina, globus pallidi and thalami (b). Cortical and sub-cortical high signal areas in the bilaterally cerebral hemispheres (c). Follow-up MR imaging after treatment (d–f) shows regression of the increased signal in the pons (d), globus pallidi, putamina and thalami (e). Pathologic changes have evolved cystic encephalomalacia in the cerebral hemispheres (f).

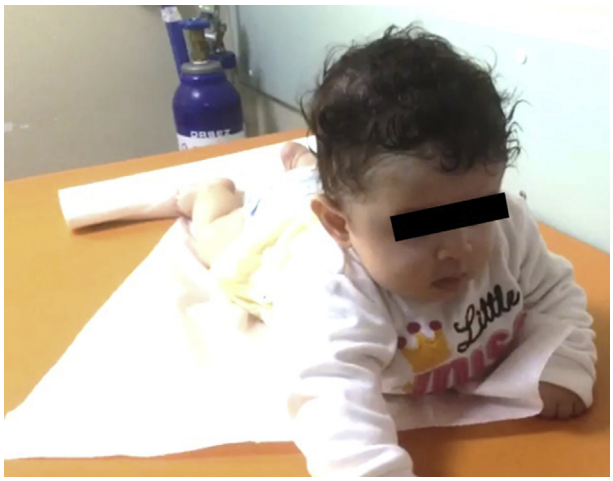

Fig. 7 – Patient 4, she was early diagnosed thanks to her sister.

Prolonged respiratory support with a mechanical ventilator in the intensive care unit was required due to pulmonary complications. No clinical improvement was achieved after starting high-dose biotin and thiamine therapy immediately after cranial MRI. On follow-up MR scans, progression was observed in all lesions despite treatment with thiamin and biotin (Fig. 10b,d). The patient died due to secondary respiratory problems.

Hypertonicity and skeletal hypotonia were described in the limbs of most patients. Genetic testing was performed with full exon sequence analysis in all six patients for mitochondrial disease and BTBGD. Newly identified mutations in the *SLC19A3* gene were detected in all patients. The same novel mutations (*c.623_624insA*) were identified in the two siblings. Their parents also had the same mutations in heterozygous form. The third and fourth patients, who were siblings, had a novel homozygous mutation (*c.620delinsAA*). The fifth and sixth patients had the same newly identified mutations (*c.894T > G* (*p.Y298**)) even though there was no familial relation between them (Fig. 11).

4. Discussion

SLC19A3 gene mutation was investigated in six patients referred to the Pediatric Neurology Clinic with recurrent encephalopathies, hypotonia, restlessness, seizures, and cranial MR findings. Three novel mutations were identified, including the same mutations in two siblings and in two unrelated patients referred from different locations. This study identified, in contrast to other cases of BTBGD in the literature, typical

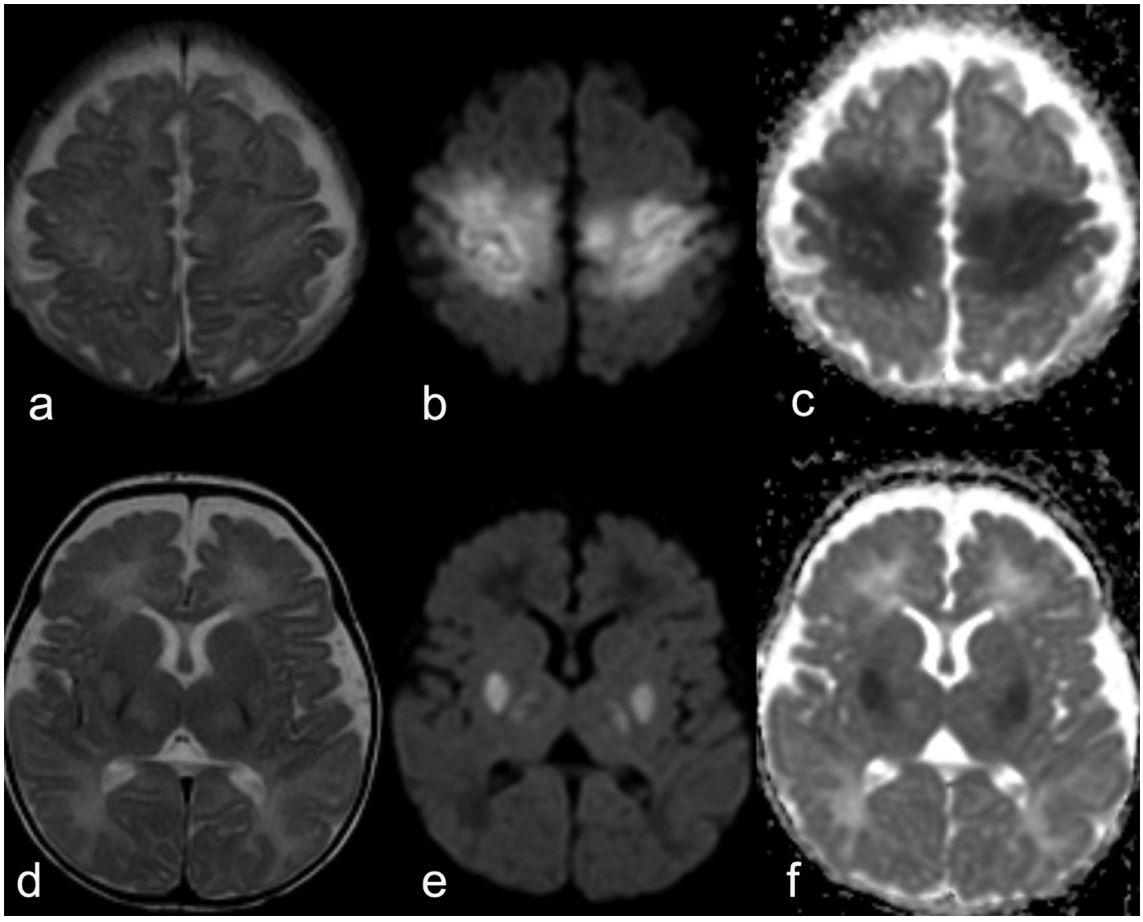

Fig. 8 – Axial T2-weighted MR images (a, d) show increased signal intensity in bilateral perirolandic area, lateral thalami and posterior putamina. There is restricted diffusion on DWI (b, e) and ADC map (c, f) in the same areas.

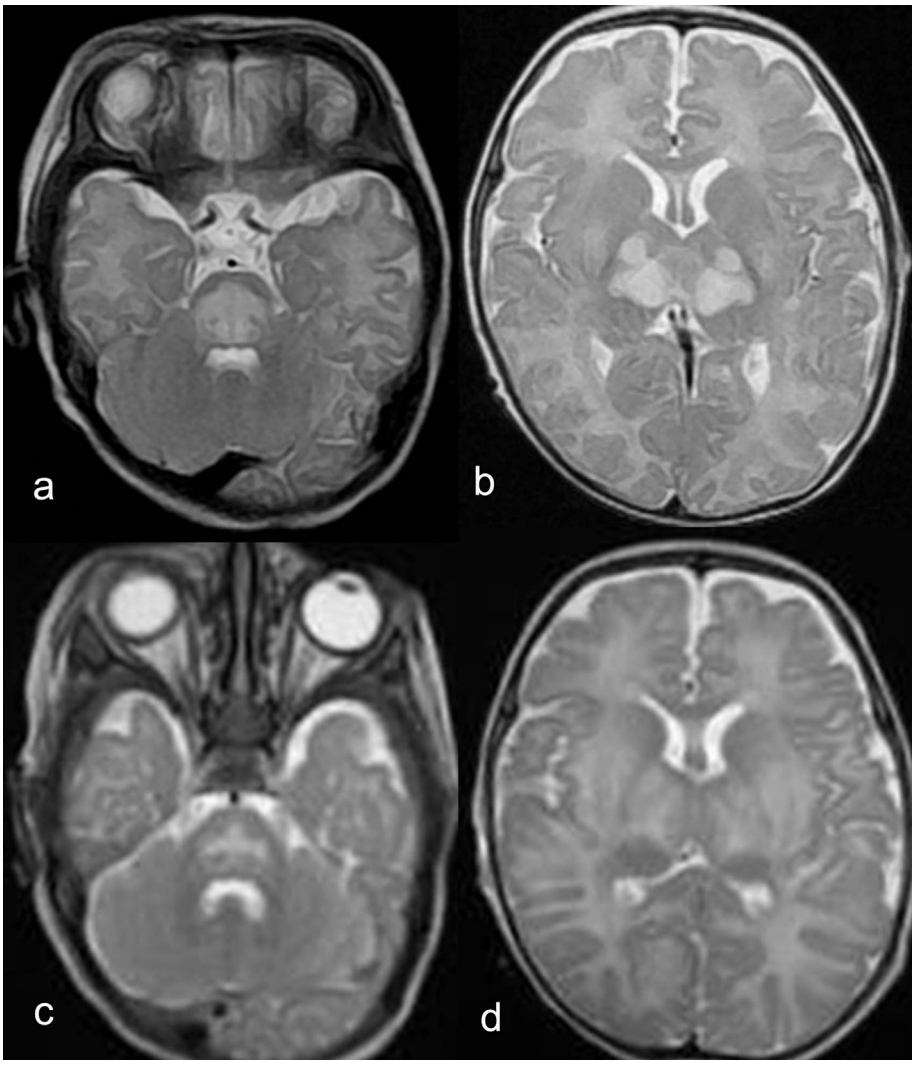

Fig. 9 – Axial T2-weighted MR images before (a, b) and 4 weeks after (c, d) treatment with thiamin and biotin. Abnormal areas of increased signal intensity in the pons (a). Swelling and abnormal hyperintense areas in the bilaterally globus pallidus, putamina and thalami (b). Follow-up MR image after treatment (c, d) reveals regression of the increased signal in the pons (c), bilaterally globus pallidus, putamina, basal ganglia and thalami (d).

MRI findings and different mutations emerging with clinical findings at early ages and a mortal course.

Clinical findings of BTBGD include subacute encephalopathy episodes, confusion, dysarthria, dysphagia, ataxia, central facial nerve palsy, external ophthalmoplegia, dystonia, rigidity, quadriparesis, associated febrile illness, coma and death. The ages at onset of 10 patients first described by Ozand et al.[4] were generally 5–6 years, and initial findings consisted of dystonia, paresis, and seizure or encephalopathic attacks triggered during infection. In their retrospective investigation of 18 BTBGD patients, Alfadhel et al.[5] reported mean ages at onset of 3–4 months. The most common clinical findings in those patients were ataxia, dysarthria, dystonia, and seizures. Age at onset of first clinical findings in the patients reviewed in our study was generally one month. The first clinical finding in all subjects was restlessness and incessant crying. Dystonic movements and tonic seizures were subsequently added to this manifestation in the first two cases, which were diagnosed late. Hypotonia, delayed motor development, and persisting seizures were recorded at follow-up. Three other patients presented with similar manifestations. The sixth patient presented with a more severe clinical manifestation and severe clonic seizures at the age of 23 days, and the subject died. We learned that this patient's two siblings had died immediately following seizures commencing at the same age. Due to the presence of such a history, the family/parents brought the patient to hospital three days after onset of seizures, and unfortunately intervention was too late.

Cranial MR findings of BTBGD in the literature generally consist of bilateral necrosis and severe edema in the basal ganglia. Caudate, nucleus, putamen and globus pallidus involvement occurs during the acute phase. Thalamus, cerebellum, brainstem and gray-white junction involvement are also seen. The changes are hyperintense on T2 images,[2–4] while the lesions generally exhibit diffuse restriction. Increased lactate levels can be observed at time of MR

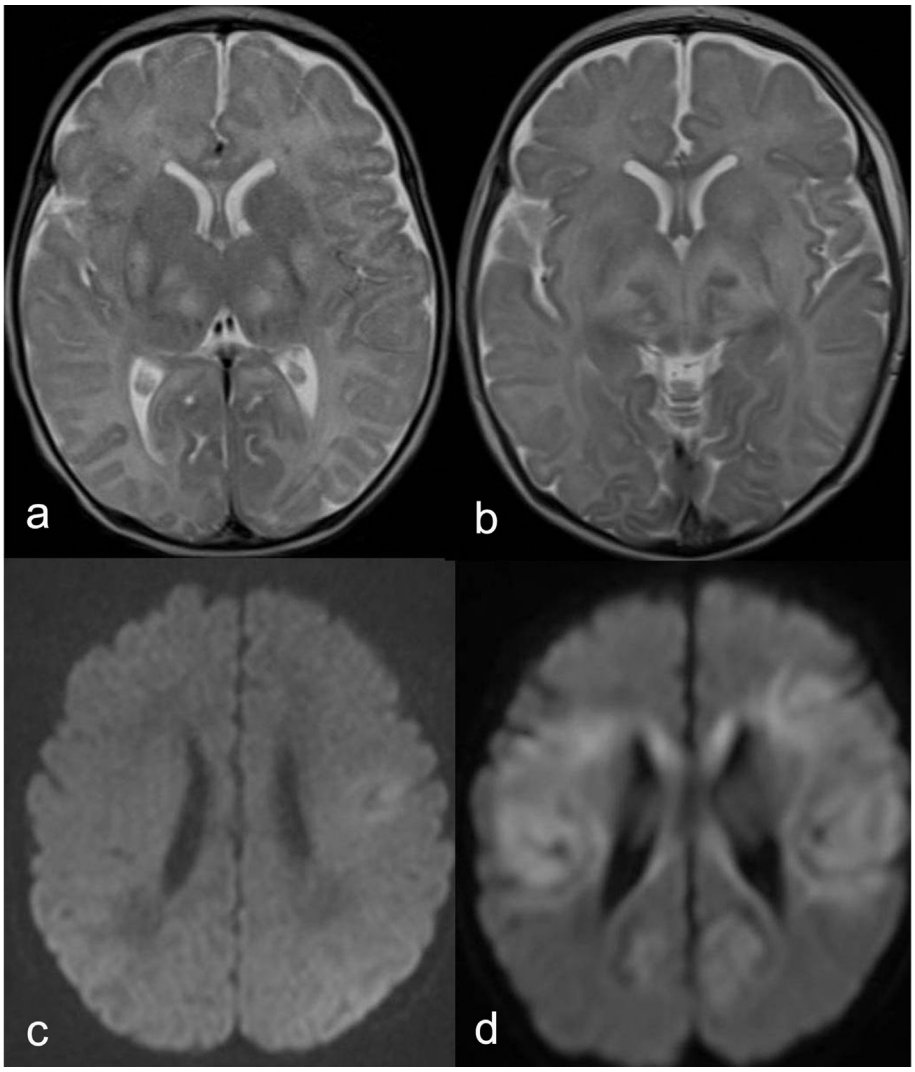

Fig. 10 – Axial T2-weighted MR images (a, b) and DWI (c, d) show increased signal intensity in the putamina and bilaterally frontal and parietal lobe. 2-week after (b, d) treatment with thiamin and biotin, swelling and abnormal hyperintense areas reveal progression.

spectroscopy, but this is not specific. Alfhadel et al.[5] reported basal ganglia, diffuse cortical, subcortical, and white matter involvement in acute attacks in 18 patients diagnosed with BTBGD, and signal increase, atrophy and necrosis in basal ganglia in the chronic period, and more rarely cerebellar and cervical spinal cord involvement. Kassem et al.[6] examined MRI findings at time of diagnosis and during follow-up in 15 patients. Complete or partial involvement in the caudate nuclei and putamen was observed in all cases. Involvement was observed in the mesencephalon, cerebral cortical and subcortical region, and the thalamus in 80% of cases. Patchy affection in white mater was seen in 53%, and cerebellar involvement in 13.3%. Regression in lesions and atrophy was observed after treatment. We observed putamen and thalamus involvement in all the cases in our study. Pons involvement was observed in two cases, and cortical involvement, particularly in the frontal and parietal region, in four. Cerebellar involvement was not present in any of our cases. Diffusion restriction was present in all cases. At follow-up of five cases, atrophy was seen at MRI in two cases, regression in three, and progression in one. Typical involvement was particularly in the putamen and thalamus in all our cases.

BTBGD is also known as thiamine transporter-2 deficiency. The general consensus is that thiamine therapy alone is not beneficial without high-dose biotin.[4,7,8] However, recent studies have reported that thiamine therapy can be beneficial by itself in these patients.[1,9] There are also studies suggesting that high doses of biotin and thiamine in BTBGD patients may lead to increased expression of the *SLC19A3* gene in vitro, which may lead to some improvement in thiamin transport. Mitochondrial disease was suspected in our two siblings, and these were given long-term thiamine (100 mg/day), but no significant benefit was achieved. High-dose biotin therapy combined with diagnosis and elevation of the thiamine dose resulted in a marked improvement in the seizures, greater interest in the surrounding environment and improved disease progression. Treatment with thiamine alone in these

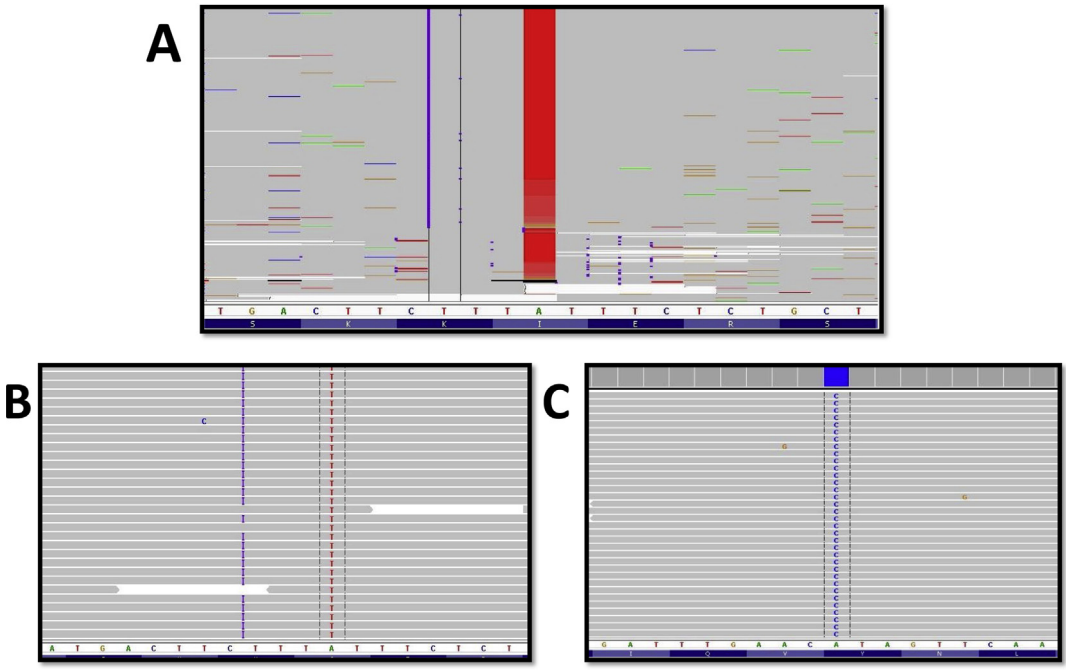

Fig. 11 – *SLC19A3* mutation was found. A: Patient 1-2 sequence result. B: Patient 3-4 sequence result. C: Patient 5-6 sequence result.

newly described mutations in our patients may therefore not have been sufficient in the first two cases. In the other four cases high-dose thiamine and biotin therapy halted the progression of the disease and reduced the seizures. The patient with refractory seizures receiving intensive care support died due to the family presenting late to hospital and to delayed treatment. We learned that this patient's two siblings had died at the same ages with similar manifestations. The clinical course was more rapid and moral in these patients.

Numerous neurometabolic diseases can be seen in childhood. Diagnosis with biochemical, genetic and metabolic tests may take a long time. MRI findings can be indicative for diagnostic and other tests in these patients. *SLC19A3* mutations, a relatively novel form of BTBGD, are noteworthy in terms of being of early onset, involving refractory seizures and progressive encephalopathy, progressing rapidly and having a fatal course. Rapid diagnosis and treatment can be life-saving in such cases. We think that if patient has *SLC19A3* mutations and also their parents have same mutations as heterozygous, may be better to given high dose thiamine and biotin treatment.

Ethical standards

This study was funded by Karadeniz Technical University Scientific Research Projects Unit. All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. The requisite permissions were received from patient-s'families before the genetic tests.

Conflict of interest

All authors declares no conflict of interest about this study.

REFERENCES

1. Subramanian VS, Marchant JS, Said HM. Biotine responsive basal ganglia disease-linked mutations inhibit thiamine transport via hTHTR2: biotin is not a substrate for hTHTR2. *Am J Physiol Cell Physiol* 2006;**291**:851–9.
2. Schänzer A, Döring B, Ondrouschek M, Goos S, Garvalov BK, Geyer J, et al. Stress-induced upregulation of *SLC19A3* is impaired in biotin-thiamine responsive basal ganglia disease. *Brain Pathol* 2014;**24**:270–9.
3. Vlasova TI, Stratton SL, Wells AM, Mock NI, Mock DM. Biotin deficiency reduces expression of *SLC19A3*, a potential biotin transporter, in leukocytes from human blood. *J Nutr* 2005;**135**:42–7.
4. Ozand PT, Gascon GG, Al Essa M, Joshi S, Al Jishi E, Bakheet S, et al. Biotin-responsive basal ganglia disease: a novel entity. *Brain* 1998 Jul;**121**(Pt 7):1267–79.
5. Alfadhel M, Almuntashri M, Jadah RH, Bashiri FA, Al Rifai MT, Al Shalaan H, et al. Biotin-responsive basal ganglia disease should be renamed biotin-thiamine-responsive basal ganglia disease: a retrospective review of the clinical, radiological and molecular findings of 18 new cases. *Orphanet J Rare Dis* 2013;**8**:83.
6. Kassem H, Wafaie A, Alsuhibani S, Farid T. Biotin-responsive basal ganglia disease: neuroimaging features before and after treatment. *AJNR Am J Neuroradiol* 2014 Oct;**35**(10):1990–5.
7. Ygberg S, Naess K, Eriksson M, Stranneheim H, Lesko N, Barbaro M, et al. Biotin and thiamine responsive basal ganglia disease – A vital differential diagnosis in infants with severe encephalopathy. *Eur J Paediatr Neurol* 2016;**4**:457–61.

8. Tabarki B, Alfadhel M, AlShahwan S, Hundallah K, AlShafi S, AlHashem A. Treatment of biotin-responsive basal ganglia disease: open comparative study between the combination of biotin plus thiamine versus thiamine alone. *Eur J Pediatr Neurol* 2015;**19**:547–52.

9. Tabarki B, Al-Shafi S, Al-Shahwan S, Azmat Z, Al-Hashem A, Al-Adwani N, et al. Biotin-responsive basal ganglia disease revisited: clinical, radiologic, and genetic findings. *Neurology* 2013;**80**:261–7.