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Rhesus E, Rhesus D and ABO Blood Groups Distribution among Indigenes of Ogoni Ethnic Group of Rivers State, Nigeria

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Authors' contributions

This work was carried out in collaboration among all authors. Authors SGC designed the study, carried out the analysis, wrote the first draft of the manuscript and performed the statistical analysis. Authors EME, ACUE and FIB approved the design of the study, supervised, managed the analyses of the study and the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Aim: The aim of the study was to determine the percentage distribution of Rhesus E, Rhesus D and ABO blood groups among indigenes of Ogoni ethnic group of Rivers State, Nigeria.
Study Design: This was a cross-sectional study carried out among indigenes of Ogoni whose origin of their first generation parents is Ogoni. A total of 101 subjects (49 females and 52 males), within the ages of 30–60 years were recruited for the study and they were apparently healthy.

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Place and Duration of Study: Ogoniland is located in an area along the Niger Delta Eastern edge, and to the north-east of the Imo River and Port Harcourt city. Ogoniland covers about 1036 Sq Km and borders the Bay of Guinea. All participants were recruited in Bori. Bori is the traditional headquarter of Ogoni. Bori is located on latitude: $4^0$40'34.64" N and longitude: $7^0$21'54.68"E. Laboratory analysis was carried out at the Post Graduate Laboratory of Rivers State University, Nkpolu-Oroworukwo, Port Harcourt, Rivers State, Nigeria. Port Harcourt, the capital of Rivers State, is located on latitude 4.75^0N and longitude 7.00^0E and lies along Bonny River in the Niger Delta. All subjects were recruited on the same day and their blood samples collected on 2nd October, 2019, and analysis performed on 3rd October, 2019.

Methodology: Determination of ABO and Rhesus D blood groups was done using Anti-A, Anti-B, and Anti-D IgD/IgM Blend Reagents, manufactured by Atlas Medicals, Cambridge-UK, Lot No[s] 19031214; 19030910; 19031921; Expiry Dates: 2021/03/16; 2021/03/10; 2021/03/20 and phenotyping of red cells was carried out manually using standard tube techniques as described by Judd, and Brecher. Determination of Rhesus E blood group was done using Anti-E Monoclonal, manufactured by Lorne Laboratories Ltd, UK. Lot No: 69189-A4; Expiry Date: 2021/03/04. Phenotyping of red cells was done using tube method as described by Lorne Laboratory Ltd.

Results: The result revealed percentage distribution of 22.77% for blood group A; 20.79% for blood group B; 52.47% for blood group O; 3.96% for blood group AB; 92.07% for Rhesus D positive blood group; and 25.74% for Rhesus E positive blood group amongst the Ogonis. The study revealed that ABO blood group percentage distribution among the Ogonis was in the order of O > A > B > AB (blood group O being the predominant ABO blood group followed by blood group A, blood group B, and blood group AB.

Conclusion: The study has revealed the presence of Rhesus E antigen in the Ogoni populace. It is therefore necessary to take into cognizance the fact that the presence of Rhesus E antigen may likely be the cause of some transfusion reactions which cannot be explained after a compatible ABO and Rhesus D cross match. We therefore recommend that routine typing of Rhesus E blood group be done on blood donors and recipients before blood transfusion.

Keywords: Rhesus E; Rhesus D; ABO blood group; Ogoni-indigenes; Rivers State; Nigeria.

1. INTRODUCTION

The ABO blood group system has an International Society of Blood Transfusion (ISBT) symbol/number which is ABO [001]. In the year 1900, Karl Landsteiner carried out an interesting work by mixing sera and red blood cells from his colleagues and he observed the presence of agglutination. Based on the agglutination pattern, he called the two first blood groups “A” and “B”. Red blood cells that were not agglutinated by any of the sera were initially called “C”, but was later called “ohne A” and “ohne B” (ohne is “without” in German), and later it was finally called “O” [1]. In the year 1907, Jansky suggested using the Roman numerals I, II, III, IV for O, A, B, and AB respectively, while in 1910, Moss suggested using I, II, III, and IV for AB, A, B, and O, respectively [1]. These numerical identifications were used in Europe and America respectively until in 1927, when Landsteiner suggested that in order not to be confused, the symbols A, B, O, and AB should be used globally [1]. When ISBT first described the nomenclature in 1982, five ABO antigens were mentioned, but now, ABO5 is obsolete after H antigen was moved to form the H blood group system in the year 1990 [1].

Among all the blood group system, the ABO blood group system is the most important amongst them all in transfusion practice. The ABO blood group system is the only blood group system in which individuals do have antibodies in their serum against antigens that are not found on their red blood cells [2]. Based on the presence of antigens and antibodies, the ABO blood groups consist of the following: (i) Blood group A, which have antigen A and antibody B, (ii) Blood group B, which have antigen B and antibody A, (iii) Blood group O, which have no antigens and have antibody A and antibody B, (iv) Blood group AB, which have antigen A and antigen B with no antibody [3]. The expression of ABO antigens in cord blood is approximately one-third of adult expression. The ABO antibodies are mostly of IgM, though there are some that are IgG. They react mostly at room temperature and at 37ºC and they activate complements except H and A1 activation of complement that is rare [4].

The ABO gene is located on chromosome 9q34.1–q34.2. Its organization is such that it has seven exons distributed over 19.5 kilobase pair (kbp) of genomic DNA (gDNA). Its gene products are 3-α-N-acetylgalactosaminyltransferase for A, and 3-α-galactosyltransferase for B. [1]. The ABO blood group system have genes that do not code for the antigens directly, which are carbohydrate in their nature, but genes for the production of glycosyltransferases. The glycosyltransferase is the enzyme that facilitate the transfer of carbohydrate molecules (sugar) onto different carbohydrate precursor molecules [5,6].

The ABO blood group system have A, B, and H genes. These genes have their respective glycosyltransferase and immune-dominant sugar. H gene have its glycosyltransferase as L-fucosyl-transferase while B gene have D-galactosyl-transferase, and A, have N-acetylgalactosaminyl-transferase. For their immune-dominant sugars, H gene have L-fucose, A gene have N-acetylgalactosamine, while B gene have D-galactose [6].

The inheritance of genes of the ABO blood group system are in accordance to Mendelian laws of genetics, and they are inherited in a codominant pattern based on the fact that both A and B alleles are fully expressed when they are present. The O gene is a silent or an amorphic gene because it appears to have a non-detectable gene product. There is no specific transferase that has been associated with the presence of the O gene [6].

The frequency distribution of ABO groups varies across ethnic populations. Asians have higher frequency occurrence of blood group B than the White-Europeans [7]. Among Africans, the distribution of ABO blood groups is B: 20%; A: 27%; AB: 4% and O: 49% [5]. Quinley and colleagues [6] reported percentage ABO blood group distribution in global population in percentage as follows: White Americans [A:42; B:10; O:44; AB:4]; African Americans [A:27; B:20; O:49; AB:4]; Native Americans [A:16; B:24; O:79; AB:<1]; Asians [A:28; B:26; O:41; AB:5]. Christian and colleagues, together with Okoroiwu and associates reported occurrence of ABO blood group in some ethnic groups of Nigeria, in the order of O>A>B>AB [8,9,10].

The Rhesus blood group system has been given the ISBT symbol and number of RH [004]. The Rhesus blood group antigen is located on chromosome 1p36.11. Its associated antigens include: D, C, E, c, e, f, Cw, Cx, V, G, hrs, Dw, Rh29, Goa, hrB, Rh32, Rh33, and 30 more (Rhnull). Its ISBT names are: RHD, RHCE (RH). The function of their red blood cell component is for the transport of carbon dioxide. Their disease association has been linked with haemolytic anaemia, hereditary stomatocytosis and haematological malignancies [11,12,13]. The Rhesus blood group have its CD numbers as CD240D (RhD); and CD240CE (RhCcEe), and its expression on tissues is erythroid specific [1].

The Rhesus blood group system is composed of forty-eight different antigens, with antigen D being the most important clinically immunogenic antigen. For clinical purposes, when testing for the Rhesus D antigen, the classification is by D + (Rhesus-positive) or D – (Rhesus-negative). Amongst the Whites, Rhesus D + in terms of frequency of occurrence is 85% while amongst the Blacks, it is 92% [4]. Most Rhesus D negative individuals will automatically produce anti-D when they are transfused with blood that is Rhesus D positive. The anti-D will then cause haemolysis of the recipient's red cells; and if it is haemolytic disease of the new born, it is as a result of a raised anti-D antibody in the mother, due to an initial transfusion of Rhesus positive blood to the negative mother or sensitization of the mother as a result of a previous pregnancy with a Rhesus positive baby [4].

The Rhesus antigens: C, c, E, and e are not so immunogenic. They only become important in patient care upon the development of the corresponding antibody. Amongst the Whites, Rhesus E + in terms of frequency of occurrence is 30% while amongst the Blacks, it is 21% [4]. Rhesus D and E antibodies are mainly IgG in nature, although there are some of them that are IgM in nature, and they mostly react at 37ºC with anti-human globulin. Based on its biochemistry, the Rhesus antigen is a multipass, non-glycosylated protein with a molecular weight of 30 to 32 kDa, and 417 amino acid residues [4]. The Rhesus antigen is possibly a cation transporter and has some roles to play in maintaining red blood cell membrane integrity. None of the Rhesus antibodies activates complement [4].

The Rhesus D antigen is a polypeptide that is produced by RHD gene, while the RHCE gene is the gene that produces a polypeptide with the C/c and E/e Rhesus antigens; the rest of the Rhesus antigens are produced as a result of

partial deletion, mutation, recombination, or polymorphisms of one or both genes [14]. Rhesus D positive individuals have RHD gene, while Rhesus D negative individuals do not have the RHD gene [15].

"The Ogonis are a minority ethnic people living in the Western Niger Delta Region of Southern Nigeria. During the 1970s, Ogoniland, or the Ogoni Nation, became part of Rivers State of Nigeria. There are approximately 500,000 Ogoni who represent less than 0.05 percent of Nigeria's 100 to 120 million people. The population density of this region equals 1,233 people per square mile, making it one of the most densely populated areas of Nigeria" [16]. "Archaeological and oral historical evidence suggests that the Ogonis have inhabited the area for over 500 years. The Ogoni people are organized into traditional political systems referred to as kingdoms. There are six kingdoms that are divided into three separate yet united divisions. First, the Khana division is situated in the eastern as well as the northern-most portions of Ogoniland. It consists of four separate kingdoms-Babbe, Ken Khana, Nyo Khana, and Tai. Each kingdom speaks a dialect of the language Khana and maintains separate territories. Second, the Gokana division and kingdom lies in the south-central part of Ogoniland where the people speak Gokana, a language similar to, but not identical to, Khana. Third, the Eleme division and kingdom is found in Western Ogoniland. Although the Eleme language is closely related to both Khana and Gokana, it is distinctly different" [16]. The Ogoni ethnic group is only found in Rivers State, Nigeria.

2. MATERIALS AND METHODS

2.1 Study Design

This was a cross-sectional study carried out among indigenes of Ogoni whose origin of their first generation parents is Ogoni.

2.2 Study Area

Ogoniland is located in an area along the Niger Delta Eastern edge, and to the north-east of the Imo River and Port Harcourt city. Ogoni land covers about 1036 Sq Km and borders the Bay of Guinea. All participants were recruited in Bori. Bori is the traditional headquarter of Ogoni. Bori is located on latitude: $4^0$40'34.64"N and longitude: $7^0$21'54.68"E. The analysis was carried out at the Post Graduate Laboratory of Rivers State University, Nkpolu-Oroworukwo, Port Harcourt, Rivers State, Nigeria. Port Harcourt, the capital of Rivers State, is located on latitude 4.75^0N and longitude 7.00^0E and lies along Bonny River in the Niger Delta [17].

2.3 Study Population

A total of 101 subjects (49 females and 52 males), within the ages of 30–60 years were recruited for the study and they were apparently healthy.

2.4 Collection of Blood Samples, Storage and Transportation

After pre-test counselling and explanations, venous blood collection was drawn from the antecubital fossa of the subject with the use of vacutainer as described by Cheesebrough [18]. Three (3.0) mL of venous blood was collected into a glass vacutainer sample bottle that contains 0.5 mL of 1.2 mg/mL of dipotassium ethylene diamine tetra-acetic acid (EDTA), it was well mixed for the serological identification of ABO, Rhesus D and Rhesus E blood group antigens. Blood samples collected in dipotassium ethylene diamine tetra-acetic were analyzed within 24 hours of collection. Collected samples were all transported under cold chain (ice packs/crushed ice in air tight and sealed thermo-container), from Bori (site of collection) to Port Harcourt (where analysis was carried out).

2.5 Methodology

2.5.1 Determination of ABO and Rhesus D blood groups using Anti-A, Anti-B, and Anti-D IgD/IgM blend reagents, Atlas Medicals, Cambridge-UK, Lot No[s] 19031214; 19030910; 19031921; Expiry Dates: 2021/03/16; 2021/03/10; 2021/03/20

Phenotyping of red cells was carried out manually using standard tube techniques as described by Judd [19], and Brecher, [20]. For ABO and Rhesus D blood phenotyping, a drop of anti-A, anti-B, anti-AB and anti-D, was placed in scrupulously cleaned test tubes labelled 1, 2, 3, 4. To each of the tube, a drop of 5% red blood cell suspension made in saline was added. The contents were gently mixed and then centrifuged for about 30 seconds at 1000 g. The cell buttons were re-suspended and then observed for the presence of agglutination. Agglutination of the tested red cells constituted a positive result while a smooth cell suspension after resuspension

followed by microscopic confirmation constituted negative test results for ABO blood group. A smooth cell suspension of the fourth tube after re-suspension followed by microscopic confirmation using bovine albumin and antihuman globulin at each stage for anti-D, constituted negative test results.

2.5.2 Determination of Rhesus E blood group using Anti-E Monoclonal, Lorne Laboratories Ltd, UK. Lot No: 69189-A4; Expiry Date: 2021/03/04

Phenotyping of red cells was done using tube method as described by Lorne Laboratory [21]. Three percent (3%) red cell suspension was prepared using isotonic saline. One volume of Lorne Anti-Rhesus E reagent was added to one volume of the prepared 3% red cell suspension and properly mixed and centrifuged for 20 seconds at 1000 g. The red cell button was gently re-suspended and read macroscopically for the presence of agglutination. Tubes that indicated a negative result were incubated for 15 minutes at room temperature, re-centrifuged again and then observed macroscopically for agglutination. Presence of agglutination indicated a positive result, while absence of agglutination, indicated a negative result.

2.6 Statistical Analysis

Data collected was statistically analyzed by simple percentage calculation.

3. RESULTS

3.1 Demographic Details of Study Population

A total of 101 subjects (49 females and 52 males), within the ages of 30 – 60 years were recruited for the study. Details are shown in Table 1.

Table 1. Demographic characteristics of study population

Parameters	Frequency	Percentage (%)
Total number of subjects	101	100
Total number of Males	52	51.5
Total number of Females	49	48.5

3.2 Percentage Distributions of ABO, Rhesus D and Rhesus E Blood Group in the Study Population

The percentage distributions of ABO, Rhesus D and Rhesus E blood groups were analysed and recorded. Details are shown in Table 2.

4. DISCUSSION

The percentage distribution of ABO blood groups amongst the Ogonis from this study were 22.77% for blood group A, 20.79% for blood group B, 3.96% for blood group AB, and 52.47% for blood group O, in the order of O > A > B > AB (blood group O being the predominant ABO blood group amongst the Ogonis, followed by blood group A, then blood group B, and blood group AB which is the least). Comparison of the findings on ABO blood group distribution with other tribes in Rivers State and other tribes in Nigeria and the Blacks reveals some similarity. This finding is similar with the finding of Christian and colleagues [8] which was on Briggs compound of another tribe (Kalabari) in Rivers State; and also with the finding of Christian and colleagues [9] which was a study carried out in Igwuruta-Ali (an Ikwerre tribe) in Rivers State; and that of Okoroiwu and colleagues [9], amongst some university students in an Eastern State of Nigeria (Imo State), and the (O > A > B > AB) order of ABO blood group distribution in all these studies were in similarity. Fang and colleagues [5], and Quinley [6], also reported similar findings amongst the Blacks in the same order of blood group distribution in ABO system.

On Rhesus D blood group percentage distribution, in this study, it was 93% in the total population. This finding is similar to that of Reid and colleagues [4], where they reported a percentage distribution among Blacks to be 92%. The study finding is also similar to that of Christian and colleagues [8] and that of Okoroiwu and colleagues [10], where they also reported that Rhesus D antigen predominates amongst the Kalabari ethnic tribe and in the Eastern state of Imo, Nigeria. Rhesus D antigen has been predominantly linked with haemolytic disease of the new-born and haemolytic transfusion reaction.

The Rhesus E blood group system have antigen that is implicated in causing both haemolytic transfusion reaction and haemolytic disease of the new-born although in a less severe form

Table 2. Percentage distributions of Rhesus E, Rhesus D and ABO blood groups in the study population

Blood Groups	Total Population N (% Distribution)	Frequency Occurrence	Percentage Distribution (%) based on Gender
A	23 (22.77)	M = 11	M = 10.98
		F = 12	F = 11.88
B	21 (20.79)	M = 10	M = 9.90
		F = 11	F = 10.89
AB	4 (3.96)	M = 4	M = 3.96
		F = 0	F = 0
O	53 (52.47)	M = 31	M = 30.69
		F = 22	F = 21.78
Rhesus D	93 (92.07)	M = 47	M = 46.53
		F = 46	F = 45.54
Rhesus E	26 (25.74)	M = 15	M = 14.85
		F = 11	F = 10.89

Key: M = Males; F = Females

when compared to Rhesus D. Rhesus E blood group distribution amongst the Ogonis from the study, revealed a percentage distribution of 25.74%. The finding in this study is higher than the percentage distribution of 21% reported by Reid and colleagues [4] and 21.74% reported by Kahar and Patel [22], amongst Indians.

5. CONCLUSION

The study revealed that ABO blood group percentage distribution amongst the Ogonis is in the order of O > A > B > AB. The study also revealed positivity for Rhesus D blood group percentage distribution as 92.07% in the total population; and positivity for Rhesus E blood group amongst the Ogonis revealed a percentage distribution of 25.74%. The study has revealed the presence of Rhesus E antigen in the Ogoni populace. It is therefore necessary to take into cognizance the fact that the presence of Rhesus E antigen may likely be the cause of some transfusion reactions which cannot be explained after a compatible ABO and Rhesus D cross match. We therefore recommend that Rhesus E blood grouping be carried out on blood donors and recipients.

CONSENT AND ETHICAL APPROVAL

Informed consent was obtained from apparently healthy subjects prior to enrolment upon ethical clearance by the Ethics Committee of the Department of Medical Laboratory Science, Rivers State University; and from the Rivers State Ministry of Health.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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