*Original Article*

# Survey on causes of hemolysis in Glucose-6-Phosphate Dehydrogenase (G6PD) deficient pediatric patients

*Fariba Eghbal[1], Mohammad Reza Fakoorziba[2], Mohammad Hossein Eghbal[3], Soheila Latifnia[4]*

**ABSTRACT**

***Objective***: Glucose-6-phosphate dehydrogenase (G6PD) deficiency is the most common enzymatic disorder of red blood cells and more than 400 million people are affected in the world.

Our objective was to evaluate the causes of hemolysis in G6PD deficient hospitalized patients.

***Methodology:*** This retrospective study was done on 196 G6PD deficient patients during 2007-2010. They were between 1-13 years of age who were admitted to Shahid Dastgheib hospital of Shiraz in Iran for management of hemolysis crisis. Patients with hemolysis but without any documents of G6PD deficiency were excluded.

***Results:*** The prevalence of G6PD deficiency was higher in boys. Seasonal variations also affect hemolysis in these patients, peak incidence was observed in spring fallowed by autumn, winter and summer respectively. Ingestion of fava beans was the major cause where as infections, drugs, exposure to henna, pollen of fava beans and diabetic ketoacidosis were the other causes of hemolysis.

***Conclusion:*** The major cause of hemolysis is ingestion of fava beans fallowed by infections and oxidative drugs consumption in this region. So, awareness of G6PD deficient subjects about these factors and an early treatment of infection is very important for prevention of hemolysis in these patients.

**KEY WORDS***:* G6PD, Hemolysis, Fava beans, Blood groups, Season.

**Pak J Med Sci July - September 2012 Vol. 28 No. 4 666-669**

***How to cite this article:***

Eghbal F, Fakoorziba MR, Eghbal MH, Latifnia S. Survey on causes of hemolysis in Glucose-6-Phosphate Dehydrogenase (G6PD) deficient pediatric patients. Pak J Med Sci 2012;28(4):666-669

1. Fariba Eghbal, GP, MD,
Pediatric Specialist, Assistant Professor,
2. Mohammad Reza Fakoorziba, Msc, PhD,
Associate Professor, Department of Medical Entomology,
College of Health & Nutrition, Research Center for Health Science,
3. Mohammad Hossein Eghbal, GP, MD,
Anesthesiologist, Assistant Professor,
Shiraz Anesthesiology & Critical Care Research Center, Medical College,
4. Soheila Latifnia, MD,
1, 4: Department of Pediatrics, Medical College, Kazerun Branch,
Islamic Azad University, Kazerun, Fars province, Iran.
2, 3: Shiraz University of Medical Sciences, Shiraz, Iran.

Correspondence:

Dr. Mohammad Reza Fakoorziba, Msc, PhD,
E-mail: mrfakoor@yahoo.com
fakoorziba@sums.ac.ir

* Received for Publication: February 12, 2012
* 1st Revision Received: February 22, 2012
* 2nd Revision Received: March 12, 2012
* 3rd Revision Received: May 21, 2012
* Final Revision Accepted: May 23, 2012

## INTRODUCTION

G6PD is an enzyme that functions throughout the body, but its deficiency is seen predominantly in its effects on the red blood cells. In affected individuals, red blood cells are prone to premature break down leading to hemolytic anemia. It is the most common human enzymopathy which affects an estimated 400 million people worldwide.[1,2] Glucse-6-Phosphate dehydrogenase (G6PD) deficiency is an X-linked recessive inherited disorder, passes from one or both parents and most often occurs in males. It may be found in both males and females but male hemizygote and female homozygote are affected more severely than female heterozygote.[3]

Patients with G6PD are at risk of hemolytic anemia in states of oxidative stress. This can be in

severe infection, medication and certain foods with high levels of oxidants. G6PD protects red blood cells form potentially harmful bio-products that can accumulate following usage of certain medications, such as Aspirin, Aantimalarias and Sulfanamids, ingestion of fava beans or inhalation of pollen of fava bean plant.[4,5]

Infections are the most common cause of hemolysis in G6PD deficient subjects in areas where favism is less prevalent. Oxidative metabolites produced by numerous bacterial, viral and rickettsial infections have been identified. Important infections that can precipitate a hemolytic episode include viral hepatitis, pneumonia and typhoid fever.[6] In addition to these agents, topical application of henna can also induce hemolysis in G6PD deficient patients.[7]

G6PD deficiency tend to affect specific ethnic groups, the type seen in Mediterranean and Middle East area is often the most severe as compared to the types found in other parts of the world.[8]

Most patients with glucose-6-phosphatase dehydrogenase (G6PD) deficiency are asymptomatic. In addition to hemolysis, some patients present with neonatal jaundice that often require exchange transfusion. Other manifestations of G6PD deficiency include abdominal and/or back pain, gallstones, splenomegaly, dizziness, headache, dyspnea and palpitation.[9]

## METHODOLOGY

This retrospective study was conducted on 196 patients (aged between 1-13 years old) admitted in Shahid Dastgheib hospital of Shiraz in southwest of Iran for management of hemolysis crisis during 2007-2010. Patients with signs and symptoms of hemolysis who were known cases of G6PD deficiency or their problems were diagnosed with G6PD test were enrolled in this study. Data were collected from records of patients who were admitted with the hemolytic through a review of medical records. Data such as age, sex, vital signs, season of admission, history of exposure to oxidative agent, blood group, laboratory results (CBC, urinalysis, urine culture, chest X-ray, Bun, electrolyte, sugar, LFT,…) and hospital courses were collected. In all patients at time of arrival, blood sampling had been taken by venupancture and qualitative fluorescent spot test used as a method of detecting G6PD deficiency. Subjects with hemolysis but without any documents of G6PD deficiency were excluded. Collected data were analyzed by using SPSS software ver 2.1 and statistical $X^2$ test. The non-parametric statistical test were applied for comparison of blood groups.

## RESULTS

Results showed that males were affected more than females, Among 196 patients, 125 persons (63.8%) were boys and 71 persons were girls (36.2%) with male to female ratio equal to 1.76/1.0 with regard to period of year, hemolytic crisis due to G6PD deficiency occurred largely in spring (173 cases equal to 88.3%) followed by autumn (10 patients equal to 5.1%), winter (7 cases equal to 3.6%) and finally in summer (6 cases equal to 3.1%) p-value=0.001. Peak prevalence of hemolysis was seen during April and March (Table-I).

In respect to causes of hemolysis in G6PD deficient patients, ingestion of fresh fava beans was the major cause of hemolytic crisis in158 patients (80.6%). Inhalation of pollen of fava beans plant also induced hemolysis in 5 patients (2.6%). Nine patients (4.6%) had history of concomitant ingestion of fava beans and also one obvious infection. In 3 patients hemolysis was recorded who ingested fresh fava beans associated with usage of aspirin (A.S.A) and in one patient, it was combination of usage of fava beans and Nalidixic Acid. Various infections were recorded in 9 patients with acute hemolysis and one patient had history of topical application of henna plant. In one patient with acute hemolytic crisis, diabetic ketoacidosis was diagnosed and indeed he was a new case of IDDM, diagnosed following an attack of hemolysis due to G6PD deficiency. In spite of extensive evaluation no causative factor could be detected for inducing hemolysis in 9 patients and was considered as unknown (Table-II). Results demonstrated that although O positive blood group was observed higher in G6PD deficient

Table-I: Monthly and seasonal distribution of hemolysis crisis in G6PD deficient Patients.

| *Season* | *Months* | *Number* | *%* |
|---|---|---|---|
| Spring | April | 80 | 40.8 |
| | May | 78 | 39.8 |
| | June | 15 | 7.8 |
| Total | | 173 | 88.4 |
| Summer | July | 2 | 1.0 |
| | August | 3 | 1.5 |
| | September | 1 | 0.5 |
| Total | | 6 | 3.0 |
| Autumn | October | 6 | 3.1 |
| | November | 2 | 1.0 |
| | December | 2 | 1.0 |
| Total | | 10 | 5.1 |
| Winter | January | 3 | 1.5 |
| | February | 2 | 1.0 |
| | March | 2 | 1.0 |
| Total | | 7 | 3.5 |

Table-II: Distribution of causes of hemolysis in G6PD deficient patients.

| *Hemolysis factors* | *Number* | *%* |
|---|---|---|
| Fava beans | 158 | 80.6 |
| Infection | 9 | 4.6 |
| Fava beans + infection | 9 | 4.6 |
| Fava beans pollen | 5 | 2.6 |
| Fava beans + Aspirin | 3 | 1.5 |
| Diabetic Ketoacidosis | 1 | 0.5 |
| Fava beans +Nalidixic acid | 1 | 0.5 |
| Application of Henna plant | 1 | 0.5 |
| Unknown | 9 | 4.6 |
| Total | 196 | 100 |

patients compared to normal population but it was not significant statistically (Table-III).

Table-III: Comparison of distribution of prevalence of blood group with G6PD deficiency.

| *Blood group* | *Prevalence in shiraz* | *Prevalence in Iran* | *z* |
|---|---|---|---|
| O+ | 36.18 | 42.3 | 1.78 |
| A+ | 25.97 | 24.0 | -1.62 |
| B+ | 24.2 | 20.4 | 1.24 |
| O- | 5.75 | 6.6 | 1.1 |
| AB+ | 3.14 | 3.1 | -0.25 |
| B- | 2.21 | 2 | -0.32 |
| A- | 2.04 | 1.5 | -0.29 |
| AB- | 0.55 | 0 | -0.99 |

## DISCUSSION

Glucose-6-phosphate dehydrogenase deficiency is an important disorder of hexos monophosphate pathway and is responsible for different clinical syndromes including neonatal jaundice, episodic hemolytic anemia induced by; infections, certain drugs and fava beans and spontaneous chronic nonspherocytic hemolytic anemia. It is common in areas where malaria has been endemic such as Mediterranean, Caribbean, Africa and south East Asia. The protective effect of certain red cell polymorphisms, such as hemoglobin S and G6PD deficiency against severe *Plasmodium falciparum* malaria has been suggested because epidemiological evidence links the distribution of these polymorphisms to areas currently or historically highly endemic for malaria.[10] The prevalence varies in different parts of the worlds. According to World Health Organization (WHO), prevalence of G6PD deficiency is 10-14% in Iran.

According to results of this investigation, hemolysis due to G6PD deficiency was more prevalent in spring. Fava beans was also reported as the major cause of hemolysis. Other causes of hemolysis were usage of oxidative drugs, infections, pollen of fava beans and diabetic ketoacidosis. There was not any correlation between blood groups and G6PD deficiency in these studied patients.

In study by Nabavizade et al,[11] the prevalence of G6PD deficiency in blood transfusion recipients in Yasuj in south of Iran was reported equal to 14.7% whereas Emamghorashi showed 13.6% results in a city of south of Iran.[12] As G6PD deficiency is an X-linked heredity disease, majority of our patients were boys but involvement of girls in this research was more than it was expected (36.2%), which is comparable with Mendalawi et al, who reported 37.8% of G6PD deficient patients in Iraq were females.[13] On the other hand, Eghbalian reported that the male to female ratio of G6PD deficiency in newborn was 5/1 in Iran.[14]

Chien et al, also reported change in the male to female ratio in screening population in Taiwan during 1996 – 2005.[15] In our study, male to female ratio among all the patients with acute hemolysis due to G6PD deficiency was 1.76 /1.00 (boys: 63.8%, girls: 36.2%). The preponderance of females noted in some studies in the Eastern Mediterranean region may be due to high rates of consanguinity, leading to increased number of homozygous females in addition to high frequency of inactivation of the normal X chromosome in female heterozygotes, which leads to disturbances in the Hardy-Weinberg equilibrium. Meloni et al, revealed that fresh fava bean is the major cause of hemolytic anemia in G6PD deficient patients like our study.[16] Pollen of the fava beans also induced acute hemolysis in 5 patients (2.6%) that is in agreement with Schilliro[17] whereas Katomis et al, never observed cases of favism that were induced by pollen inhalation.[18] Infection is also a major cause of hemolysis in G6PD deficient patients. In this research also various infections were able to induce hemolysis in patients. The mechanism of linking infection and hemolysis is complex and poorly understood. However it has been suggested that during phagocytosis, leucocytes damage erythrocytes in their vicinity by discharging reactive oxygen species. Moreover, certain infectious organisms, such as Plasmodium, Clostridium, Babesia and Bartonella spp are directly toxic to red blood cells.[19] In addition, infectious diseases appear to be a common precipitating factor.[20]

Drugs also triggered hemolysis in some of these patients. Medications that cause hemolysis in G6PD deficient patients inflict oxidative damage to erythrocyte leading to their destruction.[21] According to Youngster et al,[22] drugs can be divided into three groups: medications that should be avoided in individuals with G6PD deficiency, medications that were considered unsafe by at least one source, but

can probably be given safely in normal therapeutic dosages to individuals with G6PD deficiency, and medications where no evidence at all was found to contravene their use in G6PD-deficient patients. In this research drug-induced hemolysis was associated with other oxidative agents such as infection and usage of fava beans. So, combination of two oxidative agents caused severe hemolysis.

In our study, there was one case of hemolysis due to topical application of henna that is in line with Raupp et al, that reported henna induced hemolysis in four patients in UAE.[7] Because in some part of Asia, application of henna is a tradition, G6PD deficient patients should avoid application of this material.

In this research we also reported one case of hemolytic crisis in G6PD deficient patient who had diabetes ketoacidosis as a trigger that is in line with Messina et al who also reported hemolytic crisis in non-ketotic and euglycemic child with G6PD deficiency and onset of type 1 diabetes.[23] Eugene et al, declared that ketotic prone diabetes (KPD) display a propensity to hyperglycemic induced acute insulin stress in B cells.[24]

Regarding blood groups, results revealed that ABO blood group of our patients were comparable with normal distribution of blood groups in normal habitants in Shiraz. Although crisis was more obvious in the patients having positive blood groups but statistically this difference was not significant. Hedayat et al, also reported a higher incidence of fauvism in O positive blood group patients[25] whereas Meloni et al, disagreed any association between G6PD deficiency and blood group.[16]

As mentioned in results, the highest peak of fava beans induced hemolytic crisis was observed in spring and mostly in April and May that is in line with other studies.[16,17] It is probably due to harvest time and increased consumption of fava beans during these months in Iran.

We conclude that the most important precipitating factors for hemolysis in these patients were ingestion of fava beans, infections and oxidative drugs consumption. G6PD-deficient patients or their parents/care-givers must be counseled about avoiding trigger factors, early treatment of infections and vaccination of preventable diseases, in order to minimize hemolytic crisis and to improve the quality of life. It also warrants greater attention for the medical community as well.

## ACKNOWLEDGEMENT

Authors wish to thank all of health workers of Shahid Dastghib Hospital, Shiraz University of Medical Sciences, for accomplishing the study. The authors also thank deputy of research of Islamic Azad University, Kazerun Branch, for supporting this research that led to thesis (No: 451) for MD degree.

## REFERENCES

1. Glader BE. Glucose-6-phosphate dehydrogenase deficiency and related disorders of hexose monophosphate shunt and glutathione metabolism: In: Greer JP, Arber DA, Rodgers GM, Paraskevas F, eds. Wintrobe's Clinical Hematology. 10th ed. Baltimore: Williams & Wilkins, 2008: 1176-1190.
2. Jain SK, Palmer M. Effect of glucose-6-phosphate dehydrogenase deficiency on reduced and oxidized glutathione and lipid peroxide levels in the blood of African-Americans. Clin Chim Acta 1996;253(1-2):181-183.
3. G6PD deficiency -2nd edition. www.gosh.nhs.uk.
4. Le Pommelet C. Le Moullec N. Zuni P. Diabetic ketoacidosis revealing glucose-6-phosphate dehydrogenase deficiency. Description of an adult case. Diabetes and Metab 2006;32(6):636-637.
5. Clinical aspects of G6PD deficiency. http:/rialto.com G6PD/clinical.htm
6. Wyngaarden JB, Smith LH Jr, Bennett JC, eds. Cecil textbook of medicine. 19th ed Philadelphia: W.B. Saunders: 1992: 1539.
7. Raupp P. Hassan AJ. Varughese M. Kristiansson B. Henna Causes life threatening hemolysis in glucose-6-phosphate dehydrogenase deficiency. Arch Dis Child 2001;85:411-412.
8. Kliegman RM, Behrman, RE. Jenson, HB. Stanton BF. Nelson textbook of pediatrics. 18th ed. Sanders: Elsevier; 2008.
9. Scriver CR, Beaudet AL, Sly WS, Valle D (eds) The metabolic and molecular bases of inherited disease. 7th ed. New York: McGraw- Hill, Inc 1995: 3367-3398.
10. Flint J, Harding RM, Boyce AJ, Clegg JB. The Population genetics of the haemoglobinopathies. Baillieres Clin Haematol 1993;6:215-262.
11. Nabavizadeh SH, Anushiravani A. The prevalence of G6PD deficiency in blood transfusion recipients. Hematology 2007;12(1):85-88.
12. Emamghorashi F, Hoshmand F, Mohtashamifar A. Screening for glucose-6-phophate dehydrogenase deficiency in blood donors. Hematology 2010;15(2):122-124.
13. Al-Mendalawi MD. Epidemiological, clinical and laboratory profile of glucose-6-phosphate dehydrogenase deficiency in the middle and north of Iraq: a comparative study. East Mediterr Health J 2010;16(8):846-850.
14. Eghbalian F, Monsef AR. Evaluation of Glucose- 6-phosphate dehydrogenase deficiency without hemolysis in icteric newborn. Iranian J Pediatr 2007;17(1):36-40.
15. Chien YH, Lee NC, Wu ST, Liou JJ, Chen HC, Hwu WL. Changes in incidence and sex ratio of Glucose- 6-Phoshate dehydrogenase deficiency by population drift in Taiwan. Southeast Asian J. Trop Med Pub Health 2008;39(1):154-161.
16. Meloni T, Forteleoni G, Dore A, Cutillo S. Favism and hemolytic anemia in glucose-6-phosphate dehydrogenase-deficient subjects in North Sardinia. Acta Haematol 1983;70:83–90.
17. Schiliro G. Russo A. Curreri R, Marino S, Sciotto A, Russo G. Glucos-6-phosphate dehydrogenase deficiency in Sicily. Incidence, biochemical characteristics and clinical implication. Clin Genet 1979;15:183-188.
18. Kattamis CA, Kyriazoko M, Chaidas S. Favism: Clinical and biochemical data. J Med Gent 1969;6:34-41.
19. Berkowitz FE. Hemolysis and infection: categories and mechanisms of their interrelationship. Rev Infect Dis 1991;13:1151-1162.
20. Burka ER. Infectious disease: a cause of hemolytic anemia in glucose-6-phosphate dehydrogenase deficiency. Ann Intern Med 1969;70:222-225.
21. Manganelli G. Fico A, Martini G, Filosa S. Discussion on pharmacogenetic interaction in G6pd deficiency and methods to, identify potential hemolytic drugs. Cardiovas and Haemato DisoDrug Targ 2010;10:143-150.
22. Youngster I, Arcavi L, Schechmaster R. Medications and Glucose-6-phosphate dehydrogenase deficiency. Drug Safe 2010;33(9):713-726.
23. Messina Mf, Lombard F, Crisaflli G. Hemolytic crisis in a non-ketotic & euglycemic child with glucose-phosphate dehydrogenase deficiency & onset of type1 diabetes mellitus. Pediatr Endocrin Metab 2004;17(12):1671-1673.
24. Eugene S, Jean-François G, Jean-Philippe K. High prevalence of Glucose-6-phosphate dehydrogenase deficiency without gene mutation suggestion a Naval genetic mechanism predisposing to ketosis- prone diabetes. J Clinic Endocrinol and Metabolism 2005;90(8):4446-4451.
25. Hedayat S, Farbud DD, Montazemi K. The pattern of bean consumption laboratory findings in patients with favism, G6PD deficient, and a control group. J Trop Pediatric 1981;27:110-113.

### *Authors Contribution:*

F. Eghbal and M.R. Fakoorziba conceived, designed and editing of manuscript. M.H. Eghbal did editing and review of final approval of manuscript. S. Latifi did data collection and statistical analysis.