

Original Research Article

Haemoglobin genotype, ABO and rhesus blood group pattern among students of Bamidele Olumilua University of Education, Science and Technology Ikere, Ekiti state, Nigeria

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ABSTRACT

Background: The two most significant blood group systems of clinical importance are ABO and rhesus. An essential blood component that determines haemoglobinopathies is haemoglobin genotype. The aim of the study was to determine the distribution of haemoglobin genotypes, ABO and rhesus blood groups pattern among students of Bamidele Olumilua University of Education, Science and Technology Ikere (BOUESTI), Ekiti state.

Methods: Two thousand (2,000) samples comprising 840 (42%) males and 1,160 (58%) females were recruited for this study. The ABO blood group was determined using tile method, while the haemoglobin genotype was determined using haemoglobin electrophoresis. Data analysis was done using simple percentage and Chi square test.

Results: The results obtained showed that out of the two thousand subjects that participated in this study, 1,448 (72.4%) of the subjects had HbAA, 452 (22.6%) had HbAS, 72 (3.6%) had HbAC, 20 (1%) had HbSS and 8 (0.4%) had HbSC. The distribution of ABO blood groups of the subjects were; blood group O (53.6%), blood group B (26.6%), blood group A (17.8%) and blood group AB (2.0%). Furthermore, 96.0% of the subjects were rhesus 'D' positive, while 4.0% were rhesus 'D' negative. There was no significant difference ($p > 0.05$) in the haemoglobin genotypes and ABO blood groups of the subjects with respect to age and sex.

Conclusions: The study concludes that the distribution of ABO blood group in the study population was given by O>B>A>AB, while the genotype was given by AA>AS>AC>SS>SC respectively. Continued haemoglobin genotype test and premarital counselling of potential couples is highly recommended.

Keywords: Distribution, Blood group, Rhesus, Haemoglobin genotype, BOUESTI

INTRODUCTION

The intracellular protein found in erythrocytes called haemoglobin is made up of four polypeptide globin chains that are folded around heme molecules. It is in charge of moving carbon dioxide from the tissues to the lungs and oxygen from the lungs to the tissues.

The globin chains are known to have many alleles and are encoded by the relevant genes on chromosomes 11 and 16.

Many of these genotypes experience single amino acid alterations in the globin moiety as a consequence of point mutations in the DNA sequence. This results in the synthesis of different types of haemoglobins.¹

An essential blood component that predicts haemoglobinopathies is haemoglobin (Hb) genotype.²

The pigment of red blood cells that carries oxygen is called haemoglobin; genetic flaws can result in defective

haemoglobin, which causes a disorder known as haemoglobinopathies.³

Normal haemoglobin (Hb AA) and other abnormal haemoglobins, such as the mutant haemoglobin S (Hb S), are both part of haemoglobin genotypes. Abnormal haemoglobin genotype (Hb S) differ from normal genotype (Hb A) in that the neutral amino acid valine is substituted for glutamic acid at position 6 in the β -chain, which changes the characteristics of haemoglobin and causes red blood cells to sickle.⁴ Another variant is haemoglobin C (Hb C) which results from the substitution of lysine a basic amino acid for glutamic acid at position 6 in the β -chain.⁵

When an individual inherits mutant haemoglobin genes from both parents, such as haemoglobin S, C, D, and E, the result is an abnormal haemoglobin genotype. Different combinations can result in abnormal haemoglobin genotypes, which are inherited in an autosomal dominant manner.⁶ Several abnormal haemoglobin genotypes have been found; however, only a few are widespread and seriously detrimental to public health in various regions of the world.⁷ The most commonly occurring abnormal haemoglobin genotypes among Nigerians are; AS, AC, SC, and SS.

Blood groups are categories of inherited antigenic chemicals found on the surface of red blood cells, other cell types, and bodily secretions. These antigenic substances are coded by alleles at several loci on a chromosome.⁸ A blood group system is made up of a number of these red blood cell surface antigens that come from a single allele (or a set of closely related alleles).

The International Society of Blood Transfusion recognizes 30 different human blood group systems.⁹ The first and fourth discovered blood group systems (ABO and Rhesus), are the most commonly researched genetic markers of clinical importance in humans. They are important for blood transfusion practice, but they are also helpful for population genetic studies and for settling medical-legal disputes like parentage disputes.¹⁰ The ABO blood grouping system is divided into four types namely; A, B, AB, and O.⁸

In 1939, the Rhesus system was discovered and within a few years, it was confirmed. Due of prenatal haemolytic illness and its significance for rhesus D negative persons in later transfusions once they develop Rhesus antibodies, the Rhesus system has become the second-most important blood group system.¹¹ People are rhesus-positive if they have the D antigen, a specific rhesus antigen, on the surface of their erythrocytes, and rhesus-negative if they do not. When the mother is rhesus-negative and the fetus is rhesus-positive, rhesus incompatibility can be a serious issue.¹²

Blood groups and haemoglobin genotype distribution have been extensively studied in a variety of populations around

the globe, and their frequencies showed significant regional variation, reflecting the underlying genetic and ethnic diversity of human populations.¹³

Nigeria is a heavily populated nation made up of several ethnic groups. As with many other genetic traits, haemoglobin genotype and the gene frequency of ABO and rhesus blood group varies significantly within the six geopolitical zones in Nigeria.¹⁴

However, despite the numerous studies carried out on the distribution of haemoglobin genotypes, ABO and rhesus blood groups in Nigeria, no study has been carried out among residents in Ikere, Ekiti State.

This study was therefore carried out to determine the distribution of haemoglobin genotypes, ABO and rhesus blood groups among students of Bamidele Olumilua University of Education, Science and Technology Ikere (BOUESTI), Ekiti State, Nigeria.

METHODS

Area of study

This study was carried out in BOUESTI. Ikere is the second most populous and principal city of Ekiti state, Nigeria. The area lies between latitudes 70 30' North of the equator and longitudes 50 14' East of the Greenwich meridian. The city has an area of 262 km², of which 52.2% of the populations are females, while 47.8% are males.

Compared to the entire Ekiti as a state and Nigeria as a country, Ikere is densely populated, with a population density of 778.3/km². Ikere-Ekiti is essentially an agrarian and mining community.

According to 1991 and 2006 census, the population of was 114,780 and 147,355 respectively. There are three major types of religion in Ikere; Christianity, Islam and traditional religion.

Study design and population

The study design is a descriptive cross-sectional survey consisting of apparently healthy prospective blood donors in BOUESTI, Ikere, Ekiti State. The study was carried out from January 2022 to August 2022.

A total of two thousand (2,000) apparently healthy Students of BOUESTI within age range of 17-30 years and of both sexes were recruited for this study.

Ethical approval and informed consent

Ethical approval was obtained from the Health Research and Ethics Committee of BOUESTI, Ikere, Ekiti state.

Informed consent was sought from each participant before sample collection.

Inclusion and exclusion criteria

Apparently healthy students of BOUESTI, Ikere, Ekiti State who gave their consent were included in this study. Individuals who did not meet the inclusion criteria and those who did not give their consent were excluded from the study.

Sample collection and analysis

2 ml of venous blood was collected from the subjects via venipuncture into EDTA container.

It was mixed homogeneously and the samples were made ready for analysis. The samples were used to determine the haemoglobin genotypes, ABO and rhesus blood groups of the subjects. The ABO blood group was determined using tile method, while the haemoglobin genotype was determined using haemoglobin electrophoresis.

Statistical analysis

The data obtained in this study were presented in tables. The frequency distribution of haemoglobin genotypes, ABO and Rhesus blood groups was analyzed using the Chi square (χ^2) test. The statistically significant level was set at $p < 0.05$.

RESULTS

The distribution of haemoglobin genotypes, ABO and rhesus blood groups of the subjects studied was presented in Table 1. The results obtained showed that out of the one thousand participants that participated in this study, 1,448 (72.4%) of the subjects had HbAA, 452 (22.6%) had HbAS, 72 (3.6%) had HbAC, 20 (1%) had HbSS and 8 (0.4%) had HbSC.

The distribution of ABO blood groups of the subjects revealed that blood group O was the most prevalent with 1,072 (53.6%). This was followed by blood group B with 532 (26.6%) and blood group A with 356 (17.8%) respectively. The least prevalent ABO blood group was AB (2.0%).

The distribution of rhesus blood group among the subjects was presented in Figure 1. The results obtained showed that 1,920 (96.0%) of the subjects had rhesus 'D' positive blood group, while 80 (4.0%) had rhesus 'D' negative blood group. Blood group O rhesus 'D' positive individuals were 1,040 (52.0%), while blood group O rhesus 'D' negative individuals were 32 (1.6%).

Similarly, individuals with blood group A rhesus 'D' positive group were 332 (16.6%), while blood group A rhesus 'D' negative group were 24 (1.2%). Furthermore, individuals with blood group B rhesus 'D' positive blood group were 508 (25.4%), while those are blood group B rhesus 'D' negative were 24 (1.2%). All blood group AB subjects were rhesus 'D' positive (2.0%).

The distribution of ABO and rhesus blood group of the subjects in relation to sex was presented in Table 2.

The results obtained showed that of the 840 males who participated in this study, 492 (24.6%) had blood group O, 124 (6.2%) had blood group A, 208 (10.4%) had blood group B and 16 (0.8%) had blood group AB. Similarly, 580 (29.0%) of the female participants had blood group O, 232 (11.6%) had blood group A, 324 (16.2%) had blood group B and 24 (1.2%) had blood group AB. There was no significant difference ($p > 0.05$) in the ABO blood group distribution between male and female subjects. Furthermore, 808 (40.4%) of the male subjects had rhesus 'D' positive blood group, while 32 (1.6%) had rhesus 'D' negative blood group.

On the other hand, 1,112 (56.6%) of the female subjects had rhesus 'D' positive blood group, while 48 (2.4%) had rhesus 'D' negative blood group. There was no significant difference ($p > 0.05$) in the rhesus blood group distribution between male and female subjects.

The distribution of haemoglobin genotypes of the subjects according to sex was presented in Figure 2. The results obtained showed that 584 (29.2%) of the male subjects were HbAA, 200 (10.0%) were HbAS, 40 (2.0%) were HbAC, 12 (0.6%) were HbSS and 4 (0.2%) were HbSC. Similarly, 864 (43.2%), 252 (12.6%), 32 (1.6%), 8 (0.4%) and 4 (0.2%) of the female subjects had HbAA, HbAS, HbAC, HbSS and HbSC respectively. There was no significant difference ($p > 0.05$) in the distribution of haemoglobin genotypes in male and female subjects.

The distribution of ABO and Rhesus blood group of the subjects in relation to age was presented in Table 3. The blood group distribution of subjects in age group 16-20 years were; O (22.6%), A (9.8%), B (9.6%) and AB (1.0%). Similarly, the rhesus blood group distribution of subjects in age group 16-20 years were (41.0%) and (2.0%) for rhesus 'D' positive and rhesus 'D' negative group respectively.

Furthermore, the blood group distribution of subjects in age group 21-25 years were; O (22.0%), A (5.0%), B (12.2%) and AB (0.6%), while the rhesus blood group distribution was 383 (38.3%) and 15 (1.5%) for rhesus 'D' positive and rhesus 'D' negative antigen respectively. Age group 26-30 years blood group distribution were; 9.0%, 3.0%, 4.8% and 0.4% for blood group O, A, B and AB respectively, while the rhesus factor was 16.7% and 1.0 for rhesus 'D' positive and rhesus 'D' negative factor. There was no significant difference ($p > 0.05$) in the blood group distribution of the subjects in relation to age.

The distribution of haemoglobin genotypes of the subjects in relation to age was presented in table 4. The results obtained showed that in age group 16-20 years, 590 (29.5%) of the subjects had HbAA, 212 (10.6%) were HbAS, 40 (2.0%) were HbAC, 12 (0.6%) were HbSS and 6 (0.3%) were HbSC.

In age group 21-25 years, 608 (30.4%) of the subjects were HbAA, 160 (8.0%) were HbAS, 20 (1.0%) were HbAC, 6 (0.3%) were HbSS and 2 (0.1%) had HbSC. Finally, the haemoglobin genotype distribution in age group 26-30 years were; 250 (12.5%) for HbAA, 80 (4.0%) were

HbAS, 12 (0.6%) were HbAC and 2 (0.1%) were HbSS respectively.

There was no significant difference ($p>0.05$) in the haemoglobin genotypes of the subjects in relation to age.

Table 1: Distribution of haemoglobin genotypes and ABO blood groups of the subjects studied.

Parameters	N	%
Haemoglobin genotypes		
AA	1,448	72.40
AS	452	22.60
AC	72	3.60
SS	20	1.00
SC	8	0.40
Total	2,000	100.0
Blood groups		
O	1,072	53.60
A	356	17.80
B	532	26.60
AB	40	2.00
Total	2,000	100.0

Table 2: Distribution of ABO blood groups of subjects in relation to sex.

Sex	No. screened	ABO N (%)			Rhesus N (%)		
		O	A	B	AB	Rh+	Rh-
Male	840	492 (24.6)	124 (6.2)	208 (10.4)	16 (0.8)	808 (40.4)	32 (1.6)
Female	1,160	580 (29.0)	232 (11.6)	324 (16.2)	24 (1.2)	1,112 (55.6)	48 (2.4)
Total	1000	1,072 (53.6)	356 (17.8)	532 (26.6)	40 (2.0)	1,920 (96.0)	80 (4.0)
X ²		0.414	0.247	0.642	0.741	1.204	0.939
P value		0.368	0.102	0.454	0.360	0.260	0.357

Table 3: Distribution of ABO blood groups of subjects in relation to age.

Age (years)	No. screened	ABO N (%)			Rhesus N (%)		
		O	A	B	AB	Rh+	Rh-
16-21	860	452 (22.6)	196 (9.8)	192 (9.6)	20 (1.0)	820 (41.0)	40 (2.0)
21-25	796	440 (22.0)	100 (5.0)	244 (12.2)	12 (0.6)	766 (38.3)	30 (1.5)
26-30	344	180 (9.0)	60 (3.0)	96 (4.8)	8 (0.4)	334 (16.7)	10 (1.0)
Total	2,000	1,072 (53.6)	356 (17.8)	532 (26.6)	40 (2.0)	1,920 (96.0)	80 (4.0)
X ²		0.647	0.458	0.439	0.850	0.678	0.744
P value		0.348	0.406	0.201	0.640	0.365	0.298

Table 4: Distribution haemoglobin genotypes of subjects in relation to age.

Age (years)	No. screened	Haemoglobin genotypes				
		AA	AS	AC	SS	SC
16-21	860	590 (29.5)	212 (10.6)	40 (2.0)	12 (0.6)	6 (0.3)
21-25	796	608 (30.4)	160 (8.0)	20 (1.0)	6 (0.3)	2 (0.1)
26-30	344	250 (12.5)	80 (4.0)	12 (0.6)	2 (0.1)	-
Total	2,000	1,448 (72.4)	452 (22.6)	72 (3.6)	20 (1.0)	8 (0.4)
X ²		0.611	0.751	0.645	0.245	0.364
P value		0.351	0.501	0.396	0.410	0.244

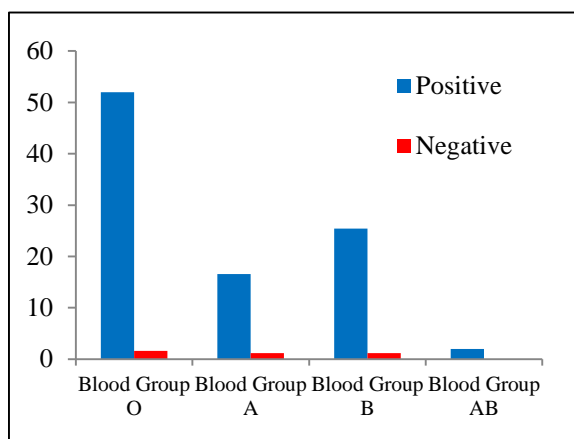


Figure 1: Bar-chart showing the percentage distribution of ABO rhesus blood group among the subjects studied.

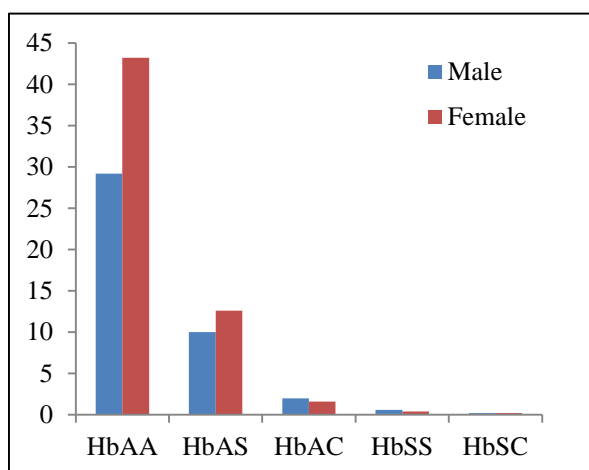


Figure 2: Distribution of haemoglobin genotypes of subjects according to sex.

DISCUSSION

In studies of the human population, helpful genetic markers include the ABO and rhesus blood groups. They are the most often used and important blood types in terms of transfusion.⁵ Haemoglobin (Hb) genotype is an important blood component that determines haemoglobinopathies. Haemoglobinopathies are among the most frequently inherited genetic disorders in the world, and they are passed down through healthy carrier/disease parents as autosomal recessive disorders.² This study was carried out to determine the haemoglobin genotypes, ABO and Rhesus blood groups of Students in BOUESTI, Ekiti state, Nigeria.

In this study, the distribution in regard to ABO blood phenotype showed that blood group O was the most common blood group (53.6%), followed by blood group B (26.6%), and blood group A (17.8%), while the least blood group was AB (2.0%). The result on the distribution of the population based on ABO blood phenotype was given by O>B>A>AB. The result of this study on the ABO blood

group phenotype frequency distribution is similar with the pattern seen in the previous studies that reported high frequencies of group O and low frequency of AB.¹⁵⁻¹⁹

The pattern of ABO blood group (O>B>A>AB) observed in this study is agreement with the study by Kooffreh et al in Calabar, Nigeria which reported that blood group O had the highest frequency (55.2%), followed by blood group B (21.6%), then blood group A (18.8%) and AB had the least (4.4%).¹⁴ In a study conducted in Oshogbo, South-West Nigeria by Muhibi, they found that blood group B (21.3%) was slightly higher than blood group A (21.1%) which corroborates our findings carried out in South-West Nigeria (Ikere-Ekiti).²⁰ Furthermore, Etim et al in a study carried out in Adamawa, North East, Nigeria, reported the blood group gene frequencies to be 56.2%, 21.3%, 17.7% and 4.7% for O, B, A and AB respectively.²¹ Another study conducted by Boskabady et al in Mashhad (North East of Iran), showed that the percentage of blood groups A, B, AB and O among the city of Mashhad population was 23.1%, 23.3%, 8.9% and 34.7% respectively.¹⁵ Likewise, in other studies conducted by Onuoha et al among residents of Yenagoa and Environs, Bayelsa State, Erhabor among students in the Niger Delta of Nigeria; and Akhigbe in Ladoko Akintola University of Technology, Ogbomosho, Nigeria, all reported higher frequency of blood group B than blood group A.^{5,22,23} Furthermore, our results also agrees with report from previous international studies carried out in Madagascar, Guinea, Pakistan and India that reported this pattern (O>B>A>AB) of ABO blood group distribution.²⁵⁻²⁸

On the contrary, the pattern of ABO blood group (O>B>A>AB) observed in this study differ from some studies in Nigeria that reported ABO blood group pattern of (O>A>B>AB). In a previous study in Ekpoma, Edo state by Nwaopara, a higher percentage of the population (63.73%) were blood group O, while those with blood group A, B and AB were (17.62%), (16.58%) and (2.07%), respectively.²⁸ Adeyemo et al in a study in Ibadan reported a frequency of 55.3%, 25.3%, 16.7% and 2.7% for blood group O, A, B and AB respectively.²⁹ A frequency of 26.7% for blood group A, 18.3% for blood group B, 2.2% for AB and 52.8% for blood group O in Port Harcourt, Southern Nigeria was reported by Jeremiah et al while Uneke et al in South-eastern Nigeria reported a frequency of 25.0% for blood group A, 16.4% for blood group B, 1.9% for AB and 56.7% for blood group O.^{4,30} Furthermore, a study conducted in Lagos, Nigeria by Odegbemi et al reported the prevalence of ABO blood group O, A, B and AB as 51.8%, 26.3%, 18.2% and 3.6% respectively.³¹ A nation-wide study by Anifowoshe et al reported similar pattern of ABO blood group distribution (O>A>B>AB: 52.93%, 22.77%, 20.64% and 3.66% respectively).³² The above previous studies supported the finding of our study; however, the only difference is the distribution of blood group A and B in some reports.

The high predominance of blood group O people in nature, as seen in this study and previous studies, is advantageous

since it indicates that there will always be blood available in an emergency. However, due to some group O blood being known to have strong immunological hemolytic antibodies (haemolysins), caution should be exercised when doing this.⁴ Every group O blood sample should undergo a routine hemolysin test to help lower the likelihood of a transfusion response. Additionally, it has been noted that the O phenotype has a parity-specific connection with malaria immunity that is protective during pregnancy, resulting in enhanced birth anthropometry.³³

In the present study the frequency distribution of ABO blood groups in both sexes was the same trend (i.e. O>B>A>AB). There was also no significant difference ($p>0.05$) in the blood group distribution of the subjects in relation to age. This finding is consistent with previous studies.^{15,18,23,31,34,35} It has been asserted that, rather than age and gender, factors such as geographic location, natural selection (infectious disease), genetic drift, gene flow between populations, and marriage distance (such as consanguineous and short marriage distance) may all play a role in the distribution of blood groups differences and similarities that exist across different populations.¹⁵

In this study the distribution of rhesus positive (Rh⁺) blood group (96.0%) was more predominant than rhesus negative (Rh⁻) blood group (4.0%). The pattern of rhesus blood group observed in our study was generally consistent with previous studies within Nigeria.^{21,23,28,29,31,32,34,35} While we reported a value of 4.0% prevalence for Rh⁻ in our study, other studies reported values as low as 2.9% in Yola, Nigeria, 2.3% in Uganda and 1.2% in Gusau, Nigeria.^{5,36,37}

The study's findings about the low gene frequency of the Rh⁻ blood type are advantageous for blood banking and the treatment of diseases like hemolytic disease of the newborn (HDN).³⁸ It reduces the need for Rh⁻ blood for transfusions, which is good news for blood bank management who often have a monumental task on their hands. In terms of Rhesus alloimmunization and attendant HDN, which frequently occur when a Rh⁻ mother becomes pregnant with a Rh⁺ fetus (acquired from the Rh⁺ father), it also gives the population certain obstetric advantages.⁵ However, it is well known that in most resource-constrained nations, rhesus alloimmunization considerably contributes to perinatal morbidity.⁵ Additionally, the fact that diverse rhesus blood groups are present in the research population necessitates the promotion of pre-marital counseling. This will allow Rh⁻ females who want to marry Rh⁺ males to take precautions against fetal loss and infant mortality as a result of HDN.

The results of this study showed that 72.4% of the subjects had HbAA, 22.6% had HbAS, 3.6% had HbAC, 1.0% had HbSS and 0.4% had HbSC. The 72.4% reported for HbAA in this study is in line with the value previously reported for Africa which is 50-75%, while the 22.6% reported for HbAS in this study is in agreement with the 20-30% recorded for Nigeria and within the value of 20-40% recorded for Africa.^{4,5,38} Generally, the pattern of Hb

genotype distribution observed in our study was similar to previous studies in Nigeria.^{23,29,36} In our study, we observed the incidences of HbAC (3.6%), HbSC (0.4%) and HbSS (1.0%) as consistent with the findings of a previous study among the Yoruba's et al in Ibadan and another study among the Ika ethnic group of Delta state.^{39,40} In our study, there was absence of HbCC which aligns with previous studies who had no report of HbCC in their respective studies.^{22,36,41} This difference could be due to the variable nature of variants of haemoglobin.

HbC is one of the most prevalent structural haemoglobin variants in humans; HbC trait (HbAC) is asymptomatic, and such heterozygote people are phenotypically normal; however, HbC disease (HbCC) patients may have mild haemolytic anaemia because of the RBCs' decreased solubility, which can result in crystal formation, splenomegaly, and borderline anaemia.⁴³ However, sickle-HbC disease (HbSC), which results from the inheritance of this HbC mutation along with HbS, may have serious clinical repercussions, including chronic haemolytic anaemia and sporadic sickle cell crises. Haemoglobin genotype screening for prospective couples is still necessary despite the low incidence of HbC trait (HbAC) in our current study (3.6%) in order to keep HbSC and HbCC out of the community.

Sickle cell trait (SCT) prevalence in the general population determines the prevalence of sickle cell disease (SCD), which is expected to be at least 2% if SCT prevalence is greater than 20%.⁴⁴ Although the frequency of SCT (HbSS) was only 1.0% in the current study, the proportion of individuals with SCT was found to be 22.6%. These results suggest that SCD was declining, as had been seen in several studies, and this could be attributed to parental Hb genotype testing before marriage, enhanced premarital counseling, increased knowledge of the risks of sickle cell anaemia, improved socioeconomic status, and increased awareness of fetal Hb genotype screening.²³ The low frequency of HbSS in the current study population could also be attributed to the study environment's disruption of the Hardy-Weinberg equilibrium with respect to the sickling gene, which could also be connected to people's growing awareness of the importance of knowing one's genotype before marriage.³⁸ With this awareness, sickle cell disease will eventually become a thing of the past.

CONCLUSION

The study concludes that HbAA and blood group O is the most predominant haemoglobin genotype and blood group respectively in the study area. Rhesus 'D' positive blood group was the most predominant as well. The result on the distribution of the population based on ABO blood phenotype was given by O>B>A>AB, while the genotype was given by AA>AS>AC>SS>SC respectively. There was no significant difference ($p>0.05$) in the haemoglobin genotypes and ABO blood groups of the subjects with respect to age and sex. Given the high incidence of sickle cell traits in our study location, it is recommended that

premarital counseling be given to prospective couples together with haemoglobin genotype testing in order to achieve the WHO goal of reducing the threat of SCD.

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