



Blood Group Antigens Phenotype Frequencies among Pregnant Women in Sokoto State, Nigeria

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Background: Blood group antigens have been used to evaluate ethnic diversity of human populations and had been related to predisposing individuals to some diseases or may protect individuals against some diseases. They also play the most important role in success of pregnancy and blood transfusion.

Aim: The aim is to determine the frequencies of some blood group antigens phenotype among pregnant women.

Materials and Methods: The study is a cross sectional study conducted in the department of obstetrics and gynaecology, antenatal clinic in Usmanu Danfodiyo Teaching Hospital Sokoto, from January 2020 to September 2020. This research included 1250 consecutively -recruited pregnant women on their first antenatal visit. The blood grouping were determined using standard tube techniques for ABO, MNSs, Duffy and Kidd antigens while column agglutination card was used for Rh C, E, c, e and Kell was utilized.

Results: The sociodemographic characteristics revealed that the age range of the subject was 17-48 years .where majority were within the age between 21-30 years (61.3%). Majority of the pregnant women had their first antenatal visit during second trimester of their pregnancy (59.7%). The distribution of the ABO blood group revealed that 48.5% were group O, 27.3% were group B, 19.4%

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were group A and 4.8% were group AB. Out the subjects investigated, 93.1%, 30.2%, 24.6% and 90.2% were RhD, RhC, RhE, Rhc and Rhe positive respectively. The prevalence of M, S and s positive were 75.5%, 31.4% and 63.3% respectively. Among the subjects studied, 97.6% were Kell positive while 2.4% were Kell negative. The prevalence of Duffy a and b antigen were 1.1% and 0.5% respectively and the prevalence of Kidd a and b phenotype positive were 15.9% and 21.7% respectively.

Conclusion: The pattern of distribution of ABO, Rh, MSs, Duffy and Kidd blood groups antigens among pregnant women in Sokoto was in agreement with other populations while that of Kell blood group antigen is at variance with other population particularly among Caucasians. Data derived from this study will help policy makers make evidenced –based decisions on management of HDFN.

Keywords: Blood group antigens; phenotypes; pregnant women; Sokoto.

1. INTRODUCTION

“Blood group systems consist of a group of antigens encoded by alleles at a single gene locus or at gene loci so closely linked that crossing over does not or rarely occur. An antigen collection consists of antigens that are phenotypically, biochemically or genetically related, but the genes encoding them have not been identified” [1]. “The Discovery of blood group antigen is usually beginning with the discovery of antibodies in the serum of multiply transfused blood recipients or the serum of a multiparous woman with a unique pattern of reactivity. The discovered antibody can then be used to study the basic biochemical properties of the corresponding antigen to enable the recognition of the pattern of the antigen in the family and population, to identify RBC that lacks the antigen and to search for antithetical antigen, and the identified characteristics are compared to existing systems and collections” [1].

“The total number of recognized blood group antigens is 346, of which 308 are clustered within 36 blood group systems and the remaining 38 serologically defined antigens have not been assigned to a blood group system yet. Six of those are in the high prevalence series (901), 17 in the low prevalence series (700) and a further 15 reside in one of six collections (the 200 series)” [2].

“The HUGO Gene Nomenclature Committee (HGNC) currently recognizes 36 blood group systems, which represent approximately 300 antigens” [2]. “The difference between red cell antigens that represent the products of alleles is small usually just one monosaccharide or one amino acid and most blood group systems have a null phenotype in which the whole blood group protein is absent from the red cells or any other cells. These usually result from homozygosity for

gene deletions or inactivating mutations within the genes” [3]. “The genes of these blood group systems are autosomal, except XG and XK which are X-linked, and MIC2 which is present on both X and Y chromosomes. The antigens can be integral proteins where polymorphisms lie in the variation of amino acid sequence (Rhesus and Kell), glycoproteins or glycolipids (ABO)” [4].

“Most blood groups are encoded by one allele but variant usually arises from single nucleotide change, for example, A and B alleles differ by amino acid substitutions in their respective transferases while some allele is silent and does not produce any recognizable antigens, for example, AA and AO. Some blood group genes are complex of several closely linked genes that evolve through duplication of an ancestral gene for example; Rh system with genes RHD and RHCE, MNS system with genes GYPA, GYPB and GYPE” [1].

“Blood group antigens have been used to evaluate ethnic diversity of human populations with similar frequencies in people ranges from 0 to 90 years and had been related to predisposing individuals to some diseases like cancer, diabetes, infectious diseases, and heart illnesses or may protect individuals against some diseases such as malaria and diabetes. The ABO and Rh blood groups are the most important antigens because their incompatibility causes transfusion reaction in recipient and haemolytic disease of the foetus and newborn Furthermore, blood antigens play an important role in the success of transfusions and organ transplants” [5,6].

2. MATERIALS AND METHODS

2.1 Study Location

The selected area for this study is Sokoto State and the area covered included Usmanu

Danfodiyo University Teaching Hospital (UDUTH), Specialist Hospital Sokoto, Maryam Abacha Women and Children Hospital, Women and Children Welfare Clinic, General Hospital Yabo and General Hospital Bodinga. "Sokoto State is located in the extreme Northwest of Nigeria, near the confluence of the Sokoto River and Rima River. With an annual average temperature of 28.30c (82.9 0F). Sokoto is, on the whole, a very hot area. However, maximum day time temperatures are for most of the year generally under 40 0C (104.0 0F). The warmest months are February to April when daytime temperatures can exceed 45 0C (113.0 0F). The rainy season is from May to October during which showers are a daily occurrence. There are two major seasons, wet and dry which are distinct and are characterized by high and low malarial transmission respectively. Report from the 2007 National Population Commission indicated that the State had a population of 3.6 million" [7].

2.2 Study Setting

The study was conducted among the pregnant women that visited various hospitals in Sokoto for their first ante-natal visit. The research laboratory analysis was done in School of Medical Laboratory Science of Usmanu Danfodiyo University in collaboration with Haematology Department of Usmanu Danfodiyo University Teaching Hospital Sokoto.

2.3 Sample Collection and Methods

A structured questionnaire was used to obtain a socio-demographic information and obstetric history of each participant. Blood samples were collected by venepuncture into ethylene diamine tetracetic acid (EDTA) anticoagulated tubes and used for the determination of ABO, Rh, MSs, Kell, Kidd and Duffy blood 1250 consecutively recruited subjects. Red cell phenotyping was carried out using standard tube techniques and column agglutination technology. The test is based on haemagglutination principle.

For ABO and Rh D blood grouping, a drop of Biorad Seralone anti-A, anti-B, and anti-D (Bio Rad Medical Diagnostics, Germany) each was placed in clean test tubes labelled 1, 2, and 3. To each tube was added a drop of 5% red blood cell suspension in saline, the contents were gently mixed together and centrifuged for 30 seconds at 1000g. The cell buttons were re-suspended and observed for agglutination.

For Rh and Kell blood group, Combined Column agglutination card consisting of antisera to C, c, E, e and K was used. A drop of washed 5% red cells was added into each column containing respected antiserum. The cards were centrifuged in Column card centrifuge for 2 min at 1500g. Each card was then read by checking whether the red cell sink to the bottom of the column or was suspended on top of the gel.

For anti-M monoclonal, a drop of 3% suspension of red cells was added to a drop of anti-M lorne reagent (Lorne Laboratories, Britain) in a tube. All tubes were centrifuge for 20 sec at 1000g. The cell buttons were re-suspended and observed for agglutination. For the anti-S and anti-s, a drop of 3% suspension of red cells was added to a drop of anti-S/anti-s lorne (Lorne Laboratories, Britain) reagent in a tube, it was then mixed thoroughly and incubated at 37^oc for 15min. The red cells was then washed once with normal saline and two drops of AHG was added, it was then centrifuged for 20 secs at 1000g, gently re-suspended and observed for agglutination.

For Kidd blood group, anti-Jk^a and anti-Jk^b (Lorne Laboratories, Britain) each was placed in clean test tubes labelled Jk^a and Jk^b. To each tube was added a drop of 5% red blood cell suspension in saline, the contents were gently mixed and incubated at room temperature for 5 minutes, centrifuged for 20 seconds at 1000g. The cell buttons were re-suspended and observed for agglutination.

2.4 Eligibility Criteria

All consenting, consecutively recruited pregnant women willing to offer a written or oral informed consent to participate in the study after counselling;

2.5 Exclusion Criteria

The pregnant women who do not meet the inclusion criteria were excluded from participating in the study that is the Non- pregnant women; Non-consented pregnant women and pregnant women attending hospitals outside the Sokoto metropolis.

2.6 Data Analysis

The data collected was recorded on an Excel spreadsheet and later subjected to statistical analysis using a statistical software SPSS

version 23.0. Statistical analysis included descriptive statistics of mean and percentage.

3. RESULTS

Among the 1250 apparently healthy pregnant women studied, the sociodemographic characteristics revealed that the age range of the subject was 17-48 years where majority were within the age bracket of 21-30 years (61.3%) and majority of the subject were Hausa (90%) followed by Igbo (5.2%), Yoruba 3.4% and others (1.4%),(Table 1). Majority of the pregnant women had their first anti-natal visit during second trimester of their pregnancy (59.8%) followed by those in third trimester (33.4%) and minority in their first trimester (6.8), (Table 1). Majority (59.6%) of the pregnant women had 1 to 4 previous pregnancy while 40.4% had more than five previous pregnancy.

Table 1. Demographic characteristics of study participants (n=1250)

Variable	n (%)
Age group	
≤20	101(8.1)
21-30	766(61.3)
31-40	366(29.2)
≥41	17(1.4)
Ethnicity	
Hausa	1125(90)
Igbo	64(5.2)
Yoruba	43(3.4)
Others	18(1.4)
Gestational age	
First	85(6.8)
Second	747(59.8)
Third	418(33.4)
Gravidity	
Multigravida (>1 pregnancy)	745(59.6)
Grand multigravida (≥ 5 pregnancies)	505(40.4)

Table 2 revealed the prevalence of ABO blood group antigens among the pregnant women were 48.5%, 27.3%, 19.4% and 4.8% for O, B, A and AB respectively. The prevalence of 93.1%, 30.2%, 24.6%, 90.2% and 97.9% were positive for RhD, RhC, RhE, Rhc and Rhe respectively and the most frequently occurring Rh phenotype was Dce. The Prevalence of The Cellano (k) of Kell blood group system was 97% and 3% for negative and positive respectively. The prevalence of positive M, S and s antigens were 75.5%, 31.4% and 63.3 respectively. The prevalence of positive Fya and Fyb antigens was

1.1% and 0.5% respectively while the prevalence of positive Jka and Jkb antigens was 15.9% and 21.7% respectively.

Table 2. The frequency distribution of blood group antigens in study participant

Blood group antigen	Positive number (%)	Negative number (%)
ABO		
A	19.4	
B	27.3	
AB	4.8	
O	48.5	
Rhesus		
RhD	93.1	6.9
RhC	30.2	69.8
RhE	24.6	75.4
Rhc	90.2	9.8
Rhe	97.9	2.1
MNSs		
M	75.5	24.5
S	31.4	68.6
s	63.3	36.7
Kell	97.6	2.4
Duffy		
Fy ^a	1.1	98.9
Fy ^b	0.5	99.5
Kidd		
Jk ^a	15.9	84.1
Jk ^b	21.7	78.3

4. DISCUSSION

This study screened one thousand, two hundred and fifty (1250) pregnant subjects for alloantibody among the women attending antenatal clinic in different hospitals in Sokoto state (Usmanu Danfodiyo University Teaching Hospital, Specialist Hospital Sokoto, Women and Children Welfare Clinic, Maryam Abacha Women and Children Hospital, General Hospital Bodinga and General Hospital Yabo) at their first booking.

Table 1 shows the socio-demographic and Obstetric characteristics of study participants at their booking. The majority were within the age range of 21-30 years (61.3%) and between 41-43 years (1.2) of age were the minority showing that the majority were within the active reproductive age of 20– 40 years. This study is in agreement with the previous research [8] in Ogun State, southwest Nigeria and also research in north Nigeria [9,10,11,12].

The distribution of study subjects based on ethnicity reveals that Hausa/Fulani were

predominant with (91%), followed by Igbo (6.8%) then Yoruba and others were the least with 3.6% and 1.4% respectively. The others are Zabarmawa, Igala, Dakarawa and Ebara. These findings are in agreement with previous reports of Sokoto [9,10]. The high prevalence of the Hausa/Fulani tribe is associated with the fact the study was carried out in northern Nigeria where the majority of the settlers are Hausa/Fulani. Also, the study covered some rural areas where the major settlers are Hausa/Fulani.

The distribution of subjects based on their gestational age shows that the majority booked within the second trimester (59.7%) followed by those that booked at the third trimester (22.4%) and the least booking is at the first trimester (6.8%). The reason for high booking at second trimester may be associated with the type of research subjects that are not primigravidae which make them reluctant to book early because of their previous pregnancy experience and also the booking at third trimester is lower than the second trimester because of the most hospital in the state frown at late booking.

The distribution of subjects studied based on their gravidity shows that 59.6% of pregnant women were Multigravida (>1 to 5 pregnancies) while 40.4% were Grand multigravida (\geq 5 pregnancies). This is in agreement with the previous reports that majority of pregnant women were having a gravida between 1 and 5 pregnancies [8,11,12].

Table 2 shows the distribution of ABO, Rh, MSs, Kell, Duffy and Kidd blood group antigens of the study participant. In this study, a prevalence of 48.5%, 27.3%, 19.4% and 4.8% was observed for blood group O, B, A and AB respectively for ABO antigens while a prevalence of 93.1% was positive for RhD while 6.9% was negative for RhD. This is in agreement with the previous reports of Northern Nigeria reported that Group O was found in 46.6% followed by 25.95% of Group B, 23.05% of Group A while the frequency of 3.64% of Rhesus negative was reported [13] and in Delta reported that blood group O was most common followed by A, B and AB respectively and Rhesus positive was more common than Rhesus negative in the rhesus system [14]. Another study also reported that 94.5% of the subjects were Rh(D) positive while 5.5% were Rh(D) negative while 46.7%, 26.4%, 22.8% and 4.1% as prevalence of O, B, A and AB blood group antigens respectively and also the prevalence of RhD antigen as (92.7%) in a multi-ethnic group in Nigeria. [15,16].

The prevalence of RhC, RhE, Rhc and Rhe was observed as follows: 30.2%, 24.6%, 90.2% and 97.9% were positive for RhC, RhE, Rhc and Rhe respectively while 69.8% 75.4% 9.8% and 2.1% were negative for RhC, RhE, Rhc and Rhe respectively and the most frequently occurring Rh phenotype was Dce. This is in line with the report [16] in the multi-ethnic group in Nigeria who reported the prevalence of Rh antigens as follow: C (20.5%), c (97.7%), E (19.5%), and e (97.4%) and another study that reported prevalence of c (99.8%), followed by e (98.7%), then D (95.0%), E (20.5%), and finally C (17.7%) [17].

It is however at a variance of the previous report that reported all the subjects to be 100% Rhc antigen-positive and only 2.78% to have RhC antigen although the prevalence of RhE and Rhe antigen is similar to that of the current research finding [18].

The Prevalence of The Cellano (k) of Kell blood group system was 97% and 3% for negative and positive respectively. The finding is in agreement with the previous finding of Erhabor *et al.*, (2015) that reported that 98% of the subject were Kell negative. This is at variance with previous findings that reported 0% -21.7% prevalence of Kell antigen [16,19]. The finding may be due to the geographical area of the research and ethnicity differences of the subject.

The prevalence of M was 75.5% and 24.5% as positive and negative respectively, S was 31.4% and 68.6% as positive and negative respectively while s was 63.3% and 36.7% as positive and negative respectively. The pattern closely resemble that of report in book 'Blood Group Antigen Facts Book' of that of Africa. Although there is a slightly non-statistical difference between their report and our findings [20]. This is at variance with the findings in Ghana that reported frequency of 39% and 94% of S and s antigen respectively in sickle cell disease patient, prevalence of 87%, 59% and 83% for M, S, and s antigens and M = 89.26%, S = 61.07%, and s = 82.55% both in Saudi Arabia [21,22,23].

Also, the frequency of M+S+s+ as 22.6% in Oman and also the frequency of 20.8% of M antigen in blood donors in Kano was reported [19,24].

The prevalence of positive Fya and Fyb antigens was 1.1% and 0.5% respectively while 98.9% and 99.5% were negative for Duffy A and B respectively. The low prevalence of the study is

in agreement with the findings of Boateng and colleagues who reported the frequency of Fya and Fyb as 4.3% and 3.8% respectively among Ghanaian sickle cell disease patient; Ouchari and colleague reported that Fy (a-b+) was the most common phenotype identified in the Tunisian population (38.1%) [21,25]. This is at variance with the report of Jamoh and colleagues in kano who reported 0% frequency in blood donors because all the donors are Fya and Fyb antigens negative [19]. Although Kano and Sokoto are within the same geographical location, the variation in the findings may be due to the sampling size of the study.

The prevalence of positive Jka and Jkb antigens was 15.9% and 21.7% respectively while 84.1% and 78.3% were negative for Jka and Jkb respectively. The findings of this study are at variance with the finding of Boateng et al., (2019) Jka and Jkb antigens as 75% and 47% in Ghana who reported higher prevalence and that of Jamoh et al., (2018) in kano that reported 0% frequency in blood donors because all the donors are Jka and Jkb antigens negative. The reason for zero frequency may also be associated with the same reason of that of Duffy antigen. It is also at variance with the findings of Osaro et al., (2015) in Sokoto that shows the prevalence Jka, Jkb and Jk(a+b+) with 8 (4.9%), 13 (8.0%) and 0 (0.0%) respectively [10,9,21].

5. CONCLUSION

The finding of this study in Sokoto state highlight the frequency of blood group antigens among the subject as previously recorded which the variation of various antigens in individual blood group systems may contribute to the development of alloantibody.

BENEFITS OF THIS STUDY

The determination of the prevalence of clinically significant red cell antigens is important to justify the need for a transfusion laboratory to stock optimum numbers of antigen-negative red cells for transfusion to antigen-negative women who may require red cell transfusion in a bid to prevent them from being sensitized and produce antibodies that can cause haemolytic disease of the foetus and newborn and also to reduce the risk of transfusion reaction.

FUTURE PLANS

Large-scale studies on blood donors and pregnant women need to be done in order to

collect sufficient evidence to formulate guidelines for premarital testing and donor screening to prevent haemolytic disease of the foetus and newborn as well as transfusion reaction.

CONSENT AND ETHICAL APPROVAL

Written and oral informed consent was obtained from all participants using a standard protocol while the ethical clearance was obtained from the Ethical Committee of Ministry of Health, Sokoto as well as the study site in accordance with Helsinki declaration (No: SMH/1580/V.IV on 08/10/2019; NHREC/30/012/2019; SHS/SUB/133/VOL 1).

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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