ORIGINAL ARTICLE

FREQUENCY OF MINOR BLOOD GROUP ANTIGENS —A STEP TOWARDS SAFER TRANSFUSION

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Background: ABO and Rh blood typing is done routinely prior to transfusion, but weaker blood group antigens like Kell, Duffy, and Kidd also have clinical relevance because of their involvement in haemolytic transfusion reactions (HTRs) and haemolytic disease of the foetus and newborn (HDFN). There is sparse data regarding the frequency of these antigens in Pakistanis. This study aimed to find the prevalence of minor blood group antigens—Kell, Duffy, and Kidd—among male O blood group donors. **Methods:** A cross-sectional study was carried out in 200 male O blood group donors from Hussaini Blood Bank during April to June 2024. Routine serological methods were used to identify Rh (excluding 'e'), Kell (K, k), Duffy (Fya, Fyb), and Kidd (Jka, Jkb) antigens. Frequencies are presented as percentages and compared with pertinent Pakistani studies of the past decade. **Results:** Of all Rh antigens, D was most common (86%), followed by C (74%), c (68%), and E (28%). 'K' antigen was present in 4% of donors, while 'k' was present in 98%. Fya and Fyb antigens were present in 78% and 64% respectively. Jka and Jkb antigens were present in 77% and 68% of donors. These results were consistent with earlier national and regional findings. **Conclusion:** A sufficiently greater prevalence of clinically significant minor blood group antigens underscores the importance of prolonged antigen characterization in blood banks for improving transfusion safety.

Keywords: Blood banks, Duffy blood group protein human, Kell blood group protein human

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INTRODUCTION

Blood transfusion remains a foundation of modern medical practice, vital for managing a variety of clinical conditions ranging from surgical interventions to traumatic injuries and haematological disorders. The selection of compatible blood for transfusion is paramount, primarily focusing on the ABO and Rh blood group systems, which play a critical role in preventing haemolytic transfusion reactions (HTRs).[1] However, beyond these major blood groups, numerous minor blood group antigens exist, which, despite their nomenclature, possess significant clinical implications.[2]

The International Society of Blood Transfusion (ISBT) recognizes approximately 43 blood group systems encompassing 349 red cell antigens. Among these, the Kell, Duffy, and Kidd systems are notable for their clinical significance.[3] Antibodies to these blood group antigens have been implicated in immediate and delayed HTRs and in Haemolytic disease of the foetus and newborn (HDFNs), frequently characterized by a decrease in post-transfusion haemoglobin levels.[4,5] Consequently, patients with a history of alloimmunization or those requiring long-term transfusion support may necessitate antigen-free blood to lessen the risk of these complications.

The prevalence of minor blood group antigens varies significantly across different ethnicities and geographical regions.[6] Therefore, establishing population-specific data is crucial for improving blood transfusion practices and ensuring patient safety. In Pakistan, there is a paucity of comprehensive data regarding the prevalence of clinically significant minor blood group antigens. Recognizing this gap, this study aimed to investigate the frequency of Kell, Duffy, and Kidd antigens in male donors having O blood group. Furthermore, this study compared the findings with other researches conducted within the Pakistani population over the past 10 years. This approach allows us to better understand the local distribution of these antigens and its implications for transfusion medicine.

The selection of O blood group donors, known for their universal compatibility due to the absence of A and B antigens, was predicated on their critical role when matching blood is unavailable. While they lack major antigens, minor blood group antigens present in O blood can still trigger alloimmunization and HTRs, particularly in patients with transfusion histories.[7] Therefore, having comprehensive data on the prevalence of these antigens in this population is essential for developing strategies to provide antigen-negative blood to sensitized patients.

METHODOLOGY

This cross-sectional study, conducted at Hussaini Blood Bank from April to June 2024, after ethical approval from Baqai Medical University (ERB# 2024/0028) included 200 blood donors. Only

individuals with O blood type were selected after taking informed consent. To ensure a homogenous study population, only male blood donors meeting standard transfusion criteria, possessing a normal haemoglobin level (13.5–18 g/dL), and a negative DAT (direct anti globulin test) were included. O blood group was the sole inclusion criterion for blood type regardless of their Rh status. Female donors were excluded due to prevalent anaemia, and non-O blood groups were excluded to maintain focus on the 'antigen free' blood group.

ABO and Rh blood typing was conducted using the tube technique. Rh antigen detection was comprehensive, with the exception of 'e' antigen (due to reagent unavailability). Indirect anti-globulin testing, using Lorne Laboratories kits, was employed to analyze the Kell (K, k), Duffy (Fya, Fyb), and Kidd (Jka, Jkb) minor blood group antigens.

For statistics, SPSS-25 was used. The prevalence of minor blood group antigens was expressed in frequencies and percentages and were compared with other studies from Pakistan conducted in the last decade.

RESULTS

The study included 200 male blood donors having mean age of 28.56±7.38. The frequency of Rh antigens in the whole population showed 86% of D antigens followed by C (74%), c (68%) and E (28%). The antigen 'e' could not be explored because of the non-availability of reagents. (Table-1).

Out of 200 blood donors, 172 (86%) were Rh-positive and 28 (14%) were Rh-negative. When the individuals were segregated into Rh^+ and Rh^- separately, the order of frequency in Rh^+ was C>c>E. However, in Rh^- donors, all individuals showed 'c' antigen on their RBCs and none had 'C' or 'E' antigen, showing association of 'c' antigen with Rh^- blood group. Similarly, 'C' antigen was found to be more associated with Rh^+ blood group than Rh^- (86% and 0% respectively).

In the Kell blood group system, the 'K' antigen was observed at a frequency of 4%, while the 'k' antigen was 98%. Within the Duffy system, the 'Fya' antigen displayed a frequency of 78%, and the 'Fyb' antigen as 64%. In the Kidd system, the 'Jka' antigen was found at 77%, and the 'Jkb' antigen at 68%. The observed frequencies of Duffy and Kidd antigens (Fya, Fyb, Jka, and Jkb), ranging from 64% to 78%, indicate a high prevalence of these antigens within our study population. For the Duffy system, a single comparable study conducted in Islamabad aligns with our finding of a higher 'Fya' prevalence compared to 'Fyb'.

Phenotype frequency analysis demonstrated that K^-k^+ was the most prevalent phenotype in the Kell blood group system, followed by K^+k^+ and K^+k^-. In the Duffy system, Fya^+Fyb^+ was the most common phenotype, observed in 38% of samples, while Fya^+Fyb^- and Fya^-Fyb^+ were found in 32% and 20% of the samples, respectively. The Jka^+Jkb^+ phenotype was the most frequent in the Kidd system, followed by Jka^+Jkb^- and Jka^-Jkb^+ in this study.

Table-1: Frequency of Rh antigens in Rh^+ and Rh^- donors separately

Antigens	Rh^+ donors [n=172 (86%)]	Rh^- donors [n=28 (14%)]	Antigen frequency in population
C	148 (86%)	0 (0%)	74%
C	108 (62.8%)	28 (100%)	68%
E	56 (32.6%)	0 (0%)	28%

Table-2: Frequency of minor blood group antigens and comparison with other Pakistani studies

Antigens	Current Study	Study from Karachi[8]	Study from Islamabad[9]	Study from Lahore[10]	Study from Rawalpindi[11]	Study from Rawalpindi[12]
(Kell) K	8 (4%)	0%	4%	3.6%	4.05%	N/A
(Kell) k	196 (98%)	100%	N/A	96.3%	98.9%	N/A
(Duffy) Fy^a	156 (78%)	N/A	58.24%	N/A	N/A	N/A
(Duffy) Fy^b	128 (64%)	N/A	39.84%	N/A	N/A	N/A
(Kidd) Jk^a	154 (77%)	N/A	65.28%	N/A	N/A	73.2%
(Kidd) Jk^b	136 (68%)	N/A	42.24%	N/A	N/A	75.4%

N/A= Not available

Table-3: Phenotype frequency of minor blood group antigens and comparison with other Pakistani studies

Phenotype	Current Study	Study from Karachi[8]	Study from Islamabad[9]	Study from Rawalpindi[11]	Study from Rawalpindi[12]
K^+k^-	4 (2%)	0%	N/A	0.3%	N/A
K^-k^+	192 (96%)	100%	N/A	95.15%	N/A
K^+k^+	4 (2%)	0%	N/A	3.75%	N/A
K^-k^-	0	0%	N/A	0.8%	N/A
$Fy^{a+}Fy^{b-}$	64 (32%)	N/A	34.56%	N/A	N/A
$Fy^{a-}Fy^{b+}$	40 (20%)	N/A	15.36%	N/A	N/A
$Fy^{a+}Fy^{b+}$	76 (38%)	N/A	27.04%	N/A	N/A
$Fy^{a-}Fy^{b-}$	0	N/A	23.04%	N/A	N/A
$JK^{a+}JK^{b-}$	44 (22%)	N/A	40%	N/A	23.6%
$JK^{a-}JK^{b+}$	16 (8%)	N/A	13.6%	N/A	25.7%
$JK^{a+}JK^{b+}$	88 (44%)	N/A	27.52%	N/A	49.6%
$JK^{a-}JK^{b-}$	8 (4%)	N/A	18.88%	N/A	1.1%

N/A= Not available

DISCUSSION

Our findings for the Rh blood group system revealed a D antigen frequency of 86%, aligning with the expected high prevalence of Rh-positivity in most populations.[13,14] The 'e' antigen unfortunately could not be assessed due to reagent unavailability, although it is the most prevalent Rh antigen in Asian population.[15] The order of frequency for other Rh antigens was C>c>E. Similar results were seen in other Asian population where among C, c and E Rh antigens, C was the most prevalent Rh antigen and E was the least.[16–18] Interestingly, when the population was segregated by Rh subgroup, all Rh^- donors exhibited the 'c' antigen and lacked 'C' and 'E' antigens. This is in line with previously reported trends in Rh^- individuals, where c antigen was the most predominant antigen suggesting an association between the 'c' antigen and the Rh^- phenotype.[13,18] This observation warrants further investigation with larger sample sizes to confirm this trend within this population and explore its potential implications for Rh typing and antibody screening in Rh^- individuals.

The prevalence of Kell blood group antigens among the Pakistani population, revealed a high frequency of the 'k' antigen (98%) and a low frequency of the 'K' antigen (4%) in the Kell system. These findings align with previous Pakistani studies and are consistent with international data. For instance, a study in Saudi Arabia reported a 'K' antigen prevalence of 6.52% and 'k' antigen of 99.28% among blood donors.[19] A research in Eastern India found the 'K' antigen in 2.4% of donors with anti-K alloantibody being the 3rd most common, detected in multi-transfused thalassemia and sickle cell (SCD) patients.[20] These similarities suggest a consistent distribution pattern of Kell antigens across these regions.

In the Duffy blood group system, our study observed 'Fya' and 'Fyb' antigen frequencies of 78% and 64%, respectively. Only 1 comparable study from Islamabad was reported from Pakistan in the last 10 years showing dominance of Fya antigen over Fyb.[9] These results are comparable to findings from a study which reported 'Fya ' and 'Fyb' frequencies of 66% and 83% among Caucasians, and 10% and 23% among Black individuals.[21] The higher prevalence of Duffy blood group antigens in our population aligns more closely with the Caucasian data, indicating potential genetic similarities or admixture.

Regarding the Kidd blood group system, our findings showed 'Jka' and 'Jkb' antigen frequencies of 77% and 68%, respectively. These frequencies are consistent with data from other Pakistani studies.[9,12] Approximately similar frequencies of Jka and Jkb antigens are seen in studies from our neighbouring countries.[22,23]

The study's strengths include its contribution to the limited local data on minor blood group antigens, offering a baseline for both clinical practice and future research. The use of standardized serological methods ensured consistent antigen detection, and ethical, systematic sample collection further ensured the validity of the findings. These results have practical implications for blood bank inventory management and donor screening policies, potentially reducing the incidence of transfusion-related complications.

Nevertheless, the study has certain limitations that include its modest sample size and lack of ethnic stratification, which may affect the generic findings across Pakistan's diverse population. Sampling from selected centres may introduce bias, and the exclusive focus on Kell, Duffy, and Kidd antigens, omits other clinically relevant groups. The use of serological methods, rather than molecular techniques, may affect detection accuracy. The absence of clinical outcome data prevents assessment of the direct impact on alloimmunized patients.

CONCLUSION

The observed antigen frequencies in our study have significant implications for transfusion medicine. This underscores the need for expanded antigen profiling in the Pakistani population and highlights the importance of region-specific data in improving transfusion compatibility. Future studies with larger sample size, incorporating molecular techniques, and ethnic diversity are warranted to build upon these findings and further optimize transfusion safety.

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SS: Drafting of manuscript and statistical analysis **RPA:** Project conceptualization and simple collection **SS:** Drafting the manuscript
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