Research Article

Prevalence of Red Blood Cell Alloimmunization among Children with Sickle Cell Disease in Southern Region of Saudi Arabia: A Single-Center Experience

Badriah Gharamah Al Asmari[1], Mohammed Alpakra[1], Ali Mujtaba Tahir[*,1], Ali Abdullah Hawan[2], Nagwan Nabil Mohamed Hossameldin[3], Mahnoor Saeed[4], Abdulaziz Mohammed Alsharaef [2], Jamilah Hadi Eissa Al Ali[5]

[1]*Department of Pediatric, Armed Forces Hospital Southern Region, Khamis Mushait, KSA.*

[2]*Department of Pathology, Armed Forces Hospital Southern Region, Khamis Mushait, KSA.*

[3]*Department of Transfusion Medicine, Blood Bank, Armed Forces Hospital Southern Region, Khamis Mushait, KSA.*

[4]*Azra Naheed Medical College, Lahore, Pakistan.*

[5]*Batterjee Medical College, Aseer, KSA.*

Abstract: Background: Sickle cell disease (SCD), carrying a varied worldwide prevalence, is a commonly encountered entity in Kingdom of Saudi Arabia and Mediterranean. It is characterized by a range of complications. Blood transfusion is essentially required for both episodic and chronic indications. These blood transfusions inherently imply various risks including alloimmunization.

Objective: To ascertain the incidence of red blood cell alloimmunization in children having sickle cell disease (SCD).

Materials and Methods: Retrospective cross-sectional study was conducted at the Department of Pediatric Medicine, Armed Forces Hospital Southern Region, Khamis Mushait, Saudi Arabia, after obtaining permission from Research Ethics Committee (REC certificate number AFHS-RMREC/2024/Pediatrics/745). The data was collected for last three years 1st July 2021 to 30th June 2024, regarding pediatric patients having sickle cell disease while aged one to fourteen years.

Result: 188 patients of sickle cell disease - all with Saudi ethnicity - were selected with 81 (43%) patients being female and 107 (57%) male. The mean age was 6 years 2 months. Each patient had an average of 2.3 admissions during the course of the disease. Out of these 188 patients, 14 (7.4%) patients were found to have alloimmunization. Mean age at which they developed alloimmunization was 6 years 8 months. 12 patients were on hydroxyurea and 7 patients (50%) underwent splenectomy. On average these cases received 13 transfusions. In contrast, only three (21.4%) patients had undergone exchange transfusion. The most commonly identified single alloantibody was anti-E as seen in 3 patients (21.4%), followed by anti-M in two cases (14.3%). In 4 cases (28.6%), there was an indeterminate antibody (with no specific pattern). Two (14.3%) children were having combination of alloantibodies.

Conclusion: Among the children from southern region of Kingdom of Saudi Arabia, the incidence of alloimmunization in SCD was 7.4%.

Keywords: Alloimmunization, Anemia, Complications of sickle cell anaemia, Hemoglobin, Red blood cell transfusion, Sickle cell disease.

INTRODUCTION

Sickle cell disease (SCD) is a disorder involving hemoglobin with autosomal inheritance pattern. SCD exhibits a varying prevalence worldwide, In USA, the reported prevalence is 329 cases in one million. In contrary, it is frequently witnessed in the African and middle-eastern population, as it affects 2400 per million Saudi children, more notably in its eastern and south-western regions.

Sickle cell disease constitutes a spectrum of disorders, encompassing varying genotypes. SS/SB0 thalassemia depicts a most severe clinical course, in contrary to S/alpha thalassemia or SB+ thalassemia exhibiting lesser severity. Hemoglobin F also contributes towards the clinical severity [1-7].

Morbidity inferring to SCD has decreased in the previous 40 years in lieu to the holistic care strategies implied in such children. One of important cornerstone in such interventions in SCD patients is blood transfusion - essentially for varying episodic or chronic indications. While lowering the level of HbS, red blood transfusion renders improved oxygen-carrying capacity. Repeated red blood cell transfusion has its inherent risks - alloimmunization being a major challenge in such individuals. Alloimmunization is associated with increased morbidity, hemolysis and delayed transfusion reactions. In United States, alloimmunization is reported in 30% of transfused sickle cell patients, in

* Address correspondence to this author at the Department of Pediatric, Armed Forces Hospital Southern Region, Khamis Mushait, KSA.
Email: aleeemujtaba@gmail.com

contrary to 2-5% of general transfusion recipients. Varying risk factors has been associated to alloimmunization including older age, frequent blood transfusions, use of non leukoreduced RBC and genetic variations [8-13].

In spite sickle cell disease being a common entity in our region, limited publications explains the prevailing alloimmunization patterns in our Kingdom. We conducted this research in our center to ascertain the incidence of red cell alloimmunization among children with sickle cell disease and various risk factors predisposing towards red cell alloimmunization. The aim was to avoid any identified risk factors preemptively in such children - limiting the morbidity and related complications.

MATERIALS AND METHODS

We conducted a retrospective cross-sectional study at Armed Forces Hospital Southern Region (AFHSR), Khamis Mushait, a tertiary care hospital and referral center, located in the southern region of Kingdom of Saudi Arabia. AFHSR serves as a prominent healthcare facility within the Armed Forces Hospital system, catering to military personnel, their families, and the civilian population residing in the surrounding area. The data was collected for last three years - 1st July 2021 to 30th June 2024 after permission was sought from Research Ethics Committee (REC certificate number AFHSRMREC/2024/Pediatrics/745). Target population was selected by convenience consecutive sampling for pediatric patients from the age of one to 14 years, who have confirmed diagnosis of sickle cell disease by hemoglobin electrophoresis and received packed red blood cell transfusion in our hospital while following with Pediatric Hematology team. However, children with comorbid requiring immunosuppressive therapy and autoimmune disease (like SLE) were excluded.

Being a retrospective study, the limitation to data collection was negotiated with thorough examination of medical archives. Confidentiality was ensured at all tiers. Detailed information from hospital electronic medical system of such children with sickle cell disease was gathered. The hospital owns a robust information and medical data management systems including archives system that aided in data collection needs for this study. A well-structured data collection pro-forma was integrated for collecting information on demographic characteristics like age, gender and residential area along with clinical presentations including alloimmunization profile, time of transfusion initiation and number of transfusions. Moreover, treatment details such as interventions administered, treatment initiation time and duration of hospital stay was recorded, in supplementation to various prevailing risk factors identified during the past years. Confidentiality measures were strictly adhered to ensure patient privacy and data was securely stored for subsequent assessment.

STATISTICAL ANALYSIS

The data was analytically evaluated for descriptive statistics, including frequency, mean and standard deviation to summarize demographic data, clinical presentations, treatment and outcomes. P value ≤0.05 regarded as significant.

RESULT

188 patients of sickle cell disease were selected with 81 (43%) patients being female and 107 (57%) male. The mean age was 6 years 2 months (±3.59). All patients were with Saudi ethnicity. Each patient had an average of 2.3 admissions during the course of the disease with quite variation observed among individual patients (maximum 18 admissions with minimum of one admission). The most common diagnosis was vaso-occlusive crisis (28%) for the inpatient care (Table **1**).

Table 1. Diagnosis for the Inpatient Admissions of Sickle Cell Disease Patients.

Diagnosis	Number of Inpatient Admissions n (%)
Vaso-Occlusive Crisis	122 (28%)
Hemolytic Crisis	97 (23%)
Acute Chest Syndrome	73 (17%)
Splenic Sequestration	60 (14%)
Sepsis	36 (9%)
Osteomyelitis	34 (8%)
Stroke	4 (1%)
Total	**426 (100%)**

All of these patients received folic acid and oral penicillin V. Hydroxyurea (hydroxycarbamide) was advised to all the patients, however, it was discontinued in 17% of the patients in lieu of drug induced thrombocytopenia and parents of 3% refused for hydroxyurea administration.

Out of these 188 patients, 14 (7.4%) patients were found to have alloimmunization. Mean age at which they developed alloimmunization was 6 years 8 months (± 3 years 1 month). 12 patients were on hydroxyurea (as parents of two patients refused for hydroxyurea). 7 patients (50%) had undergone splenectomy. On average these cases received 13 transfusions - with quite variation ranging from 2 times blood transfusion to 33 times blood transfusion. In contrast, only three (21.4%) patients had under gone exchange transfusion.

The most commonly identified single alloantibody was anti-E as seen in 3 patients (21.4%). In 4 cases (28.6%), there was an indeterminate antibody (with no specific pattern), as displayed in Table **2**. Two (14.3%) children were having combination of alloantibodies. The distribution of characteristics observed among the studied cases is shown in Table **3**, which presents the demographic and clinical characteristics of patients with detected alloantibodies.

Table 2. Alloantibodies Identified in Children with Sickle Cell Disease.

Type of Alloantibodies		No. of Patients (n=14)	Percentage
Single Alloantibody (n=12)	Anti-E	3	22
	Anti-M	2	14
	Anti-K	1	7
	Anti-Fyb	1	7
	Anti-C	1	7
	Indeterminate antibody (with no specific pattern)	4	29
Combination Alloantibodies (n=2)	Anti-E, Anti-C	1	7
	Anti-Lea, Indeterminate antibody	1	7

Table 3. Characteristics Distribution among Patients with Detected Alloantibodies.

Characteristics of the Patients		Detected Alloantibodies		p-value
		Positive n=14 (%)	Negative n=174 (%)	
Sex	Male	6 (43%)	96 (55%)	<0.001
	Female	8 (57%)	78 (45%)	
Age at Diagnosis	<1 year	0 (0%)	38 (21%)	<0.001
	1-2 years	1 (7%)	102 (59%)	
	≥ 2 years	13 (93%)	34 (20%)	
Rhesus D (RhD) Antigen	Positive	14 (100%)	167 (96%)	0.375
	Negative	0 (0%)	7 (4%)	
Received Hydroxyurea	Yes	12 (86%)	138 (79%)	0.161
	No	2 (14%)	36 (21%)	
Splenectomy	Yes	7 (50%)	29 (17%)	<0.001
	No	7 (50%)	145 (83%)	
Age of First Blood Transfusion	<1 year	0 (0%)	4 (2%)	<0.001
	1-2 years	1 (7%)	25 (15%)	
	2-3 years	2 (14%)	49 (28%)	
	≥ 3 years	11 (79%)	96 (55%)	
Phenotype tested before transfusion	Yes	11 (79%)	149 (86%)	0.172
	No	3 (21%)	25 (14%)	

Only one patient was given steroid for 6 weeks with diagnosis of AIHA (autoimmune hemolytic anaemia). Five patients were on regular blood transfusion program with indications for adhering to program are shown in Table **4**.

Table 4. Indications for Chronic Blood Transfusion Program (n=5).

Indication for Chronic Blood Transfusion Program	No. of Patients (n=5)
High indices of TCD (Transcranial Doppler)	2
Repeated splenic sequestration (awaiting for splenectomy)	2
History of stroke	1

DISCUSSION

Sickle cell disease (SCD) is a usual entity in selected populations. It is implicated as a disorder pertaining to hemoglobin with autosomal recessive inheritance. Prevalence of SCD exhibits a quite variation worldwide. In the United States, 72,000 people bears a homozygous sickle cell gene, while 2 million carry sickle cell trait. In contrary, the incidence is comparatively higher in African and middle-eastern ethnicity, as 1.4 % of population in the Kingdom of Saudi Arabia (KSA) affected by SCD, notably in its eastern and southwestern regions [14-17].

Sickle cell hemoglobin causes sickling of RBCs with eventually their premature destruction. This can infer as sludging and occlusion of blood vessels and resulting in tissue ischemia. SCD encompasses whole range of complications - from hemolytic and vaso-occlusive crisis to splenic sequestration and stroke. A comprehensive care for such children is essential in limiting its morbidity and mortality. Blood transfusion is essentially required for both episodic and chronic indications in SCD. These blood transfusions inherently imply various risks including alloimmunization. Erythrocyte alloimmunization can be a major barrier to transfusion as it predisposes to transfusion incompatibility and life-threatening hemolytic transfusion reactions [18-21]. Despite SCD being common in Saudi Arabia and blood transfusions frequently practiced in such individuals, data is lacking in Southern region of Saudi Arabia regarding the incidence of alloimmunization. We did a retrospective review and determined the prevalence of alloimmunization and to ascertain its related risk factors. Moreover, the study outcomes may contribute to the development of evidence-based protocols for such patients.

We enrolled 188 patients of sickle cell disease who received blood transfusion during the last 3 years. Out of these, there was a male preponderance (57%). However, one of the study have reported that female and male children were equally affected [22]. The mean age of our SCD children was 6 years 2 months and all patients were with Saudi ethnicity.

Each patient had an average of 2.3 admissions during the course of the disease with quite variation observed among individual patients (maximum 18 admissions with minimum of one admission). The most common diagnosis was vaso-occlusive crisis

(28%) for the inpatient care. Moreover, children with frequent hospitalization (≥5 times annually) tend to have more alloantibodies [23, 24].

In our observation of these 188 SCD patients, 7.4% patients were found to have alloimmunization. Many studies have been conducted worldwide to identify its incidence. A multicenter registry identified the incidence as 5-75% [23]. In USA, 30% of SCD patients who underwent transfusion were noted to have alloimmunization - a significant higher frequency than the transfusion recipients among the general population (2% to 5%) [24]. This study has surfaced the prevalence towards the lower grid. A study conducted in Jeddah KSA from 2007 to 2011 revealed 12.8% were having alloantibodies [22]. A study had also reported identical results to our study as alloimmunization of 6.6% in 60 children with SCD [25]. Homogeneity in RBC antigens in lieu to racial similarity between donors and recipients can be hypothesized for lower alloimmunization incidence.

Our patients developed alloimmunization at mean age of 6 years 8 months (± 3 years 1 month). Studies have documented average age of 7-9 years for presentation of alloantibody [26].

12 patients were on hydroxyurea (as parents of two patients refused for hydroxyurea). 7 patients (50%) had undergone splenectomy. On average these cases received 13 transfusions - with quite variation ranging from 2 times blood transfusion to 33 times blood transfusion. In contrast, only three (21.4%) patients had undergone exchange transfusion. One of the study reported that exchange transfusion did not predispose towards alloimmunization [27]. Five of our cases were on regular blood transfusion program - signifying the frequent transfusions. Our data coincides with other studies that correlated alloimmunization with frequency of blood transfusions [7, 9, 16, 28].

The most commonly identified single antibody was anti-E as seen in 3 (28.6%) patients, followed by anti-M in 2 patients (14.3%). The alloantibodies observed in our study (E, M, K, C, Fyb, C+E, Lea+Indeterminate) were in conflict with other studies, pointing towards ethnicity variation. The publications have mentioned alloantibodies against antigens of Rh and Kell systems to be most usual, followed by the Kidd and Duffy [23-25]. Two (14.3%) of our children were having combination of alloantibodies (C+E, Lea+Indeterminate). In contrast, other studies documented multiple antibodies in 50% cases [26]. This highlights the divergent alloantibody responses in distinctive regions, steering towards the need for further probing into the underlying influencing circumstances.

The potential racial homogeneity in our study suggests comparatively lower alloimmunization rate than global prevalence. We failed to establish any link of age at first transfusion with alloimmunization. Patients hospitalized frequently showed increased alloantibody responses. In summary, our research is distinguished in regards to its eccentric emphasis on pediatric cases.

Being a tertiary referral center in Southern Region of Saudi Arabia, its gives a validity and strength to the results of our study. The findings disclosed valuable insights into factors underlying alloimmunization. The apparent limitation of our study is its retrospective design.

CONCLUSION

Our study demonstrated that 7.4% children with sickle cell developed alloantibodies, which is an appreciable lower rate of alloimmunization as observed in other ethnic populations. Extended matching in term of Fy, Jk, and Ss blood groups can aide towards curtailing alloimmunization. High-resolution RBC genotyping technology for patients as well as donors can surely help in such endeavors. Furthermore, any efforts with multicenter randomized trial can shed light towards the path of limited alloimmunization.

AUTHORS' CONTRIBUTION

Badriah Gharamah Al Asmari and **Mohammed Alpakra**: Conceptualization, Study Design.

Ali Mujtaba Tahir: Conceptualization, Study Design, Methodology, Data analysis and interpretation, Writing draft, Critical review and revision the manuscript.

Ali Abdullah Hawan and **Nagwan Nabil Mohamed Hossameldin**: Methodology, Data analysis and interpretation.

Mahnoor Saeed and **Jamilah Hadi Eissa Al Ali**: Writing draft.

Abdulaziz Mohammed Alsharaef: Writing draft, Critical review and revision the manuscript.

FUNDING DISCLOSURE

Declared none.

CONFLICT OF INTEREST

Declared none.

ACKNOWLEDGEMENTS

We are really indebted to Miss Maria Baig, Editorial Manager Publication (NJHS) for her sincere guidance, help and support during the submission and approval of the manuscript.

REFERENCES

[1] Switzer JA, Hess DC, Nichols FT, Adams RJ. Pathophysiology and treatment of stroke in sickle-cell disease: Present and future. Lancet Neurol 2006; 5(6): 501-12.

[2] Piel FB, Steinberg MH, Rees DC, Longo DL. Sickle cell disease. N Engl J Med 2017; 376(16): 1561-73.

[3] Al-Qurashi MM, El-Mouzan MI, Al-Herbish AS, Al-Salloum AA, Al-Omar AA. The prevalence of sickle cell disease in Saudi

children and adolescents: A community-based survey. Saudi Med J 2008; 29(10): 1480-3.

[4] Memish ZA, Saeedi MY. Six-year outcome of the national premarital screening and genetic counseling program for sickle cell disease and βthalassemia in Saudi Arabia. Ann Saudi Med 2011; 31(3): 229-35.

[5] Pirenne F, Floch A, Habibi A. How to avoid the problem of erythrocyte alloimmunization in sickle cell disease. Hematology Am Soc Hematol Educ Program 2021; 2021(1): 689-95.

[6] Thein SL, Pirenne F, Fasano RM, Habibi A, Bartolucci P, Chonat S, *et al*. Hemolytic transfusion reactions in sickle cell disease: Underappreciated and potentially fatal. Haematologica 2020; 105(3): 539-44.

[7] Tormey CA, Hendrickson JE. Transfusion-related red blood cell alloantibodies: Induction and consequences. Blood 2019; 133(17): 1821-30.

[8] Chou ST, Alsawas M, Fasano RM, Field JJ, Hendrickson JE, Kameka M, *et al*. American Society of Hematology 2020 guidelines for sickle cell disease: Transfusion support. Blood Adv 2020; 4(2): 327-55.

[9] Boateng LA, Schonewille H, Lightard PC, Javadi A, Veldhuisen B, Osei-Akoto A, *et al*. One third of alloantibodies in patients with SCD transfused with African blood are missed by the standard red blood cell panel. Haematologica 2021; 106(8): 2274-6.

[10] da Cunha Gomes EG, Machado LAF, de Oliveira LC, Neto JFN. The erythrocyte alloimmunisation in patients with sickle cell anaemia: A systematic review. Transfus Med 2019; 29(3): 149-61.

[11] Stef M, Fennell K, Apraiz I, Arteta D, Gonzalez C, Nogues N, *et al*. RH genotyping by nonspecific quantitative next-generation sequencing. Transfusion 2020; 60(11): 2691-701.

[12] Gleadall NS, Veldhuisen B, Gollub J, Butterworth AS, Ord J, Penkett CJ, *et al*. Development and validation of a universal blood donor genotyping platform: A multinational prospective study. Blood Adv 2020; 4(15): 3495-506.

[13] Noizat-Pirenne F, Bachir D, Chadebech P, Michel M, Plonquet A, Lacron J-C, *et al*. Rituximab for prevention of delayed hemolytic transfusion reaction in sickle cell disease. Haematologica 2007; 92(12): 132-5.

[14] Dean CL, Maier CL, Chonat S, Chang A, Carden MA, El Rassi F, *et al*. Challenges in the treatment and prevention of delayed hemolytic transfusion reactions with hyperhemolysis in sickle cell disease patients. Transfusion 2019; 59(5): 1698-705.

[15] Pal M, Bao W, Wang R, Liu Y, An X, Mitchell WB, *et al*. Hemolysis inhibits humoral B-cell responses and modulates alloimmunization risk in patients with sickle cell disease. Blood 2021; 137(2): 269-80.

[16] Alboog AS, Tayeb TM, Alsager MO, AlNajjar SA, Damanhouri GA, Jarullah JS, *et al*. Red blood cell alloimmunization in sickle cell disease patients in Jeddah, Saudi Arabia: A pilot study. JKAU Med Sci 2015; 22(3): 17-22.

[17] Jastaniah W. Epidemiology of sickle cell disease in Saudi Arabia. Ann Saudi Med 2011; 31(3): 289-93.

[18] Fortin PM, Hopewell S, Estcourt LJ. Red blood cell transfusion to treat or prevent complications in sickle cell disease: An overview of Cochrane reviews. Cochrane Database Syst Rev 2018; 8(8): CD012082.

[19] Kangiwa U, Ibegbulam O, Ocheni S, Madu A, Mohammed N. Pattern and prevelence of alloimmunization in multiply transfused patients with sickle cell disease in Nigeria. Biomark Res 2015; 3(26): e4603770.

[20] Josephson CD, Su LL, Hillyer KL, Hillyer D. Transfusion in the patient with sickle cell disease: A critical review of the literature and transfusion guidelines. Transfus Med Rev 2007; 21(2): 118-33.

[21] Desai PC, Deal AM, Pfaff ER, Qaqish B, Hebden LM, Park YA, *et al*. Alloimmunization is associated with older age of transfused red blood cells in sickle cell disease. Am J Hematol 2015; 90(8): 691-5.

[22] Halawani HM, Al-Shahrani RS, Al-Harbi HA. Prevalence of stroke among patients with SCD at King Abdulaziz university hospital, Jeddah, Saudi Arabia. Med Sci 2020; 24 (102): 464-71.

[23] Natukunda B, Schonewille H, Ndugwa C, Brand A. Red blood cell alloimmunization in sickle cell disease patients in Uganda. Transfusion 2010; 5(1): 20-5.

[24] Alkindi S, AlMahrooqi S, AlHinai S, AlMarhoobi A, Al-Hosni S, Daar S, *et al*. Alloimmunization in patients with sickle cell disease and thalassemia: Experience of a single centre in Oman. Mediterr J Hematol Infect Dis 2017; 9(1): e2017013.

[25] Sharma S, Parikh L, Shah N, Waghela S, Ghildiyal R. Alloimmunization in children with sickle cell disease: A tertiary care experience. Pediatr Hematol Oncol J 2023; 8(2): 140-4.

[26] Linder GE, Chou ST. Red cell transfusion and alloimmunization in sickle cell disease. Haematologica 2021; 106(7): 1805-15.

[27] Venkateswaran L, Teruya J, Bustillos C, Mahoney Jr. D, Mueller BU. Red cell exchange does not appear to increase the rate of allo- and auto-immunization in chronically transfused children with sickle cell disease. Pediatr Blood Cancer 2011; 57: 294-6.

[28] Pirenne F. Heme control to major B (cell): are you listening? Blood 2021; 137(2): 153-4.

Received: December 09, 2024 Revised: February 17, 2025 Accepted: February 20, 2025

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