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Evaluation of Alpha (α) and Beta (β) Haemolysin Antibodies Incidence among Blood Group 'O' Donors in ATBUTH Bauchi- Nigeria

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Abstract **Background**: Antibodies with haemolytic properties are common within the ABO system. These lytic antibodies are immunoglobulin G (IgG) and in high titres cause haemolysis during blood transfusion. Information on Immunoglobulin types and concentration of ABO haemolysins in Nigerian population is not readily available especially in Northern Nigeria. **Methods:** Serum samples from 200 males and 25 females O group blood donors were screened for A and B haemolysins. Forty two positive samples (38 males and 4 females) were treated with dithiothretiol (DTT) for characterization of IgG class. Antibody titre was compared with grade of haemolysis. **Results:** Out of 50 positive samples 42 highly haemolytic serum samples had IgG titres of > or = 64 after treatment with DTT. There was 18.6% occurrence of α and or β haemolysins in blood group O donors also with average titre values of α-haemolysins (68.2) and β-haemolysins (67).**Conclusion:** Results showsclear indication of high incidence of α and β haemolysins among the healthy blood group O donors hence, Haemolysin test was found to be a useful screening test to identify group O donors with high levels of IgG anti A and/or anti B as far as safe blood transfusion is concerned. **Aim:** The study was determined the incidence of ά and β haemolysins in healthy blood group O donors in the Blood Bank Unit of ATBUTH Bauchi.

Keywords: *alpha (α) and beta (β) haemolysin, group "O", gender, prevalence, Bauchi-Nigeria*

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1. Introduction

Antibodies with haemolytic properties are common within ABO system. These lytic antibodies are immunoglobulin G (IgG) and in high titres cause haemolysis during blood transfusion. In our country as adequate supply of blood and component separation facility is not always available, so it is not uncommon to transfuse group O blood to non-group O individuals..With limited pre-transfusion workup if the transfused blood to the non-group O individual happens to be from these group O donors with lytic antibodies, it can result in intravascular haemolysis in such patients. Hence it is important to avoid transfusion of blood containing high titres of immune anti-A and anti-B antibodies to non-group 'O' individuals.

Many patients often suffer severe blood transfusion reactions from the donated blood. This incident was not explained until 1900 – 1901 when Landsteiner discovered that the blood of another might cause visible clumping of blood (agglutination) or hemolysis [1]. Good practice in blood transfusion testing requires compatibility testing and for this reason, the practice of transfusing group O blood to non O recipient should be discouraged [2,3]. Haemolysins are antibodies which in the presence of complement will cause the haemolysis of Red blood cells possessing their corresponding antigen. They are l gMor IgG immunoglobin which can lyse alpha(ά) and beta (β) cells respectively [4,5,6]. Other mechanism that might considerably be responsible for the development of immune A or B antibodies in a significant proportion of eligible donors includes infection such as pneumococcus and possibly other bacteria which might produce heterophylic stimulation with the formation of antibodies capable of destroying red blood cells [7]. The presence of strong heamolysin has removed the concept of universal donors and universal recipient from the terminology used in blood banking [8]. There is a wide variation in the titre

of ABO iso agglutinin in a random population on the occurrence of haemolysin antibodies among sickle cell anaemic patients within Calabar metropolis of Nigeria as out of 54 sickle cell anaemic patients sera and 100 normal patient's sera analyzed, haemolysin antibodies was detected in 24 out of 54 sickle cells anaemic patients, which is equivalent to 44% of the total samples analyzed and 25 out of 100 normal group O donors showed positive ABO haemolysin antibodies which is equivalent to 25% of the total normal patients analyzed. However, the incidence of ά and β haemolysin in group O sickle cells anaemic patient is more prevalent than that of the normal individuals [9]. Study on prevalence and titre of ά and β haemolysin in blood group O donors in Ilorin, Kwara State of Nigeria revealed that out of 250 subjects analyzed, the prevalence of ά and β haemolysin was 23.2%. Haemolytic anti B occurs twice as frequent at haemolytic anti A. Titre value of anti A was higher than haemolytic anti B and there was no relationship between the age and sex and prevalence of heamolysin [10]. Prevalence of ά and β heamolysin only was10.3% and 12.6% respectively while those donors having both ά and β haemolysins in their sera was 32.5% from an overall prevalence of 55.4% of haemolysin in group O donors in a study carried out in northeastern Nigeria 2007 and 2010 [11].

1.1. Aim

To determine the incidence of ά and β haemolysins in healthy blood group O donors in the Blood Bank Unit of ATBUTH Bauchi.

2. Materials and Method

Blood samples of 225 of blood group O were collected in the blood bank of AbubakarTafawaBalewa University Teaching Hospital (ATBHUTH), Bauchi, Nigeria between February and October 2011. Blood group was determined by cell and serum grouping. Group O serum samples were tested for haemolysins. The haemolysin test and scoring was done as per standard procedures. Serum samples having a score of 2+ and 3+ were considered strongly positive while 1+ partially positive. All the samples were tested within 24 hours of collection to ensure the adequate levels of complement. All strongly positives samples for haemolysis were treated with dithiothretiol (DTT) for characterization of Immununoglobulin class.

Ethical Issues: Ethical clearance was obtained from ATBUTH Bauchi, Nigeria.

2.1. ABO and Rhesus Blood Group Analysis

Potent anti - A, anti - B anti - AB and anti D sera were used to group the subjects red cells as described by Dacie and Lewis [10,12].

Pooled cells A and B were also re-grouped for confirmation.

2.2. Heamolysin Test

One volume of serum and one volume of 5% red cells suspension of A and B cells were placed into each test tube respectively. The tubes were then incubated at 37°C for 2hrs after which all tubes were then centrifuged. They solutions were then examined macroscopically in a bright light background for heamolysis and / or agglutination. Degree of Heamolysis was graded as follows

Complete Heamolysis …….. (3+)
Partial Heamolysis (more than 50%) …….. (2+)
Trace Heamolysis ……..... …… (1+)
No visual Heamolysis ……. Negative

Known A and B red cells were used to test the subject's sera for agglutination and subsequent hemolysis of the red cells in the presence of haemolysin and complement in the fresh sera samples [10,12].

Samples were observed both physically and via the microscope (visual haemolysis and microscopic agglutination).

3. Results

Table 1. General Distribution of Haemolysins

Subject	Number	Percentage (%)
Positive heamolysin	50	22.2
Negative Heamolysin	175	77.8
Total	225	100

Table 2. General Pattern of Distribution ofHaemolysins

Haemolysin Type	A	B	α +β	Total
Numbers of positive	12	22	16	50
Percentage (%)	24	44	32	100

Table 3. Distribution of Highly Positive Haemolysins

Haemolysin Type	A	B	α +β	Total
Numbers of positive	7	22	13	42
Percentage (%)	16.7	52.3	30	100

Total Number of donors 225.

Table 4. Degree of Haemolysis (Alpha (α) Haemolysin)

Degree of traced Heamolysis	1+	2+	3+	Total
Numbers of positive	0	5	7	12
Percentage (%)	0	41.7	58.3	100

Table 5. Degree of Haemolysis (Beta (β) Haemolysin)

Degree of traced Heamolysis	1+	2+	3+	Total
Number of positive	0	7	15	22
Percentage (%)	0	31.8	68.2	100

Table 6. Degree of Haemolysis (Alpha (α) and Beta (β) Haemolysins)

Degree of traced Heamolysin	1+	2+	3+	Total
Number of positive	0	6	10	16
Percentage (%)	0	37.5	62.5	100

Table 7. TitrationResults

Immunoglobulin	Average Titre
Alpha (α)	68.2
Beta (β)	67

4. Discussion

From the 225 donors screened for haemolysins,50 were positive out of which 42 (18.67%) were strongly positive (α = 7, β = 22, and α + β = 13), 8 (3.56%) were partially positive while 175 (77.8%) negative Furthermore, the degree of heamolysis in β- heamolysin (68.2%) is higher compared to those of α- heamolysin (58.3%) and α + β

haemolysin (62.5%) respectively.Result shows high incidence of both strongly positive α and or β haemolysins (18.6%) in blood group O donorsand average titre values of α-haemolysins (68.2) andβ-haemolysins (67). The foregoingpointsto high risk of blood transfusion if not properly handled as it is a common practice to transfuse ABO compatible group O blood in emergency. Haemolysis following such transfusion due to passively transfused antibodies can cause significant morbidity even when plasma volume is reduced. Haemolysis has also been reported following transfusion of other products containing ABO incompatible plasma. Hence it is important to avoid transfusion of blood containing high titres of immune anti-A and anti-B antibodies to non-group O recipients. Keeping in view the above facts and the findings of the present study it is suggested that Haemolysin Test should be performed when group O whole blood or components containing plasma needed to be transfused to non-group O recipients. This will also identify the dangerous universal donors or the donors with high levels of anti-A and anti-B haemolysins. It is therefore concluded that haemolysin test is a useful screening test to identify group O donors with high levels of anti-A and/or anti-B antibodies for safe blood transfusion.

However studies have shown that any sera that produces (2+) heamolysin should be considered as having significant lytic antibodies and those with (3+) (complete heamolysin) are dangerous for transfusion to a non – group O recipients [14]. Those with (1+) heamolysin may be considered as not having significant lytic antibodies but as much as possible should not be transfused to a non – group O recipient. Gender status has no significant effect on the prevalence of heamolysin in this study. This conforms to the findings of [9,10].

Absence of α and β Haemolysins cannot be totally ruled out in group 'O' blood donors as blood not screened for haemolysins before day of transfusion runs in the recipient.

5. Conclusion

Blood group O could not be regarded as universal donors unless it has been successfully screened for presence of Heamolysin and found Negative.The observed 18.6% occurrence of α and or β haemolysins in blood group O donors also with average titre values of α-haemolysins (68.2) and β-haemolysins (67) are clear indications of high incidence of α and β haemolysins among the healthy blood group O donors in the Blood Bank Unit of ATBUTH Bauchi.

Acknowledgment

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Conflict of Interest

No conflicting interest

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