Original Article

CULTURAL PRACTICES AND THE USE OF ICTEROGENIC AGENTS IN GLUCOSE 6 PHOSPHATE DEHYDROGENASE DEFICIENT NEONATES: ANY EFFECT?

[1]Jatau, E.D, [1]Zakari A., [1]Damulak O. D, [2]Toma, B.O., [1]Egesie, O. J ,[1]Akor E. A .
[1]Department of Haematology and Blood Transfusion, Jos University Teaching hospital.
[2]Department of Paediatrics, Jos University Teaching Hospital.

ABSTRACT

Background: Cultural practices involving the use of certain agents known to cause haemolysis in Glucose-6-phosphate dehydrogenase (G6PD) deficient individuals are common during perinatal and neonatal periods. This study was targeted at identifying some of these agents and their role in the development of hyperbilirubinaemia in G6PD deficient neonates in our environment with a view at raising public awareness for an acceptable neonatal outcome.
Materials and methods: One hundred and fifty neonates admitted into the Special Care Baby Units (SCBUs) of the Jos University Teaching Hospital, Bingham University Teaching Hospital, and the Plateau State Specialist Hospital with neonatal jaundice were enrolled for this study. Information on age, sex, history of drugs, chemicals and herbs used during or after pregnancy were obtained using a questionnaire. Five millilitres of the blood sample was collected into anticoagulated and plain sample bottles for Full Blood Count (FBC), Reticulocyte Count, Serum Bilirubin (SB) and G6PD assay.
Results: Mean age at presentation was 3.28 ± 3.11 days. Mean haemoglobin concentration of the neonates was 15.90 ± 2.23 g/dL while mean reticulocyte count, total leukocyte and platelet count were 2.42 ± 0.71%, 7.10 ± 2.76 (x 10^9/L) and 228.45 ± 85.57 respectively. Sixty-one (40.7 %) of the studied neonates were G6PD deficient with mean G6PD activity of 3.79 ± 1.37 IU/gHb. Mean total serum bilirubin was 205.01 ± 96.57µmol/L. Icterogenic agent use was identified in 70 (46.7%) of the study subjects with naphthalene balls used in 19 (12.7%) study subjects.
Conclusion: Icterogenic agents are being used for neonatal care in our environment despite the consequences of hyperbilirubinaemia in those with G6PD deficiency.
Keywords: Glucose-6-phosphate dehydrogenase deficiency, Hyperbilirubinaemia, Icterogenic agents, Neonates

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INTRODUCTION

Neonatal jaundice is a term used to describe the yellowish discolouration of the skin, sclera, and mucous membranes due to the deposition of bile pigments.[1] It develops when the rate of bilirubin production exceeds the rate of its elimination primarily by conjugation.[1] It is a common clinical presentation of varying aetiology during the neonatal period affecting about 60-70% of otherwise healthy full-term neonates and 80% of preterm neonates. [2]Clinical jaundice is noticed when total serum bilirubin (TSB) exceed 85.5µmol/L (5mg/dL) but not all jaundiced neonates become endangered by hyperbilirubinaemia.[2] Bilirubin encephalopathy or permanent, irreversible brain damage is known to occur when free bilirubin crosses the blood-brain barrier of a full-term or preterm neonates and is deposited in the basal ganglia and other areas of the brain. Neonatal jaundice has been reported to be a significant cause of morbidity and mortality in paediatric age group and an important contributing cause of cerebral palsy in the country.[3-7]

Several factors are associated with neonatal jaundice among which is prematurity, sepsis, ABO and Rh incompatibility as well as Glucose-6-phosphate dehydrogenase (G6PD) deficiency an X-linked recessive disorder and the commonest inherited red blood cell enzymopathy Worldwide. G6PD deficiency has the highest prevalence in the tropics, subtropics and in the Mediterranean countries.[8] In the United States of America, 2.5% of males and 1.6% of females are deficient, with most having only moderate deficiency. [8] The prevalence rate of G6PD deficiency in West Africa varies depending on the location. [8-9] In Nigeria, the prevalence of G6PD deficiency ranges from 4-26% in the general population and up to 40-43% among G6PD deficient jaundiced neonates.[9-11]

Reports from many countries of the world including Nigeria have indicated G6PD deficiency to be a major risk factor for neonatal jaundice, which may occur after exposure to oxidant agents like infection, drugs or fava beans ingestion, and consequently haemolysis with excess bilirubin production.[12-15]

Correspondence to :
Dr. Jatau, E.D
Department of Haematology and Blood Transfusion,
Jos University Teaching Hospital.
Email: ezradjatau@gmail.com
Phone: +2348065403375

This study is aimed at identifying icterogenic agents commonly use in our environment with a view of raising awareness at avoiding them within the perinatal as well as the neonatal period for better neonatal outcome.

Materials and Methods

Study design and setting

A cross-sectional study involving neonates with jaundice admitted into the Special Care Baby Units of the Jos University Teaching Hospital, Bingham University Teaching Hospital, and Plateau State Specialist Hospital between March 2013 to February 2014. They were recruited using the non-probability convenience sampling technique. A standardized questionnaire was administered to get information on age, weight, and length of the neonate at birth estimated gestational age, at booking, and history of drugs/chemicals used in pregnancy or after, as well as other risk factors for developing neonatal jaundice.

Laboratory Procedures

Three millilitres of venous blood was taken into an EDTA sample bottle for full blood count (FBC) auto analysis using the 3-part Sysmex haematology auto-analyser (KX-21N 2007 model). Reticulocyte count was manually performed using freshly prepared methylene blue as described by Dacie and Lewis. [16] G6PD enzyme activity was determined using reagents manufactured by the Pointe Reagent Company (USA) with G6PD deficiency defined by enzyme activity < 6.0 IU/gHb. Two millilitres of the blood sample was also collected into a plain bottle for bilirubin assay by the Jendrassik and Grof method using the Roche/Hitachi 902 SN 1694-019-1996 auto analyzer.

Statistical Analysis

Epi Info Version 6 software was used for the data analysis. The results were reported in tables, proportions, and percentages while mean, median, mode and standard deviation (SD) were used to describe continuous variables. Students' t-test showed the significance between the means of two groups and p-value $\leq$ 0.05 was considered statistically significant. Spearman's rank correlation coefficient (r_s) was used to determine the strength of the relationship between the icterogenic agents and development of hyperbilirubinaemia.

Ethical Consideration

Ethical approval was obtained from the Health Research Ethics Committees of Jos University Teaching Hospital, Bingham University Teaching Hospital, and Plateau State Specialist Hospital while written informed consent was obtained from parents/caregivers of the participating neonates

Results

A total of 150 icteric neonates (92 males and 58 females) were studied. Mean age at presentation was 3.28 ± 3.11 days with 78 (84.4%) of the males and 55 (94.8%) of the females in the 0-5 days age group (Table 1).

Table 1: Age at presentation and sex distribution of study subjects

Age (days)	Sex		Total
	Male n (%)	Female n (%)	
0-5	78 (84.4)	55 (94.8)	133
6-10	11 (12.0)	2 (3.4)	13
11-15	1 (1.1)	0 (0.0)	1
16-20	2 (2.2)	0 (0.0)	2
21-25	0 (0.0)	1 (1.7)	1
26-28	0 (0.0)	0 (0.0)	0
Total	92 (100)	58 (100)	150

Parenthesis ≡ Percentage total

Age at detection of jaundice

The mean age at detection of jaundice amongst the icteric neonates recruited for this study was 2.86 ± 1.67, a median of 2.00 days within a range of 1-12 days (Table 2).

Icterogenic agents used

Icterogenic agent use was identified in 70 (46.7%) of the study subjects with naphthalene balls in 19 (12.7%), menthol powder 8 (5.3%), traditional herbs 6 (4.0%), Mentholatum 5 (3.3%), and Aboniki in 3 (2.0%). Combinations of two or more were used on 29 (19.3%) of these neonates while there were no identifiable icterogenic agents used on 80 (53.3%) of these subjects (Table 2).

Haematological parameters, total serum bilirubin and G6PD activity of all subjects

The haemoglobin concentration of the study subjects was in a range of 11.33-22.30 g/dL with a mean of 15.90 ± 2.23 g/dL, and a median of 15.69 g/dL while the mean haematocrit was in a range of 0.34-0.67 with a mean of 0.47 ± 0.06 and median of 0.45. The study subjects had reticulocyte count in the range of 0.50-3.90 %, mean count of 2.42 ± 0.71% and a median of 2.50 %.

The total leucocyte count was in the range of 2.00-16.00 (x 10^9/L) with a mean of 7.10 ± 2.76 and median of 6.75 x 10^9/L while the platelet count (x 10^9/L) was in the range of 80.00-446.00 with a mean of 228.45 ± 85.57 and median of 200.00.

The mean serum bilirubin of the 150 study subjects was found to be 205.01 ± 96.57µmol/L with a mode of 184.50µmol/L and a range of 86.70-606.00µmol/L.

Eighty-nine (59.3%) of the icteric neonates were G6PD normal with a mean G6PD activity level of 10.92±4.24IU/gHb while 61 (40.7%) were G6PD deficient with a mean G6PD activity level of 3.79±1.37 IU/gHb (Table 2).

Table 2: Relevant clinical data and Laboratory parameters of study subjects

Parameters	n; 150
Age jaundice was noticed (days; mean + SD)	2.86 ± 1.67
Icterogenic agent use; n (%)	70 (46.7)
Haemoglobin concentration (g/dL; mean ± SD)	15.90 ± 2.23
Haematocrit (mean ± SD)	0.47 ± 0.06
Retics (%; mean ± SD)	2.40 ± 0.73
Leucocyte count ($x10^9$/L; mean ± SD)	7.10 ± 2.76
Platelet ($x10^9$/L; mean ± SD)	228.45 ± 85.57
Serum bilirubin (µmol/L; mean ± SD)	205.01 ± 96.57
G6PD activity level (n) (IU/gHb; mean + SD)	
G6PD Normal	(89) 10.92±4.24
G6PD Deficient	(61) 3.79±1.37

G6PD status, exposure to Icterogenic agents and severity of hyperbilirubinaemia

Seventy (46.7%) of the mothers whose babies were enrolled in this study use certain agents for baby care and as medication during or after delivery. There was no identifiable risk factor in 80 (53.3%) of the neonates.

Table 3 shows the frequency of the different agents the neonates were exposed to in relation to G6PD activity level and degree of hyperbilirubinaemia. Seven (11.5%) of the G6PD deficient neonates were exposed to naphthalene balls and had mean total serum bilirubin of 168.1 ± 48.5 µmol/L while 1 (1.1%) of them had exposure to traditional herbs with mean total serum bilirubin of 518.6 ± 0.00 µmol/L. Twelve (13.5%) of the G6PD normal neonates exposed to naphthalene balls had a mean total serum bilirubin of 213.0 ± 78.1 µmol/L while 5 (5.6%) of the G6PD normal neonates were exposed to traditional herbs and had a mean total serum bilirubin of 239.1 ± 107.3 µmol/L. Thirty-seven (60.7%) of the G6PD deficient neonates in this study were not exposed to any icterogenic agent. The use of icterogenic agents in relation to G6PD status and degree of hyperbilirubinaemia showed no statistically significant difference with P-values > 0.05 while a Spearman's rank correlation coefficient (r_s) between the icterogenic agents and degree of hyperbilirubinaemia was 0.99 depicting a strong positive correlation.

Table 3: G6PD status, exposure to icterogenic agents and hyperbilirubinaemia

Icterogenic agent	G6PD Deficient		G6PD Normal				
Exposure	n (%)	SB (µmol/L)	n (%)	SB (µmol/L)	t	p value	Total
Abk	2 (3.3)	166.3 ± 88.5	1 (1.1)	188.6 ± 00.0	0.21	0.75	3
MP	3 (4.9)	137.4 ± 53.5	5 (5.6)	186.3 ± 36.9	1.55	0.17	8
Mt	3 (4.9)	346.6 ±193.2	2 (2.3)	149.5 ± 19.0	1.37	0.27	5
Np	7 (11.5)	168.1 ± 48.5	12 (13.5)	213.0 ± 78.1	1.37	0.18	19
TH	1 (1.6)	518.6 ± 00.0	5 (5.6)	239.1 ± 107.3	2.38	0.08	6
MP + Mt	-	-	1 (1.1)	120.8 ± 00.0	-	-	1
Np + MP	3 (4.9)	296.7 ± 152.6	6 (6.7)	172.8 ± 57.6	1.85	0.11	9
Np + Mt	2 (3.3)	227.1 ± 171.7	2 (2.3)	110.4 ± 33.5	0.94	0.45	4
Rb + Mt	-	-	1 (1.1)	200.2 ± 00.0	-	-	1
TH + MP	-	-	3 (3.4)	171.3 ± 97.9	-	-	3
TH + Np	-	-	1 (1.1)	326.2 ± 00.0	-	-	1
TH + MP + Abk	-	-	1 (1.1)	184.0 ± 00.0	-	-	1
Np + MP + Abk	-	-	2 (2.3)	152.0 ± 29.8	-	-	2
Np + MP + Mt	1 (1.6)	141.1 ± 00.0	2 (2.3)	171.5 ± 8.8	-	-	3
Np + MP + TH	1 (1.6)	154.7 ± 0.00	1 (1.1)	185.0 ± 0.00	-	-	2
Np + Mt + TH	1 (1.6)	203.0 ± 00.0	-	-	-	-	1
Np+MP+Mt+TH	-	-	1 (1.1)	606.0 ± 00.0	-	-	1
Not exposed	37 (60.7)	223.5 ± 103.0	43 (48.3)	185.1 ± 67.5	2.00	0.05	80
Total	61 (100)		89 (100)				150

Abk-Aboniki, MP-Menthol Powder, Mt-Mentholatum, Np-Naphthalene balls, TH-Traditional Herbs, Rb-Robb

Discussion

Frequency of exposure to icterogenic agents in the G6PD deficient neonates was not too different when compared to the G6PD normal neonates as obtained from history. The study showed that both the G6PD deficient and G6PD normal neonates had similar exposure to icterogenic agents with no remarkable difference in the severity of hyperbilirubinaemia, but a strong positive correlation was established between the use of icterogenic agent and severity of hyperbilirubinaemia (r_s 0.99). This report is in contrast to studies from other countries who had documented that hyperbilirubinaemia were significantly higher among G6PD deficient neonates exposed to certain icterogenic agents compared to G6PD normal neonates who had also been exposed. [17- 20] However, same studies have agreed with the fact that hyperbilirubinaemia often occurs with no apparent offending factor or known triggers of haemolysis giving such agents a doubtful role. [17-20] They further stated that G6PD deficiency could act as an independent icterogenic factor increasing the risk of developing significant hyperbilirubinaemia in the absence of other causes. [20] In contrast to the finding in this study, Owa*et al.* and Sodeinde*et al.* in studies conducted in Nigeria declared a direct association between exposure to icterogenic agents and severe hyperbilirubinaemia with its attendant consequences. [21, 22] Furthermore, other previous investigations have documented an association between exposure to certain icterogenic agents, haemolysis and G6PD deficiency, but not all of the G6PD deficient individuals exhibited haemolysis. [23] This finding may be attributed to the fact that G6PD variants play a significant role in the development of haemolysis and hyperbilirubinaemia even in the presence of an icterogenic agent. [23] Haemolysis is known to be common in persons with G6PD Class II variant and rarely does it occur in individuals with the G6PD A^+ variant even after exposure to chemicals like naphthalene and menthol or drugs like primaquine. [23] However, the limitation of this study and those with the contrasting views is that no genetic testing aimed at identifying the predominant G6PD variants was done. Genetic testing is therefore important as it will assist in characterizing the G6PD variants in each population group and help in determining the clinical course of the disorder with the view of instituting appropriate management strategy early.

Conclusion

The frequency of exposure to the icterogenic agent was generally high in the deficient and normal neonates exposing the level of ignorance about the dangers of such agents in this environment. It is also worthy of note that neonates exposed to traditional herbs alone or in addition to other icterogenic agents had higher serum bilirubin concentration. There is, therefore, the need for public health education on the dangerous effect of these icterogenic agents with women of childbearing age and pregnant mothers being the target population. A further study on G6PD variants and the effect of the different icterogenic agents' especially traditional herbs available in our environment is also advocated.

Conflict of interest

None declared

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