案报告

儿童患者因酮症性低血糖诊断为糖原合成酶缺陷

由于新识别的GYS2基因突变

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1. Introduction

糖原合成酶缺陷，也称为糖原沉积病(GSD) 0型是一种由GYS2基因突变引起的先天性错误，该基因位于肝脏中，传递为常染色体显性遗传 [1]。它是一种罕见的肝脏糖原储存疾病，小于30例报告在文献中[2,3]。该障碍是由于糖原合成酶缺陷，几乎没有肝脏的糖原合成，肝脏不能看到在患者中糖原合成酶缺陷。

症状的倒影性低血糖症在患者中GSD 0通常出现为第一次在婴儿期在喂养从食物中醒来。儿童可能有早晨的疲劳、皮肤、呕吐和疲劳。低血糖症与低血糖症的出现相关，但它们是罕见 [4]。通常低血糖症在实验室测试中出现为在诊断过程中发生的差异常态过程 [5]。

背景：糖原合成酶缺陷（糖原存储疾病0—GSD 0）由于在GYS2基因上的突变而引起。它是一种由于缺乏肝脏中糖原合成的罕见疾病。它是一个在文献中所报道的罕见条件。在文献中报道的文献中，该障碍被识别为GYS2基因上的新同源突变：NM_021957.3:p.Phe574Leu/c.1720T>C在ex. 14。

A random, asymptomatic hypoglycemia with ketonuria was found in this patient at the age of 7. His developmental parameters were within normal ranges. Oral glucose tolerance test showed normal baseline blood levels of glucose, insulin and lactate, and their increase following glucose intake. Eight-hour fasting plasma glucose test, revealed glucose blood level of 34 mg/dl with no clinical symptoms. The results of these tests suggested GSD 0. Molecular analysis of the GYS2 gene was not feasible, but this particular gene was included in the panel of hypoglycemia of whole exome sequencing (WES) which was at our disposal.

A 7-year-old boy was referred to the Department of Endocrinology and Metabolic Disorders for the diagnosis of hypoglycemia. A random hypoglycemia of 32 mg/dl with no clinical symptoms, and massive ketonuria were found during his hospitalization at the pediatric ward due to infection.

In general, GSD 0 has no impact on physical or mental development, only some children may have a mild growth delay [5].

Alike other hepatic glycogenosis, the goal of management for GSD 0 is to prevent hypoglycemia by avoiding fasting. Clinical management is therefore based on frequent meals composed of high protein intake during the day and addition of uncooked cornstarch in the evening [6].

Next-generation sequencing techniques including whole-exome sequencing (WES) have now opened promising possibilities to identify the molecular background of rare metabolic disease [7]. Through the use of WES technology with one experiment, it is possible to identify pathological mutations in an ample number of selected genes that are potentially associated with the disorder being tested.

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He was born at term, as the first child of parents of Polish origin who declared non-consanguinity, however having lived in the neighboring villages. His body mass was 3150 g, head circumference 34 cm, and Apgar score 10p.

Boy’s psycho-motor and physical development were normal; 50th weight-for-age percentile and 25th height–for-age percentile, but his mid parental height was 167.5 cm.

Patient’s glucose profile studied during hospitalization revealed a random hypoglycemia without post-prandial hyperglycemia. Lactates were measured during function tests. Glucose challenge showed baseline blood levels of glucose, insulin and lactate within normal values, and their increase following glucose intake. After 8 h of prolonged fasting test, hypoglycemia of 34 mg/dl with no clinical symptoms, and poor response to glucagon were found (Table 1). The glucose and lactate monitoring was performed using both laboratory readings (glucose and lactate) and meter determinations (glucose).

Outcomes of both function and laboratory tests suggested the diagnosis of glycogen synthase deficiency.

Whole exome sequencing (WES) was conducted to identify the molecular basis of the hypoglycemia in the patient. The study was approved by the Ethics Committee of the CMHI. Informed consent from the guardians of the patient undergoing large-scale sequencing was obtained.

WES was performed on a HiSeq 1500 using an Exome Enrichment Kit (Illumina) according to published protocol [8]. Generated reads were aligned to the hg19 reference human genome. Alignments were viewed with Integrative Genomics Viewer v.2.3.40. The mutations

![Integrative Genomics Viewer view of GYS2 homozygous mutation found in the proband by whole-exome sequencing. Nomenclature of mutation followed the guidelines of the Human Genome Variation Society using NM_021957.3 as a reference cDNA sequence for GYS2 gene. Gen GYS2 (RefSeq NM_021957.3; NP_068776.2; CCDS8690.1). Genotyp zgodny z HGVS v.2.0: NM_021957.3: c.|1720T→C||1720T→C]. Protein gene description: NP_068776.2: p.[Phe574Leu].[Phe574Leu].](image)
A 8-month-old infant
A 9-year-old girl
A boy of Italian ancestry at the age 21 months
3 children from 2 German families
A 15 month-old child
A 7 year-old Canadian girl
2 children
Our patient

| Patient | Symptoms and metabolic profile | Symptoms | Ketonuria | Baseline lactate and glucose | References |
|---------|--------------------------------|----------|-----------|-----------------------------|------------|
| An 8-month-old infant | Transient neurological symptoms improved after the feed | - | N | Lewis et al. [9] |
| A 9-year-old girl | Hypoglycemic seizures at the age of 7 years | P | N | Aynsley-Green et al. [10] |
| A boy of Italian ancestry at the age 21 months | Signs of hepatic deficiency with mild clinical symptoms contrasted with a remarkable fatty liver degeneration + atypical reaction to fructose overload | P ↑ (L)/(N)/G | de Kremer et al. [14] |
| 3 children from 2 German families | - 2 cases: morning fatigue rapidly disappearing after heating - 1 case: asymptomatic | P | N | Gitzelmann et al. [11] |
| A 15 month-old child | Generalized tonic–clonic seizures after night fasting | P | N | Rutledge et al. [12] |
| A 7 year-old Canadian girl | Asymptomatic | P | N | Laberge et al. [13] |
| 2 children | Glucosuria and hyperglycemia | P | N | Bachrach et al. [15] |
| Our patient | Asymptomatic | P | N | Our patient |

P – present.
N – normal.

identified as pathologies were confirmed using the Sanger method following the standard protocol (BigDye® Terminator v3.1 Cycle Sequencing Kit, Applied Biosystems®).

Homozygous molecular variant c.1720T>C (p.Phe574Leu) in the GYS2 gene was identified (Fig. 1). It is a novel missense substitution, which was not observed in a control group of 800 alleles and was not identified in the different Exome Sequencing Projects (e.g. ESP 6500, ExAC 65000). In silico prediction by various algorithms was assessed and pathogenic effect of mutation was revealed: CADD (result: 34), MetaSVM (Deleterious), Polyphen2 HDIV (Deleterious) and HVAR (Deleterious), Mutation Assessor (Medium), LRT (Deleterious), MetaLR (Deleterious), SIFT (Deleterious), and MutationTaster (Deleterious). In order to predict the pathogenicity of the mutation, a bioinformatic analysis was conducted instead of functional studies, which are time-consuming and long-term techniques.

Parents have not been tested to confirm the significance of a homozygous variant, because the patient’s father was not available for blood sample collection as he had emigrated.

Dietary management with frequent meals (every 3–4 h with late dinner and early breakfast), protein supplementation and administration of uncooked cornstarch (1–1.5 g/kg) at bedtime was introduced. Currently the patient is 9 years old and has been maintaining a fairly constant growth curve since the therapy was initiated. His baseline glucose and ketone monitoring, and function tests including prolonged fasting test and glucose challenge may be recommended to screen for this disorder.

4. Conclusion

Glycogenosis type 0 should be suspected in children, who usually present with fasting hypoglycemia and urinary ketons.

However, the disorder may also be asymptomatic, and since it is considered a very rare form of hepatic GSDs, its prevalence may be underestimated then. Moreover, any genetically determined disease requires a confirmed molecular diagnosis.

Compliance with ethics guidelines

The following authors: Edyta Szymańska, Urszula Wątrobinska, Dariusz Rokicki, Elżbieta Ciara, Paulina Halat, Rafał Płoski, and Anna Tylki-Szymańska declare that they have no conflict of interest.

This article does not contain any studies with human or animal subjects performed by the any of the authors — it is not a study/trial but a case report.

Details of the contributions of individual authors

Edyta Szymańska — planning, conduct, and reporting.
Urszula Wątrobinska — conduct.
Dariusz Rokicki — conduct, and reporting.
Elżbieta Ciara — conduct, and reporting.
Paulina Halat — conduct.
Rafał Płoski — conduct.
Anna Tylki-Szymańska — planning, conduct, and reporting.
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