Case Report

Yasemin Ardicoglu Akisin*, Zafer Arslan, Serdar Ceylaner and Nejat Akar

Glucose-6-phosphate dehydrogenase gene Ala365Thr mutation in an Iraqi family with confusing clinical differences

<https://doi.org/10.1515/tjb-2021-0088>

Received April 15, 2021; accepted November 15, 2021;

published online December 20, 2021

Keywords: favism; female; glucose-6-phosphate dehydrogenase deficiency; hemolytic anemia; Iraqi population.

Abstract

Objectives: Glucose-6-phosphate dehydrogenase (G6PD) has role in the Embden Meyerhof road. Any loss of its function causes NADPH to cease, leaving erythrocytes susceptible to oxidative damage resulting in acute hemolytic anemia attacks secondary to drugs or infection and favism. Because of X-linked recessive inheritance males are mainly affected. Being heterozygous, females have less severe clinical presentation.

Case presentation: G6PD deficiency was suspected in a six-year-old girl from an Iraqi family with a history of yellowing of skin and darkening of urine after eating broad beans. Besides the patient, G6PD levels were found low in the father and in two sisters who showed no symptoms. The father was found hemizygous and the three sisters were found heterozygous for NM_000402.4c.1093G>A (p.A365T)(6.Ala365Thr) mutation while the mother was normal.

Conclusions: G6PD enzyme deficiency can be seen in both genders, and it may be presented with different clinical manifestations even within the people having the same mutation.

Introduction

Glucose-6-phosphate dehydrogenase (G6PD) enzyme is an important enzyme that has role in the first step of the Embden Meyerhof road. Any loss of function in this enzyme causes NADPH to cease, leaving erythrocytes susceptible to oxidative damage. G6PD deficiency is seen as acute hemolytic anemia attacks secondary to drugs or infection, favism, increased risk of neonatal jaundice and chronic non-spherocytic hemolytic anemia [1].

It is the most common erythrocyte enzyme deficiency and has X-linked recessive inheritance. The incidence is high in Mediterranean countries, Africa and China, but has been identified in all ethnic groups. Although, males are mainly affected, females are heterozygous and have less severe clinical presentation. The frequency of G6PD deficiency is 0.5% across Turkey and 8.2% in Cukurova region. Over 400 variants and approximately 170 mutations of the enzyme have been identified [2, 3].

Erythrocytes, which are unable to synthesize the enzyme with reduced activity, undergo hemolysis due to toxic oxygen radicals formed as a result of their metabolism. This phenomenon may cause mild and persistent, or sudden and severe hemolytic crisis due to some triggering factors (some drugs, broad beans, some chemicals). Although favism caused by G6PD deficiency is always seen in males, neonatal jaundice may also occur in females [4].

In heterozygote female patients, according to Lyon hypothesis, there are two separate erythrocyte groups. One of these groups has low enzyme levels (10–60% of normal enzymatic activity) and the other has normal levels. Rarely, heterozygous women may be symptomatic due to excessive lyonization. When all genetic combinations are examined, the male-female ratio of deficiencies is 2:1 [5]. Here, we present a family from Iraq, to emphasize the

***Corresponding author: Yasemin Ardicoglu Akisin**, Assistant Professor, Department of Biochemistry, TOBB Economy and Technology University Faculty of Medicine, Ankara, Turkey, E-mail: yardicoglu@gmail.com. <https://orcid.org/0000-0002-4109-0220>

Zafer Arslan and Nejat Akar, Department of Pediatrics, TOBB Economy and Technology University Faculty of Medicine, Ankara, Turkey. <https://orcid.org/0000-0002-2610-4217> (Z. Arslan). <https://orcid.org/0000-0001-8228-8885> (N. Akar)

Serdar Ceylaner, Intergen Genetic Centre, Ankara, Turkey. <https://orcid.org/0000-0003-2786-1911>

clinical importance of G6PD deficiency causing confusing different clinical findings.

Case presentation

A family with three daughters (aged 2, 5 and 6 years) from Iraq was presented. G6PD deficiency was suspected when a six-year-old girl admitted to TOBB Economy and Technology University Faculty of Medicine, Department of Pediatrics with a history of yellowing of skin and darkening of the urine after eating broad beans.

When detailed information was taken, the presented patient had the history of yellowing of skin and darkening of the urine encountered 1–2 times in the last 4 years after eating broad beans. She was given iron therapy during this period in Iraq. Besides, she has dermatitis and allergic asthma triggered by physical activity. It was found that there was no similar history or sensitivity to broad beans in the other members of the family and also stated that the children did not encounter any conditions neither in the newborn period nor after. The physical examination of the patient was normal.

The complete blood count (CBC) was analysed using Sysmex XN-1000 (Sysmex Co., Japan) and G6PD levels were measured by using a commercial kit (Randox Laboratories Ltd., United Kingdom) in Cobas 6000 (Roche Diagnostics Co., Mannheim, Germany).

The same tests were analyzed in the mother, father and two sisters when patient's G6PD level was found low (5.48 U/g Hb; normal 7.0–20.5 U/g Hb). On the day of admission, the erythrocyte count, hemoglobin level, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) were found normal in all family members due age and gender. Besides the patient, G6PD level was found low in the father and in other two sisters (0.52 U/g Hb; 5.48 U/g Hb; 4.57 U/g Hb; 5.53 U/g Hb, respectively) (Table 1). No pathological results were seen in other biochemical and hematological tests.

Table 1: Laboratory findings of family members.

Tests	Mother	Father	Sister 1 (6 year old)	Sister 2 (5 year old)	Sister 3 (2 year old)
Erythrocyte ($\times 10^6/\mu\text{L}$)	4.5 (4–5.2)	4.36 ^a (4.5–5.9)	4.6 (4.1–5.2)	4.53 (4.1–5.2)	4.44 (3.9–5.3)
Hemoglobin, g/dL	12.1 (12–16)	13.5 (13.5–17.5)	12.3 (11.5–14.5)	12.0 (11.5–14.5)	11.5 (10.5–13.5)
Hematocrit, %	37.1 (36–46)	40.5 (40–53)	37.2 (25–45)	37.8 (25–45)	35.8 (33–39)
MCV, fL	82.4 (80–100)	92.9 (81–101)	80.9 (77–87)	83.4 (77–87)	80.6 (77–87)
MCH, pg	26.9 ^a (27–31)	31 (27–31)	26.7 (25–29)	26.5 (25–29)	25.9 (25–29)
MCHC, g/dL	32.6 (32–36)	33.3 (32–36)	33.1 (32–36)	31.7 (32–36)	32.1 (32–36)
G6PD, U/g Hb	13.2 (7.0–20.5)	0.52 ^a (7.0–20.5)	5.48 ^a (7.0–20.5)	4.57 ^a (7.0–20.5)	5.53 ^a (7.0–20.5)

Normal reference ranges due to age and gender were given in parentheses. ^aLow values due to reference range.

The mutation analysis of the whole family was performed. Written informed consent for genetic analysis was obtained both from the parents. The father was found hemizygous and three sisters were found heterozygous for the NM_000402.4 c.1093G> A (p.A365T) (6.Ala365Thr) mutation while the mother's result was normal.

Discussion

Being the most common intra-erythrocyte enzyme deficiency, G6PD deficiency affects approximately 3% of the world population. The disease is more common in the Mediterranean region and the African American and the Chinese population. The incidence was 0.5% in Turkey and has risen to a rate of 8.2% to Cukurova region [6].

The G6PD gene is located at the q28 locus in the subtelomeric region of the X chromosome. More than 400 variants have been identified, mostly due to point mutations. Thus, different variants and clinical phenotypes emerged [5, 6]. The World Health Organization (WHO) categorized G6PD enzyme deficiency into five classes according to the degree of enzyme activity and clinical findings [3] (Table 2).

G6PD B is the most common variant and seen in Caucasians, Asians and Africans (class-IV) and G6PD A is the normal variant seen in 10–20% of Africans (class-IV). G6PD A- is the most common variant of G6PD deficiency and

Table 2: WHO G6PD deficiency classification.

Class I	Enzyme activity is 10% below normal. Chronic hemolytic anemia is seen.
Class II	Severe enzyme deficiency. Often intermittent (secondary to drugs, infection, and chemicals) hemolytic anemia is detected.
Class III	Moderate (10–60%) enzyme deficiency. Intermittent hemolytic anemia.
Class IV	No enzyme deficiency and hemolysis.
Class V	Enzyme activity is high.

causes moderate to severe hemolysis. G6PD Mediterranean is the most common variant in Caucasians. It causes severe hemolysis and is classically associated with favism. It is the most common form seen in Turkey [3].

Males can have normal gene expression or be G6PD deficient because they are hemizygous for the G6PD gene. Having two copies of the G6PD gene on each X chromosome, females can have normal gene expression, be heterozygous or uncommonly be homozygous for the mutations. The clinical manifestations in heterozygous females are less severe than in G6PD deficient males. The degree of inactivation of one of the X chromosomes (lyonization) causes a variability in enzyme activity (normal to complete deficiency) [5, 6].

In Iraq, the prevalence of hemizygous G6PD deficiency in male population is 6.1%. The prevalence in female population can be estimated by using the Hardy–Weinberg equilibrium and it is found that the prevalence of heterozygous G6PD deficiency in Iraqi females would be 11.5% vs. 0.37% for homozygous females [7]. In Baghdad, Iraq, the Mediterranean (74.3%), Chatham (5%), and A- (2%) variants are found in males [8]. The prevalence differs in different parts of Iraq due to genetic heterogeneity (Baghdad 6.1–12.4%, Basra city 15.3%, Sulymania 6%). Higher prevalence can be seen in nearby countries like Saudi Arabia, Oman, UAE, and Kuwait, while it found lower in Ethiopia, Iran and Egypt [9].

Malbora et al. reported a similar case about a 26 month old girl and her father diagnosed with G6PD deficiency. The girl was presented with hemolytic anemia after eating broad beans, however, his father has no history of hemolytic anemia despite multiple exposure to broad beans [10].

In our case, carrying the same mutation, the father shows severe enzyme deficiency (0.52 U/g Hb-Class II according to the WHO classification) while the three sisters have moderate (5.48 U/g Hb; 4.57 U/g Hb; 5.53 U/g Hb-Class III according to the WHO classification). However, the clinical manifestations occurred only in the presented patient while the other family members showed no symptoms.

For G6PD deficiency, females are expected to be carriers without any clinical symptoms if the father has the deficiency and the mother is normal. On the other hand, enzyme deficiency in all three girls and only one of them having favism symptoms is unexpected. It is thought that this situation is due to the difference in the average enzyme

activity depending on the lyonization. It is noteworthy that despite the lack of enzyme in the father, there is no sensitivity of broad beans.

Conclusions

This case is presented to emphasize that G6PD enzyme deficiency can be seen in both genders, and that enzyme deficiency may be presented with different clinical manifestations even within the people having the same type of mutation.

Competing interests: Authors state no conflict of interest.

Ethical approval: The local Institutional Review Board deemed the study exempt from review.

References

1. Tetik E, Aral YZ, Kaynak Türkmen M, Bozkurt G. Glukoz-6-Fosfat Dehidrogenaz Enzim Eksikliği Olan Çocuklarda G6PD S218F Akdeniz Mutasyonu Sıklığı. *ADÜ Tıp Fakültesi Dergisi* 2014;15: 1–8.
2. Human Gene Mutation Database (HGMD). 2020. [HGMD Professional 2020.4 Release]. Available from: <http://www.hgmd.cf.ac.uk/ac/index.php>.
3. Glukoz 6 Fosfat Dehidrogenaz Enzim Eksikliği Tanı ve Tedavi Kılavuzu. *Türk Hematoloji Derneği, Ulusal Tedavi Kılavuzu*; 2011.
4. Günay İ. Yenidoğan sarılığına G6PD eksikliğinin etkisi. *İzmir Dr. Behçet Uz Çocuk Hast Dergisi*. 2011;1:115–20.
5. Luzatto L, Paggi V. Glucose 6 phosphate dehydrogenase deficiency. *Hematol Oncol Clin North Am* 2016;30:373–93.
6. Altay Ç, Gümrük F. Red cell glucose-6-phosphate dehydrogenase deficiency in Turkey. *Turk J Hematol* 2008;25:1–7.
7. van den Broek L, Heylen E, van den Akker M. Glucose-6-phosphate dehydrogenase deficiency: not exclusively in males. *Clin Case Rep* 2016;4:1135–7.
8. Al-Musawi BM, Al-Allawi N, Abdul-Majeed BA, Eissa AA, Jubrael JM, Hamamy H, et al. Molecular characterization of glucose-6-phosphate dehydrogenase deficient variants in Baghdad city – Iraq. *BMC Blood Disord* 2012;12:4.
9. Alezzi JI, Sharef AJ, Hassan EW, Latif RA. Screening for G6PD enzyme deficiency among children aged five years and below in Diyala Province/Iraq. *Pediatr Pract Res* 2019;7:9–14.
10. Malbora B, Koca SB, Tanyildiz HG. Glucose-6-phosphate dehydrogenase deficiency cases with different clinical findings: daughter with hemolytic anemia and asymptomatic father. *Turkish J Pediatr Dis* 2015;3:211–3.