Original Article

Frequency of Subgroups of Blood Group "A" and "AB" amongst the Blood Donors in a Regional Cancer Institute of North East India and its Importance: A Retrospective Study

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ABSTRACT

Introduction: ABO Blood group system was discovered by Karl Landsteiner in the year 1900 who classified the blood groups into A,B,AB and O on the basis of the presence or absence of the antigens A and B on the cell surface of Red blood cells.Further ,in the year 1911, Von Dungern and Hirszfeld divided the group A into A1 and A2 hence total of six blood groups A1,A2,B,A1B,A2B,O. Aim of the study is to find out the frequency of subgroups of A and AB Blood groups and its importance in transfusion practice.

Methods: A total of 5594 blood donors' samples were collected in a one-year period who came to donate blood. Blood grouping was done by the conventional tube method both forward and reverse grouping. Sub grouping of A and AB was done by using the commercially available Lectin Anti A1 (Tulip diagnostics, Goa ,India) .Group A red blood cells which agglutinate with Anti A1 lectin are classified as subgroup A1,whereas which do not agglutinate are classified as A2.

Result: Out of total 5594 blood donors, 1354 belonged to group A and 413 constituted group AB. The percentage of A1 amongst the A blood group is 93.94% (1272) and A2 is 6.056 (82). The percentage of A1 amongst the AB blood group is 91.04% (376) and A2 is 8.95% (37)

Conclusion: The frequency of A1 subgroup is more in comparison to A2 in both A and AB blood groups amongst the blood donors. Identification and recording of subgroups is important in blood bank in respect to blood group discrepancy, blood transfusion reactions as well as Organ transplantation.

Key words: blood group, subgroup A1, A2, Transfusion reaction

INTRODUCTION:

ABO Blood group system was discovered by Karl Landsteiner in the year 1900 who is considered as the father of transfusion medicine. The ABO antigens are defined by carbohydrate moieties on the extracellular surface of the red blood cell (RBC) membranes. They are also highly expressed on the surface of a variety of human cells and tissues, including epithelium, sensory neurons, platelets, and vascular endothelium. The ABO and Rh blood group system remain the most important blood group systems.^{2,3} The frequency of ABO blood groups varies greatly in different races and populations. The prevalence of blood group O is highest in most of the population followed by B, A and AB.⁴ A and AB have been divided into subgroups A1, A2, A1B and A2B depending on the reaction with anti A1 Lectin, the extract from the seed of lectin Dolichos biflorus or human anti A1 serum.Anti A1 agglutinates A1 and A1B cells but not A2 and A2Bcells.Subgroups weaker than A2 occur infrequently. They are characterized by the declining

NJMR Volume 10 Issue 3 July –Sept 2020

number of A antigen sites on red cells and reciprocal increase in H reactivity. Weaker variants of A are A3, Ax, Am and A Intermediate. Anti A1 reacting at 220C or lower has no clinical significance but if reacts at 370C is significant resulting in the blood group discrepancy, incompatibility and transfusion reaction.^{1,5}

MATERIALS AND METHODS

A total number of 5594 blood samples were collected from the blood donors who came to blood bank as replacement or voluntary donors for a period of two years from January to December 2016-2017 with their due consent according to the NBTC guidelines following the donor questionnaire. Ethical clearance for the study was obtained from the Institutional Ethical Committee. The identity and the gender of the donors was not revealed in any manner in the study conducted. Blood grouping was done by the conventional tube method. Both the forward and re-

verse blood grouping were done by using the commercially available monoclonal antisera anti A, anti B and antiAB (Tulipdiagnostics, Goa, India) for forward grouping. The reverse grouping was done by in house prepared pooled A cell, B cell, and O cell. Blood groups were interpreted based on agglutination pattern in both forward grouping and reverse grouping. In forward grouping, A or B antigen agglutination was observed with the corresponding antisera & circulating anti-A or anti-B were detected by reverse typing using pooled cells. Rh typing was not done as the study was mainly to determine the subgroups. For forward grouping, first 2-5% cell suspension of the test sample in normal saline is prepared after cell wash. In our study we prepared 3% red cell suspension by transferring 0.3 ml of washed red cells to a test tube with 9.7 ml of saline. 1 drop of anti-A, anti-B & anti-AB were placed in labelled tubes. To each test tubes 1 drop of 3% suspension of the red cells were added. After mixing, centrifugation, red cell buttons were resuspended & examine for agglutination. For reverse grouping, two drops of the test serum were placed in the test tubes labelled as A, B, O cells. 1 drop of A, B, and O pooled cells were added to the labelled test tubes. After mixing, centrifugation, the serum was examined for agglutination. Agglutination was graded according to AABB guidelines. Single clump of agglutination with no free cells was graded 4+, three or four agglutinates with few free cells as 3+, many fairly large agglutinates with many free cells as 2+ and small agglutinates with a turbid background as 1+, very small agglutinates with a turbid background were graded as weak reaction (wk) and mixtures of agglutinated and un-agglutinated red blood cells as mixed field (mf). Samples of group A and AB were further tested with anti-A1 lectin (Tulip Diagnostic, Goa, India) to classify them into A1, A2 subgroups. When the sample showed 4+ agglutinates with anti-A antisera but negative anti-A1 lectin, the sample was considered as A2 subgroup. A or AB blood group samples showing agglutination with pooled A cells were tested with A1 cells to confirm presence of anti-A1 antibodies.

RESULT

Out of the total 5594 blood samples, "O" Blood group constituted 36.03%(2016),followed by blood group "B" 32.37%(1811),blood group "A" 24.20%(1354) and blood group "AB" 7.38%(413) (Table 1). Blood groups "A" and "AB" constituted a total of 1767 out of the total donors. Out of 1354 "A" blood group donors, sub group A1 constituted 1272(93.94%) and subgroup A2 82(6.056%).Out of 413 "AB" blood group donors, subgroup A1B showed a number of 376 (91.04%) and A2B was 37(8.95%).

Table 1. Blood Groups with Subtypes

Blood Group	Cases (%)
0	2016 (36.03%)
В	1811 (32.37%)
А	1354 (24.20%)
Subtype A1	1272 (93.94%)
Subtype A2	82 (6.06%)
AB	413 (7.38%)
Subtype A1B	376 (91.04%)
Subtype A2B	37 (8.95%)
Total	5594 (100.0%)

DISCUSSION

Various studies have been done till date regarding the distribution of the various blood groups in different parts of the world including India. It has been seen that there is variation in the distribution of the blood groups according to region, races, and ethnicity. Our study is mainly confined to a small group of population of the North Eastern part of India coming to donate blood voluntarily or as replacement in a regional cancer Institute. The study has shown that the prevalence of blood group "O" is highest (36.03%) followed by blood group "B"(32.37%), then "A"(24.20%) and lastly "AB"(7.38%) which is similar to studies conducted by Sunder Periyavan et al, the most common blood group was O (39.81%) followed by B (29.95%), A(23.85%) and AB (6.37%)[6], conducted by Chapagain RH et al (2005)7, AnjuVerma et al (2011)8, and Das PK et al(2001)9, Mathur Niharika et al.¹⁰ It is seen that the most common blood group in India is O followed by B which can also be seen in our study. However, a study conducted in Uttarkhand by Garg P et al (2014)11 showed that Blood group B was more common than O followed by A and AB. The ABO blood group system was discovered, a century ago in the year 1900. Gradually it was discovered that there are weaker variants due to the heterogenosity of the A and B alleles and which play an importance in the immunohaematology. The blood group A can be subclassified as A1, A2 and weak A subgroups (Ax, A3, Aend, etc.) based on red cell agglutinability and various serological reactions.12 Approximately 80% of Blood Group A or AB are classified as A1 or A1B the remaining 20% are either A2 or A2B.In our study we have seen that the percentage of A1 in blood group A is 93.94% and in AB it is 91.04% whereas the percentage of A2 in blood group A is 6.056% and in AB it is 8.95% which is similar to the studies done in other parts of India as well as at international level.¹²⁻¹⁷ A2 and A2B are rare subgroups as evident. But the anti-A1 antibodies which are present in the sera of A2 groups and in A2B subgroups can cause discrepancy in blood grouping and compatibility testing resulting in transfusion reactions and complicate organs transplantation. Around 1% to 8% of A2 individuals produce anti-A1 in the serum and 22% to

35% of A2 B individuals produce anti-A1.16 A2 and A2B individuals may be immunologically stimulated to produce specific anti-A1 antibody that does not cross react with A2 red cells, but reacts with A1 red cells since they cannot recognize A1 antigens as being part of their own red cell make up. Weaker variants like A2, A2B and other subgroups of A and AB may become clinically significant when they have antiA1antibody reacting at 37 C. This sometimes leads to incorrect ABO blood typing. AB group maybe mistyped as B group and A group as O group. Approximately 0.4% of A2and 25% of A2B individuals have anti-A1 in the serum.Individuals with an A2B phenotype are more likely than A2 individuals to produce anti-A1 because of the relative reduction of A antigens on A2B cells.18

CONCLUSIONS

This study shows that the frequency of subgroup A1 is more than A2 in blood group A and AB but the frequency of A2 in AB is more than in A. Identification of these subgroups is important in respect to blood group discrepancy ,blood transfusion reactions as well as Organ transplantation .Subgrouping should be done routinely in the blood banks so as to avoid the transfusion reactions and the incompatibilities. Further studies are required at the molecular level for safe and proper blood transfusion services which will lead to greater benefit of patients.

Limitation: Study population is limited to the blood donors coming to donate blood in the regional cancer institute, Guwahati. Molecular studies were not done due to resource constraints.

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