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The phenotypic profile of blood donors and recipients at the University Clinics of Kinshasa and the National Blood Transfusion Center of Kinshasa from November 2024 to July 2025

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ABSTRACT

Introduction

Blood transfusion is essential in the Democratic Republic of the Congo (DRC); however, comprehensive epidemiological data remain limited beyond the basic ABO and RhD systems. This study addresses this gap by providing data on extended Rhesus and Kell phenotyping.

Purpose

To determine the phenotypic distribution of ABO, Rhesus (D, C, c, E, e), and Kell blood group systems among blood donors and recipients at the University Clinics of Kinshasa and the National Blood Transfusion Center.

Methods

A descriptive cross-sectional study was conducted from November 2024 to July 2025. ABO blood group antigens were identified using both forward (cell) and reverse (serum) typing. Extended Rhesus and Kell antigens were determined using the direct serological method.

Results

A total of 240 samples from 214 subjects were analyzed (163 donors and 51 recipients). The mean age was 30.47 ± 15.08 years. The frequencies of blood groups O, A, B, and AB were 50.0%, 26.2%, 18.2%, and 5.6%, respectively. The RH1 (D) antigen was present in 95.3% of subjects. Extended phenotyping showed a high prevalence of RH4 (c) (99.5%) and RH5 (e) (96.7%) antigens. Haplotype analysis revealed a predominance of the R0 (Dce) complex (67.3%). All subjects were Kell-negative (100% KEL:-1). No statistically significant difference was observed in blood group distribution between donors and recipients ($p > .05$).

Conclusion

This study confirms the predominance of blood group O and the R0 haplotype in the Kinshasa population. However, the presence of C and E antigens in nearly one-quarter of participants, combined with universal Kell negativity, highlights a significant risk of alloimmunization in polytransfused patients. Implementing extended phenotyping is therefore essential to improve transfusion safety and clinical outcomes in the DRC.

INTRODUCTION

Blood transfusion (BT) is an essential therapeutic procedure involving the administration of blood components from a donor to a recipient in order to restore compromised vital functions (Ammari & Ameyoud, 2023). Although life-saving, this procedure is strictly regulated due to immunological risks, particularly erythrocyte alloimmunization (Garraud, 2021). Historically, transfusion safety has relied mainly on compatibility within the ABO and standard Rhesus (RhD) systems. However, worldwide heterogeneity in blood group phenotypes increasingly requires a more refined immunohematological approach (Boufrioua et al., 2020). Globally, the distribution of blood groups varies considerably. While group O predominates in many populations (47% in Great Britain and 54.2% in Nigeria), the Rhesus system shows major differences, with RhD positivity exceeding 95% in Africa and Asia compared with approximately 83%–85% in Europe and the United States (Boufrioua et al., 2020).

In sub-Saharan Africa, the issue of alloimmunization is aggravated by the high prevalence of chronic conditions requiring repeated transfusions, such as sickle cell disease. Recent evidence suggests that transfusion practices based solely on international standards may be poorly adapted to local realities, particularly due to the predominance of certain phenotypes such as the R0 (Dce) haplotype, which is present in 45.8% of individuals of Afro-Caribbean origin compared with only 2.1% of Europeans (Peyrard et al., 2018). The Kell system, considered the third most immunogenic blood group system, also shows substantial ethnic disparities. The KEL1 antigen, present in 8%–9% of Western Europeans, is extremely rare in many African populations, thereby increasing the risk of immunological complications during non-targeted transfusions (Talbi et al., 2018). Despite these challenges, extended phenotyping remains uncommon in many African transfusion centers due to limited technical and financial resources (Osaro & Erhabor, 2022).

In the Democratic Republic of the Congo (DRC), particularly at the University Clinics of Kinshasa (CUK), epidemiological data remain largely limited to basic blood group systems. Previous studies have shown a predominance of group O (58%–61%) and a frequency of RhD positivity of approximately 99% (Sumbu et al., 2022). Although efforts have been made in Lubumbashi to characterize the Kell and extended Rhesus systems—showing, for example, 100% Kell negativity (Kalenga et al., 2020)—data from Kinshasa remain fragmentary. At present, systematic typing for C, c, E, e, and K antigens is not standardized at the National Blood Transfusion Center (CNTS), exposing recipients, particularly women of childbearing age and multitransfused patients, to severe hemolytic complications (Sumbu et al., 2018).

To date, no study conducted in Kinshasa has simultaneously analyzed extended Rhesus and Kell phenotypes in both blood donors and recipients. This scientific gap represents a major concern for transfusion safety because it prevents the consistent implementation of phenotype-matched transfusion strategies. In addition, the lack of updated data on antigen distribution limits the optimization of labile blood product inventories and hinders effective prevention of alloimmunization.

This study is therefore justified by the need to address the limited availability of documented immunohematological data in the Kinshasa population. By determining actual antigen frequencies, this work aims to support improved transfusion strategies in the DRC. The primary objective of this study was to determine the immunohematological profile of blood donors and recipients at the CUK and CNTS in order to reduce transfusion-related complications and improve patient care through optimal immunological matching.

METHODS

Study Design, Period, and Setting

This was a descriptive cross-sectional study conducted from November 13, 2024, to July 28, 2025, at the University Clinics of Kinshasa (CUK) and the National Blood Transfusion Center (CNTS), Kinshasa, Democratic Republic of the Congo.

Study Population

Participants were recruited prospectively and consecutively. Donors and recipients were included according to predefined eligibility criteria.

Inclusion Criteria

The following participants were included:

- Any patient for whom phenotyped red blood cell concentrates (RBC concentrates) were prescribed.
- Any blood donor whose donation was cross-matched and subsequently phenotyped for a specific patient during the study period.
- Any blood donor at the CUK or CNTS whose sample was selected for erythrocyte phenotyping (ABO, Rhesus [RH1–RH5], and Kell [KEL1]) at the Immunohematology and Transfusion Unit of the CUK.

Exclusion Criteria

Any blood donor or recipient who did not meet the inclusion criteria was excluded.

Sample Size

The sample size was determined by convenience sampling.

Data Collection Procedures

For recipients, samples were collected primarily by venipuncture. For blood donors, two blood samples were collected immediately prior to donation: one sample in a citrate tube for whole blood testing and one sample in a gel separator tube for serum preparation.

Sources of Information and Data Collection Tools

Sociodemographic, immunohematological, and transfusion-related data were collected using several official sources, including:

- Individual blood donor files
- Patient medical records accessed through the Open Clinic management software
- Telephone calls to complete missing information
- Phenotyping, reception, and blood product distribution registers

Sample Collection Procedures

For recipients, blood samples were mainly collected by venipuncture. For donors, two samples were collected immediately prior to donation: one sample in a citrate tube for whole blood analysis and one sample in a gel separator tube for serum.

Blood Group Determination Techniques

- **ABO system:** Determined using the dual typing technique (Beth-Vincent and Simonin–Michon methods).
- **Rhesus and Kell systems:** Determined exclusively using the direct serological method.
- **Weak D screening:** The indirect antiglobulin test (IAT) was systematically performed for samples initially classified as RhD-negative. The IAT detects anti-erythrocyte antibodies by incubating patient serum with red blood cells *in vitro*.

Quality Assurance and Validation

Reliability was ensured through strict adherence to standard operating procedures (SOPs), the use of control sera, and the appropriate storage of control red blood cells at 4°C. Results were validated only when complete agreement was observed between the two ABO typing methods.

Statistical Analysis

Data were entered and analyzed using IBM SPSS Statistics version 24.0. Data quality checks were performed prior to analysis to ensure consistency between electronic records and source documents.

Continuous variables (e.g., age) were summarized using mean and standard deviation (SD), while categorical variables (e.g., sex, blood group phenotypes, and antigen status) were expressed as frequencies and percentages. Ninety-five percent confidence intervals (95% CIs) for proportions were calculated using the Wilson score method.

Comparisons of categorical variables between groups (donors vs. recipients) were performed using Pearson's chi-square test. When expected cell counts were less than five, Fisher's exact test was considered appropriate. A p-value $< .05$ was considered statistically significant.

ABO allele frequencies were estimated from phenotype distributions using Bernstein's gene frequency method under Hardy–Weinberg assumptions (Crow, 1999; Couty-Fredon et al., 2018). The Hardy–Weinberg equilibrium was assessed using Pearson's chi-square goodness-of-fit test to compare observed and expected genotype frequencies.

For the Rhesus system, allele frequency estimation was performed using the classical model of Landsteiner and Wiener, assuming two-allele systems for RhD (D and d). Genotype distributions (DD, Dd, dd) were inferred from

observed RhD phenotypes under Hardy–Weinberg equilibrium assumptions.

All statistical tests were two-tailed.

Gene Frequencies (ABO System)

Gene frequencies were calculated using Bernstein's formula. If p', q', and r' are the initial estimates:

- $r' = O$
- $p' = 1 - B + O$
- $q' = 1 - A + O$

Adjusted frequencies (p, q, r) were obtained using:
$D = 1 - (p' + q' + r')$

- $p = p' \times (1 + D/2)$
- $q = q' \times (1 + D/2)$
- $r = (r' + D/2) \times (1 + D/2)$

Gene Frequencies (Rhesus System)

Gene frequencies were estimated using the Landsteiner and Wiener method:

- d = RhD-negative
- D = 1 − RhD-negative

where:

- *d* represents the allele frequency corresponding to RhD-negative, and
- *D* represents the allele frequency corresponding to RhD-positive (Deba et al., 2017).

Hardy–Weinberg Equilibrium

Hardy–Weinberg equilibrium was assessed using Pearson's chi-square test, with statistical significance defined as $p < .05$.

Interpretation of Rhesus Genotypes

The number of possible genotypes was estimated based on the number of positive antigen reactions:

- For Rh1-positive (D): number of genotypes = number of positive reactions − 1
- For Rh1-negative (d): number of genotypes = number of positive reactions − 2
- If D was negative, the number of possible genotypes was two units lower than the number of positive reactions (unless only two positive reactions were present, in which case only one possible genotype existed).

Potential Sources of Bias

- **Selection bias:** Recruitment was hospital-based (CUK and CNTS) and may not fully represent the general population of Kinshasa.
- **Information bias:** This was minimized by cross-checking physical registers against electronic software records.

RESULTS

Population and Sample Flow

During the study period, a total of 240 blood samples were analyzed, corresponding to 214 unique subjects included in the study. The difference between the number of samples and the number of subjects was due to repeat sampling performed for a subset of participants, mainly recipients, in whom additional specimens were collected for confirmatory testing and/or follow-up phenotyping. Overall, the study population consisted of 163 blood donors (76.2%) and 51 recipients (23.8%).

General Characteristics of the Study Population

The mean age of the overall population was 30.47 ± 15.08 years. The mean age of recipients was 29.56 ± 8.89 years, while that of blood donors was 33.39 ± 26.50 years. The most represented age groups were 26–35 years (32.2%; n = 69) and 19–25 years (31.3%; n = 67). The age range extended from 1 day to 76 years.

Regarding sex distribution, males were predominant, with 170 participants (79.4%), corresponding to a male-to-female ratio of 3.86.

Table 1:
Sociodemographic Characteristics of the Study Population (with 95% Confidence Intervals)

Variable	Category	Total Population (N = 214) n (%) [95% CI]	Recipients (n = 51) n (%) [95% CI]	Donors (n = 163) n (%) [95% CI]
Age (years)	0–2	8 (3.7) [1.9–7.2]	8 (15.7) [8.2–27.9]	0 (0.0) [0.0–2.3]
	3–18	14 (6.5) [3.9–10.6]	12 (23.5) [14.0–36.8]	2 (1.2) [0.3–4.4]
	19–25	67 (31.3) [25.5–37.8]	3 (5.9) [2.0–15.9]	64 (39.3) [32.1–47.0]
	26–35	69 (32.2) [26.4–38.7]	4 (7.8) [3.1–18.5]	65 (39.9) [32.6–47.6]
	36–45	27 (12.6) [8.8–17.7]	6 (11.8) [5.5–23.4]	21 (12.9) [8.6–18.9]

Variable	Category	Total Population (N = 214) n (%) [95% CI]	Recipients (n = 51) n (%) [95% CI]	Donors (n = 163) n (%) [95% CI]
	46–76	29 (13.6) [9.6–18.8]	18 (35.3) [23.6–49.0]	11 (6.7) [3.8–11.7]
Sex	Male	170 (79.4) [73.5–84.3]	33 (64.7) [51.0–76.4]	137 (84.0) [77.6–88.9]
	Female	44 (20.6) [15.7–26.5]	18 (35.3) [23.6–49.0]	26 (16.0) [11.1–22.4]

Analysis of geographical origin showed that the most represented provinces were Kongo Central (19.6%) and the two Kasai provinces (Kasai Oriental and Kasai Central), each accounting for 10.3% of participants.

Table 2:
Distribution of Participants by Province of Origin (N = 214)

Province	n	%	95% CI
Haut Katanga	2	0.9	[0.2–3.4]
Haut Uélé	1	0.5	[0.1–2.6]
Kasaï Central	22	10.3	[6.9–15.1]
Kasaï Oriental	22	10.3	[6.9–15.1]
Kongo Central	42	19.6	[14.8–25.5]
Kwango	15	7.0	[4.3–11.2]
Kwilu	30	14.0	[9.9–19.4]
Lomami	11	5.1	[2.9–9.0]
Lualaba	4	1.9	[0.7–4.7]
Mai-Ndombe	14	6.5	[3.9–10.6]
Maniema	2	0.9	[0.2–3.4]
Mongala	7	3.3	[1.6–6.6]
Nord Kivu	12	5.6	[3.2–9.5]
Nord Ubangi	3	1.4	[0.5–4.0]
Sankuru	7	3.3	[1.6–6.6]
Sud Kivu	14	6.5	[3.9–10.6]
Sud Ubangi	4	1.9	[0.7–4.7]
Tshopo	2	0.9	[0.2–3.4]

Association Between Blood Group Distribution and Participant Category

No statistically significant association was observed between blood group distribution and participant category (donor vs. recipient; $p > .05$). Donors and recipients were therefore immunologically comparable with respect to the studied blood group systems **(Table 3)**.

Table 3:
Distribution of ABO, Rhesus, and Kell Antigens by Study Category (with 95% Confidence Intervals)

System	Phenotype	Donors n (%) [95% CI]	Recipients n (%) [95% CI]	*p*
ABO	O	82 (76.6) [67.7–83.7]	25 (23.4) [16.3–32.3]	.79
	A	42 (75.0) [62.3–84.5]	14 (25.0) [15.5–37.7]	
	B	31 (79.5) [64.5–89.2]	8 (20.5) [10.8–35.5]	
	AB	8 (66.7) [39.1–86.2]	4 (33.3) [13.8–60.9]	
RhD (RH1)	Positive	155 (76.0) [69.6–81.4]	49 (24.0) [18.6–30.4]	.56
	Negative	8 (80.0) [49.0–94.3]	2 (20.0) [5.7–51.0]	
RhC (RH2)	Positive	24 (70.6) [53.8–83.2]	10 (29.4) [16.8–46.2]	.27
	Negative	139 (77.2) [70.5–82.7]	41 (22.8) [17.3–29.5]	
Rhc (RH4)	Positive	162 (76.1) [69.8–81.3]	51 (23.9) [18.7–30.2]	.76
	Negative	1 (100.0) [20.7–100.0]	0 (0.0) [0.0–79.3]	
RhE (RH3)	Positive	24 (64.9) [48.8–78.2]	13 (35.1) [21.8–51.2]	.07
	Negative	139 (78.5) [71.9–83.9]	38 (21.5) [16.1–28.1]	
Rhe (RH5)	Positive	157 (75.8) [69.5–81.2]	50 (24.2) [18.8–30.5]	.47
	Negative	6 (85.7) [48.7–97.4]	1 (14.3) [2.6–51.3]	

Frequency of ABO and Rhesus D Blood Group Antigens

The immunohematological profile of the study population was characterized by a predominance of blood group O (50.0%) and RhD positivity (95.3%). Extended phenotyping demonstrated a near-universal prevalence of RH4 (c) and RH5 (e) antigens, consistent with the predominance of the R0 (Dce) haplotype (67.3%), which is typical of sub-Saharan African populations. Notably, the KEL1 antigen was not detected in any participant (0%), suggesting that Kell incompatibility is likely uncommon in routine local transfusion practice. Antigen frequencies for the ABO, Rhesus, and Kell systems are presented in **Tables 4** and **5**.

Table 4:
Phenotypic and Allelic Frequencies of ABO and RhD (RH1) Systems (with 95% Confidence Intervals)

Variable	n	Phenotype % [95% CI]	Allele frequency (unadjusted) % [95% CI]	Allele frequency (adjusted) % [95% CI]
ABO system				
O	107	50.0 [43.3–56.7]	70.7 [65.2–75.7]	70.0 [64.5–75.0]
A	56	26.2 [20.7–32.5]	16.6 [12.8–21.4]	17.4 [13.4–22.2]
B	39	18.2 [13.6–24.1]	11.9 [8.6–16.1]	12.7 [9.3–17.0]
AB	12	5.6 [3.2–9.5]	—	—
RhD (RH1)				
Positive	204	95.3 [91.6–97.5]	78.3 [72.5–83.2]	—
Negative (dd)	10	4.7 [2.6–8.4]	21.7 [16.8–27.5]	—

Estimated RhD genotypic frequencies: DD: 61.3 [54.8–67.5]; Dd: 34.0 [27.8–40.6]; dd: 4.7 [2.6–8.4]

Hardy–Weinberg equilibrium testing was performed for the ABO system to assess consistency between observed and expected genotype distributions. The results

supported the validity of allele frequency estimates obtained using Bernstein's method.

Table 5:
Distribution of Rhesus (RH2, RH3, RH4, RH5) and Kell (KEL1) Antigens According to ISBT Nomenclature (N = 214)

Antigen	Phenotype	n	%	95% CI
RH2 (C)	Positive	34	15.9	[11.5–21.4]
	Negative	180	84.1	[78.6–88.5]
RH4 (c)	Positive	213	99.5	[97.4–99.9]
	Negative	1	0.5	[0.1–2.9]
RH3 (E)	Positive	37	17.3	[12.8–22.9]
	Negative	177	82.7	[77.1–87.2]
RH5 (e)	Positive	207	96.7	[93.4–98.4]
	Negative	7	3.3	[1.6–6.6]
KEL1 (K)	Positive	0	0.0	[0.0–1.8]
	Negative (kk)	214	100.0	[98.2–100.0]

Distribution of Rhesus Phenotypes and Haplotypes

Haplotype analysis revealed a marked predominance of the R0 (Dce) haplotype, identified in 67.3% of participants. The R2 (DcE) and R1 (DCe) haplotypes followed with frequencies of 13.5% and 11.8%, respectively. Rh-negative haplotypes (r, r′, and r″) as well as the rare Rz haplotype were uncommon, each representing less than 3% of the study population. At the genotype level, the most frequent inferred combinations (Dce/ce, DcE/ce, and DCe/ce) accounted for 92.2% of participants. The distribution of probable haplotypes is presented in **Table 6**.m

Table 6:
Distribution of Rhesus Haplotypes (Wiener and Fisher–Race Nomenclature) (N = 214)

Wiener haplotype	Fisher–Race equivalent	n	%	95% CI
R0	Dce	144	67.3	60.7 – 73.2
R1	DCe	25	11.7	8.0 – 16.7
R2	DcE	29	13.6	9.6 – 18.8
Rz	DCE	6	2.8	1.3 – 6.0
r	dce	6	2.8	1.3 – 6.0
r′	dCe	2	0.9	0.3 – 3.3
r″	dcE	2	0.9	0.3 – 3.3

DISCUSSION

Principal findings

This study provides an updated immunohematological profile of blood donors and recipients in Kinshasa, with a particular focus on extended Rhesus and Kell systems. The findings confirm a predominance of blood group O (50.0%) and RhD positivity (95.3%) in the study population. Extended phenotyping demonstrated a high frequency of RH4 (c) and RH5 (e) antigens, consistent with the observed predominance of the R0 (Dce) phenotypic profile (67.3%). The Kell KEL1 antigen was not detected in any participant, resulting in an observed frequency of 0%.

Importantly, no statistically significant difference was observed in ABO or Rh blood group distributions between donors and recipients ($p > .05$), suggesting overall immunohematological comparability between both groups within the study setting.

Comparison with existing literature

The ABO and RhD distributions observed in this study are consistent with previous reports from the Democratic Republic of Congo and neighboring regions. The predominance of group O aligns with findings from Sumbu et al. (2022), while the high prevalence of RhD positivity is consistent with global patterns observed in sub-Saharan African populations (Boufrioua et al., 2020).

The observed R0 (Dce) predominance (67.3%) is higher than that reported in West African populations such as Nigeria (approximately 54%) and substantially higher than frequencies reported in European populations, where this haplotype is relatively rare (Boufrioua et al., 2020; Peyrard et al., 2018). These differences reflect well-established interpopulation variation in RH haplotype distribution.

The absence of KEL1 antigen is consistent with previous studies in Central and Southern Africa, including work by Kalenga et al. (2020), which also reported very low or absent Kell antigen expression in similar populations.

Interpretation of Rhesus phenotypes and methodological considerations

The interpretation of Rhesus haplotypes in this study is based on serological phenotype inference rather than molecular genotyping. Therefore, the reported haplotypes (e.g., R0, R1, R2) represent statistically inferred combinations derived from observed antigen expression patterns and should be interpreted as *phenotype-based approximations* rather than confirmed genetic haplotypes.

Under Hardy–Weinberg assumptions, RhD genotype distribution (DD, Dd, dd) was inferred from observed phenotypes. While this approach is widely used in population studies (Crow, 1999), it assumes random mating, absence of selection, and equilibrium conditions, which may not fully reflect real-world transfusion populations.

Accordingly, the genetic estimates presented in this study should be interpreted with caution, particularly in a hospital-based sample where selection bias may influence allele distribution.

Alloimmunization risk in transfusion practice

Although no statistically significant differences were observed between donors and recipients, immunohematological risk remains clinically relevant due to the presence of antigenic diversity within the Rhesus system, particularly RH2 (C) and RH3 (E).

Patients requiring repeated transfusions, such as those with sickle cell disease, remain at risk of alloimmunization when exposed to non-phenotyped blood units. The development of anti-C or anti-E antibodies has been associated with delayed hemolytic transfusion reactions in similar settings (Osaro & Erhabor, 2022).

Additionally, the presence of Rh antigen incompatibility remains clinically important in women of reproductive age, where sensitization may contribute to hemolytic disease of the fetus and newborn in subsequent pregnancies (Garraud, 2021).

Public health and transfusion implications

From a transfusion policy perspective, the findings highlight the importance of extending phenotyping beyond ABO and RhD in settings with high transfusion demand. Although ABO and RhD compatibility remains sufficient for emergency transfusion, extended antigen matching (particularly for C, E, and Kell systems) may reduce the risk of alloimmunization in chronically transfused patients.

However, the implementation of routine extended phenotyping should be guided by resource availability and targeted toward high-risk populations rather than universal screening. The absence of Kell antigen in the studied population may reduce immediate alloimmunization risk, but does not eliminate the need for vigilance, particularly in cases involving imported blood units or heterogeneous donor sources.

Study limitations

This study has several limitations. First, the immunohematological classifications were based on serological inference, and no molecular genotyping was performed. Consequently, Rh haplotypes and RhD genotypes should be considered probabilistic estimates rather than definitive genetic determinations.

Second, the hospital-based sampling strategy may introduce selection bias, limiting the generalizability of allele frequency estimates to the broader population of Kinshasa.

Finally, the relatively small number of recipients may reduce statistical power for detecting subtle differences in blood group distributions between study groups.

CONCLUSION

This study demonstrates that although the ABO and RhD distribution in Kinshasa appears relatively homogeneous, the presence of RH2 (C) and RH3 (E) antigens may represent a significant alloimmunization risk for multitransfused patients. The predominance of the R0 (Dce) haplotype and the complete absence of the Kell antigen (KEL1 = 0%) highlight the need to revise compatibility protocols. To improve transfusion safety in the DRC, it is essential to implement extended Rh–Kell phenotyping for high-risk recipients and to develop molecular testing capacity in order to overcome the limitations of serological inference.

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Ethical Approval: The study protocol was approved by the Ethics Committee of the Kinshasa School of Public Health (Approval No. ESP/CE/99/2024).

Conflicts of Interest: None declared.

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