Frequency Of ABO And Rh Blood Groups In Blood Donors: A Study From A Tertiary Care Teaching Hospital In India

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Abstract : Background & Objectives: Out of total 328 antigens recognized by the International Society of Blood Transfusion (ISBT), ABO and RhD antigens are the most important from the transfusion medicine perspective. The present study was conducted with larger sample size than prior studies to determine frequencies of ABO and Rh alleles and obtain distribution of ABO and RhD blood group pattern among blood donors. Methods: A retrospective study was conducted in the Department of IHBT, Civil Hospital, Ahmedabad from October 2007 to September 2012. ABO grouping and RhD typing was done using conventional tube technique on a total of 109771 donors. Commercial anti-sera and in-house prepared cells were used for cell and serum grouping respectively in those tests. Departmental Standard Operating Procedures (SOPs) were followed for each aspect of testing. Observed ABO and RhD antigen frequencies were noted. Bernstein and Hardy-Weinberg equations were applied to determine the allele frequencies of ABO and RhD respectively. Results: Blood group B has the highest prevalence (35.81%) in the population under study followed by O (32.74%), A (22.68%) and AB (8.77%). Female donors comprised only 1.75% of the sample size. Rh D positivity was noted in 94.48% donors. Conclusion: Results obtained were quite similar to prior studies from Ahmedabad with smaller sample size. Remarkable differences were noted as compared with western population. The data generated in the present study combined with several other studies of different geographical region of India has significant implications in inventory management of blood transfusion services. [Sonani R et al NJIRM 2013; 4(4): 69-73] Key Words: ABO, Rh D, Antigens, Distribution.

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Introduction: At present, a total of 328 erythrocyte antigen specificities have been recognized by the International Society of Blood Transfusion (ISBT) and many antigens are in the provisional list ¹. These antigens are organized into 30 blood group systems, high and low incidence series and collections of blood group antigens by the Working party of ISBT². Among those antigens ABO and RhD are most important from the transfusion and transplantation perspective. The discovery of the ABO blood groups by Karl Landsteiner was an important achievement in the history of blood transfusion which was followed by discovery of Rh (D) antigen ^{3,4} Blood group antigens play a vital role in transfusion safety, understanding genetics, inheritance pattern, and disease susceptibility. All human populations share the same blood group systems; although they differ in the frequencies of specific types. The incidence of ABO and Rh groups varies markedly in different races, ethnic groups in different part of the world⁵. The antigen phenotype is determined by the genetic make-up of the alleles of a system. The vast majority is inherited in a simple Mendelian fashion and is stable characteristics which are useful in paternity

testing⁶. Blood group antigens are known to have some association with diseases like duodenal ulcer, diabetes mellitus, urinary tract infection, Rh incompatibility and ABO incompatibility of newborn⁷. The frequencies of ABO and Rh blood groups vary from one population to another and time to time in the same region. Knowledge of the distribution of ABO and Rh blood group is essential for effective management of blood bank inventory, be it a facility of a smaller local transfusion service or a regional or national transfusion service. It is, therefore, imperative to have information on the distribution of these blood groups in any population⁸. Knowledge of blood group distribution is also important for clinical studies, for reliable geographical Information. Apart from their importance in blood transfusion practice, the ABO and Rh blood groups are useful in population genetic studies, researching population migration patterns as well as resolving certain medico legal issues, particularly of disputed paternity cases. The present study was conducted with larger sample size than prior studies to determine frequency of alleles and distribution of ABO and Rh blood group patterns among blood donors in and around

69

Ahmedabad, Gujarat and compare with other data from similar studies within India and other parts of the world.

Material & Methods: A retrospective study was conducted in the Department of IHBT, Civil Hospital, Ahmedabad during five year period from October 2007 to September 2012. Study subjects included all donors who donated blood at our Centre, both in-house as well as in outdoor blood donation camps. (n=109771)

ABO and Rh blood grouping was done by tube technique by using commercially available antisera anti-A, B, AB, H and Rh (two different Rh antisera, monoclonal IgM and monoclonal IgM+IgG), and known cells prepared, in-house, from pooled blood units. In Rh system, only D antigen was typed as it is the most antigenic among other antigens of the system .Hence those who tested positive with anti-sera D were considered to be Rh positive and those who did not, weak D was tested with Indirect anti-globulin test. Negative Indirect anti-globulin test was typed as Rh negative. These anti-sera were validated at our centre before using them. For determination of ABO blood groups, both cell and serum grouping was carried out using conventional tube technique. For cell grouping, all donor samples were washed with normal saline and 5% saline suspended samples prepared. Cell grouping was done using commercial anti-sera. For serum grouping, donor's serum was added to freshly prepared 5% suspension of washed pooled A-cells, B-cells and O-cells prepared in-house. Tests were incubated at room temperature for 20-30 minutes & centrifuged at 1000 rpm for 1 minute. The results were noted and all the negative results were confirmed by microscopy. Anti-D negative donors were tested for weak D by Indirect anti-globulin test with Column Agglutination Technology (Biorad). Weak D positive results were considered Rh positive.

Male:Female ratio for different blood groups was analyzed. ABO Rh allele frequencies were analyzed using statistical equation. Results of the study were compared with results from other geographical locations. Bernsteins equation is a simplified form of Hardy Weinberg equation for triallelic inheritance. ABO allele frequencies were estimated according to a Bernsteins equation which yields results that are close to maximum likelihood estimates⁹. Preliminary estimates were calculated as:

p = 1 - v(B)+(O), q = 1 - v(A)+(O), r = v(O)

[p, q, r denote allele frequencies of A,B,O and (A),(B),(O) denote observed frequencies of blood groups A, B and O].

A correction factor (θ) was calculated according to $\theta = 1 - p - q - r$. The final estimate of allele frequencies was then calculated as follows: p1 = p (1 + $\theta/2$); q1 = q (1 + $\theta/2$); r1 = (r + $\theta/2$) (1 + $\theta/2$) [where p1, q1, and r1 denote corrected allele frequencies]. Rh D allele frequencies were calculated according to the Hardy-Weinberg equation¹⁰.

Results: <u>ABO Blood Groups:</u> In present study, Blood group B was the most common and blood group AB was the least common amongst the blood donors in Ahmedabad. The distribution of phenotypes and allelic frequencies of ABO blood groups are listed in Table 1. The frequencies of alleles p, q, and r for the ABO blood group system were found to be 0.1721, 0.2556, 0.5722 respectively for the total data.

<u>Rh Blood Groups:</u> In our study, Rh positive donors were 94.48% and Rh negative donors were 5.52%. The frequencies of alleles *D* and *d* for the Rh blood group system were found to be 0.765 and 0.235 respectively for the total data.

Discussion: In the present study, the ABO blood group typing in the total sample showed the same trend of prevalence as in the general Indian subcontinent ($B \ge O > A > AB$). The same trend was found among males but the order was different among females ($O \ge B > A > AB$). However, the allelic frequencies were in the order of O>B > A.

NJIRM 2013; Vol. 4(4).July - August

70

Blood	Male (%)	Female	Total
groups		(%)	population (%)
A	24722	173	24895 (22.68)
	(22.92)	(8.98)	
В	38687	620	39307 (35.81)
	(35.87)	(32.19)	
0	34987	952	35939 (32.74)
	(32.44)	(49.42)	
AB	9449	181	9630 (8.77)
	(8.76)	(9.39)	
Total	107845	1926	109771 (100)
	(100)	(100)	

Table 1: ABO blood groups frequencies in blooddonors

Table 2: ABO alleles frequencies in blood donors

ABO alleles	Male	Female	Total population
А	0.1735	0.0967	0.1721
В	0.256	0.2359	0.2556
0	0.5695	0.7029	0.5722

Table 3: Rh blood group frequencies

Rh blood	Male (%)	Female (%)	Total (%)
group			
Rh Positive	101844	1865	103709
	(94.44)	(96.84)	(94.48)
Rh Negative	6001 (5.56)	61 (3.16)	6062 (5.52)
Total	107845	1926 (100)	109771
	(100)		(100)

Table 4: Rh alleles frequencies

Rh	Male	Female	Total	
alleles			population	
Rh D	0.764	0.822	0.765	
Rh d	0.236	0.178	0.235	

Hardy Weinberg equilibrium is a useful indicator of genotype frequencies within a population and whether they are based on a valid definition of alleles and a randomly mating sample. Hardy Weinberg equilibrium assumes a stable population of adequate size without selective pressures and is used in human genetic studies as a guide to data quality by comparing observed genotype frequencies within a population. Bernsteins equation is a simplified form of Hardy Weinberg equation for triallelic inheritance.

Table 5 : Frequency of weak D in blood donors

	Weak D	Rh D	Total
	positive	negative	(%)
	(%)	(%)	
Male	206 (3.32)	6001(96.68)	6207 (100)
Female	3 (4.69)	61 (95.31)	64 (100)
Total	209 (3.33)	6062(96.67)	6271 (100)

Although prior two studies from Ahmedabad had smaller sample size than the present study, the prevalence of blood groups A, B, O and AB as well as those of RhD Positive and RhD Negative was quite similar to these two studies^{13,14}. There are remarkable differences in the distribution of ABO blood group antigens and RhD antigen when the population under the study is compared to other countries such as Australia, Britain and USA.

In modern medicine, blood groups are important in evolution and their relation to disease and environment is being increasingly important ^{22,23}. It is, therefore imperative to have information on the distribution of ABO and RhD antigens in any population group from the public health perspective as well.

Conclusion: The study of blood group distribution has significant implications in the management of blood bank and transfusion services. Blood grouping should be done for each newborn at the time of birth and repeated later. Blood group of an individual indicated on national identity cards, driving licenses and school/office identity cards, will be of tremendous use in case of acute hemorrhage or anemia in children when urgent transfusion of yet to be cross-matched blood is required.

Place of Study	А	В	AB	0	Rh Positive	Rh Negative
Within India						
Shimoga-Malnad ¹¹	24.27	29.43	7.13	39.17	94.93	5.07
Davanagere ¹²	26.15	29.85	7.24	36.76	94.8	5.52
Eastern	23.3	35.5	8.8	32.5	94.2	5.8
Ahmedabad ¹³						
Western	21.94	39.4	7.86	30.79	95.05	4.95
Ahmedabad ¹⁴						
Punjab ¹⁵	21.85	37.6	9.3	29.3	97.3	2.7
Bangalore ¹⁶	23.85	29.95	6.37	39.82	94.2	5.79
Chittoor ¹⁷	18.95	25.79	7.89	47.37	90.6	8.42
Vellore ¹⁸	18.85	32.69	5.27	38.75	94.5	5.47
Present Study	22.68	35.81	8.77	32.74	94.48	5.52
Outside India						
Pakistan	23.8	38	10	27.2	89.1	10.9
Nepal	34	29	4	33	96.7	3.33
Australia ¹⁹	38	10	3	49		
Britain ²⁰	41.7	8.6	3	46.7	83	17
USA ²¹	41	9	4	46	85	15

Table 6: Comparison of frequency of ABO and Rh phenotypes from different geographical areas (%)

It is necessary to conduct similar well designed studies in other states of India in order to determine the blood group frequencies in them. The data generated in the present study and several other studies of different geographical region of India will be useful for health planners while making efforts to face the future health challenges in the region.

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