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**Original Research Article**

# Distribution of RH and Kell (K) blood group antigens among blood donors in a tertiary care hospital of Jammu region, India

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## ABSTRACT

**Background:** Knowledge about the frequency of red cell antigen phenotype is very important for the creation of donor data bank and to minimize the risk of alloimmunization. This requires the determination of immunological characteristics of blood products and blood recipients by performing phenotyping of clinically significant blood group antigens. The aims and objectives were to study the distribution of Rh and Kell (K) antigen among blood donors of different ethnic groups in a tertiary care hospital.
**Methods:** This was prospective observational cross sectional one-point analysis study which was carried out over a period of one year with effect from November 2015 to October 2016 in the Postgraduate Department of Immunohematology and Blood Transfusion Medicine, Shri Maharaja Gulab Singh (SMGS) Hospital, Government Medical College, Jammu and Kashmir, India. It comprised of voluntary and replacement donors and categorized into different ethnic groups i.e Dogras, Gujjar Muslims, Non-Gujjar Muslims, Kashmiri Pandits, Sikhs and Christian. Donors selection criteria was as per Drug and Cosmetic Act.
**Results:** A total of 500 (Five hundred) blood samples from the donors of all blood groups were typed for the presence of Rh (D, C, E, c, e) and Kell (K) antigens. Out of these 500 samples, 420 were antigen typed by conventional tube technique and 80 samples were typed by column agglutination technique using glass beads. As per ethnicity, maximum donors were Dogras (74%) followed by Non-Gujjar Muslims (9.4%), Gujjar Muslims (9%), Sikhs (5.6%), Kashmiri Pandits (1.4%) and Christians were the least in frequency (0.6%). On phenotyping for Rh and Kell antigens 'e' antigen have the ubiquitous distribution and was found to have the highest frequency 486 (97.2%) followed by 'D' antigen 472 (94.4%), 'C' antigen 426 (85.2%), 'c' antigen 320 (64.0%) and 'E' antigen 103 (20.6%). Overall frequency of Kell (K) antigen was 2.6%.
**Conclusions:** Knowledge of red cell antigen phenotype frequencies in a population with different ethnic groups can help in creating donor data bank and database for the distribution of blood groups for preparing inhouse cell panels and providing proper antigen compatible blood for patients with multiple alloantibodies and also reduce the risk of RBC antigen alloimmunization along with their complications.

**Keywords:** Alloimmunization, Antibodies, Hemolytic transfusion reaction

## INTRODUCTION

The ultimate goal of blood transfusion services is to ensure safe and adequate blood supply to the patient and to minimize the development of transfusion reactions. In red blood cell transfusion, transfused RBCs should have an acceptable survival rate, and there should be no significant destruction of the recipient's own RBCs.

Blood transfusion can carry immediate or delayed immunological reactions, the most common and most serious is hemolytic transfusion reaction by antibody incompatibility. Knowledge about the frequency of red cell antigen phenotype is very important for the creation of donor data bank and to minimize the risk of alloimmunization. This requires the determination of immunological characteristics of blood components and blood recipients by performing immune-haematology analysis such as phenotyping.[1] Antibodies directed against Rh, Kell, Kidd, Duffy and MNSs blood group systems are implicated in cases of hemolytic transfusion reactions (HTRs) and hemolytic disease of fetus and newborn (HDFN) and therefore, regarded as clinically significant if these react in the indirect antiglobulin test at 37ºC.[2] In multi transfused patients and antenatal patients it is recommended that Rh and Kell matched blood should be transfused. This is possible only if know the frequencies of the antigens in the donor population. Hence, the present study was undertaken to determine the distribution of Rh and Kell among different ethnic groups in this tertiary care hospital. The aims and objectives were to study the distribution of Rh and Kell (K) antigen among blood donors of different ethnic groups in a tertiary care hospital.

## METHODS

This study was a prospective observational cross sectional one-point analysis study and was carried out over a period of one year in the Postgraduate Department of Immunohematology and Blood Transfusion Medicine SMGS Hospital of Government Medical College, Jammu and Kashmir, India which is a tertiary care hospital. This study was duly approved by Institutional Ethics Committee. This was the first report describing the frequencies of clinically significant blood groups like Rh (D, C, E, c, e) and Kell (K) from Jammu and Kashmir, India.

It comprised of voluntary and replacement donors of SMGS Hospital, belonging to different ethnic groups i.e., Dogras, Gujjar Muslims (GMs), Non-Gujjar Muslims (NGMs), Kashmiri Pandits (KPs), Sikhs and Christians. Donors who fulfill inclusion criteria specified by DGHS and Departmental SOPs were selected and written informed consent was taken from them.[3]

### *Sample size and sample collection*

Samples were selected by simple random sampling because it eliminates bias by giving all individuals an equal chance to be chosen. 500 (Five hundred) samples were collected from voluntary and replacement donors. Consent of the donor was taken prior to phlebotomy. Blood collection procedure (phlebotomy) was performed as per transfusion medicine, technical manual, DGHS and this Departmental SOP.[3] About 6 ml of blood was collected in the pilot tube of which about 3 ml in red top vial and 3 ml in purple top vial (K3EDTA). Tube containing K3EDTA as anticoagulant were chosen for antigen typing, as K3EDTA samples up to 14 days old are suitable for typing when stored at 2-8ºC. All samples were processed for antigen typing within 48 hours of collection. Out of '500' samples, '420' samples were antigen typed by conventional tube technique using anti seras from ortho-clinical diagnostics and '80' samples were antigen typed by column agglutination technique using glass beads Rh-Kell phenotyping cassettes from ortho-clinical diagnostics as the Ortho TM Workstation was introduced at that time in this blood bank.

The data was analysed with the help of computer software SPSS for Windows version 17.0.The data was presented as percentage in tabular and appropriate diagrammatic forms.

## RESULTS

A total of 500 donors belonging to different ethnic groups i.e., Dogras, Non-Gujjar Muslims (NGM), Gujjar Muslims (GM), Sikhs, Kashmiri Pandits (KPs) and Christians as shown in Table 1.

**Table 1: Distribution of different ethnic groups of study population.**

| Ethnic groups | No. of donors | % Donors |
|---|---|---|
| Dogras | 370 | 74% |
| Non-Gujjar Muslims | 47 | 9.4% |
| Gujjar Muslims | 45 | 9% |
| Sikhs | 28 | 5.6% |
| Kashmiri Pandits | 07 | 1.4% |
| Christians | 03 | 0.6% |
| Total | 500 | 100% |

On phenotyping for Rh antigens frequency of 'e' antigen 97.2% (486) followed by 'D' antigen 94.4% (472), 'C' antigen 85.2% (426), 'c' antigen 64.0% (320) and 'E' antigen 20.6% (103) (Table 2).

Eight of phenotypes Rh blood group system were found. DCe/DCe ($R_1R_1$) has the highest incidence (36.0%) followed by $R_1r$ (34.6%), $R_1R_2$ (14%), rr (5.0%), $R_2r$ (3.8%), $R_0r$ (3.2%), $R_2R_2$ (2.8%) and least common phenotype among Rh antigens was dCe/dce (r'r) i.e., 0.6% in this study.

Thus, as evident from Table 3, $R_1R_1$ phenotype was highest among Sikhs (39.28%), $R_1r$ among Christians (66.6%), $R_1R_2$ among Gujjar Muslims (20%), R2r among Kashmiri Pandits (14.28%), R2R2 among Christians (33.3%), R0r among Gujjar Muslims (4.44%) andrr among Gujjar Muslims (11.11%), r'r (0.81%) among Dogras population.

Overall frequency of Kell (K) antigen was 2.6% (Table 4). So, percentage of Kell (K) positive antigen was highest among Gujjar and Non-Gujjar Muslims (4.25%).

In this study, the percentage of different ABO blood groups among 500 blood donors was 'A' 122 (24.4%), 'B' 180 (36%), 'O'134 (26.8%) and 'AB' 64 (12.8%). Kell (K) antigen was most frequent among 'A' blood group donors (3.27%) followed by 'AB' group (3.1%), 'O' group (2.98%) and 'B' group (1.66%). In this study, all K antigen positivity was seen among RhD positive donors (2.6%).

**Table 2: Distribution of Rh antigens among different ethnic groups.**

| Antigen present | Dogras (n=370) | NGM (n=47) | GM (n=45) | Sikhs (n=28) | KPs (n=7) | Christians (n=3) | Total |
|---|---|---|---|---|---|---|---|
| D | 352 (95.1%) | 42 (89.36%) | 40 (88.8%) | 28 (100%) | 7 (100%) | 3 (100%) | 472 |
| C | 323 (87.29%) | 38 (80.85%) | 34 (75.55%) | 24 (85.71%) | 5 (71.4%) | 2 (66.6%) | 426 |
| E | 73 (19.72%) | 9 (19.14%) | 13 (28.8%) | 5 (17.8%) | 2 (28.5%) | 1 (33.33%) | 103 |
| c | 227 (61.35%) | 32 (68.08%) | 36 (80%) | 17 (60.7%) | 5 (71.4%%) | 3 (100%) | 320 |
| e | 362 (97.8%) | 46 (97.8%) | 44 (97.77%) | 26 (92.8%) | 6 (85.71%) | 2 (66.66%) | 486 |

NGM-Non-Gujjar Muslims, GM- Gujjar Muslims, KPs- Kashmiri Pandits.

**Table 3: Distribution of Rh phenotypes among different ethnic groups.**

| Ethnic groups | $R_1R_1$ | $R_1r$ | $R_1R_2$ | $R_2r$ | $R_2R_2$ | $R_0r$ | Rr | r'r | Total (n) |
|---|---|---|---|---|---|---|---|---|---|
| Dogras | 38.6%% | 33.7% | 14.05% | 3.51% | 2.16% | 2.97% | 4.05% | 0.81% | 370 |
| NGMs | 31.91% | 34.04% | 14.89% | 2.12% | 2.12% | 4.25% | 10.6% | 0 | 47 |
| GMs | 20% | 35.55% | 20% | 6.66% | 2.22% | 4.44% | 11.11% | 0 | 45 |
| Sikhs | 39.28% | 39.2% | 7.14% | 3.57% | 7.14% | 3.57% | 0 | 0 | 28 |
| KPs | 28.57% | 42.8% | 0 | 14.28% | 14.28% | 0 | 0 | 0 | 7 |
| Christians | 0 | 66.6% | 0 | 0 | 33.3% | 0 | 0 | 0 | 3 |

NGM-Non-Gujjar Muslims, GM- Gujjar Muslims, KPs- Kashmiri Pandits.

**Table 4: Distribution of Kell (K) antigen among different ethnic groups.**

| Ethnic groups | Kell (K) positive (%) | Kell (K) negative (%) | Total |
|---|---|---|---|
| Dogras | 2.16% | 97.8% | 370 |
| NGM | 4.25% | 95.7% | 47 |
| GM | 4.25% | 95.5% | 45 |
| Sikhs | 3.57% | 96.4% | 28 |
| KPS | 0 | 100% | 7 |
| Christians | 0 | 100% | 3 |
| Total | 2.6% | 97.4% | 500 |

NGM-Non-Gujjar Muslims, GM- Gujjar Muslims, KPs- Kashmiri Pandits.

## DISCUSSION

Knowledge of prevalence of different blood group antigens in any population is very important for blood banks and Transfusion services policies. The distribution of ABO, Rh and Kell antigen varies from race to race and among different ethnic groups. It is always helpful in managing the cases of multiple transfusions like thalassemia, sickle cell anemia, cancer patients, patients on dialysis etc., who are prone to develop antibodies against blood group antigens as it is not practically feasible to match all the blood group antigens before transfusion. Antigens of the major blood group systems plays a very important role in determining the outcomes of transfusion in recipients of blood and blood components. In those cases, where clinically significant antibodies are identified in patient's serum, antigen negative donor unit can easily be retrieved from the donor data based available blood groups in blood banks.

In this study, blood grouping is extended to major alloantigens of Rh (D, C, E, c, e) and Kell (K) systems where allogenic differences persists and multi transfused patients are not spared of alloimmunization. Present study focused on 500 voluntary and replacement donors who have made donations at SMGS Hospital Blood Bank. All these results have been compared with the data from other published articles from India and abroad.

The prevalence of ABO blood groups varies from race to race. It is the most clinically significant blood group system because of regular occurrence of antibodies of the

blood group system and ability of antibodies to cause hemolytic transfusion reaction and HDFN. The Rh blood group system is the most polymorphic next to ABO blood group system. It is the most clinically significant blood group system in transfusion medicine. The frequency of D positive (D+) in this study was 94.4% and D negative frequency was 5.6% which is in concordance with other studies conducted in India like Garg N et al, on healthy donors in Delhi showing the frequency of 93.8% D positive antigen and 6.2% D negative antigen.[4] In Agarwal N et al, study on blood donors at AIIMS, New Delhi, India shows that Rh (D) antigen was positive in 94.36% and negative in 5.64%.[5] Nanu A et al, shows the frequency of D+ in 95.37% and D- in 4.63%.[6] However, in Whites the frequency of D positive was 85% and D negative was 15% and in blacks, 92% population was D positive and 8% was D negative. The frequencies of other Rh antigens in this study were 'C' was positive in 426 (85.2%) donors, 'E' was positive in 103 (20.6%) donors, 'c' was positive in 320 (64.0%) donors and 'e' was positive in 486 (97.2%) donors. Thus, the frequencies of 'e' and 'C' antigens in this study were high similar to the study conducted by Thakral B et al, on north Indian blood donors at PGIMER Chandigarh, Punjab, India showing the frequency of 'e' antigen 98.3% and 'C' antigen 84.76%.[7] Present study results of Rh (C, E, c, e) antigen are also similar to the study conducted by Kahar MA et al, on donors of South Gujarat, India showing the frequency of 'e' antigen 100% and 'C' antigen 81.74%.[8] In Whites and Blacks, the frequencies of 'e' and 'c' was highest when compared to other antigens (Table 5). The study conducted by Gajjar M et al, on voluntary donors at Ahmedabad, Gujarat, India also showed high frequency of 'e' (98.65%) as this study followed by 'D' (94.76%), 'C' (88.82%),'c' (58.47%) and 'E' (17.18%).[9]

**Table 5: Comparison of distribution of Rh antigens (C, E, C, E) among blood donors.**

| Antigens | Present study | Kahar MA et al[8] | Thakral B et al[7] | Whites | Blacks |
|---|---|---|---|---|---|
| C | 85.2% | 81.74% | 84.76% | 68% | 27% |
| E | 20.6% | 21.74% | 17.9% | 29% | 22% |
| c | 64.0% | 56.52% | 52.82% | 80% | 96% |
| e | 97.2% | 100% | 98.3% | 98% | 98% |

**Table 6: Comparison of Kell (K) antigen among different studies.**

| Antigen | Present study (%) | Kahar MA et al[8] (%) | Agarwal N et al[5] (%) | Thakral B et al (%)[7] | Nanu A et al (%)[6] | Whites (%) | Blacks (%) |
|---|---|---|---|---|---|---|---|
| K+ | 2.6 | 6.09 | 1.97 | 5.68 | 4.04 | 8.80 | 2 |
| K- | 97.4 | 93.9 | 98.03 | 94.32 | 95.96 | 91.2 | 98 |

The frequencies of various phenotypes were not different from those conducted in various parts of India and abroad. In this study, $R_1R_1$ phenotype was the most common (36.0%) followed by $R_1r$ (34.6%) which are similar to other studies i.e., Garg N et al, shows the highest prevalence of $R_1R_1$ (44.60%) followed by $R_1r$ (32.60%).[4] Kahar MA et al, shows the highest frequency of $R_1R_1$ (40.87%) followed by $R_1r$ (23.48%).[8] Agarwal N et al, shows the frequency of $R_1R_1$ (42.93%) followed by $R_1r$ (35.60%).[5] Thakral B et al, shows the highest frequency of $R_1R_1$ (43.8%) followed by $R_1r$ (30%).[7] However, the $R_1R_1$ phenotype is present in only 17.6% of White population and 2.9% of Black population.

In Kell blood group system, Kell (K) antigen was found positive in 2.6% blood donors and negative in 97.4% in this study. This is in concordance with other studies i.e. Agarwal N et al, shows the frequency of 1.97% of K positive antigen and 98.03% of K negative antigen and K antigen was positive in 2% and negative in 98% among black population.[5] But this study results are lower than the studies conducted by Kahar MA et al, which shows the frequency of 6.09% of K positive antigen and 93.91% of K negative antigens and in Whites, the frequency of K positive was 8.80% and K neg was 91.2% (Table 6).[8]

Thus, the distribution of different blood group in this study were ABO (A=24.4%, B=36%, O=26.8%, AB=12.8%), rhesus (positive =94.4%, negative=5.6%) and Kell (K=2.6%). B blood group was found to be most common similar to other studies from North India, Agarwal N et al.[5] In Rh blood group system, D antigen frequency was found to be 94.4% which was comparable with other studies from India, Thakral B et al.[7] The most common phenotype found in our study was $R_1R_1$ (36.0%) followed by $R_1r$ (34.6%) and similar findings were obtained in the studies of Agarwal N et al, (42.93% and 35.60% respectively) and Thakral B et al, (43.8% and 30% respectively).[5,7] r'r (dCe/dce) phenotype frequency was least common (0.6%) similar to the study conducted by Thakral B et al, (0.56%).[7] The most common phenotypes in Whites was $R_1r$ (31.1%) and in Blacks $R_0r$ phenotype was found to be highest (22.9%).

The K antigen frequency was 2.6%, similar to that was reported in Blacks (2%) and Agarwal N et al, (1.97%) and Garg N et al, (1.6%) but lower than the studies conducted by Thakral B et al, (5.68%), Kahar MA et al, (6.09%) and that in Whites (8.8%).[4,5,7,8] Kell antigen frequency in this study was much lower than the study conducted in Maldives by Mohamed S et al, (5.7%).[10] The knowledge of antigen frequency in a given population is clinically important as one can predict the common alloantibodies that could be formed in patients who had received multiple transfusions such as in patients with Thalassemia, dialysis patients, cancer patients etc. and also helps in selection of antigen negative blood units for these patients who had developed the alloantibodies. It may also reduce the reported RBC antigens alloimmunization along with their possible complications such as hemolytic transfusion reactions (HTRs) and hemolytic disease of fetus and newborn (HDFN).

In this region (Jammu, India), majority of the population was formed by Hindus (62.55%), followed by Muslims (33.45%), Sikhs (3.3%), Christians (0.28%) and others (0.70%). Majority of the Hindus belongs to ethnicity called Dogras (67%) cited on November 2016 available at website: /wiki/Jammu%20and%20Kashmir.

Present study included very less number of some ethnic groups like Kashmiri Pandits and Christians, so, some Rh antigens and Kell (K) antigen were not found among these ethnic groups. So, larger number of samples of these ethnic groups were needed to include in this study.

## CONCLUSION

Thu knowledge of red cell antigen phenotype frequencies in a population with different ethnic groups can help in creating donor data bank and database for the distribution of blood groups for preparing inhouse cell panels and providing proper antigen compatible blood for patients with multiple alloantibodies and also reduce the risk of RBC antigen alloimmunization along with their complications.

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