

## Prevalence of Rh phenotype in voluntary blood donors of Uttarakhand

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**Abstract:** Karl Landsteiner and Wiener discovered Rhesus (Rh) system in the year 1940. Due to its immunogenicity along with A, B antigens, Rh D antigen testing was made mandatory in pre-transfusion testing. Presently there are more than 50 antigens in Rh blood group system but major ones are D, C, E, c, and e. The most important irregular red blood cell alloantibodies in daily transfusion practice are directed towards the Rh(antiD, -C, -E, -c and -e), KEL Therefore it is important to match the rare antigens before issuing blood so as to prevent complications. Our study aims to find out the Rh phenotype among the voluntary blood donors of Uttarakhand. This is a prospective cross-sectional study was carried out on 500 healthy voluntary blood donors from January 2016 to July 2017 at our blood bank. Donors were grouped and typed for ABO and Rh major antigens using monoclonal blood grouping reagents and statistical analysis was carried out. A total of 500 blood samples from voluntary blood donors were phenotyped. Among Rh antigens, Rho was the most common antigen (98.2%), followed by RhD-94.2%, RhC-88.6%, Rhc-54.8% and RhE-18.6% with DCe/DCe (36.2%) being the most common phenotype and the least common phenotype is dce/dce and dCE/dCE (0.2%). Database for antigen frequency of Rh blood group system in local donors helps to provide antigen negative blood to patients and thereby improve blood safety.

**Key-Words:** Alloimmunization, Antigen, Blood group, Phenotype, Rh

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### I. Introduction

Blood transfusion is an important part of supportive care in every hospital. The blood transfusion services aim to ensure adequate and safe blood to minimize the transmission of transfusion associated infections and development of transfusion reactions.

The credit of safe blood transfusion goes to discovery of ABO blood group system by Karl Landsteiner in 1900-1901 and later on the discovery of RhD blood groups in association with Alexander S Wiener after 30 years<sup>1</sup>. Today at least 33 blood group systems and more than 600 different blood group antigens are known<sup>2</sup>. But, ABO and Rh systems are the most widely used and the clinically most significant blood group systems. Out of all the 58 Rh antigens, 5 main Rh red cell antigens D, C, c, E, and e which causes clinically most significant transfusion complications are determined<sup>3</sup>. After anti-D Rh antibodies, the most commonly found antibody in the sera of allo-immunized individuals are anti-E > anti-c > anti-e > anti-C<sup>4</sup>. The aim of this study is to find the prevalence of these important Rh antigens in the population of Uttarakhand, a small hilly state of northern India, so that the feasibility of providing antigen compatible blood to those needy patients can be worked out. It will also help in reducing the time lag in providing immunologically safe blood to such recipients. Few studies are available from North India, this would be first of its kind in Uttarakhand region.

### II. Material and method

This study was conducted in the blood bank of a tertiary care hospital Shri Mahant Indires Hospital (SMIH) Dehradun of Uttarakhand. This was a prospective study conducted over a period of two years from June 2015 to June 2017. The study included 500 healthy donors from in-house as well as the donors who donated blood in various blood donation camps conducted by the blood bank.

**Study Design:** Prospective study

**Study Location:** This was a tertiary care teaching hospital based study done in blood bank of Shri Mahant Indires Hospital under the department of pathology Shri Guru Ram Rai Institute of Medical and Health Sciences Dehradun Uttarakhand.

**Study Duration:** June 2015 to June 2017

**Sample size:** 500 voluntary blood donors.

**Sample size calculation:** Blood bank of shri Mahant IndiresH Hospital collects about 10,000 units of blood annually from voluntary blood donors in house or by holding voluntary blood donation camps. Out of 20,000 donors 500 sample were selected randomly for study.

**Subjects & selection method:** The study population was drawn from about 20,000 voluntary blood donors attended the in house or voluntary blood donation camps organized by the SMI Hospital from June 2015 to June 2017. Five hundred sample were randomly selected for studding the Rh phenotyping.

**Inclusion criteria:**

1. Voluntary blood donors
2. Either sex
3. Aged 18 - 60 years,
4. Weight > 45 Kg
5. Haemoglobin > 12.5gm%
6. Afebrile and pulse rate 70-100/minute
7. Blood pressure - normotensive
8. Time interval between two donations at least 12 weeks.

**Exclusion criteria:**

1. Pregnant women;
2. Positive history (H/O) of jaundice in the past
3. Positive history of epilepsy,
4. H/O allergic condition,
5. H/O unexplained weight loss or sexually transmitted disease (STDs)
6. H/O tuberculosis, hypertension, diabetes, malignant disease
7. H/O long term fever or typhoid in the past 1year or ,malaria in the past 6 months
8. H/O surgeries in past 6 months
9. H/O recent drug intake in last 72 hrs
10. H/O of any recent blood or blood component transfusion in the past 6 months
11. H/O abnormal bleeding tendencies or blood coagulation disorders like haemophilia
12. H/O taking anti- arrhythmic drugs, anticonvulsants, anticoagulants, anti-thyroid drugs, digitalis, immunosuppressant, sedatives or tranquilizers in high doses, vasodilators etc.

**Procedure methodology :**

After written informed consent obtained for voluntary blood donation , a well-designed questionnaire was used to collect all the required information. Donor selection is strictly based on standard donor selection criteria which include brief medical history, limited physical examination and simple laboratory tests. Donor's age ranges from 18 to 60 years, weight >45 kg, Hb>12.5 gm/dl, afebrile, pulse rate 70-100/minute, normotensive and a time interval between two donations at least 12 weeks. Persons with history of jaundice or hepatitis in the past, epilepsy, allergic condition, unexplained weight loss, sexually transmitted disease (STDs), tuberculosis, hypertension, diabetes, malignant disease, long term fever or typhoid in the past 1year,malaria in the past 6 months, surgeries in past 6 months, recent drug intake in last 72 hrs, any recent blood or blood component transfusion in the past 6 months; were excluded along with permanent deferral of donors with abnormal bleeding tendencies or blood coagulation disorders like haemophilia, donors on anti-arrhythmic drugs, anticonvulsants, anticoagulants, anti-thyroid drugs, digitalis, immunosuppressant, sedatives or tranquilizers in high doses, vasodilators etc. Weight, pulse rate, blood pressure and oral temperature were determined appropriately . Hb was measured by finger prick on ' Hemocue' haemoglobinometer .

In the laboratory ABO blood groups were determined by forward and reverse blood grouping using blood group sera from Orthodiagnositics.The Rh blood grouping of blood donors were determined by test tube method using antisera RhD, Rhc, RhC, RhE and Rhe from Tulip. Agglutination is indicative of a positive test result and confirms the presence of C or c or E and / or e antigen in donor serum.. No agglutination means a negative test result and indicated the absence of C or c or E and / or e antigen.

**Statistical analysis:**

Statistical analysis was carried out using SPSS version 16.Descriptive statistics for categorical variable were performed computing the frequencies (percentages) in each category. Chi-square test was performed wherever required.

### III. Results

Maximum number of blood donors were in 18-25 years of age-group accounting for 399/500 (79.8 %), followed by 26-35 years 51/500(10.2%), 36-45 years 38/500 (7.6%) and  $\geq 46$  years of age 12/500 (2.4 %). Four hundred and forty one. (88.2 %) were males and 59 (11.8%) were females (**Table 1**).

**Table 1:** Showing the age and sex distribution of donors (N=500)

Age Group frequency in years	Male	Female	Total number of blood donors
18-25	350	49	399
26-35	45	6	51
36-45	35	3	38
$\geq 46$	11	1	12
TOTAL	441	59	500

ABO blood group frequency pattern was as follows B>O>A>AB. The commonest B group contributing **36.2%** followed by **O (29%)**, **A (24.8%)** and **AB (10%)** respectively. Rh antigen frequency pattern was as follows Rhe>RhD>RhC>Rhc>RhE. The commonest Rh antigen was 'Rhe' contributing **98.2%** followed by **RhD (94.2%)**, **RhC (88.6%)**, **Rhc (54.8%)** and **RhE (18.6%)** respectively (**Table 2**).

**Table 2:** Distribution of various Rh antigens in the donor population.

Rh antigens	Number of blood donors (N=500)	Percentage (%)
D	471	94.2
C	444	88.6
c	273	54.8
E	93	18.6
e	491	98.2

Rh antigen 'e' had the maximum frequency in both genders (98.2%) while 'E' showed the minimum frequency (18.6 %). Rh antigen frequency pattern was e>D>C>c>E in both males and females. In RhD positive donors the most common phenotype was DCCee which had 36.2% incidence rate and DCe/DCe was the most common genotype. In RhD negative donors, the most common phenotype was dCcee with the prevalence rate of 2.6% and dCe/dce was the most common genotype (**Table 3**).

**Table 3:** Showing incidence of phenotypes and genotypes of various Rh antigen in donor population.

D	C	C	E	e	Phenotype	Incidence (N=500)	Incidence (%)	Most probable genotype
+	+	-	-	+	DCCee	181	36.2%	DCe/DCe
+	+	+	-	+	DCcee	162	32.4%	DCe/Dce
+	-	+	-	+	Dccee	41	8.2%	Dce/Dce
+	+	+	+	+	DCcEe	37	7.4%	DCe/DcE
+	+	-	+	+	DCCEe	31	6.2%	DCe/DCE
+	-	+	+	+	DccEe	11	2.2%	DcE/Dce
+	-	+	+	-	DccEE	03	0.6%	DcE/DcE
+	+	+	+	-	DCcEE	03	0.6%	DcE/DCE
+	+	-	+	-	DCCEE	02	0.4%	DCE/DCE
-	+	+	-	+	dCcee	13	2.6%	dCe/dce
-	+	-	-	+	dCCee	09	1.8%	dCe/dCe
-	+	-	+	+	dCCeE	03	0.6%	dCe/dCE
-	+	+	+	+	dCcEe	02	0.4%	dCE/dCe/dcE/dce
-	+	-	-	+	dCCEE	01	0.2%	dCE/dCE
-	-	+	-	+	dccee	01	0.2%	dce/dce
-	-	+	+	+	dccEe	Nil		dce/dcE
-	-	+	+	-	dccEE	Nil		dcE/dcE
-	+	+	+	-	dCcEE	Nil		dcE/dCE

#### IV. Discussion

Karl Landsteiner's discovery of ABO blood group system opened the door to the birth of a wide spectrum of discoveries in the field of immunohaematology. After the ABO system, the Rh system is regarded as the second most important blood group system because of its association with hemolytic transfusion reactions and hemolytic disease of the new born.

During the study, most common age group blood donors was 18-25 years contributing about 79.8 % (399/500) of the total blood donors. It was followed by 26-35, 36-45 and  $\geq 46$  years of age group contributing 10.20 % (51/500), 7.60 % (38/500) and 2.4 % (12/500) of the total donors respectively. In a study by **Patel et al.** about 51.9 % (2764/5316) of blood donors belonged to 31- 40 years of age group<sup>5</sup>. **Kotwal et al.** also reported that 89.09% donors were below 40 years of age with mean age of 29.87 years<sup>6</sup>. In a study done in Pakistan by **Karim et al.** the median age of donors was 35 years<sup>7</sup>. This is due to fact that the younger age groups are main strength of any society and many blood donation camps are organised in educational institutions. Moreover, younger people have greater awareness, are more fit and therefore, less number of deferrals. So, they are the most common age group encountered for donating blood. Demographic variation also plays an important role eg, in India most of the population is young as compared to that in Japan where the major population belongs to older age group.

In our study, the commonest blood group was 'B' contributing **36.2 %** (181/500) followed by blood groups 'O', 'A' and 'AB'. Blood group 'O' was found in **29%** (145/500), 'A' in **24.8%** (124/500) and 'AB' in **10%** (50/500) of the total 500 donors included. In studies from various parts of India, like by **Patel et al., Aggarwal et al., Garg et al., Sarkar et al. and Itgappa et al.** blood group 'B' was found to be the commonest<sup>5,8-11</sup>. **Karim et al. from Pakistan** documented that blood group O was the most common accounting for 37% followed by B, A and AB comprising of 31%, 21% and 11% respectively<sup>7</sup>. This may be because of geographical variation.

In the present study frequency of Rhe antigen was the highest accounting to 98.2% (491/500) in the study population (Table 2). High frequency of Rhe antigen is seen in most of the population of the world including whites and blacks accounting to approximately 98%<sup>12</sup>. **Aggarwal et al., Garg et al., Sarkar et al. and Thakral et al.** reported similar frequency of Rhe antigen<sup>8-10, 13</sup>. **Sharma et al.** from Central India also documented the incidence of Rhe as 78.5%<sup>14</sup>.

The overall positivity of Rh D antigen was **94.2%** (471/500) while RhD negative blood group accounted for **5.8%** (29/500) of the total donors (Table No. 2). Most of the studies in India showed the Rh positivity within the range of 85- 99%. **Aggarwal et al., Thakral et al., Sharma et al.** documented RhD positivity more or less similar to our study i.e. 94.36%, 84.76% and 91.6% respectively [8, 13, 14]. The prevalence of RhD antigen has a huge variation in the world ranging from 60% to 99%. In people of Japan and Myanmar, Rh D has the highest incidence accounting for 99-100% while it is minimum in the population of Southern France and Northern Spain which ranges from 60-80%<sup>10</sup>. In Pakistan, **Karim et al.** reported 97% RhD positive donors<sup>7</sup> while in Bangladesh it was 94.6% as reported by **Shil et al.**<sup>15</sup>.

RhE is the least common Rhesus antigen worldwide. In the present study prevalence of RhE is 18.6% (93/500) (Table No. 2) which is in concordance to other studies from the rest of India **Thakral et al., Sharma et al. and Sarkar et al.** reported the incidence of RhE antigen as 17.9%, 25.6% and 26.55% respectively<sup>10,13, 14</sup>.

Prevalence of RhC antigen in this study was 88.6% (444/500) (Table No. 2) which is in concordance to other studies reported from Pakistan (87%), India (89.5%) and China (93%)<sup>7</sup>.

Prevalence of Rhc varies among different population of the world. In the present study its incidence is 54.8% (Table No. 2). In studies from Asia its incidence is reported as 52.8%, 57%<sup>17</sup>. In people of Africa and Europe it occurs in a frequency of 96% and 80% respectively<sup>12</sup>.

**Garg et al.** reported that the most common Rh antigen was Rhe (98.7%) which was followed by RhD (93.8%), RhC (91.8%), Rhc (55.2%) and RhE (21.1%) which were more or less similar to the present study<sup>9</sup>. **Gundrajukuppam et al., Thakral et al.** also documented the frequency of Rh antigen as follows Rhe>RhD>RhC>Rhc>RhE which was in concordance to our study<sup>4,13</sup>. In Pakistan **Karim et al.** reported that the most common Rh antigen was e [99%] followed by D [97%], C [87%], c [57%] and E [19%] which was similar to the present study<sup>7</sup>.

Phenotype was computed of the study subjects depending upon the expression of different Rh antigens. The most common phenotype was DCCee accounting for 36.2% (181/500) of total and the least common was DCCEE comprising 0.4% (2/500) among RhD positive subjects. dCcee (2.6%) was the most common and dccee (0.2%) was the least common among Rh negative donors [Table no. 3]. Most common phenotype reported in whites was DCCee - 42% while it was Dccee - 44% in black. The results found in the present study was in concordance to the studies carried out in different parts of India as well as other parts of Asia. **Thakral et al., Garg et al.** from northern India, **Sharma et al.,** from central India, **Kahar et al. and Gundrajukuppam et al.,** from the Southern part of India reported that DCe/DCe was the most common genotype which is similar to the

present study<sup>4, 9,13,14,16</sup>. In Pakistan, **Karim et al.** documented that DCE/DCE was the most common phenotype which is similar to the present study<sup>7</sup>.

## V. Conclusion

The study has a significant implication regarding the management of blood bank and transfusion services in this area. Knowledge of ABO and Rh blood group distribution is also important for clinical studies, for reliable geographical information and for forensic studies in the population. Multiple transfusions may develop alloantibody against some of the rare antigens (CcEe and kell). The most important irregular red blood cell alloantibodies in daily transfusion practice, in terms of frequency of occurrence, are directed towards the RH (anti-D, -C, -E, -c and -e), KEL. These irregular allo-antibodies can cause immunogenic haemolytic reactions following the subsequent transfusions. Therefore it is important to properly match the Rh antigens before issuing of the blood. Antigen typing of the various blood group systems will help in preparation of indigenous screening cell preparation and identification panels. Antigen phenotype frequencies and awareness of incidence of irregular antibodies in a given population is helpful in creating a blood donor data base which not only provides data about the availability of human blood in case of regional calamities, but is also helpful for preparation of indigenous cell panels, and also providing the compatible and safe blood to the needy patients.

## References

- [1]. Owen R. Karl Landsteiner and the first human marker locus. *Genetics* 2000;155(3):995-8.
- [2]. Lögdberg L, Reid ME, Lamont RE, Zelinski T. Human blood group genes 2004: chromosomal locations and cloning strategies. *Transfus Med Rev* 2005 Jan 31;19(1):45-57.
- [3]. Flegel WA. The genetics of the Rhesus blood group system. *Blood Transfusion* 2007;5(2):50.
- [4]. Gundrajukuppam DK, Vijaya SB, Rajendran A, Sarella JD. Prevalence of Principal Rh Blood Group Antigens in Blood Donors at the Blood Bank of a Tertiary Care Hospital in Southern India. *J Clin Diagn Res* 2016;10(5):EC07-EC10.
- [5]. Patel Piyush A, Patel Sangeeta P, Shah Jigesh V, Oza Haren V. Frequency and distribution of Blood Groups in Blood Donors in Western Ahmedabad – A Hospital based study. *National J Med Res* 2012; 2(2):207-10.
- [6]. Kotwal U, Raina TR, Sidhu M, Dogra M. Distribution of ABO & Rh (D) Blood Groups Among Blood Donors of Jammu Region with Respect to Various Ethnic Groups. *Journal of Medical Thesis* 2014;2(1):31-4.
- [7]. Karim F, Moiz B, Muhammad FJ, Ausat F, Khurshid M. Rhesus and Kellphenotyping of voluntary blood donors: foundation of a Donor data bank. *Journ Coll Physicians Surg Pak* 2015;25(10):757-60.
- [8]. Agarwal N, Thapliyal RM, Chatterjee K. Blood group phenotype frequencies in blood donors from a tertiary care hospital in north India. *Blood Res* 2013;48:51-4.
- [10]. Garg N, Singh DK, Tomar R, Singh B. Phenotype Prevalence of Blood Group Systems (ABO, Rh, Kell) in Voluntary, Healthy Donors- Experience of a Tertiary Care Hospital in Delhi, North India. *J Blood Disord Transfus* 2015;6:297.
- [11]. Sarkar RS, Philip J, Mallhi RS, Yadav P. Proportion of Rh phenotypes in voluntary blood donors. *Med J Armed Forces India* 2013;69(4):330-4.
- [12]. Itgappa M, Chandrappa MS, VidyaNadigere M, Raju GM, Dinesha R, Thammannagowda SS. Variation of ABO and Rh Blood Groups Among Medical Students of SSIMS & RC, Davanagere, Karnataka, India. *International Journal of Pharmaceutical and Clinical Research* 2011;3(4):70-2.
- [13]. Daniels G. Human blood groups. Chichester, West Sussex: John Wiley & Sons; 2013.
- [14]. Thakral B, Saluja K, Sharma RR, Marwaha N. Phenotype frequencies of blood group systems (Rh, Kell, Kidd, Duffy, MNS, P, Lewis, and Lutheran) in north Indian blood donors. *Transfus Apher Sci* 2010;43(1):17-22.
- [15]. Sharma DC, Singhal S, Rai S, Iyenger S, Sao S, Jain B. Incidence of Rh antigens, phenotype and probable genotype in the population of Gwalior and Chambal region, Central India. *Int Blood Res Rev* 2013;1:29-43.
- [16]. Shil N, Sultana N, Sormin S. Study of Rhesus Genotype and Phenotype in Bangladeshi Population Attended in a Tertiary Care Hospital Transfusion Medicine. *AKMMC J* 2017;7(2):25-8.
- [17]. Kahar MA, Patel RD. Phenotype frequencies of blood group systems (Rh, Kell, Kidd, Duffy, MNS, P, Lewis, and Lutheran) in blood donors of south Gujarat, India. *Asian J Transfus Sci* 2014;8(1):51-5.

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