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Correlation between ABO/Rhesus Blood Group and a Sickle Cell Disease among Sicklers at the Sickle Cell Center, Alkuwity Teaching Hospital, Elobeid City, North Kordofan Estate, West of Sudan: A Cross-Sectional Study

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Abstract

Aims: Sickle Cell Disease (SCD) is a genetic blood disorder affecting red blood cells, with high morbidity and mortality rates. Many attempts being made to determine the relationship between blood antigens, Hemoglobin (Hb) genotypes.

Objective: The study was carried out to determine any potential correlation between ABO/Rhesus D Blood Groups and SCD and SCD and to provide data on the distribution of ABO/Rhesus D Blood Groups among Sicklers attending the Sickle Cell Center, Alkuwity Teaching Hospital, Elobeid City, North Kordofan Estate, and West of Sudan.

Materials and Methods: A total of 230 Sicklers (Hemoglobin SS) patients, 111 males, 119 females were enrolled in the study after signing the written informed consent form, all the participants were interviewed by a questionnaire which contains their basic sociodemographic and medications uses. ABO/Rhesus blood typing was done by classic (Antigen-antibody agglutination test).

Results: The ABO blood group distribution indicated that blood group O was the most prevalent (56.5%) among our subjects, followed by A (27%), B (13%), and AB (3.5%). O was the predominant blood group followed by group A, B and AB (p=0.001). A significant number of subjects were Rhesus D positive (89.1%) compared to 10.9% who were Rhesus D negative (p=0.001). Blood group O^{+ve} is the dominant blood group. There was a correlation between ABO/Rhesus blood group and sickle cell disease (p<0.05).

Conclusion: The results from this study also show that blood group O^{+ve} is the predominant blood group in the Sickle Cell Center, Alkuwity Teaching Hospital, Elobeid City, North Kordofan Estate, and West of Sudan. Evidenced based data obtained from this study will facilitate optimal stocking of blood and blood products in blood banks in the area and the management of Hemolytic Transfusion.

Keywords: Sickle cell disease; Hemoglobinopathies; ABO; Rhesus d blood groups; Haemoglobin genotypes; SUDAN

Introduction

Blood group system is well-defined based on presence or absence of antigens existing on the surface of red blood cells [1]. In spite of the presence of more than 400 blood group antigens, the ABO and Rh have been recorded as the major clinically significant blood group antigens and the main human blood type system with principal importance in clinical practice and constitutes the fundamental basis of blood banking and modern transfusion medicine [2]. ABO blood group antigens are glycoproteins. Their expression is partly dependent on racial origin [3]. "ABO" blood type system comprises of four major "ABO" phenotypes "A", "B", "O", and "AB" depending on the presence or absence of gene "A" and "B" positioned on short arm of chromosome 9 (9q34) [4,5]. The ABO blood group system was the first human blood group system discovered by Karl Landsteiner in 1900.

The Rhesus factor is another form of blood belongings associated with ABO blood grouping. Rhesus factors were discovered in 1937 by Karl Landsteiner and Alexander Solomon Wiener who first discovered them in blood of a rhesus monkey. After its discovery, Rh D blood group was subsequently confirmed to be of importance in blood group serology. The importance attached to Rh D blood group system is due to its immunogenicity in negative individuals with multiple blood transfusions and pregnancies [6]. Concerning the rhesus blood group system, there are only two Rh phenotype Rh positive and Rh negative, depending on the presence or absence of the Rh antigen on the RBC [7]. Clinically, the Rhesus blood group system is second in importance to the ABO system.

Haemoglobin (Hb) is the oxygen carrying pigment of the RBCs. Faults in its genes can produce abnormal haemoglobin which leads to disorders known as hemoglobinopathies for example sickle cell disorders which is the most predominant hemoglobinopathies in human people. Sickle cell disease disturbs millions of people throughout the

globe. Sickling disorders include the heterozygous form for haemoglobin S or the sickle cell trait (AS), the homozygous state for HbS or sickle cell anemia (SS), and the compound heterozygous form for HbS together with other haemoglobin (C,D,E) or other structural variants [8]. SCD is an inherited blood disorder/disease caused by abnormal hemoglobin. It is genetically transmitted as autosomal recessive mode [9]. The Haemoglobin S (HbS), which is a structural variant of the normal Haemoglobin (HbA), be the result of a genetic mutation in the β-globin gene where thymidine replaced adenine, resulting in the substitution of glutamic acid by valine, a neutral amino acid in position 6 of the β-globin chain. Sickle cell disease was a well-known disorder in West Africa for many years before it was discovered in America in November 1910 by Dr. James Herrick, a Chicago physician [10]. Sickle Cell Anemia (SCA) is the homozygous state for the HbS gene (HbSS), and it is the commonest and severest type of Sickle Cell Disease (SCD). The haematological feature of SCD is characterized by red cell sickling, haemolytic anaemia, leukocytosis and thrombocytosis. The clinical course of SCD is typically characterized by variable periods of painless steady state that is periodically break off by painful Vaso-Occlusive Crisis (VOC) resulting from deoxygenation of HbS i.e. red cell sickling, tissue necrosis, chronic anemia, ischemic organ damage, infections, short stature, and delayed puberty. Anemia in SCD could be distressing and may lead to repeated hospitalization and, transfusion dependency [11]. The frequencies of abnormal haemoglobin variants vary from one population to another people in Africa constitute a major part of the population that is susceptible to haemoglobinopathies. It is well-known that ABO/Rhesus blood group and SCD are genetically inherited. There are inconsistent reports about the correlations between ABO/Rhesus blood group and sickle cell disease. Many attempts being made to find an answer about the relationship between blood antigens, Hemoglobin (Hb) genotypes and increased susceptibility to certain diseases are unending. Therefore, the present study was conducted to determine if there is any correlation between ABO/Rhesus D blood group and SCD among Sicklers at the Sickle Cell Center, Alkuwity Teaching Hospital, Elobeid City, North Kordofan Estate, and West of Sudan.

Materials and Methods

Ethical approval

Two hundred and thirty (230) apparently healthy subjects (Males-111, Females-119) were randomly selected for this study. Ethical approval was obtained from the Alkuity Teaching Hospital Ethical Committee Board, as well as the principal and the Head of Medical Laboratory units of Alkuity Teaching Hospital approved the study. All the participants gave their verbal informed consent before sample collection and they willingly presented themselves at the Medical Laboratory for sample collection and donated their blood sample for the study. Their results were confidentially given to them.

Study design

The aim of this prospective cross-sectional study was to determine the distribution of ABO and Rhesus D blood group among sicklers at Alkuwity Teaching Hospital, Sickle Cell Center, Elobeid City, North Kordofan Estate, and West of Sudan during the period March-December 2022.

Study subjects

A total of 230 Sickle Cell Disease (Hemoglobin SS) patients consecutively sicklers with homozygous haemoglobinopathies HbSS (with a diagnosis of SCA i.e. known patients with SCA (HbSS) as diagnosed by cellulose acetate electrophoresis at pH 8.6) attending the consultant outpatient clinic at the Sickle Cell Center, Alkuwity Teaching Hospital, Elobeid City, North Kordofan Estate, West of Sudan, they constituted the subjects for this study between March 2022 and December 2022 in their steady state were recruited into the study after obtaining informed consent. They included 111 males and 119 females. The SCD patients used for this research were both children and adults in stable hemodynamic states and with no apparent sickness, with age range of 1–29 years (mean age 9.24 ± 6.570 years). Structured questionnaires with questions to suit the population of study needs were in use for the study. After signing the written informed consent form, all the participants were interviewed by a questionnaire which contains their basic sociodemographic and medications uses (e.g., age, gender, residence, education level, tribe, medications used such as folic acid and hydroxyurea). Questionnaires were completed *via* face-to-face interviews to gather information.

ABO and Rh blood groups tests

Red cell phenotyping was carried out with standard Antisera to determined antigens-antibodies reaction on the red blood cell. Aseptic measures were ensured, and blood samples were taken by finger pricking with sterile lancet. ABO/Rhesus blood typing was done by classic (antigen-antibody agglutination test) method by slide method using Anti Sera-A, Anti Sera-B and Anti Sera-D marketed by Diagnostics Ltd (Crescent Company) KSA. Blood obtained by finger pricking was used for each of the depression on the tile, with the used of Pasteur pipette three different spots of blood was placed on the tile, equal volume of anti-A (blue) was placed on the first drop and anti-B (yellow) to the second drop as well as anti-D (colorless) was placed on the last spot. Clean glass slide was used to mixed each spot respectively, it was shaken for about two minutes and agglutination were observed and result was interpreted.

Statistical analysis

The data obtained were analyzed statistically to determine any relationship between ABO/Rhesus blood group and Sickle Cell Disease (SCD). The data were analyzed by the Statistical Package of Social Science (SPSS) software version 24.0 in windows 10. Demographic data of the study population was evaluated by descriptive statistics. Categorical variables were reported as number and percentages, whereas continuous variables like age, were expressed as Mean (M) ± Standard Deviations (SD). Proportions of the studied groups were expressed in percentages (%) and absolute number (n) of frequencies. The Chi-square test was used to determine the relationship of ABO/Rhesus blood group and Sickle Cell Disease (SCD). The confidence limit was kept at 95%, hence values less than 0.05 on the Chi-square distribution table is statistically significant (a P-value<0.05) was considered to be statistically significant. While greater than 0.05 (a P-value>0.05) on the Chi-square distribution table is not significant.

Results

Two hundred and thirty sicklers (HbSS) were enrolled in this study. Out of the 230 sicklers; 111 (48%) were males and 119 (52%) were females (Figure 1).

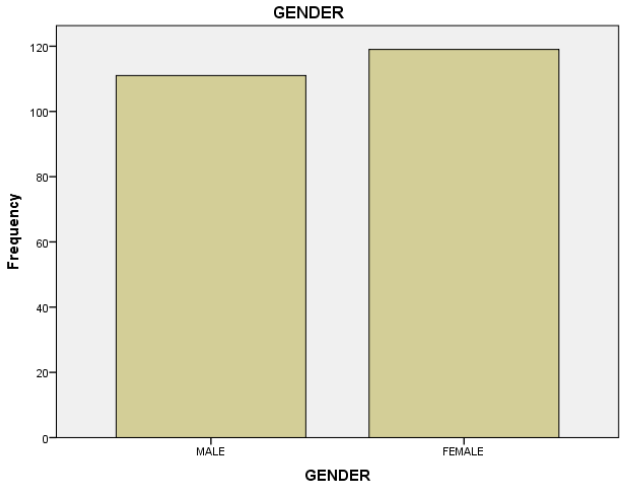

Figure 1: Demographic Characteristics of Study Participants.

The mean age of the study group 9.24 ± 6.570 years. 25.9% of sicklers were Falata, 15.8% were Misaria, 8.3% were Bidariaa, 3.5% were Gawamaa, 3.9% were Barno, 2.6% were Bargo, 35.5% were Hausa 2.6% were Silihab, 2.6% were Shanabla (Figure 2).

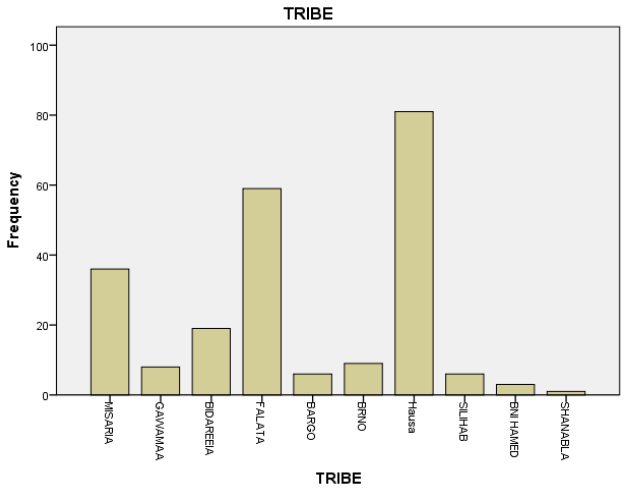

Figure 2: Demographic and Ethnic Characteristics of the Study Group.

The frequencies of all the ABO blood groups are shown in Table 1.

ABO blood group	Frequency (n)	Percent (%)
A	62	27
B	30	13
AB	8	3.5
O	130	56.5
Total	230	100

Table 1: ABO Blood Groups Pattern of Sicklers (HbSS).

Table 1 shows the distribution of the ABO blood groups among 230 sicklers. Blood group O was the most prevalent (56.5%) among our subjects, followed by A (27%), B (13%), and AB (3.5%). Blood group O, it was more prevalent (56.5%) (Figure 3).

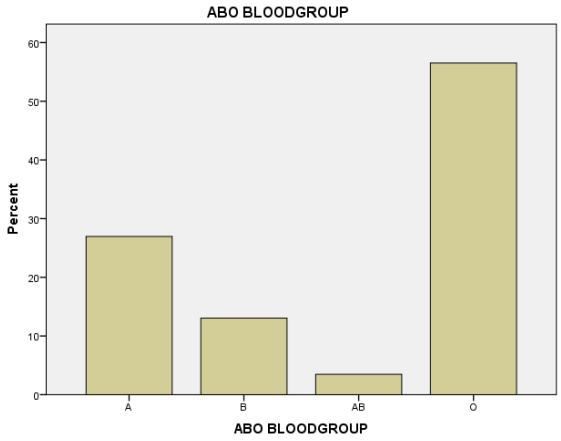

Figure 3: Distribution of Blood Groups among Study Subjects.

The most and least prevalent blood groups were blood group O and AB, 56.5% and 3.5%, respectively. The O blood group had the highest prevalence (56.5%) while the AB blood group was the least (3.5%) in this study. Hb SS was more prevalent among group O subjects followed by A, B and AB. The ABO distribution among the subjects followed the pattern O>A>B>AB. This distribution is statistically significant (P=0.000) i.e. $p<0.01$

The frequencies of all the Rh D blood groups are shown in Table 2.

Rhesus blood group	Frequency(n)	Percent (%)
Rhesus Positive	205	89.1
Rhesus Negative	25	10.9
Total	230	100

Table 2: Rhesus factor Blood Groups Pattern of Sicklers (HbSS).

Table 2 shows the distribution of the Rhesus Factor (D) blood groups among 230 sicklers. Rhesus negative was found to be less common (10.9%), in comparison, Rhesus positive was found to be more common in sicklers (89.1%) in this study Figure 4.

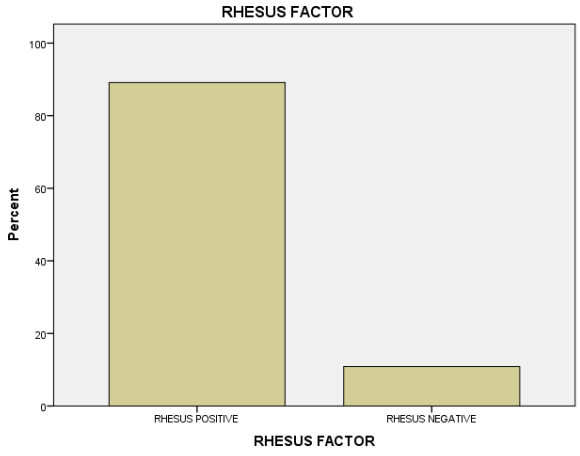

Figure 4: Distribution of Rhesus factor Among Study Participants.

Rh D^{+ve} was the most prevalent blood group while Rh D− was the least prevalent blood group. This distribution is statistically significant (P=0.000) i.e. p<0.01. The frequencies of Rh D groups among the four ABO groups are shown in Table 3. Out of which 62 (27.0%) were A Rhesus D positive, 28 (12.2%) were B Rhesus D positive, 130 (56.5%) were O Rhesus D positive and 3 (1.3%) B rhesus D negative blood group,3 (1.3%) AB Rhesus D negative blood groups.

ABO/Rhesus Factor blood group	Frequency (n)	Percent (%)
A^{+ve}	62	27%
A^{-ve}		
B^{+ve}	28	12.2
B^{-ve}	3	1.3
AB^{+ve}	4	1.7
AB^{-ve}	3	1.3
O^{+ve}	130	56.5
O^{-ve}		
Total	230	100

Table 3: ABO Blood Groups Pattern of Sicklers (Hb SS).

This distribution is statistically significant (P=0.000) i.e. p<0.01. There was neither A Rhesus D negative blood group (0.0%) nor O Rhesus D negative blood group (0.0%) from this study. Rh D^{-ve} was recorded most in group O (56.5%) and least (1.3%) in group AB and in group B.

Table 4 shows Gender ABO/Rhesus cross tabulation.

Discussion

The ABO system encompasses 4 groups, A,B,AB and O, determined genetically by 3 allelic genes located on chromosome 9. The distribution of blood groups varies both regionally and ethnically. The distribution of ABO and Rh blood groups varies greatly in different geographical and ethnic groups [12]. Haemoglobin genotypes and blood groups are all inherited blood types. The inherited disorders of haemoglobin are the most communal gene disorders with 7% of the earth population being carrier. Blood group distribution among various population groups is an important consideration in health care. This study was designed to determine the ABO and Rhesus blood group distribution among sicklers in at the Sickle Cell Center, Alkuwity Teaching Hospital, Elobeid City, North Kordofan Estate, West of Sudan. In gender distribution, it was observed that females were found to be more in sicklers (52%) than males (48%). The most common blood group was O (56.5%); AB was the least common (3.5%). Our results revealed that ABO blood group distribution is in the following order O>B>A>AB i.e. blood group O is most distributed and AB blood group is least distributed. The prevalence of Rh-positive and Rh-negative was 81.9% and 10.1% respectively. Blood group O has 56.5% of HbSS genotype. In rhesus blood group, the prevalence of rhesus positive among the patients of HbSS genotype were 81.9%. This study therefore showed significant correlation between ABO/Rhesus blood group and sickle cell disease. It is therefore concluded that ABO/Rhesus blood group has correlation with sickle cell disease (p<0.01) and genomic expression is in this order O>B>A>AB. Rhesus positive blood group is also more prevalent in sickle cell patients and Rhesus negative is less prevalent in sickle cell disease. It is therefore concluded that ABO/Rhesus blood group has correlation with sickle cell disease.

It is well-known that ABO/Rhesus blood group and SCD are genetically inherited. There are inconsistent reports about the correlations between ABO/Rhesus blood group and sickle cell disease. The people in Africa constitute a major part of the population that is susceptible to many erythrocytic hereditary and haematological disorders for example haemoglobinopathies. The frequencies of abnormal haemoglobin variants vary from one population to another. The prevalence of ABO/Rhesus blood groups reported in our study is in agreement with other studies from different parts of Nigeria. Many previous reports are in agreement with the frequencies obtained in this study and it suggest that group O appears to display predominance over the other blood groups. For example, findings of this study were in agreement with findings of Alagwu et al., study Correlation between ABO/Rhesus blood group, Sickle Cell Disease (SCD) which includes 100 proven cases of sickle cell patients (HBSS) from the sickle cell clinic in the General Hospital, Okwe, Asaba, Delta State, Nigeria.2016 [9]. In the ABO/Rhesus blood group and SCD, the result showed that there was a correlation between ABO/Rhesus blood group and sickle cell disease (p<0.05). It was also observed that blood group O has the highest frequency distribution among the sicklers (63%), followed by blood group B (20%), then blood group A (17%), the least was AB blood group with (0%) distribution. For Rhesus blood system, the prevalence of Rh positive was 96% and Rh negative was 4%. Again in accordance with our results the study by Medugu, et al., investigations carried out in the Department of Hematology, Federal Medical Centre, Yola, North-Eastern Nigeria, between March, 2010 and December, 2012. The study was carried out to provide information on the distribution of ABO, Rh blood groups, and Hb phenotypes in pregnant women in Yola, North-Eastern Nigeria. A total of 2226

Gender* ABO Rhesus cross tabulation								
Count								
		ABO Rhesus						Total
		A^{+ve}	B^{+ve}	B^{-ve}	AB^{+ve}	AB^{-ve}	O^{+ve}	
Gender	Male	24	12	2	2	2	69	111
	Female	38	16	1	2	1	60	118
Total		62	28	3	4	3	129	229

Table 4: Gender ABO/Rhesus Blood Groups.

(n2226) records of women attending antenatal care was obtained and analyzed. The most and least prevalent blood groups were blood group O and AB, 47.7% and 3.5%, respectively. Rh D^{+ve} and Rh D^{-ve} were recorded in 97.1% and 2.9% of studied subjects, respectively. Aligned with our results was study conducted by Ahmed et al., [13]. This is a retrospective cross-sectional cohort study conducted in the University of Maiduguri Teaching Hospital, Maiduguri, Borno State, northeast Nigeria, 2006–2007 (2 years) and in Aminu Kano Teaching Hospital, Kano, northwest Nigeria, 2009–2010 in 352 adult patients with SCD in Nigeria. The most and least prevalent blood groups were blood group O and AB, 53.1% and 3.1%, respectively. In harmony with our results was study conducted by Nwabuko et al., which includes a total of 52 SCD pregnant women with a mean age of 27 were seen at the antenatal clinic of BMSH, Port Harcourt, between January 2004 and December 2013, out of which 14 (27.0%) were A Rhesus D positive, 5 (9.6%) were B Rhesus D positive, 32 (61.5%) were O Rhesus D positive and 1 (1.9%) O Rhesus D negative blood groups. There was no AB Rhesus blood group from this study. The O blood group had the highest prevalence (63.4%) while the AB blood group was the least (0.0%) from this study. The gene frequency with respect to ABO system can be shown as O>A>B>AB. 98.1% of the women were Rhesus D positive blood group while 1.9% (1) was Rhesus D negative blood group. In Consistent with our findings study conducted by Amodu et al., [14]. The study was carried out in Ibadan, Southwest Nigeria. The participants in this study comprised 3100 unrelated children were recruited from the Children's Emergency ward of the University College Hospital, Ibadan and the Adeoyo Maternity Hospital Ibadan and the Oni Memorial Hospital, Ibadan, which are all within the same area in Ibadan. The percentage distribution of the ABO group in the entire study population was A: 22.5%, B: 25.2%, AB: 4.6%, O: 47.7% [14]. Again, in line with our results was the result of study entitled Abnormal haemoglobin variants, ABO and Rh blood groups among student of African descent in Port Harcourt, Nigeria which includes six hundred and twenty (620) apparently healthy subjects (males-166, females-454) were selected randomly from a cross-section of college students. Of the 620 subjects screened, 80.32% were HbAA, 19.68% were HbAS. 22.9% were of blood group A, 17.10% group B, 4.84% group AB and 55.16% group O. 96.77% were Rh D positive while 3.23% were Rh D negative [15]. The study of ABO Blood Groups and Haemoglobin Genotypes in Hassan Usman Katsina Polytechnic, Katsina, Nigeria. Which includes a total of 100 volunteer randomly selected for this work. The volunteers were sent to General hospital Katsina for blood groups and haemoglobin genotype tests. The results show that 67 out of 100 have the normal haemoglobin (AA), 39 of which belong to group O^{+ve}. 30 have the sickle cell trait (AS) of which 16 belong to group O^{+ve}. Only 2 individuals have the sickle cell disease (SS), and they also belong to group O^{+ve}. Blood group O^{+ve} is the dominant blood group in Hassan Usman Katsina Polytechnic, Katsina [16]. Also, our findings were in agreement with results of the study entitled Prevalence of sickle cell disease/ABO blood group among secondary school students: A case study of Government Day Secondary School (GDSS) mile six Jalingo, Taraba state, Nigeria. One hundred and thirty-nine (139) students selected at random constituted the subjects for this study. The result showed that; (106) 76.26% have HbAA haemoglobin, (32) 23.02% have HbAS haemoglobin, (1) 0.72% have HbSS haemoglobin. ABO blood group screened were as follows: Group A^{+ve} (35) 25.18%, group B^{+ve} (30) 21.58%, group AB^{+ve} have (8) 5.76%, group O+ have (55) 39.57% were Rh. 'D' positive while the rest are Rh 'D' negative 2.87% (4) were group B^{-ve} 1.44% (2) were group AB^{-ve} 3.59% (5) were group O. Also, our findings were in concordance with results of the study entitled Haemoglobin Electrophoretic Patterns, ABO and Rhesus D Blood Groups Distribution among Antenatal Women in Sokoto, Nigeria, includes three hundred consecutively recruited pregnant women attending antenatal clinic in Sokoto, North Western Nigeria. The age range and mean age of these subjects was 18-45 years and 27.62 ± 3.6 years respectively constituted the subjects for this study. Among the 300 pregnant women studied, 221 (73.7%) were HbAA, 67 (22.3%) were HbAS, 11 (3.7%) were HbAC and 1 (0.3%) was HbSC (p=0.01). The ABO blood group distribution indicated that blood group O was the predominant blood group followed by group A, B and AB (p=0.01). The ABO distribution among the subjects followed the pattern O>A>B>AB. A significant number of subjects were Rhesus D positive (94%) compared to 6% who were Rhesus D negative (p=0.001) [17]. Likewise in study by Olorunshola and Audu entitled ABO (H) secretor status of sickle cell disease patients in Zaria, Kaduna State, Nigeria,which includes a total of 64 Sickle Cell Disease (Hemoglobin SS) patients (23 males and 41 females) All the subjects were of Hausa tribe from northern Nigeria. Blood group O had the highest frequency in SCD patients (39.1%). In contrast to our result and many previous studies Olorunshola and Audu, found that the least blood group frequency was blood group B (10.9%). O (39.1%), and A (21.9) AB (12.5) and B (10.9%) (Figure 5).

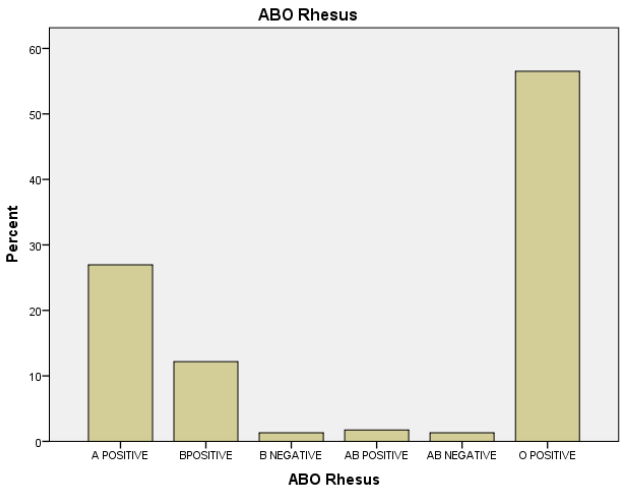

Figure 5: Comparison of Blood Group Frequencies with Previous Studies.

The ABO distribution among the subjects followed the pattern O>A>AB>B. There is a statistically significant correlation (p<0.001) [18].

Therefore, the frequencies of ABO and Rh blood groups in our study appeared to be constant and consistent with previous published data.

This study shows very high prevalence of Blood group O with low Rh D negative and rare AB blood groups among SCD patients. This same as findings of the study entitled Assessment of ABO-rhesus blood groups and hemoglobin concentrations of sickle cell disease pregnant women at booking in Nigeria. O blood group (Universal blood donor) are of advantage in terms of availability of blood during emergency, that of SCD patients would not provide such advantage as SCD patients' blood fall inside the category of permanently deferred blood as they are not qualified donors. Therefore, they cannot donate but can just receive blood. The scarcity of AB blood group among our study population means that much caution is needed during grouping

and cross-matching blood for transfusion to SCD patients as there are rare universal recipients in this population of patients. The support in this case will be more of group-specific blood transfusion so as to avoid adverse blood transfusion reaction. The high frequency of group O in this population provides an advantage in terms of availability of blood for blood transfusion especially in emergencies. However, there is a warning to this practice particularly since the plasma of some group O blood individuals are known to contain high titer of potent A and B immune haemolytic antibodies (haemolysins). One of the major challenges in sub Saharan Africa is accessing safe and adequate quantity of blood for transfusion [19].

Our result shows that sicklers with rhesus negative blood group were fewer than those with rhesus positive blood. This gives reasons for why people with rhesus negative blood do not get blood donors easily when they need. Fortunately, our result also shows that only two women (1.7%) from all women were Rh D negatives. This low percentage of D negative women protects from the risk of developing anti–D which can cause both moderate and severe form of Hemolytic Disease of Newborn (HDN).

Conclusion

In conclusion, it can be said that the majority were of blood group O^{+ve} was the predominant blood group among sicklers at Sickle Cell Center, Alkuwity Teaching Hospital, Elobeid City, North Kordofan Estate, and West of Sudan. Knowledge of the frequency of ABO and Rh D blood groups will help in the formulation of genetic counseling policies. Evidenced based data obtained from this study will facilitate optimal stocking of blood and blood products in blood banks in the area and the management of Haemolytic Transfusion.

The recommendations of this study were a compulsory premarital screening for sickle cell disease i.e. Blood test for genotype to intending couples to avoid sickle cell disease. People with rhesus negative blood should lead a very cautious life so as to avoid blood transfusion.

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