CONGENITAL DISORDER OF GLYCOSYLATION TYPE Ia

A non-progressive encephalopathy associated with multisystemic involvement

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Congenital disorders of glycosylation (CDG) are a group of severe, autosomal recessive, multisystemic diseases, characterized by abnormal glycosylation of glycoproteins and glycolipids[1]. The first disease of this group was reported in 1980[2]. Since then, at least 16 entities were recognized and were classified in two groups (CDG-Ia to IL and CDG-IIa to IId). Each disease is caused by a specific enzymatic deficiency[3]. CDG-Ia is related to deficient phosphomannomutase activity and represents 80% of CDG type I[4]. The enzyme is coded by PMM2, a gene situated in chromosomal segment 16p13. The estimated prevalence of CDG-Ia is as high as 1:20000 newborns among caucasians[5], therefore making this disease one of the most common metabolic disorders. CDG-Ia has a broad spectrum of age-related clinical findings[6] with variation even within the same sibship[7]. Table 1 lists the most important clinical features. Failure to thrive, gastrointestinal and neurologic symptoms predominate in the neonatal period and infancy. The most important features during childhood are hypotonia, ataxia, mental retardation, joint contractures, and pigmentary retinopathy. It is estimated that 20% of patients die during the first year of life due to severe infection, liver insufficiency or cardiomyopathy[8]. Some patients develop multiorgan system failure. There is no neurologic regression. Muscle atrophy, peripheral neuropathy and secondary skeletal deformities ensue during adolescence. Some patients survive into adulthood[9]. Adults have premature aging and hypergonadotropic hypogonadism occurs in females[10]. When CDG is suspected, a serum sample is analyzed for transferrin hypoglycosylation by isoelectric focusing or other available method. Diseases like galactosemia and hereditary fructose intolerance must be ruled out, as they are known to cause false-positive results on transferrin isoelectric focusing[11,12]. The diagnosis of CDG-Ia is confirmed by assay of phosphomannomutase, which is deficient in either leukocytes or fibroblasts. In some undefined cases, direct gene sequencing is necessary to establish the diagnosis.

Table 1. Clinical manifestations of CDG-Ia.

Neonatal and infancy	Central nervous system
	Psychomotor delay
	Hypotonia
	Abnormal eyes movements
	Squint
	Cerebellar hypoplasia (especially cerebellar *vermis*)
	Dysmorphisms
	Bitemporal narrowing
	Almond-shaped eyes
	Inverted nipples
	Gluteal, perineal and digital fat pads
	Gastrointestinal
	Anorexia, vomiting, diarrhea
	Others
	Failure to thrive, osteopenia
Childhood	Central nervous system
	Mental retardation
	Epilepsy
	Ataxia
	Stroke-like episodes
	Pigmentary retinopathy
	Hemostasis: coagulopathy (thrombotic tendency)
	Joint contractures
	Multi-organ failure (liver, kidney, heart)
Adolescence	Peripheral neuropathy
	Muscle atrophy
	Kyphoscoliosis
Adult	Premature aging
	Hypergonadotrophic hypogonadism (in females)

The aim of this report is to increase health professionals' awareness of this disease, which is clinically nonspecific and relatively frequent. This is the first description of a CDG-Ia Brazilian patient, although other patients' data

DISTÚRBIO CONGÊNITO DA GLICOSILAÇÃO TIPO Ia: ENCEFALOPATIA NÃO-PROGRESSIVA ASSOCIADA A ENVOLVIMENTO MULTI-SISTÊMICO.

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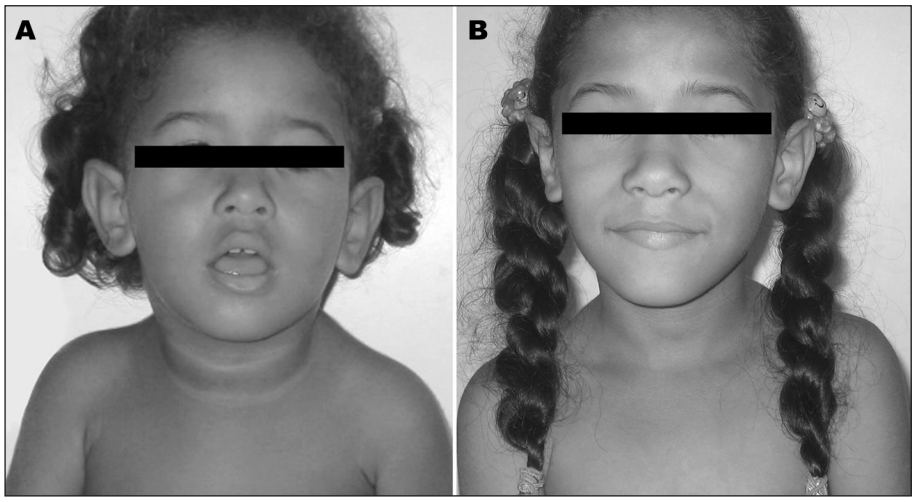

Fig 1. Mild facial dysmorphism. (A) Note bitemporal narrowing, up-slanting palpebral fissure, inverted nipples and almond-shaped eyes (19 months old) and (B) Almost normal facial features (6 years old).

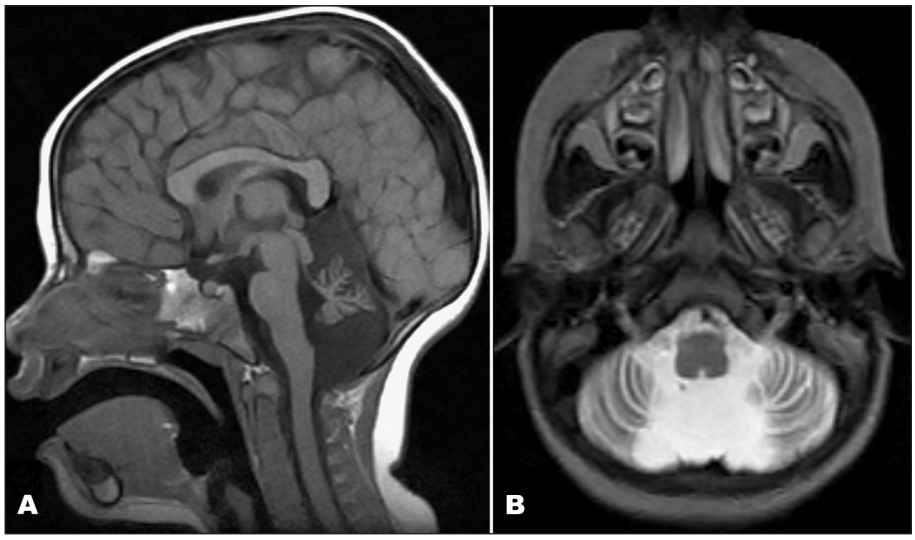

Fig 2. Magnetic resonance imaging (22 months old). (A) Cerebellar vermis and brain stem atrophy. (B) Cerebellar folia widening, suggestive of atrophy.

have been presented at national and international congresses, including those presented by our own group[13-17].

CASE

The patient, a six year old girl, is the third child of a healthy nonconsanguineous couple. She was born by cesarean delivery, due to postdate pregnancy, otherwise normal. Birth lenght was 53 cm and weight was 3,535 gm, both on 75th percentile, and head circumference was 33.5 cm, on 25th percentile. Apgar scores were 2 and 7 at one and five minutes, respectively. She was submitted to cardiopulmonary resuscitation, followed by oxygen therapy for four hours in intensive care unit. The girl was discharged from hospital after five days. The diagnoses were meconium aspiration syndrome and neonatal sepsis. During a gastroenteritis episode, at two months of age, the patient presented seizures successfully treated with phenobarbital. She was first seen in our service at the age of four months, due to the history of seizures, and showed low weight and normal neurologic examination. Mild facial dysmorphisms, inverted nipples and convergent squint (Fig 1A) were present during the first year of life, when cognitive impairment, failure to thrive and microcephaly (42 cm; below percentile 5) became noticeable. At follow-up she had development delay: head control at 11 months of age, monosyllabic babble at 19 months, first real words at 29 months, and walking with support at the age of 36 months. She never reached independent walking, and became wheelchair-bound at four years of age. She developed ataxia and dysarthria at the age of three years. At five years of age deep tendon areflexia and generalized muscle hypotrophy were noted. Facial dysmorphisms were less evident at the age of six years (Fig 1B). The child presented recurrent episodes of upper airways infection and diarrhea, as well as low weight gain. Stature as well as muscle tonus remained normal. The child had cognitive impairment, according to Bayley Scales of Infant Development (below 5th percentile for cognitive and expressive language skills and between 5th and 10th percentiles for receptive language). Hepatomegaly and high plasma transaminases levels led to the discontinuation of phenobarbital therapy at six months of age. As these findings persisted, they were con-

Table 2. Patient's hormone and coagulation tests results.

Test	Result	Reference range
FSH	146.5 UI/L	1.2–5.7*
LH	5.8 UI/L	0.1–1.3*
Factor VIII	>200%	50–150
Factor XI	20%	60–140
Protein S	49%	70–130
Protein C	34% and 32%	70–130
Antithrombin III	52% e 37%	80–120
PTTa	1.5	<1.3

* Values refer to 10th and 90th percentiles.

sidered to be independent of the drug use. At the age of three years, there was recurrence of seizures, which were controlled with phenobarbital therapy, without evidence of hepatopathy. Initial diagnoses were static chronic encephalopathy due to perinatal anoxia and cow milk intolerance. The patient was referred since admission to motor and cognitive rehabilitation therapy with partial improvement.

Laboratory workup showed normal results for: hepatitis and other congenital infections serology, blood gases, plasma ammonia, lactate and screening for inborn errors of metabolism. Serum alanine aminotrasferase (ALT), aspartate aminotransferase (AST), and gamma-glutamyltransferase levels persisted elevated until 18 months of age. Screening for lysosomal storage diseases was also normal, except for plasma hexosaminidases, which showed a significant increase in activity: total hexosaminidases=4.472 nmol/h/mL (normal range=520–1.420) and hexosaminidase A (with sulfated substrate)=39 nmol/h/mL (normal range=12–27). These results led us to suspect of CDG. Plasma transferrin isoelectric focusing displayed CDG I pattern. Determination of leukocyte phosphomannomutase showed deficient activity: 0.17 nmol NADPH/min/mg of protein (controls=0.96–3.56). This result confirmed the diagnosis of CDG Ia. Some serum glycoproteins, especially FSH and LH, showed increased activity or levels, while most glycoproteins involved in coagulation had decreased activity (Table 2). Additional 19 tested glycoproteins showed normal values. The patient had progressive lymphopenia: 3.500 to 1.792 cells/mm^3 (normal range=4.000–13.500). HIV test was negative.

Electroencephalogram was normal at five months of age. At 3.5 years it showed abnormal patterns suggesting diffuse cortical and subcortical dysfunction. Electroneuromyography revealed generalized demyelinating sensitive and motor polyneuropathy at 22 months and at 4.5 years of age. Auditory, visual and somatosensory evoked potential were normal. Magnetic resonance imaging (MRI) performed at 22 months of age disclosed myelination delay, cerebellar *vermis* atrophy, mild IV ventricle widening and brain stem atrophy (Figs 2A and 2B).

This study was approved by the Ethics Committee of the hospital and written informed consent was obtained from the parents.

DISCUSSION

The patient showed nonspecific signs (e.g., seizures, developmental delay and recurrent infections) common to many diseases, including CDG. Initially, chronic encephalopathy secondary to perinatal anoxia and lactose intolerance were considered as diagnoses. Later, we had some clues that made us suspect of CDG. The first puzzling finding was the fluctuating and unexplained increase in serum levels of gamma-glutamyltransferase, ALT, AST, and plasma hexosaminidases. Increased activity of hexosaminidases in serum has been reported in CDG-Ia[18]. The association of increased plasma hexosaminidases activity, liver dysfunction and cerebellar atrophy in a child with inverted nipples and mild facial dysmorphisms led us to suspect of CDG. The pattern of transferrin by isoelectric focusing was compatible with CDG type I. Markedly deficient phosphomannomutase activity in the patient (7.5% of the mean control) and intermediate (heterozygote) levels in his parents (47.8% e 36.3% of the controls' levels) established the diagnosis of CDG-Ia.

The diagnosis can be difficult due to the broad spectrum of clinical presentations. The patient did not have hypotonia or short stature, two of the most frequent findings in CDG-Ia[8,19]. In addition, she did not have abnormal subcutaneous fat deposition, with "fat pad" on the buttocks, which suggests the diagnosis[7]. The deficiency of several glycoproteins (e.g., hormones, transport protein, and membrane receptors) is the biochemical basis of the involvement of several organs seen in this disease. In our patient, glycoproteins involved in hemostasis were abnormal, although without overt clinical manifestation so far. Increased FSH and LH levels in the present patient can be considered as an early symptom of hypergonadotrophic hypogonadism, a typical finding in girls with CDG[20]. The results of these tests must be carefully interpreted, as most laboratories provide only adult reference ranges. Pediatric values have been published, but results vary according to the laboratory method used for determination[21-24].

Our patient showed hepatomegaly regression and normalization of liver enzymes at 18 months of age. She has had recurrent diarrhea with partial improvement with age. Gastrointestinal symptoms in CDG type I could be secondary to inflammatory disease in liver and small intestine[25], and usually resolve spontaneously after some years[26]. Neuropathy has been addressed as a medical issue of adults with CDG-Ia[27]. Our patient showed, at 22 months of age, early signs of peripheral neuropathy, which is thought to be the cause of her muscle hypotrophy. It is interesting to note that in spite of the existing neuropathy

and cerebellar atrophy, the patient did not have hypotonia. Depression of immunity, which has been shown to be secondary to neutrophilic dysfunction and to low immunoglobulin levels, is also a feature of CDG-Ia[28]. Although we were not able to obtain the patient's data on immune function, there was evidence of immune impairment (e.g., recurrent episodes of infection). Furthermore, our patient had lymphopenia, a feature not reported in CDG type Ia yet. This finding in our patient does not seem to be due to protein-losing enteropathy (a feature of CDG Ia), as diarrhea became less frequent with age, in contrast with the progressive lymphopenia and the persistent normal levels of plasma albumin.

The aim of this report is to draw attention of health professionals on CDG as a cause of neurologic impairment. CDG is underdiagnosed due to lack of health professionals' awareness, its nonspecific symptoms, and unavailability of widespread laboratory testing. CDG should always be investigated in any patient with any unexplained multisystem involvement, especially if there is neurologic impairment[3]. Diagnosis allows genetic counseling and prenatal diagnosis. There is not a specific treatment available for CDG-Ia, but motor and cognitive rehabilitation is advisable in order to improve patient's quality of life.

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