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## RESEARCH ARTICLE

### DISTRIBUTION OF ABO, RHESUS BLOOD GROUPS AND HAEMOGLOBIN GENOTYPE IN ENUGU NORTH AND ENUGU SOUTH LOCAL GOVERNMENT AREA, ENUGU STATE, NIGERIA

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#### ABSTRACT

**Background:** ABO, Rhesus blood groups and Haemoglobin genotype are inherited characters. These may vary from one population to another. **Aim:** This study is to update information on the distribution of ABO, Rh blood group and haemoglobin genotype in Enugu North and Enugu South local Government Area, Enugu State. **Methods:** Eight hundred participants from each of the zones in the two local government areas were selected randomly. Two milliliters of blood collected from the cubital fossa of the subjects into an EDTA bottle was used for the determination of ABO, Rhesus blood group and Hb genotype using the standard tile technique and alkaline cellulose acetate electrophoresis technique. Data obtained were analysed using SPSS version 20. **Results:** The result showed that the pattern of distribution of ABO in each of the local government areas was in the order O>A>B>AB. In Enugu North, the pattern of distribution of ABO blood group were observed as O (57.5%), A (23.0%), B (17.0%) and AB (2.5%) respectively. The percentage frequency of Rh positive and Rh negative was 90.8% and 9.1% respectively. The percentage frequency of HbAA, HbAS and HbSS was 68.5%, 30.0% and 1.5% respectively. In Enugu South, the pattern of distribution of ABO groups were observed as O (58.2%), A (21.8%), B (14.5%) and AB (5.5%). The Rh positive and Rh negative distribution in the studied population was 93.8% and 6.2% respectively. The haemoglobin genotype, 73% were HbAA, 26% HbAS, 1% HbSS were obtained. **Conclusion:** Blood group O and Rh positive blood groups were the most common blood group obtained in each of the two local government areas and blood group AB and Rh negative blood groups was the least common in this study. The percentage frequency of HbSS obtained in this study was low and this indicates a low prevalence of Sickle cell disease in this area.

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## INTRODUCTION

Whole blood is composed of blood cells suspended in plasma. The Red blood cells constitute about 45% of the whole blood, their membrane are marked by glycoproteins that define the different blood group antigens. Thirty-five human blood group systems with over 600 different blood group antigens have been recognized by The International Society of Blood Transfusion (ISBT) as at November 2014 (ISBT, 2014). The ABO is clinically the most important amongst the other blood groups discovered (Ukaejiofor, 2009). Karl Landsteiner first discovered the ABO blood group in 1900 and it as served the beginning of blood banking and transfusion medicine (Landsteiner and Weiner, 1940).

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Individuals are divided into four major blood groups- A, B, AB and O according to the presence or absence of the antigen (Dacie and Lewis, 2012). The prevalence of blood type is determined by the frequency of inheritance of the 3 alleles of the ABO gene which is determined by social tendencies like marriage and migration (Ali et al., 2005). The Rhesus (Rh) system, discovered in 1940 by Landsteiner and Weiner is the second most important blood group system due to its immunogenicity in RhD negative individuals during pregnancy and blood transfusion (Kaur, 2015). Individuals are positive (Rh+) if they have Rh D antigen on their erythrocytes and are negative (Rh-) if they do not have this RhD antigen (Adeyemo and Soboyejo, 2015). Rh incompatibility can pose a major problem in some pregnancies involving a Rh-negative mother with a Rh-positive foetus (Advent, 1999), this could lead Haemolytic Disease of the Newborn (HDN).

The higher the proportions of RhD negative individuals in a population, the more likely the incidence of Haemolytic Disease of the Newborn in that population. The genetic defects of haemoglobin are the most common genetic disorders worldwide (Hoffbrand and Moss, 2011). Haemoglobin is the oxygen carrying pigment of the erythrocytes and defects in its genes can produce abnormal haemoglobin which leads to conditions known as haemoglobinopathies (Hoffbrand and Moss, 2011). The World Health Organization (WHO) figures estimates that 6% of the world's population is a carrier for haemoglobin disorders (WHO, 2006). Haemoglobin genotypes are inherited characters determined by different combinations of the four polypeptide globin chains in a haemoglobin molecule, the normal haemoglobin-HbA and abnormal HbS. The abnormal haemoglobin occur as a result of a mutations in the B-chain gene, this causes a change in the properties of haemoglobin which results in the sickling of erythrocytes. Haemoglobin S disorders can exist in heterozygous-the sickle cell trait (HbAS) or the homozygous state- HbSS or sickle cell anemia (HbSS), and the compound heterozygous state for HbS together with other haemoglobin (HbSC, HbSD, HbSE) (Patel et al., 2009). These variants cause moderate to severe haemolytic anaemia leading to high degree of morbidity and mortality (Akhigbe, 2009) (Patel et al., 2009).

Many studies have been published on ABO, Rh blood types and haemoglobin genotype link with increase or decrease susceptibility to a particular disease (Sharara, 2006) (Shimazu et al., 2000). Blood group A individuals are at greater risk for carcinoma of the stomach, pernicious anaemia, coronary thrombosis in men and venous thrombo-embolism in women (Albert and Khan, 2012). People of blood group O are at higher risk of contracting malaria, duodenal ulcer and some infectious diseases, such as cholera (Albert and Khan, 2012). Studies have also shown the relationship between haemoglobin variants and the malaria parasite, Plasmodium falciparum (Edozien et al., 1960). The sickle cell heterozygote (HbAS) has resistance to malaria thus in areas where malaria is common, the prevalent genotype appears to be the sickle cell heterozygote (HbAS). The ABO blood groups, Rh blood groups and Haemoglobin Genotype are inherited characters and are known to vary across various ethnic populations in different geographical areas. This could be due to environmental factors. The knowledge of ABO and Rh blood groups are useful in blood transfusion services as it is essential for effective management of blood banks, population genetic studies, researching population migration patterns as well as resolving certain medico-legal issue. In modern medicine besides their importance in evolution, their relationship to disease and environment has been increasingly important. Nigeria ranks first as the sickle cell endemic country in the world with an annual infant death of 100,000- representing 8% of infant mortality in the country and also it is reported that an average of 150,000 infants are born with sickle cell disease (WHO, 2006).

Enugu North and Enugu South local government areas in capital city of Enugu State, Nigeria was the capital of the defunct eastern region of the country. The inhabitants are mainly non-indigenes that have migrated from all parts of Nigeria. Information on the distribution of these blood groups- ABO and Rh blood group in this area is necessary for effective blood transfusion service. Also haemoglobin genotype determination can serve as a basis for genetic counseling to help in making crucial decisions like marriage, which can

contribute to ultimately curb the occurrence of haemoglobinopathies especially sickle cell disease.

## MATERIALS AND METHODS

This is a cross sectional study. Eight hundred participants from the eight districts in the two local government participated in the research. This comprises of one hundred participants from each zone. Ethical clearance approval was given by the Enugu State University of Science and Technology Teaching Hospital Ethics Committee prior to this research. Two milliliters of blood was collected in sodium EDTA container. ABO and Rh blood typing was done using the ABO grouping and Rhesus Typing Antisera (www.biotech.com 2017). The procedure was in accordance with the manufacturer's guidelines test for both ABO and Rh-D grouping. A drop of anti-A, anti-B, anti-AB and anti-D was placed on a labeled tile and a drop of blood sample was added to it and mixed. The presence agglutination after mixing indicates a positive result (www.biotech.com 2017). Haemoglobin genotype was done using cellulose acetate electrophoresis at PH 8.6 using Helena Biosciences equipment (Model SAS MX Chamber, Cat No. 4063). A small quantity of haemolysate prepared by adding three drops of water to one drop of venous blood in a tube and mixed, was placed on the cellulose acetate paper with the aid of an applicator. The cellulose acetate paper was placed in the electrophoresis chamber and connected to a power supply of e.m.f 230V for 20 minutes. Haemolysates from blood samples of known genotypes (AA, AS and SS) were run as control.

### Statistical analysis

The data was analyzed using computer database software from the Statistical Package for Social Sciences (SPSS version 20; SPSS Inc., Chicago) to generate frequency distribution and percentage prevalence scores of the various parameters.

## RESULTS

A total of 800 subjects, comprising of 400 participants from each of the zones (Amigbo, Ihenwuzi, Onueto, and Umunevo) in Enugu North and 400 donors from Enugu south (Ugwuaji, Obeagu, Akwuke and Amechi) Local Government Areas, were selected randomly in order to determine the distribution of ABO, Rh blood group and Hb genotype in the area.

The results obtained in Enugu North local Government Area were as follows

Table 1 shows that blood group O has the highest frequency of 230 (57.5%) blood group AB occurs with the least frequency of 27(2.5%). Table 2 shows that RhD+ has the most frequency among all Rh blood group with a frequency of 375(90.8%) whereas the percentage frequency of RhD – is 9.3% Table 3 shows that the frequency of haemoglobin genotype- HbAA is 274 (68.5%), HbAS is 120 (30.5%) and HbSS is 6(1.5%). The results observed in Enugu South Local government area shows a similar pattern. Table 4 shows that the highest value was blood group O which is 233(58.2%) followed by blood group A which is 87(21.8%) Table 5 shows that Rh+ is more frequent among all Rh blood group with a frequency of 363(93.8%) whereas the percentage frequency of Rh – is low (6.2%) Table 6 shows that the frequencies of haemoglobin genotype- HbAA is 292(73%), HbAS is 104(26%) and HbSS is 4(1.0%).

**Table 1. Frequency Distribution of ABO blood group in Enugu North Local Government Area**

Blood group	Number	Percentage frequency
A	92	23.0
AB	10	2.5
B	68	17.0
O	230	57.5
TOTAL	400	100

**Table 2. Frequency Distribution of Rhesus D blood group in Enugu North Local Government Area**

Rh blood group	Number	Percentage frequency
Rh+	363	90.8
Rh-	37	9.2
TOTAL	400	100

**Table 3. Frequency Distribution of Haemoglobin genotype in Enugu North Local Government Area**

Hb genotype	Number	Percentage Frequency
AA	274	68.5
AS	120	30.0
SS	6	1.5
TOTAL	400	100

**Table 4. Frequency Distribution of ABO blood group in Enugu South Local Government Area**

Blood group	Number	Percentage Frequency
A	87	21.8
Ab	22	5.5
B	58	14.5
O	233	58.2
Total	400	100

**Table 5. Frequency Distribution of Rhesus,D blood group in Enugu South Local Government Area**

Rh blood group	Number	Percentage Frequency
Rh+	375	93.8
Rh-	25	6.2
Total	400	100

**Table 6. Frequency Distribution of Haemoglobin genotype in Enugu South Local Government Area**

Hb Genotype	Number	Percentage Frequency
AA	292	73
AS	104	26
SS	4	1
TOTAL	400	100

## DISCUSSION

The clinical importance of ABO and Rhesus blood groups in the transfusion of blood/blood products, organ transplant and their relation to certain disease conditions has made the pursuit of these blood groups in different populations an important and very significant area of investigation over the years. They are also useful in forensics and for resolving some medico-legal issues. It is therefore very crucial to have accurate information on the current distribution of these blood groups as they may vary from one population to another. From this study, In each of the two local government areas, the pattern of distribution of ABO was in the order O>A>B>AB. The same trend of prevalence of blood groups (O>A>B>AB) was observed and

reported by some other studies in Nigeria. For example, Adeyemo and Soboyejo reported percentage frequency of 55.3%, 25.3%, 16.75 and 2.7% for blood group O, A, B and AB respectively among 150 students selected randomly in University of Lagos (Adeyemo and Soboyejo, 2006). Bakare and his colleagues also reported percentage frequency of 50%, 22.9%, 21.3% and 5.9% for O, A, B and AB respectively among 7653 individuals in Ogbomoso (Bakare, 2006) Another study done at Ibadan by Omotade and his colleagues reported percentage frequencies of 54.2%, 21.6%, 21.4% and 2.8% for O, A, B and AB respectively (Omotade *et al.*, 1999). These studies are in concert with the fact that Nigerian populations are characterised by high frequencies of the O allele and an average of about 14% each of the A and B genes (Ukaejiofor, 2009). This could be due to some environmental factors. It has been discovered that some bacterial enzymes can convert red blood cells of types A, B, and AB into O by stripping away their identifying surface antigens (Sharara, 2006). Deviations from the findings in this study and other findings as regards the trend of prevalence of these blood groups were observed. For instance in a study carried out in Adamawa, it was observed that blood groups O>B>A>AB with a percentage frequency of 50.6%>21.3%>16.5%>11.7% (Omotade *et al.*, 1999). The high prevalence of group O individuals in this study is of great advantage because of their status as 'universal donors' as this implies availability of blood in cases of emergency. However, caution should be taken in doing this as some group O blood is known to have potent immune haemolytic antibodies (haemolysins) (Jeremiah, 2006). In addition, a study conducted by Kaur on the relationship of blood groups to anaemia reported that blood group O individuals have resistance to anaemia (Kaur, 2015). Some research have also shown that individuals with blood group O had the smallest percentage of severe malaria when compared with other blood groups such as A, B and AB (Edozien *et al.*, 1960). The reason for less severe malaria attack seen in blood group O individuals may be due to mechanism of reduced rosettes formation by parasitized RBCs of blood group subjects. It is also important to bear in mind that there could be high incidence of cholera and duodenal ulcer in this area because blood group O individuals have a higher risk of contracting these diseases (Albert and Khan, 2012).

The Rhesus blood group distribution may also vary from one human population to another. Rhesus positive and Rhesus, D negative blood groups were obtained in Enugu North and Enugu South Local Government Area In this study, Rhesus, D positive was most prevalent in each of the two local government areas while Rhesus negative was least prevalent. This is in agreement with what is expected from the Hardy Weinberg equilibrium. A similar pattern of the distribution was also found in an Iranian study in which the Rhesus positive and Rhesus negative frequencies were 90.45% (2001)/91.63% (1982) and 9.54% (2001)/8.37% 1982 respectively (Pourfath, 2004). To compare, the result obtained from this study is in contrast to that of Salmon and his colleagues and Njoku and his colleagues who reported Rhesus positive values of 100% for Eastern Highlands of Papua Guinea and Nigeria respectively (Salmon *et al.*, 1988) (Njoku *et al.*, 1996). Also, the percentage frequency (90.8%) of Rhesus positive obtained in this study was observed to be lower than some previous studies in Nigeria and across the world. 96.7% positive rate was recorded in Igbos by Ukaejiofor in 2009 (Ukaejiofor, 2009). The percentage frequency of Rhesus negative obtained in this study is observed to be low. The percentage frequency

of Rhesus negative is higher for instance amongst the British, Rhesus negative blood group represents 17% of the population and in Caucasians, it represents 15% ((Giri et al 2011). The low prevalence of Rhesus negative blood group in this study is an advantage because the incidence of Haemolytic Disease of the Newborn in this area would be equally low. Haemoglobin genotypes are inherited blood characters. The inherited disorders of haemoglobin are the most common gene disorders with 7% of the world population being carriers (Weatherall, 2001). The observed high incidence of HbAA and HbAS in this study is in agreement with previous reports that the normal haemoglobin (HbAA), range from 55% to 75% (Nwafor and Banigo, 2001) and the sickle cell trait (HbAS), 20% to 30% in Nigeria (Reid and Famod, 1998). Jeremiah also reported a frequency of 80.2% for HbA and 19.68% for HbAS in the Niger Delta area (Jeremiah, 2006). Esan and his colleagues reported 72.8% for HbAA and 23.5% for HbAS in Ekiti State. Akhigbe and co-workers also reported a prevalence of 71.03% for HbAA, 22.19% for HbAS, in Ogbomoso South West Nigeria (Ukajeiofor, 2009). Comparatively, the proportion of HbAS found in this study (30.0%) is slightly higher than the 29.4% observed in the Southern part of Nigeria among students in the Niger Delta (Reid and Famod, 1998).

The observed frequency of HbAS is consistent with the 20 – 30% frequency of HbAS observed by other researchers in other parts of Nigeria (Advent, 1999). This finding is also consistent with frequency of 20% – 40% observed in previous report in other parts of Africa (Reid and Famod 1998) (Erhabor et al., 2006). The frequency of HbAS was reported as follows; 8% – 16% for Black Americans, 8% – 10% for white Americans, 6% – 15% for Europeans, 7% – 8% for Middle Easterns (Sinou, 2003). HbAS has been thought to offer some protective role against Plasmodium falciparum malaria (Edozien et al., 1960) Akhigbe and his co-workers in their work attributed this to the substitution of hydrophilic amino acid glutamate with hydrophobic amino acid valine (as in HbSS) or lysine (as in HbC) causing increasing binding affinity between haemoglobin molecules with polymerization of haemoglobin deforming red blood cells which is rapidly cleared from the circulation (Akhigbe, 2009). The percentage frequency of sickle cell disease (HbSS) in these areas was observed to be low (1.5% in Enugu North and 1% in Enugu South) from this study. In a previous report, the geographical distribution of Sickle cell anaemia (HbSS) was given as 3-9% for USA black Americans, 1-8% for US whites, 3-7% for Europe (UK, Pakistan-blacks), 2-8% for other European countries (Mediterranean), 1-3% for Caribbean, 1-3% for Middle East, 1-10% for Africa (Erhabor et al., 2006). The low percentage distribution of HbSS in this study can be due to the adaptation of carrier testing programs as well as premarital counseling/testing for prospective couples prior to marriage in the area.

## Conclusion

This is a comprehensive study on the distribution of ABO, Rhesus blood group and Haemoglobin genotype in Enugu North and Enugu South Local Government Area. The result obtained reveals that the frequency of ABO and Rhesus blood group in this area is stable and consistent with other reports from previous studies in other tribes in Nigeria. Blood group O and Rhesus positive blood were the most prevalent while blood group AB and Rh negative blood groups remains the least prevalent. This also means there is a large pool of universal

blood donors in this population. The frequency of HbAA and HbAS obtained in each of the local government areas in this local government was in tandem with previous studies in Nigeria. The prevalence of sickle cell disease obtained in this study was low. The knowledge of ABO, Rh blood groups and haemoglobin genotype are useful in planning efficient and safe blood transfusion services, for medical diagnosis, genetic information, genetic counselling and also assessment of the general wellbeing of individuals.

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