

# A retrospective study: ABO and Rh phenotype blood group distribution among blood donors in H.N.B. Base Hospital, Srinagar, Uttarakhand, India

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## ABSTRACT

**Objective:** To determine the prevalence of ABO and Rh blood groups based on the antigenic presence on the surface of red blood cells with respect to gender and calculate allele frequency of the blood groups. Globally, approximately 700 type red cell antigens have been identified till now. ABO and Rh blood groups play an important role in the process of blood transfusion, resolving certain medicolegal issues, parental testing, and various genetic studies. **Methods:** This study was conducted in H.N.B. Base Hospital, Srinagar, Uttarakhand, from January 2012 to December 2016. Relevant data of blood donors were collected from blood bank department of the hospital. Blood grouping was conducted using commercially available standard monoclonal antisera applying test tube and column agglutination techniques. **Results:** Out of 9883 individuals, 9333 (92.4%) were males and 750 (7.6%) were female individuals. The most common blood group found was B (31.68%) and least common being AB (11.70%). The prevalence of Rhesus positive and negative distribution in the present studied population was found as 93.51% and 6.49%, respectively. Overall, male ABO group pattern found was shown by formula B > A > O > AB which was similar among Rh-positive male individuals while Rh-negative males' pattern was found as A > B = O > AB. In females, ABO group pattern was B > O > A > AB which was similar to Rh-positive female pattern while differs in Rh negative. The estimated allele frequencies were found as 0.2403, 0.2475, and 0.5122 for I<sup>A</sup> (p), I<sup>B</sup> (q), and I<sup>O</sup> (r), respectively. **Conclusion:** The most common blood group found among the Gharwali donors was B positive while the least common was AB negative, which plays an important contribution for making government policies to develop National Health Program.

**Keywords:** ABO and Rh blood group, allele frequency, blood donor

## Introduction

Different types of blood groups are hereditary and determined based on the presence of surface antigens in the RBC and these groups play a vital role during transfusion. In the year 1900, Karl

Landsteiner discovered ABO blood group system, which became an important milestone in the history of blood transfusion followed by discovery of Rh (D) antigen.<sup>[1]</sup> Distribution of ABO and Rh (D) blood groups varies between populations and races. The studies of blood groups are important parameters in various genetic studies for reliable geographical information and in blood transfusion process with associated diseases, which will eventually help in reducing morbidity and mortality rate. Knowledge of blood

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grouping is also essential for effective management of blood bank inventory.<sup>[1,2]</sup>

Subgroups of A have been further classified as A1, A2, Aint, A3, Ax, Am, Aend, Ael, and Abantu 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-A1 are classified as A1. A1 constituted approximately 80% of entire A blood group population, and Group A cells which react with anti-A and not agglutinated with anti-A1 are designated as A2, making up of remaining 20%.<sup>[3]</sup> Subgroup of B is very rare and occurs less frequently than subgroup of A and which are B3, Bx, Bm, and Bh.<sup>[4]</sup>

The cause of Hemolytic Disease of the Fetus and Newborn was linked to the Rh blood group system by Levine and Stetson in the year 1940.<sup>[5]</sup> The significant Rh antigens are referred to as D and related ones are C and E while antithetically related antigens are designated as c and e.<sup>[5]</sup> Now, nearly 29 blood group systems and approximately 700 different types of blood group antigens were discovered so far but ABO and Rhesus are the most significant blood group systems.<sup>[6]</sup> This system is also useful in different genetic studies, relation to certain diseases, different migration patterns of population as well as helps in resolving certain medicolegal issues, basically paternity dispute.<sup>[7]</sup>

## Methods

The present retrospective study was carried out in the blood banks of H.N.B. Base Hospital, under Veer Chandra Singh Garhwali Government Medical Science and Research Institute, Srinagar, Uttarakhand, during the last 5 years from January 2012 to December 2016. Srinagar city is an important cultural and educational center of the Garhwal hilly region located in the foothills of the Sivalik Ranges of the Greater Himalayas. It is nearly 156 km away from its state capital, Dehradun, located on the left bank of river Alaknanda with an average elevation of 560 meters (1837 feet) above sea level.<sup>[8,9]</sup>

Total numbers of 9883 individuals were considered medically fit for donating their blood. All individuals are more than 18 years in age. All blood collections had been taken either from voluntary donors at blood donation camps or as replacement donors at blood bank of the hospital. The blood samples were collected in vacutainer containing Ethylenediaminetetraacetic acid by the method of venepuncture. The tests were purely based on antigen-antibody agglutination test.

In ABO blood grouping (forward and reverse), monoclonal anti A, anti-A1, anti B, anti AB, anti H antisera, and A, B, O pooled cells are used. For Rh typing, anti-D (R0 and R1, anti IgM and blend of anti IgM and anti IgG) antisera were used. Finally, blood groups were selected only when both forward and reverse groups are identical. Rh-negative blood groups were confirmed

by anti-globulin technique and remaining all weak D groups were considered as Rh positive.

## Statistical analysis

Collected data were entered into Microsoft Excel and analyzed using Demo version. Descriptive statistical measures such as percentage and confidence interval were applied to identify relationship between the variables.

Allele frequencies were calculated under the assumption of Hardy-Weinberg equilibrium and the results were expressed as percentages, using the following equations:

$$p = 1 - \sqrt{(B + O)}, q = 1 - \sqrt{(A + O)}$$

$r = \sqrt{O}$ ,  $E = 1 - e$ ,  $e = \sqrt{dd}$  where p, q, r, E, and e represents the frequencies of the genes for A, B, O, Rh +ve, and Rh -ve, respectively.<sup>[10]</sup>

## Results

Observed and expected phenotype frequency patterns were found approximately same. Tables 1 and 2 shows that out of 9883 individuals of blood donors, majority were male individuals, i.e., 9133 (92.4%) and females were 750 (7.6%).

Maximum 58% blood donors were found to be in the age group of  $\leq 30$  years whereas 42% blood donors were in the age group  $> 30$  years. Blood group B was found highly prevalent (31.68%) followed by blood group A (30.39%), blood group O (26.24%), and AB (11.7%), respectively, i.e.,  $B > A > O > AB$  [Figure 1]. The positive Rhesus factor was shown to be the most prevalent (93.51%) and negative Rhesus factor was found in 6.49% [Figure 2].

Table 3 shows distribution of blood donors according to rhesus phenotype. Rhesus-positive male pattern was found to be shown as  $B > A > O > AB$ , which is similar to overall ABO blood group pattern but female pattern was found to be  $B > O > A > AB$ . Prevalence of Rhesus negative patterns in male and female were found to be  $B > A = O > AB$  and  $A > O > B > AB$ , respectively.

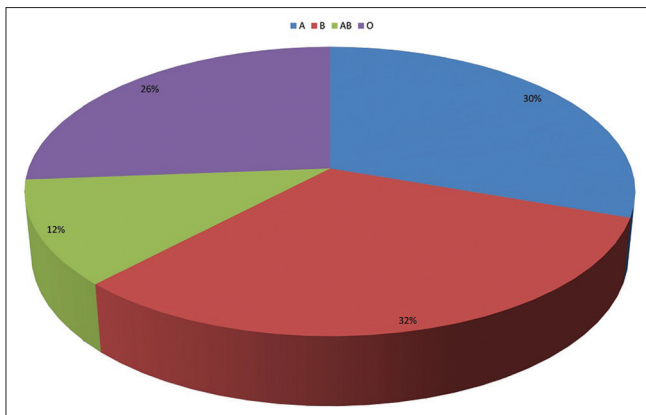
**Table 1: Comparison of observed and expected phenotype frequency among blood donors**

Blood group system	Phenotype	Observed frequency	Genotype	Expected frequency
ABO	A	0.3039	AA	0.3038
			AO	
	B	0.3168	BB	0.3147
			BO	
O	0.2624	OO	0.2623	
		AB		0.1170
		AB		
Rh	D+	0.9351	DD	0.9549
			Dd	
	D-	0.0649	dd	0.0649

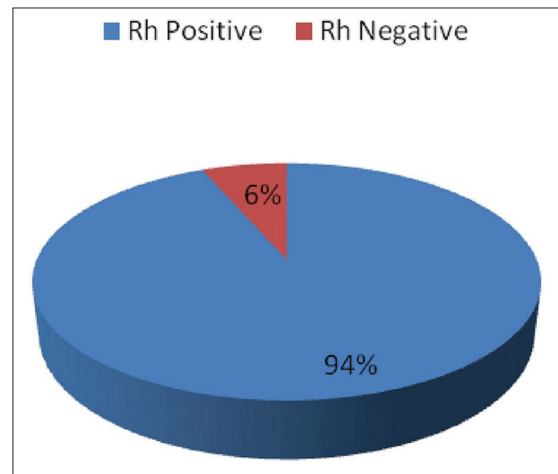
**Table 2: Distribution ABO and Rh blood group systems with respect to gender**

Blood group	Cases (%)		Total	Total cases		95% CI
	Male	Female		Prevalence rate		
A	2797 (30.6)	206 (27.5)	3003	30.39		29.5-31.3
B	2881 (31.5)	250 (33.3)	3131	31.68		30.8-32.6
AB	1079 (11.8)	77 (10.3)	1156	11.70		11.1-12.4
O	2376 (26.0)	217 (28.9)	2593	26.24		25.4-27.1
Total	9133 (92.4)	750 (7.6)	9883	100		NA
Rhesus (D) blood group						
Rh positive	8542 (93.5)	700 (93.3)	9242	93.51		93.1-93.9
Rh negative	591 (6.5)	50 (6.7)	641	6.49		06.1-06.9

CI: Confidence interval



**Figure 1: ABO blood group frequency with %**



**Figure 2: Rhesus factor frequency with %**

The present study has calculated the allele frequency of ABO and Rh blood group by following Hardy–Weinberg equation as shown in Table 4. The calculated allele frequency was found to be 0.2403 for I<sup>A</sup>, 0.2475 for I<sup>B</sup>, and 0.5122 for I<sup>O</sup>. In present study, Rhesus group allele frequencies for I<sup>D</sup> = 0.7452 and for I<sup>d</sup> = 0.2548 were found.<sup>[11-14]</sup>

Table 5 shows the variation in ABO blood group and Rhesus factor in Indian and International studies.<sup>[15-26]</sup> The present study exhibits that A, AB, and Rh negative blood groups are having the highest prevalence of 30.39%, 11.70%, and 6.49% respectively which is comparatively higher than previous similar studies. Another blood group in the present study B is having last second lowest prevalence (31.39%) when compared among Indian studies but international studies were found second highest prevalence percentage. Next blood group O and Rhesus positive factor are having lowest prevalence of 26.24% and 93.51% when compared among Indian studies but in international comparison were found lowest and second highest position in prevalence percentage, respectively, for the year 2016 onward.

### Discussion

There are large number of male donors compared to female donors; this has been observed in most of the studies in India being a developing nation. The main reasons behind it

were lack of education, social taboo, cultural habits, lack of motivation, and fear of blood donation.<sup>[27]</sup> A large section of female from the menstruating age groups were occasionally found anemic with low body weight, so they are considered unfit for donating blood and usually eliminated during the predonation screening and counseling. In this regard, the general health status of the female needs to be improved by providing proper nutritional diet and iron supplements. The fear regarding blood donation among Indian females needs to be driven out by educating them with the advantages of blood donations. Most of the older people suffer from hypertension, diabetes mellitus, low hemoglobin, and ischemic heart diseases and found unfit during predonation counseling.<sup>[28]</sup> In present study, we found significantly higher contribution of female donors. This may be because of higher literacy rate among the females of Uttarakhand in comparison to rest of India as exhibited in Tables 2 and 3.

The phenotype and genotype frequencies of ABO and Rh groups vary widely across different races and geographical areas of the world. Few studies have been done across India to find the variation. 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 4. The gene frequencies of ABO and Rh blood group found in

other four studies done by Raja *et al.*, Suresh *et al.*, Agrawal *et al.*, and Sidhu *et al.* are shown in Table 4.<sup>[11-14]</sup> The actual distribution of ABO blood group did not differ significantly from the calculated gene frequencies in Table 1.

Distribution pattern of blood groups of the present study was compared with the recent studies done across India at Andhra Pradesh,<sup>[15]</sup> Jharkhand,<sup>[16]</sup> Delhi and Uttarakhand,<sup>[17]</sup> Madhya Pradesh,<sup>[18]</sup> Karnataka,<sup>[19]</sup> and Assam.<sup>[20]</sup> The present study revealed that the most common blood group was “B” and the least common was “AB” which is similar to result found in

Jharkhand, Delhi, Uttarakhand, and Madhya Pradesh but differs from the other studies. In context to the recent international studies from African countries such as Ethiopia,<sup>[21]</sup> Uganda,<sup>[22]</sup> and Libiya,<sup>[23]</sup> from western Asian countries such as Iran<sup>[24]</sup> and Iraq,<sup>[25]</sup> and from south Asian country Bangladesh,<sup>[26]</sup> the result of the present study is similar to the study of Bangladesh specifying blood group “B” as the most common and “AB” as the least prevailing blood group, but in rest of the countries, the frequency of blood group O is highest which differs from the present study. Rh negativity status was 6.49% which is relatively higher as compared to the recent other studies across India but least among all the foreign nations except Uganda as shown in Table 5.

**Table 3: Distribution of ABO blood group with Rhesus factors with respect to gender**

Blood group	Cases (%)		Total cases	95% CI
	Male	Female		
A positive	2620 (28.7)	189 (25.2)	2809	27.5-29.3
B positive	2702 (29.6)	238 (31.7)	2940	28.9-30.7
AB positive	1020 (11.2)	71 (9.5)	1091	10.4-11.7
O positive	2200 (24.1)	202 (26.9)	2402	23.5-25.2
A negative	177 (1.9)	17 (2.3)	194	01.7-02.6
B negative	179 (2.0)	12 (1.6)	191	01.7-02.2
AB negative	59 (0.6)	6 (0.8)	65	0.52-0.84
O negative	176 (1.9)	15 (2.0)	191	01.7-02.2

CI: Confidence interval

**Table 4: ABO system and Rh phenotype allele distribution in different studies**

Authors	Year	I <sup>A</sup>	I <sup>B</sup>	I <sup>O</sup>	I <sup>D</sup>	I <sup>d</sup>
Present study	2017	0.2403	0.2475	0.5122	0.7452	0.2548
Raja <i>et al.</i> <sup>[11]</sup>	2016	0.1844	0.2477	0.5679	0.7794	0.2206
Suresh <i>et al.</i> <sup>[12]</sup>	2015	0.1398	0.2148	0.6454	0.7321	0.2679
Agrawal <i>et al.</i> <sup>[13]</sup>	2014	0.1653	0.2254	0.6093	0.7679	0.2321
Sidhu <sup>[14]</sup>	2003	0.1710	0.2700	0.5590	0.8360	0.1640

**Table 5: ABO and Rh phenotype comparison between national and international level studies for the year 2016 onwards**

Author	Sample size	Blood group (%)						
		A	B	O	AB	Rh+	Rh-	
<b>Indian studies</b>								
Srinagar, Uttarakhand	Present study	9883	30.39	31.68	26.24	11.70	93.51	6.49
Andra Pradesh <sup>[15]</sup>	Bhavani <i>et al.</i>	6942	20.00	35.80	36.90	7.30	96.28	3.72
Jharkhand <sup>[16]</sup>	Singh <i>et al.</i>	2055	22.09	35.15	34.73	8.03	96.46	3.54
Delhi <sup>[17]</sup>	Kaur <i>et al.</i>	15,446	22.60	37.80	29.50	10.10	94.47	5.53
Madhya Pradesh <sup>[18]</sup>	Mehta and Mehta	800	25.63	39.25	28.63	6.50	94.88	5.12
Uttarakhand <sup>[17]</sup>	Kaur <i>et al.</i>	6350	23.40	35.60	29.50	11.40	94.55	5.45
Karnataka <sup>[19]</sup>	Anushree <i>et al.</i>	1959	21.40	34.80	38.80	5.00	97.10	2.90
Aasam <sup>[20]</sup>	Islam Barbhuiya <i>et al.</i>	334	21.60	29.30	44.30	4.80	98.50	1.50
<b>International studies</b>								
Ethopia <sup>[21]</sup>	Zerihun and Bekele	6922	31.90	21.50	43.10	3.50	92.80	7.18
Uganda <sup>[22]</sup>	Apecu <i>et al.</i>	23,504	25.00	20.39	50.36	4.25	97.97	2.03
Libiya <sup>[23]</sup>	Saad	1306	31.17	23.43	37.44	8.96	83.92	16.07
Iran <sup>[24]</sup>	Torabizade Maatoghi <i>et al.</i>	29,922	28.48	24.71	40.21	6.60	92.38	7.62
Iraq <sup>[25]</sup>	Saleh and Abood	1268	23.11	21.45	48.03	7.41	88.56	11.44
Bangla Desh <sup>[26]</sup>	Verma <i>et al.</i>	937	26.57	34.15	29.67	9.61	90.82	9.18

### Conclusion

The present study concludes that the most common blood group is “B” and “AB” is the least common among the donors in the region of Garhwal hills at Srinagar, Uttarakhand, India. Rh positive were 93.51% and Rh negative were 6.49%. Awareness about donation of blood has to be created in order to increase the number of female donors. The data obtained in our present study when read in context of several other studies of different regions of India and abroad may be useful in policy making and policy implementation to face the future health challenges.

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### Conflicts of interest

There are no conflicts of interest.

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