



Haemoglobin Electrophoresis Pattern among the Children Attending Paediatrics Department of Usmanu Danfodiyo University Teaching Hospital (UDUTH), Sokoto, North-Western Nigeria

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Authors' contributions

This work was carried out in collaboration between all authors. Authors KKI and NMJ designed the study, wrote the protocol and wrote the first draft of the manuscript. Authors AG and AU managed the literature searches and analyses of the study. Authors JMB and MLJ concerned with the logistics. Author MOM concerned with data interpretation. All authors read and approved the final manuscript.

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ABSTRACT

Aim: The aim of this study was to determine the Haemoglobin Electrophoresis pattern among the children attending Paediatrics Department of Usmanu Danfodiyo University Teaching Hospital (UDUTH), Sokoto, North-Western of Nigeria.

Materials and Methods: A total of three hundred and Ninety-five (395) children aged 0-15 years were recruited for this study. About 3 milliliter of blood was collected into the K₂EDTA container. The haemoglobin electrophoresis was carried-out at alkaline pH (8.6) using the method described by Helena Biosciences procedure.

Results: The present study recorded the prevalence of HbAA, HbAS, HbSS, HbAF, HbSS+F, HbAC and HbSC to be 70.0, 15.2, 5.0, 0.3, 6.0%, 2.0, and 1.5% respectively with a prevalence of 11% sickle cell anaemia among the study subjects.

Conclusions: HbAA is the predominant haemoglobin in our environment, there is also a significant number of other haemoglobin variants. Many children with other haemoglobin variants are surviving to adulthood due to advances in medicine, a larger number of pregnant women with sickle cell disease with all the attendant challenges, it should be expected in our environment. It is necessary therefore, to keep abreast with developments in the area of its management in order to cope with the challenges.

Keywords: Hemoglobin electrophorsis; prevalence; children; Sokoto; Nigeria.

1. INTRODUCTION

Haemoglobin genotypes are inherited blood characters. The inherited disorders of haemoglobin are the most common genetic disorder worldwide, with 7% of the world's population being carriers [1]. It is on record that about 300.000 children are born with sickle cell disease (SCD) worldwide every year [2]. The most common types of hemoglobinopathies are the sickle cell disorder and the thalassaemias, occurring in people of African, Asian, South European and Middle Eastern descent [1]. These sickling disorders include the heterozygous state for haemoglobin (HbAS), the homozygous for haemoglobins (HbSS) and the compound heterozygous state for haemoglobins together with haemoglobin C, D, E or other structural variants. Haemoglobins S differs from haemoglobin A by the substitution of valine for glutamic acid at position 6th in the β chain ($2\beta_2^{6\text{Glu}} \dots \text{Valine}$) [3]. Substitution of glutamic acid for lysine at position 6 of the globulin chain ($2\beta_2^{6\text{Glu}} \dots \text{Lys}$) also occurs in haemoglobin (HbC) genotype [4]. In haemoglobin D-punjab (HbA) glycine in being substituted for glutamic acid at position 121st in the β globulin chain ($2\beta_2^{121\text{Glu}} \dots \text{Gly}$) while in haemoglobin E (HbE) lysine is substituted for glutamic acid at the 26th position of the β globulin chain ($2\beta_2^{26\text{Glu}} \dots \text{Lys}$) [4]. Subjects heterozygous for HbC (HbAC) are asymptomatic and may present with a mild microcytosis with relative red cell resistance to haemolysis [5]. Subject homozygous for HbC (HbCC) have usually compensated haemolysis with splenomegaly. There is an increased risk of hypersplenism,

biliary lithiasis, folate deficiency and worsening of anaemia following some infections (parvovirus) [6]. The association of HbCC with β thalassaemia, especially with β^+ thalassaemia (more common than β^0 thalassaemia) in the ethnic groups concerned by (HbCC) results in a clinical picture similar to that of HbCC. $C\beta^0$ thalassaemia is more severe and can exceptionally mimic beta thalassaemia intermedia. Compound heterozygotes HbSC present with a sickling disorder similar to sickle cell anemia (SCA), although it is generally milder than in the HbSS form. However, 2% of HbSC patients have more severe disease with frequent Vaso-occlusive crisis (VOC) and acute chest syndrome (ACS) [7]. Priapism may occur, mainly in adults. Aseptic necrosis of femoral heads and, in a lesser extent, of shoulder, and proliferative retinopathy with risk of vitreous hemorrhages are common complication of the HbSC genotype. Splenomegaly persists after 5 years, often in adulthood, leading to recurrent spleen infarcts or sequestration hypersplenism. Hemoglobinopathies are among the most common genetic disorders worldwide, inherited as autosomal recessive disorders. The frequencies of these inherited characters have severally been reported to vary significantly in various populations and ethnic groups around the world. It is anticipated that the global economic burden of the hemoglobinopathies on public health will increase over the coming decades [8]. Children and pregnant women with hemoglobinopathies are at greater risk for anaemia. In Sub-Saharan Africa; foetal and maternal mortality rates are particularly high

among children and pregnant women with hemoglobinopathies [9]. In Nigeria, few recent published data have been encountered, but none in Sokoto, North-West of Nigeria. This study was therefore designed to provide the frequencies of haemoglobin variants for reference purposes using children attending the paediatrics department of UDUTH, Sokoto, Nigeria.

2. MATERIALS AND METHODS

2.1 Study Area

This study was carried out in Paediatrics Department of Usmanu Danfodiyo University Teaching (UDUTH) Sokoto, North-Western Nigeria. Sokoto State is located in the extreme North- West of Nigeria, near to the confluence of the Sokoto River and the Rima River. The State is located between longitude 11°30', 13°50' East and latitude 4°0' to 6°0' North. It is bordered to the North by Niger Republic, Zamfara State to the East while Kebbi State borders most of the South and Western parts [10]. The major indigenous tribes in the state are the Hausa and Fulani and other groups such as Gobirawa, Zabarmawa, Kabawa, Adarawa, Arawa, Nupes, Yorubas, Ibos and others. Occupation of city inhabitants include trading, commerce, with a reasonable proportion of the population working in private and public sectors. Majority of the Hausas' are farmers while Fulanis are nomadic and are engaged in animal rearing [11]. Based on 2006 population census, Sokoto State had a population of 3,696,999 [12], with an average estimate of 4,806,098 in 2015 based on the population annual growth rate of 3% [13].

2.2 Study Population

A total of 395 children aged 0.0-15.0 years whose parents offered a written informed consent for their wards to participate in the study were recruited for this cross-sectional study. The subjects were recruited from Paediatrics Department of Usmanu Danfodiyo University Teaching Hospital, Sokoto, North- Western Nigeria. The age of 15 years is considered as the limit for Paediatrics patients in UDUTH, Sokoto [14].

2.3 Research Design

This was a cross-sectional study designed to investigate the prevalence of Sickle Cell Anaemia and other variants of abnormal haemoglobin among 395 children attending

Paediatrics Outpatients Department of UDUTH, Sokoto, North Western Nigeria, between January and April, 2016. About 3 mL of blood was collected from the subjects in to the K₂EDTA containers. Haemoglobin electrophoresis was carried on the samples at alkaline pH 8.6 using the method as described in the Helena BioSciences procedure [4]. Leishman's stained blood films were also examined for red cell morphology; these would give additional information on Sickle Cells Anaemia.

2.3.1 Inclusion criteria

The inclusion criteria adapted for this study is; Children aged (0.5 -15 years), attendance to the paediatrics department of the study hospital and written informed consent obtained from their parents/guardians.

2.3.2 Exclusion criteria

Individuals who did not meet the inclusion criteria; individuals above 15 years and children whose parents/guardians refused to offer a written informed consent for their wards to participate in the study as subjects.

2.4 Statistical Analysis

The data collected were entered into the data editor of statistical package for social sciences (SPSS Version 22) software. Analysis was based on simple percentages, or proportions and values were expressed as Mean \pm SD. A Chi-square test at a 95% confidence level was also used to test for association between age groups, anaemia and gender. A p-value of < 0.05 was considered as significant in all statistical analysis.

3. RESULTS

Out of the three hundred and ninety-five children tested, 223(56.5%) were male while 172(43.5%) were female.

4. DISCUSSION

Three hundred and ninety-five (395) children were tested for haemoglobin Electrophoresis. This present study recorded the prevalence of HbAA, HbAS, HbSS, HbAF, HbSS+F, HbAC and HbSC to be 70.0%, 15.2%, 5.0%, 0.3%, 6.0%, 2.0%, and 1.5% respectively. These findings are consistent with the previous reports, in which the prevalence of 70%, 23.25%, 4.75%, 1.25%, 0.75% were observed for HbAA, HbAS, HbSS,

HbAC, and HbSC respectively [15]. Similarly, a prevalence of 69.35%, 26.94%, 3.54%, 0.12%, and 0.02% of HbAA, HbAS, HbSS, HbAC, and HbSC respectively observed in Anambra [16]. Also in Ogbomoso a prevalence of 71.03%, 22.11%, 0.54%, 5.26%, and 0.805 was observed for HbAA, HbAS, HbSS, HbAC, and HbSC respectively [17]. The observed prevalence of HbAA is within the normal range of 55 – 75% earlier reported for Blacks [18].

The results of this finding revealed a prevalence of 5.0% HbSS + 6.0% HbSS+F (11.0%) among the study subjects. This finding is also consistent with other published reports in Nigeria; 3.0% in the South-West region of Nigeria [19], 2% among undergraduate students in Bayelsa State [20] and 3% in Rivers State [21] both in the South-South of Nigeria. Our finding is however at

Table 1. Age and gender distribution of the study population

Age (years)	Gender		Total
	Male	Female	
0.5 – 5.0	125(31.7%)	100(25.3%)	225(57.0%)
5.1 – 10.0	62(15.7%)	35(8.9%)	97(24.6%)
10.1 - 15.0	36(9.1%)	37(9.3%)	75(18.4%)
Total	223(56.5%)	172(43.5%)	395(100.0%)

$\chi^2 = 4.47; df = 2; p\text{-value} = 0.107; df = \text{degree of freedom}$

Table 2. Prevalence of haemoglobinopathies among the study population

Electrophoretic pattern	Frequency	Percent
AA	276	70.0
AC	8	2.0
AF	1	0.3
AS	60	15.2
SC	6	1.5
SS	20	5.0
SS+F	24	6.0
Total	395	100.0

A = Normal Haemoglobin, S = S Haemoglobin (trait), C = C Haemoglobin (trait), F = Foetal Haemoglobin, AA = Healthy, AS = Carrier, SS = Sickle Cells, SC = Sickle Cells Disease

Table 3. Gender comparison of haemoglobinopathies among the study population

Haemoglobin	Gender		Total
	Male	Female	
AA	150(38.0%)	126(32.0%)	276(70.0%)
AS	40(10.1%)	20(5.1%)	60(15.2%)
AC	5(1.2%)	3(0.8%)	8(2.0%)
AF	1(0.2%)	0(0%)	1(0.2%)
SC	4(1%)	2(0.5%)	6(1.5%)
SS	12(3%)	8(2%)	20(5.0%)
SS+F	8(2%)	16(4.1%)	24(6.1%)
Total	220(55.5%)	175(44.5%)	395(100.0%)

$\chi^2 = 10.782; df = 8, p\text{-value} = 0.214$

Table 4. Age comparison of haemoglobinopathies among the study population

Age (years)	Haemoglobin electrophoretic pattern							Total
	AA	AS	AC	AF	SC	SS	SS+F	
0.5 - 5.0	164	28	7	1	2	7	14	223
5.1 – 10.0	63	20	0	0	1	8	5	97
10.1 – 15.0	49	12	1	0	3	5	5	75
Total	276	60	8	1	6	20	24	395

$\chi^2 = 20.01; df = 16, p\text{-value} = 0.220$

variance with previous report in Kenya, East Africa [18] and among 620 University students in Port Harcourt Nigeria [22] on which both reported a 0% prevalence of HbSS. The zero prevalence observed in these studies, possibly imply that the sickling gene pool is gradually reducing in some African populations due to increase awareness and pre-marital counselling. The low prevalence of HbSS observed in these studies also could be attributed to increased awareness of the disease, improved socio-economic conditions, improved pre-marital counselling, environmental and genetic factor which have an overall effect on the sickling gene pool. These zero prevalences may also be attributed to an active program of prenatal diagnosis among pregnant women in Nigeria. The finding from this study is consistent with prevalence of HbSS observed among the Black population in the United States which was reported to be 9% and 30%–40% generally for Africans [23-24]. Sick cell haemoglobin (HbS) is the most common and clinically significant haemoglobin structural variant.

This present study recorded zero percent of HbCC which is inconsistent with the previous findings: 0.24% in Akwa Ibom [25], 0.18% in Ogbomoso [20] and 0.01% in Anambra [19]. The zero frequencies observed in our studies, may be attributed to sample size and other environmental conditions. The number of people with homozygous HbSS for both male and female respectively in Sokoto, is high. The reason for this high prevalence may be due to the absence of carrier testing programs and premarital counselling/testing for prospective couples prior to marriage. Other factors such as persisting high concentration of foetal haemoglobin could invariably influence the prevalence of SCA in the population.

Age and gender comparison did not show any statistically significance difference ($p>0.05$). The reason for this might be linked to the facts that hemoglobinopathies are inherited characters determined by the different combinations of globin chains [6]. Evidenced-based data from Belgium, a country with universal neonatal screening programme has shown that neonatal screening is an excellent health education tool [26]. The Nigerian government can benefit by implementing a carrier testing programs and premarital counselling/testing for prospective couples prior to marriage in a bid to reduce the prevalence of haemoglobinopathies in the area. Sokoto State in particular and Nigeria in general

can benefit from universal neonatal screening program. It can be an effective way to diagnose and monitor the trend of haemoglobinopathies in the state. There is also a need for a sickle cell disease clinical care programs which should include: infection prophylaxis with penicillin and malarial prophylaxis; family training to identify early, severe, or persistent symptoms and increased awareness of the gravity of malarial crises; the evaluation of the patient's nutritional status and fluid intake; and education about the importance of regular medical visits.

5. CONCLUSION

This study indicated that HbAA is the predominant haemoglobin in our environment, while other variants are also found with little proportions. Many children with other haemoglobin variants are surviving to adulthood due to advances in medicine. This implies that, a larger number of pregnant women with sickle cell disease with the entire attendant challenges it poses should be expected in our environment. It is necessary therefore, to keep abreast with developments in the area of its management in order to cope with the challenges.

ETHICAL CONSIDERATION

This study was approved by Health Research and Ethics Committee of UDUTH, Sokoto, North-Western Nigeria.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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