Original Article

Hemovigilance and Red Cell Phenotyping among Patients with Hereditary Blood Diseases in Basra, Southern Iraq

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ABSTRACT

Objective: To assess the incidence of direct antiglobulin test positivity and to profile severity and demographic parameters of acute transfusion reactions

Study Design: Cross sectional descriptive study

Place and Duration of Study: This study was conducted at the Basra Centre for Hereditary Blood Diseases, Basra Southern Iraq, from 15th September 2025 to 15th February 2026.

Methodology: 1426 patients were registered. The blood bank electronic real-time database under the supervision of qualified blood bank laboratory technicians was used to obtain the data. The transfusion events monitored were 108 and analysed were ABO/Rh(D) typing, extended Rh phenotyping (D, C, c, E, e antigens), direct antiglobulin test and hemovigilance data.

Results: There were 38.43% of peoples have blood group O+ and Rh(D)-positive rate was 92.78%. The most frequent extended Rh phenotype was the R1r (31.41%). The overall percentage of direct antiglobulin test positivity was 40% and was the highest in β-thalassaemia major (55.6%). Grade B (moderate) transfusion reactions were the most common (67.6%), and 69.5% of the reactions were Grade A (mild) or B (moderate).

Conclusion: The transfusion-dependent population extended Rh phenotyping and systematic hemovigilance is necessary. The high DAT positivity and moderate to severe reaction rates highlight the need for antigen matched blood strategies and formal hemovigilance systems.

Key Words: Hemovigilance, Red blood cell, Phenotyping, Rh antigens; Direct antiglobulin test, Transfusion reactions.

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INTRODUCTION

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Blood transfusion continues to play a critical role in the management of most of the hereditary blood disorders, such as β-thalassemia major (β-TM), sickle cell disease (SCD). Although the introduction of transfusion therapy has had a positive impact on the survival and quality of life of those who suffer from it, repeated exposure to donor red blood cells has been associated with certain immunological complications including alloimmunization, autoimmunization, and transfusion-related adverse reactions (TRA).[1,2] Such complications can lead to increased transfusion needs, may make the determination of compatibility more difficult and can be a source of morbidity in chronically transfused patients.

ABO and Rhesus (Rh) blood group systems are the two most clinically important antigen systems in transfusion medicine. Compatibility testing of ABO and Rh(D) blood group types is regular before transfusion but incompatibilities with minor Rh antigens like C, c, E and e are still significant causes of alloantibody formation in transfusion-dependent patients.[3]

Extended red cell phenotyping is therefore becoming an important tool for decreasing alloimmunization and improving transfusion safety, particularly in patients that have a need for transfusion on a life-long basis.[4]

Hemoglobin disorders are a significant burden in the public health sector in Iraq. Southern Iraq, including Basra Province, has high prevalence of β-thalassemia major, sickle cell disease and related hemoglobinopathies, partly due to the high prevalence of consanguineous marriages.[5] Epidemiologic studies carried out recently in Basra have identified an increasing number of patients who need to receive regular transfusion therapy and long-term hematological care.[6] In addition, research conducted in Basra has shown the clinical use of Rh and Kell antigen typing to improve transfusion support for this high-risk population.[7]

ABO and Rh blood groups are distributed differently in various ethnic groups and populations. However, several studies in Iraq have indicated that the major ABO blood group phenotype is O and the major Rh(D) blood group phenotype is Rh(D)+ (over 90%).[8,9] One of the major complications of repeated transfusion therapy is red cell alloimmunization. Alloantibodies are produced when a person is exposed to foreign erythrocytes, most often Rh and Kell antigens. These antibodies can cause delayed hemolytic transfusion reactions, problems obtaining blood that matches patients, and higher health cost.[10,11] In the past, Rh and Kell antigens have been found as the main antigens involved in alloimmunisation in multi-transfused patients in Iraq[12,13] and this is the reason for the necessity of developing better compatibility methods. The Direct Antiglobulin Test (DAT) is a commonly employed technique to identify the presence of in vivo sensitization of red blood cells with immunoglobulin or complement components. DAT positivity has been described on a regular basis in multi-transfused patients who have thalassemia and sickle cell disease and could be a result of the presence of alloantibodies, autoantibodies or continued immune mediated hemolysis.[14,15] Therefore, monitoring of DAT status could be useful in understanding the immunologic effects of chronic transfusion therapy. Hemovigilance is becoming an integral part of today's transfusion medicine. It is described as the systematic reporting and monitoring of adverse events throughout the transfusion chain, from blood collection to follow up with recipients.[16] Hemovigilance programs worldwide have shown that the most frequent adverse events associated with blood transfusion are febrile non-hemolytic and allergic reactions.[17,18] Good hemovigilance systems enhance patient safety, support quality assurance and reachable evidence to promote transfusion optimization. Although the incidence of hereditary blood disorders has been rising in Basra, detailed examination of blood group distribution and extended Rh phenotypes, DAT positivity, and blood transfusion adverse events are still limited. The purpose was to find ABO and Rh blood group distribution, characterize extended Rh phenotypes, evaluate the positive DAT and evaluate hemovigilance outcomes among patients of hereditary blood diseases.

METHODOLOGY

This retrospective cross sectional study was carried out at Basra Centre for Hereditary Blood Diseases (BCHBD), Basra Southern Iraq from 15th September 2025 to 15th February 2026 vide letter No. 456778/Approval/DFTBV dated 2nd April 2024. The BCHBD electronic blood bank database, a real-time information system that records patient demographic, clinical diagnosis, transfusion history, blood group phenotypes, serological investigations and adverse events related to transfusion were used to obtain the data. Qualified blood bank laboratory staffs routinely perform data entry verification to insure completeness and accuracy of data. Overall, 1,426 patients were confirmed to have a hereditary blood disorder and were registered. Diagnoses included β-thalassemia major (β-TM), thalassemia intermedia (β-TI), sickle cell disease (HbSS), HbS/β-thalassemia, pure red cell aplasia, aplastic anemia and coagulation factor deficiency. Patients were included if all of their ABO/Rh(D) typing and Rh phenotyping data were stored in the database. Specific analyses were performed excluding records with incomplete serological data.

Standard tube agglutination and gel-card methods were used to detect ABO and Rh(D) blood grouping, using commercially available antisera. Extended Rh phenotyping involved the identification of the 5 main Rh antigens, D, C, c, E and e. The Fisher–Race nomenclature system was used to classify the Rh phenotype combinations. The Direct Antiglobulin Test (DAT) was only ordered when patients had clinical features such as history of hemolysis, transfusion reactions, or physician request to undergo an immunohematologic work up. Anti-human globulin (AHG) method was used for testing. A positive result was defined as any agglutination that was seen.

Patients' clinical staff recorded transfusion reactions, and these were entered into an electronic database, prospectively, during and after transfusion episodes. The hemovigilance analysis included a total of 108 transfusion episodes. Adverse reactions were graded as follows: Grade A: Serious reaction that needs prompt action, Grade B: Moderate reaction that may be treated clinically, Grade C: Mild reaction which requires little intervention to resolve and Grade D: No RATE. The data was analyzed through SPSS-25.

RESULTS

The most common phenotype was blood group O+ (38.43%) followed by blood group A+ and B+ (both 24.54%). The population was predominately Rh-positive with 93.3% of the patients having the Rh(D) positive phenotype and 6.7% having the Rh(D) negative

phenotype (Table 1). The major Rh-antigens revealed high prevalence for e (94.44%), D (92.78%), C (70.80%) and c (70.53%), while E antigen showed the lowest prevalence (22.49%).The results are useful in planning strategies of antigen matching for chronically transfused patients (Table 2).

Extended phenotypic analysis revealed that the most frequent Rh phenotype was R1r (31.41%) followed by R1R1 (25.85%).These had more than 50% of patients. The other phenotypes were less common: R1R2, R2r, R2R2 and rr (Table 3). The clinical indications for DAT were immunological transfusion complications, and it was performed in 35 patients. Overall, 14 (40%) patients were DAT-positive. Patients with β-thalassemia major showed the highest positivity rate while patients with thalassemia intermedia showed the next highest positivity rate (37.5%). Lower positivity rates were observed in sickle cell disease patient (Table 4).

All 108 monitored transfusion episodes were used for hemovigilance analysis. The majority of episodes were in children and adolescents (ages 5–14).The most common reaction category for all age groups was moderate transfusion reactions (Grade B) and severe transfusion reactions (Grade A) occurred in only a few patients (Table 5).

Transfusions for thalassemia patients were the most common monitored transfusion events (78.7%).Moderate transfusion reactions were the most common adverse events reported for all disease categories. The incidence of severe reactions was low and was rarely seen in highly transfusion dependent patients. The numbers of patients with sickle cell disease, aplastic anaemia and other hereditary disorders were fewer as contributors to transfusion episode (Table 6).

Moderate reactions were the most common adverse events reported; mild reactions were present in a smaller number of the monitored transfusion episodes (108). Severe reactions were rare, suggesting that the majority of transfusion-related problems were clinically controllable. However, the number of transfusion reactions observed underlines the need for ongoing hemovigilance monitoring and measures to improve transfusion safety in people with hereditary blood diseases.

Table No. 1: ABO and Rh(D) blood group distribution

Blood Group	ABO Phenotype	Rh(D) Status	No.	%	Rh(D) Sub-total
O+	Group O	Rh(D)+	548	38.43%	1,331 (93.3%)
O-	Group O	Rh(D)−	41	2.88%	95 (6.7%)
A+	Group A	Rh(D)+	350	24.54%	1,331 (93.3%)
A-	Group A	Rh(D)−	15	1.05%	95 (6.7%)
B+	Group B	Rh(D)+	350	24.54%	1,331 (93.3%)
B-	Group B	Rh(D)−	30	2.10%	95 (6.7%)
AB+	Group AB	Rh(D)+	83	5.82%	1,331 (93.3%)
AB-	Group AB	Rh(D)−	9	0.63%	95 (6.7%)

Table No. 2: Blood group antigen frequency (Frequencies are listed by the order of the extended blood group system)

Rh Antigen	Antigen Symbol	Tested (n)	Antigen-Positive	Antigen-Negative
D	RH1	1426	1323 (92.78%)	103 (7.22%)
C	RH2	1421	1006 (70.80%)	415 (29.20%)
c	RH4	1422	1003 (70.53%)	419 (29.47%)
E	RH3	1423	320 (22.49%)	1103 (77.51%)
e	RH5	1422	1343 (94.44%)	79 (5.56%)

Table No. 3: Most prevalent Rh blood group phenotype combinations

Rank	DCcEe Phenotype	Common Nomenclature	No.	%
1	D+ C+ c+ E− e+	R1r (DCe/dce)	446	31.41
2	D+ C+ c− E− e+	R1R1 (DCe/DCe)	367	25.85
3	D+ C+ c+ E+ e+	R1R2 (DCe/DcE)	155	10.92
4	D+ C− c+ E− e+	R2r (DcE/dce) / R0r	145	10.21
5	D+ C− c+ E+ e+	R2R2 (DcE/DcE)	95	6.69
6	D− C− c+ E− e+	rr (dce/dce)	84	5.92
7	D+ C− c+ E+ e−	R2R2 (DcE/DcE) homozygous	46	3.24
8	D+ C− c− E− e+	Other combination	24	1.69
9	D+ C+ c− E+ e+	R1r″ (DCe/DCE)	9	0.63
10	D+ C− c+ E− e−	R0r (Dce/dce)	9	0.63

Table No. 4: The direct antiglobulin test performed as part of the haematological diagnosis

Diagnosis	No. (Total)	DAT Positive	% Positive	DAT Negative	Clinical Implication
β-Thalassaemia Major (β-TM)	18	10	55.6%	8	High DAT positivity reflects alloimmunisation from chronic polytransfusion; warrants extended matching
Thalassaemia Intermedia (β-TI)	8	3	37.5%	5	Moderate DAT positivity; less frequent transfusion than β-TM but alloimmunisation risk persists
Sickle Cell Disease (HbSS)	6	1	16.7%	5	Lower positivity rate; anti-erythrocyte antibodies may complicate delayed haemolytic transfusion reactions
HbS/β-Thalassaemia (S/β)	2	0	0.0%	2	All negative in this series; small sample size limits interpretation
HbE/β-Thalassaemia (E/β)	1	0	0.0%	1	Single case; insufficient data for pattern analysis
Total	35	14	40.0%	21	—

Table No. 5: Distribution of transfusion reaction grades by age group

Age Group	No.	Grade A (Severe)	Grade B (Moderate)	Grade C (Mild)	Grade D (None)	Total	% A+B
Infant (<1 yr)	2	-	1	1	-	2	50.0%
Early Childhood (1–4 yrs)	10	-	5	5	-	10	50.0%
Middle Childhood (5–9 yrs)	47	1	32	14	-	47	70.2%
Early Adolescence (10–14 yrs)	43	1	30	12	-	43	72.1%
Late Adolescence/Adult (≥15 yrs)	6	-	5	0	1	6	83.3%
Total	108	2	73	32	1	108	—

Table No. 6: Distribution of transfusion reaction grades according to type of the disease

Diagnosis (consolidated)	Sub-group (n)	Grade A	Grade B	Grade C	Grade D	Total	% A+B
Thalassaemia (Thal / TM / β-TM)	F=82;TM=1;β-TM=2	1	57	26	1	85	68.2%
Thalassaemia Intermedia (IF)	IF=12	0	7	5	0	12	58.3%
Sickle Cell Disease/Feature (SCD/SCF)	SS=2; SCA=1; SF=3	1	5	0	0	6	100.0%
Pure Red Cell Aplasia (PRCA)	PRCA=3	0	2	1	0	3	66.7%
Aplastic Anaemia (AA)	AA=1	0	1	0	0	1	100.0%
Factor X Deficiency	n=1	0	1	0	0	1	100.0%
Total	—	2	73	32	1	108	—

DISCUSSION

This study is the first comprehensive assessment of the practice of hemovigilance, Positive Direct Antiglobulin Test and extended Rh phenotypes in patients with hereditary blood diseases in Basra, Southern Iraq. The blood group O frequency, along with the high frequency of Rh(D) positive individuals in this population, is similar to the other studies published in Iraq and neighbouring Middle Eastern countries.[7-9] The distribution of hereditary blood disease patients is similar to that of blood donors and other regional populations, indicating that patients with blood diseases are representative of the regional population. However, local knowledge of the blood group frequency is still a key element in blood inventory management and donor recruitment strategy, especially in chronic transfusion centers.

This indicates that R1r and R1R1 were the most common combinations of Rh phenotypes when extended Rh phenotyping was performed. Similar results have been documented in Saudi Arabia and Kuwait and these phenotypes are also common in the Arab populations of these countries.[19,20] Low frequency of the E antigen found in this study is clinically relevant as anti-E antibodies are one of the most common transfusion dependent patients' alloantibodies. Many E-negative patients may be susceptible to alloimmunization and extended antigen matching should be routinely used.

In 40% of the patients tested, DAT positivity was observed, being the highest in patients of β-thalassemia major. This rate is higher than several previously published studies and is possibly due to the cumulative immune response from repeated exposure to transfusion.[14,15] Chronic antigenic stimulation can lead to the formation of both alloantibodies and autoantibodies, which can lead to positive DATs and make future transfusion management more complex. The high rate of positivity is more likely in β-

thalassemia major as the subjects usually undergo regular transfusion from childhood.

The hemovigilance analysis showed that moderate transfusion reaction was the predominant adverse event. There were very few severe reactions, less than 2% of all the responses monitored. These results are comparable to those reported internationally, where febrile non-hemolytic, and allergic reactions are the most common complications of transfusion.[21,22] A large proportion of the moderate reactions were not life-threatening, but there was a significant burden of moderate reactions in terms of patient and healthcare service consequences.

This study showed that majority of reactions was in the younger age groups, which is consistent with the age distribution of the transfusion dependent population. Likewise, the majority of transfusion episode and adverse reaction occurred in thalassemia patients, pointing to the high clinical impact of this disease in southern Iraq. The fact that the reactions were more frequent in the male patients is intriguing, and previous reports on sex differences in transfusion immunology have been inconsistent. The results of this study are in favor of extended Rh and Kell antigen testing of chronically transfused patients. The previous studies have shown that antigen matched transfusion programs can decrease the incidence of alloimmunisation and enhance the long-term outcome of transfusion.[10,11,13]

Further improving transfusion safety can be achieved through routine leukoreduction, frequent DAT testing and organised reporting of adverse events. The advantages of this study are that a large number of children were included, the Rh phenotyping was comprehensive, a real-time electronic database was used and a hemovigilance assessment was integrated. Several constraints must however be noted. This study was performed at a single tertiary center and detailed identification of all alloantibodies was not available in all DAT-positive patients. In addition, the retrospective study design restricted the evaluation of long-term clinical results related to alloimmunisation.

In general, we achieved a good baseline data for developing evidence-based transfusion policies and national hemovigilance initiatives in Iraq. The results highlight the need for improved transfusion safety by broadening Rh and Kell antigen matching, implementing routine leukoreduction, systematic monitoring of DAT and structured reporting of hemovigilance.

CONCLUSION

The extended Rh phenotyping revealed R1r and R1r1 as the major Rh phenotypes, whereas the presence of the E antigen was relatively low, it could be an additional risk factor responsible for alloimmunization in chronically transfused patients. DAT positivity was frequently seen, especially in the cases of patients with β-thalassemia major, showing the significant immunological burden of repeated transfusion therapy. Evaluation of hemovigilance adverse events revealed that moderate transfusion reactions were most common while severe reactions were rare.

Author's Contribution:

Concept & Design or acquisition of analysis or interpretation of data:	Basim A. Al-Hijaj, Ali K. Jumaa
Drafting or Revising Critically:	Zaynab R. Qasim, Qutaiba M. Dawood
Final Approval of version:	All the above authors
Agreement to accountable for all aspects of work:	All the above authors

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