## **Original Research Article**

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# Frequency and distribution of ABO and Rh blood groups among blood donors in tertiary care hospital of South Gujarat, India

Kruti A. Raja\*, Gopi H. Dobariya, Chirag A. Unagar, Amrish N. Pandya, Jitendra N. Patel, Sangita J. Wadhwani

Department of Immuno-Haematology and Blood Transfusion, Government Medical College, Surat, Gujarat, India

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\***Correspondence:** Dr. Kruti A. Raja, E-mail: seemadeepakmysore@gmail.com

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## ABSTRACT

**Background:** Since it was discovered by Karl Landsteiner, the ABO blood group system is the most important blood group system in Transfusion Medicine. The blood group systems are also very important in population genetic studies, researching population migration patterns as well as resolving certain medico-legal issues, particularly disputed parentage. This study was carried out with an objective to provide data regarding gene frequency and distribution of ABO and Rh blood groups among blood donors in South Gujarat, India.

**Methods:** Data of 40732 blood donors were retrospectively collected and analyzed regarding ABO and Rh blood groups from May-2011 to April-2016 and reported in simple numbers and percentage. Blood group of the blood donors was determined by forward and reverse methods with the help of commercially available standard monoclonal antisera by test tube and column agglutination techniques in required cases.

**Results:** The most common blood group among donors was B (34.43%) followed by O (32.26%), A (24.35%), while the least prevalent blood group was AB (8.94%). Rh positivity among donors was (95.12%). Rests were Rh Negative (4.87%). The estimated gene frequencies were 0.1844 for  $I^{A}(p)$ , 0.2477 for  $I^{B}(q)$  and 0.5679 for  $I^{O}(r)$ .

Conclusions: The most common blood group in donors was B positive and least common was AB negative.

Keywords: ABO & Rh blood group, Blood donor, Gene

## **INTRODUCTION**

It was not until the year 1900, when Karl Landsteiner discovered why some blood transfusions were successful while others could be deadly. Landsteiner discovered the ABO blood group system by mixing the red cells and serum of each of his staff. He demonstrated that the serum of some people agglutinated the red cells of other. From these early experiments, he identified three types, called A, B and C (C was later to be re-named O for the German "Ohne", meaning "without", or "Zero", "null"). The fourth less frequent blood group AB, was discovered a year later. In 1930, Landsteiner received the Nobel Prize in physiology and medicine for his work.<sup>1</sup> The A subgroups have been classified as  $A_1$ ,  $A_2$ ,  $A_{int}$ ,  $A_3$ ,  $A_x$ ,  $A_m$ ,  $A_{end}$ ,  $A_{el}$ , and  $A_{bantu}$  based on the reactivity of red

cells with human anti-A and anti-AB. Group A red cells which react with both anti-A and Anti-A<sub>1</sub> are classified as A<sub>1</sub>. A<sub>1</sub> constituted approximately 80% of entire A blood group population and group A cell which react with anti-A and not agglutinated with anti-A<sub>1</sub> are classified as A<sub>2</sub>, making up of remaining 20%.<sup>2</sup> Subgroup of B are very rare and occur less frequently than subgroup of A and they are B<sub>3</sub>, B<sub>x</sub>, B<sub>m</sub> and B<sub>h</sub>.<sup>3</sup>

In 1940, the cause of Haemolytic Disease of Foetus & New-born (HDFN) was linked to the Rh blood group system by Levine and Stetson. Rh was the name given to the system because of the similarity of this antibody to one made from stimulating guinea pigs and rabbits with rhesus monkey cells. This Rh antibody, described by Landsteiner and Wiener, agglutinated 85% of human red cells tested and was nonreactive with 15%.<sup>2</sup> The original Rh antigen was referred to as D and related ones are C and E; the antithetically related antigens are called c and e.<sup>4</sup>

Among a total of 29 blood group systems and over 600 different blood group antigens discovered so far, ABO and Rhesus are the most important blood group systems.<sup>5</sup> There is occurrence of antibodies of these blood group system and ability of antibodies to cause hemolytic transfusion reaction and HDFN. Apart from their importance in blood transfusion practice, ABO and Rh blood groups are useful in population genetic studies, relationship to certain diseases, researching population migration patterns as well as resolving certain medicolegal issues, particularly disputed parentage.<sup>6</sup>

The prevalence of ABO blood groups varies from race to race and region to region. For effective management of blood banks inventory, the knowledge of the distribution of ABO and Rh blood groups is also essential.<sup>7</sup> It is therefore important to have an information on the distribution of these blood groups in the population of this study region and hence this study has been carried out to determine the gene frequency and distribution of ABO and Rhesus blood groups in blood donors of tertiary care hospital of South Gujarat and to compare it with the other studies.

## **METHODS**

In this retrospective study, data of 40732 blood donors (aged between 18-65 years) were analysed. These donors had donated blood in the blood bank and in camps conducted by the blood bank during the five year time period from May 2011 to April 2016. Each donor, at the time of donation, was interviewed before screening for his/her particulars such as address, age and sex. Information regarding medical history including a history of previous transfusion was also obtained. After blood donation, blood group was determined by forward and reverse grouping by conventional tube method from the pilot samples of the donors following standard operative procedures of the blood bank.

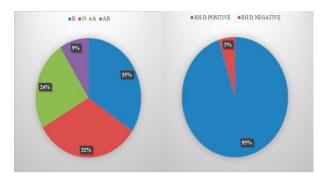
For complete ABO grouping (forward and reverse) mono clonal anti A, anti-A<sub>1</sub>, anti B, anti AB, anti H antisera and A, B, O pooled cells were used. For Rh typing anti-D ( $R_0$  &  $R_1$ , anti Ig M and blend of anti Ig M and anti Ig G) antisera were used. Final blood group was confirmed only if forward and reverse groups are identical. Rh negative blood groups were confirmed by antiglobulin technique. All weak D groups were considered as Rh positive.

The Hardy-Weinberg formula with Ceppilini correction & with required assumptions were used to calculate a determination of the gene frequencies. Data on frequency of ABO and Rh-D blood groups were reported in simple

percentages and compared with the similar studies by other authors.

#### RESULTS

ABO blood groups of 40732 healthy young adults from 18 to 65 years of age was carried out. We found that the percentage of ABO blood groups in donors in descending order was as follows: B>O>A>AB. Out of 40732 blood donors, 14025 (34.43 %) were of blood group B, 13144 (32.26 %) of O, 9920 (24.35 %) of A and 3643 (08.94 %) were of blood group AB (Figure 1).



#### Figure 1: Frequency of ABO and Rh blood group.

Out of 3643 donors of group AB; 02 (0.05 %) were of  $A_2B$ . One donor was typed for having Parabombay Blood Group. While further investing this donor of Parabombay blood group, the author founded his sibling to have same Parabombay blood group. In 40732 blood samples, incidence of Rh-D positive was 95.12 % (38746) and Rh-D negative was 04.87 % (1986) (Figure 1).

## Calculation of expected phenotype frequency

Calculation of 'O' allele frequency (r)

 $\begin{aligned} r^2 &= \text{frequency of O phenotype} \\ r &= \sqrt{r^2} \\ r &= \sqrt{0.3226} \\ r &= 0.5679 \end{aligned}$ 

Calculation of 'A' allele frequency (p)

p = frequency of A phenotype + frequency of O phenotype

 $\begin{array}{l} p^2 + 2pr + r^2 = (p+r)^2 \\ p = \sqrt{(p^2 + 2pr + r^2) - r} \\ p = \sqrt{(0.2435 + 0.3226) - 0.5679} \\ p = 0.1844 \end{array}$ 

Calculation of 'B' allele frequency (q)

q = 1 - (p+r) q = 1 - (0.1844 + 0.5679) q = 0.2477*Calculation of 'd' allele frequency (u)*   $u^2$  = frequency of d phenotype  $u^2 = 0.0487$ u = 0.2206

Calculation of 'D' allele frequency (v)

v+u=1 v = 1 - u v = 1 - 0.2206v = 0.7794

#### Calculation of genotype frequency

 $AA = p^{2} = 0.0340$  AO = 2pr = 0.2094 BB = q2 = 0.0613 BO = 2qr = 0.2813  $OO = r^{2} = 0.3225$  AB = 2pq = 0.0913  $DD = v^{2} = 0.6074$ Dd = 2vu = 0.3438

## $dd = u^2 = 0.0487$ Distribution of ABO and Rh (D) allele frequency in South Gujarat

 $I^{A}$  allele = 0.1844  $I^{B}$  allele = 0.2477  $I^{O}$  allele = 0.5679  $I^{D}$  allele = 0.7794  $I^{d}$  allele = 0.2206

The Chi-square test for goodness of fit between the observed and expected phenotypes in case of ABO group was not found to be statistically significant (P = 0.99).

#### DISCUSSION

The phenotype and genotype frequencies of ABO and Rh groups vary widely across different races and geographical areas of the world. There were few studies have been done across the India to find out this variation.

## Table 1: Comparison of observed and expected phenotype frequency among blood donors.

Frequency of phenotype and genotype of ABO and Rh system								
Blood group system	Phenotype	<b>Observed frequency</b>	Genotype	Expected frequency				
ABO	А	0.2435	AA	0.0340	0.2434			
			AO	0.2094				
	В	0.3443		0.0613	0.3426			
			BO	0.2813				
	0	0.3226	00	0.3225	0.3225			
	AB	0.0894	AB	0.0913	0.0913			
Rh	D positive	0.9512	DD	0.6074	0.9512			
			Dd	0.3438				
	D negative	0.0487	dd	0.0487	0.0487			

The most common blood group in the present study was B (34.43%) followed by O (32.26%) and A (24.35%) and blood group AB was least common (8.94%). In Rh group, frequency of Rh D positive was 95.12% and Rh D negative was 4.87%.

Combining ABO and Rh blood groups, frequencies of different blood groups were in the following order: B positive (32.65%)>O positive (30.73%)>A positive (23.24%)>AB positive (8.48%)>B negative (1.77%)>O negative (1.53%)>A negative (1.10%)>AB negative (0.45%). Same blood group frequency was found in males. But in females O negative is more common than B negative. On further analysis, female donors showed a relatively higher incidence of Rh negativity (68, 5.5%) as compared with males (1918, 4.85%).

As shown in Table 2, there are variations in ABO blood group frequency across the India.<sup>8-16</sup> According to the

studies done in Northern India and Western India, B group was having highest prevalence (34-40 %) followed by O, A and AB blood group.<sup>8-12</sup> The present study also showed similar findings with all studies except for the multi-centric study done by Agrawal A et al which concluded that O blood group was having highest prevalence in the western India.<sup>8</sup>

Studies done in the Eastern India and Southern India showed that blood group O has highest prevalence (34-40 %) followed by B, A & AB blood group.<sup>14-16</sup> The study done by Agrawal A et al showed O blood group with highest frequency while the study done by Gupta NK et al showed B as the leading blood group for the Central India.<sup>8-13</sup>

In the present study, 95.12 % of blood donor population showed Rh positivity for D antigen while 04.87% showed Rh D negativity. The Rh D positive blood group was found in the range of 91 % to 97 % across the India in different studies.<sup>8-16</sup> The present study correlate with

these findings of all studies.

## Table 2: Region wise distribution of ABO & Rh blood group.

Region	Author	Study	Blood group frequency (%)					
		size (n)	Α	В	0	AB	Rh D positive	Rh D negative
North India								
North zone	Agrawal A et al	2042	24.54	34.47	29.43	11.55	94.8	5.19
Lucknow	Chandra T et al	140320	21.38	39.92	29.27	9.43	95.71	4.29
	Nanu and Thapliyal	6334	24.7	37.5	32.5	5.3	95.37	4.63
Punjab	Sindhu S et al	1150	21.91	37.57	31.22	9.3	97.3	2.7
West India								
West zone	Agrawal A et al	2220	23.69	32.74	36.75	6.8	92.97	7.02
Rajasthan	Behra R et al	83631	22.2	36.4	31.7	9.4	91.75	8.25
Present Study		40732	24.35	34.43	32.26	8.94	95.12	4.87
Central India								
Centre zone	Agrawal A et al	2021	23.1	26.57	43.24	7.07	96.23	3.72
Indore	Gupta NK et al	17080	24.15	35.25	31.5	9.1	95.43	4.57
East India								
East zone	Agrawal A et al	1595	21.88	33.85	37.55	6.7	95.23	4.76
Durgapur	Nag I et al	3850	23.9	33.6	34.8	7.7	94.7	5.3
South India								
South zone	Agrawal A et al	1808	20.68	33.07	38.99	6.25	93.91	6.08
Karnataka	Periyavan S et al	36964	23.85	29.95	39.81	6.37	94.21	5.79
South India	Das et al	150536	18.85	32.69	38.75	5.27	94.53	5.47

Table 3: ABO & Rh D allele distribution in different studies.

Author	Calculated ABO & Rh D allele frequencies							
	I <sup>A</sup>	$I^{B}$	Io	ID	$\mathbf{I}^{d}$			
Amit Agrawal et al	0.1653	0.2254	0.6093	0.7679	0.2321			
Sindhu S et al	0.171	0.27	0.559	0.836	0.164			
Present Study	0.1844	0.2477	0.5679	0.7794	0.2206			

In the present study two donors (0.05 %) were detected as  $A_2B$  blood group and not a single donor was found to have  $A_2$  blood group. The frequencies of  $A_2$  blood group was 3.01 % and 1.07 % and frequencies of  $A_2B$  was 1.43 % and 8.99 % in the studies done by Das PK et al and Shastry S et al respectively.<sup>16</sup>

The present study has also calculated the gene frequency of ABO and Rh blood group by following Hardy Weinberg equation and frequencies are shown in Table 3. The gene frequencies of ABO and Rh blood group found in other two studies done by Agrawal A et al and Sindhu S et al are also shown in table 3.<sup>8-11</sup> The actual distribution of ABO blood group did not differ significantly from the calculated gene frequencies.

#### CONCLUSION

To conclude, the commonest blood group was B in both males and females in the South Gujarat. Regarding

Rhesus group Rh D negativity was found in only 4.87 % population. Based on the findings of the present study and other reference studies, it can be concluded that B blood group has highest frequencies in the Northern and Western India while O blood group has highest frequency in the Eastern and Southern India.

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Conflict of interest: None declared Ethical approval: The study was approved by the Institutional Ethics Committee

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