

ORIGINAL ARTICLE

The Prevalence of Lewis, Lutheran, and P1 Antigens and Phenotypes in Southwestern Saudi Arabia

Amr J. Halawani¹, Hisham I. Abu-Tawil^{2,3}, Yahya M. Dawood², Mohammed A. Ageeli²,
Essam A. Hamzi⁴, Fawaz I. Muyidi⁵, Saeed M. Kabrah¹, Hani Alnadawi¹,
Mohammed Almehmadi¹, Assaf Alnufai¹, Abdulrahman Alzubaidi¹,
Rama M. Chandika⁶, Haneen O. Alhaj⁷

¹ Department of Clinical Laboratory Sciences, Faculty of Applied Medical Sciences, Umm Al-Qura University, Makkah, Saudi Arabia

² Department of Laboratory and Blood Bank, Prince Mohammed bin Nasser Hospital, Ministry of Health, Jazan, Saudi Arabia

³ Department of Laboratory and Blood Bank, King Faisal Medical City for Southern Regions, Ministry of Health, Abha, Saudi Arabia

⁴ Department of Laboratory and Blood Bank, Altuwal Hospital, Ministry of Health, Jazan, Saudi Arabia

⁵ Department of Laboratory and Blood Bank, Alhurrath Hospital, Ministry of Health, Jazan, Saudi Arabia

⁶ Department of Clinical Nutrition, Faculty of Applied Medical Sciences, Jazan University, Jazan, Saudi Arabia

⁷ Kholoud Laboratory, Makkah, Saudi Arabia

SUMMARY

Background: Inherited hemoglobinopathies are common in Jazan Province, Saudi Arabia, and some patients may frequently require a blood transfusion. Therefore, the provision of compatible units using extended phenotypes is necessary to preclude the risk of alloimmunization. This study aimed to investigate the frequencies of the Lewis (LE), Lutheran (LU), and P1 antigens, as well as determine the prevalence of LE and LU phenotypes.

Methods: This study collected 150 blood samples from Saudi Arabian anonymous volunteering blood donors at Prince Muhammed bin Nasser Hospital in Jazan Province, Saudi Arabia. Serotyping was performed using antigen profile-II based on gel card technology to determine LE, LU, and P1 antigens.

Results: The prevalence of antigens was as follows: Le^a (n = 37, 24.6%), Le^b (n = 87, 58%), Lu^a (n = 6, 4%), Lu^b (n = 150, 100%), and P1 (n = 120, 80%). Regarding the LE phenotypes, Le (a⁺b⁻) was 24.7%, Le (a⁻b⁺) was 58%, and Le (a⁻b⁻) was 17.3%.

The frequencies of only observed LU phenotypes Lu (a⁻b⁺) and Lu (a⁺b⁺) were 96% and 4%, respectively.

Conclusions: In summary, this study reports LE, LU, and P1 antigen prevalence. Moreover, LE and LU phenotype frequencies were investigated. This study may help establish a national database of blood group antigens in Jazan Province, Saudi Arabia. Additionally, it may provide better transfusion practice to avoid the alloimmunization risk.

(Clin. Lab. 2023;69:xx-xx. DOI: 10.7754/Clin.Lab.2023.230609)

Correspondence:

Amr J. Halawani
Department of Clinical Laboratory Sciences
Faculty of Applied Medical Sciences
Umm Al-Qura University
Makkah
Saudi Arabia
Email: ajjhalawani@uqu.edu.sa

KEYWORDS

blood groups, blood transfusion, immunohematology, Saudi Arabia

INTRODUCTION

Jazan Province of Saudi Arabia is a region where hemoglobinopathies are endemic, including patients with thalassemia and sickle cell disease (SCD) [1]. These patients normally require frequent blood transfusions.

Blood banks are always challenged to provide matching and safe blood units to blood recipients. Hemolytic transfusion reactions, including alloimmunization, are major problems for transfusion-dependent patients [2]. Therefore, extending blood unit matching between blood donors and their recipients is crucial.

The International Society of Blood Transfusion (ISBT) identified 44 blood group systems, comprising 354 antigens [3]. These systems are encoded by 49 genes. Further, the ISBT maintained three categories for antigens that have yet to be associated with these systems. Antigens that are biochemically, serologically, or genetically related but lack a genetic foundation were grouped by creating collections (the 200 series) [4]. A system or collection cannot include the 700 Series, which contains antigens with an incidence of 1% across all ethnic backgrounds, and the 901 Series, which contains antigens with an incidence of > 90% [4].

ABO is the most important blood group system, followed by the RH system [5]. Frequencies of antigens, phenotypes, and haplotypes vary between different ethnicities [6,7]. Most of the blood group studies in Saudi Arabia were restricted to the ABO and RH blood group systems [8-12]. The national database is required to be updated regarding the other blood group systems, especially in areas where SCD and thalassemia are endemic. Therefore, many studies were conducted in Jazan Province regarding the other blood groups, including Kell (KEL), Duffy (FY), Kidd (JK), and MNS [13-16].

This study aimed to demonstrate the frequencies of antigens of Lewis (LE), Lutheran (LU), and P1 in Jazan Province, Saudi Arabia. Moreover, the prevalence of the LE and LU phenotypes was investigated. This may assist in establishing a database in Jazan region to provide antigen-negative blood units to transfusion-dependent patients, including patients with SCD and thalassemia. Furthermore, this will help to reduce the risk of alloimmunization in those patients.

MATERIALS AND METHODS

Blood samples

This study obtained 150 blood samples in EDTA (ethylenediaminetetraacetic acid) tubes from anonymous Saudi citizen blood donors living in Jazan Province, Saudi Arabia. The Jazan Hospital Institutional Review Board approved this study (NO. 2017). Participants donated blood voluntarily at the blood bank in Prince Muhammad bin Nasser Hospital after signing an agreement and filling out the donation consent form.

Inclusion criteria

Participants were voluntary Saudi citizens who meet the criteria of Saudi Arabia's Ministry of Health regarding official blood transfusion standards, including requirements for age, medical history, and being free from any transfusion-transmitted diseases.

Immunoematology

Serotyping was performed using a commercial kit based on the ID card antigen profile-I (P1, Le^a, Le^b, Lu^a, and Lu^b) based on gel card technology (DiaMed GmbH, Cressier, Switzerland) according to the manufacturer's instructions. Phenotyping was conducted using a 5% red cell suspension in ID-Diluent-1. Briefly, 12.5 µL of the 5% red cell suspension was added into all six microtubes containing specific test anti-sera. All cards were spun for 10 minutes at 85 x g using the ID-Centrifuge.

Interpretation of results

A pellet at the bottom of the microcells resulting from the reaction demonstrated a negative result and relevant antigen nonappearance. Conversely, a positive result, which was classified (1+, 2+, 3+, and 4+) based on the strong reaction point, making a distinct red line or cell diffusion, specified the presence of the corresponding antigen.

Statistical analyses

The sample size was calculated based on our previously reported study [17]. LE, LU, and P1 antigen prevalence, as well as LE and LU phenotypes, were presented and standardized as a percentage. A chi-squared test was performed. p-values of < 0.05 and < 0.01 indicated significant and highly significant differences, respectively.

RESULTS

This study analyzed 150 samples of LE, LU, and P1 antigens among Saudi blood donors living in Jazan Province, Saudi Arabia. Table 1 demonstrated the frequencies of these antigens. The most prevalent antigen observed among the entire samples of our study was Lu^b (100%). Conversely, the Lu^a antigen was the least prevalent antigen and found only in 6 (4%) samples. The frequencies of Le^a, Le^b, and P1 antigens were 37 (24.6%), 87 (58%), and 120 (80%), respectively.

Tables 2 and 3 demonstrate the LE and LU phenotypes determined in the Jazan population in comparison with other ethnicities, respectively. Regarding the LE blood group system, the most observed phenotype was Le (a⁻b⁺) at 58%. Homozygous phenotype Le (a⁺b⁺) was not observed. The most common phenotype detected concerning the LU phenotypes was Lu (a⁻b⁺) at 96%. A total of 4% was seen regarding the homozygous phenotype Lu (a⁺b⁺). Interestingly, Lu (a⁺b⁻) and Lu (a⁻b⁻) were not observed.

DISCUSSION

Widespread information on blood group frequency is crucial in transfusion practice. Therefore, several investigations on the prevalence of different blood kinds have already been conducted in southwestern Saudi Arabia [13-18]. These studies on blood groups may assist to

Table 1. The frequency of the LE, LU, and P1 antigens in Jazan Province, Saudi Arabia.

Antigen	Observation (n)	Frequency (%)
Le ^a	37	24.6
Le ^b	87	58
Lu ^a	6	4
Lu ^b	150	100
P1	120	80

Table 2. The frequencies of the LE phenotypes in the Jazan population compared to other ethnic backgrounds.

Phenotype	Current study (%)	Eastern Province of Saudi Arabia [21]	Indian [22]	Caucasian [6]	African [6]	Chinese [23]
Le (a ⁺ b ⁻)	24.7	29	21	22	23	14
Le (a ⁻ b ⁺)	58	49	61	72	55	59
Le (a ⁺ b ⁺)	0	3	0	rare	rare	4
Le (a ⁻ b ⁻)	17.3	19	19	6	22	23
[§] p-values		Jazan/Eastern p = 0.244	Jazan/Indian p = 0.767	Jazan/Caucasian p = 0.03 *	Jazan/African p = 0.007 **	Jazan/Chinese p = 0.04 *

[§] - Chi-squared test, * - significant, ** - highly significant.

Table 3. The prevalence of the LU phenotypes in the Jazan population compared to other ethnic backgrounds.

Phenotype	Current study (%)	Eastern Province of Saudi Arabia [21]	Indian [22]	Caucasian [6]	African [6]	Chinese [23]
Lu (a ⁺ b ⁻)	0	0	0	0	0	0
Lu (a ⁻ b ⁺)	96	97	96	92	92	98
Lu (a ⁺ b ⁺)	4	2	2	7	7	1
Lu (a ⁻ b ⁻)	0	1	1	Rare	Rare	0
[§] p-values		Jazan/Eastern p = 0.700	Jazan/Indian p = 0.710	Jazan/Caucasian p = 0.343	Jazan/African p = 0.343	Jazan/Chinese p = 0.177

[§] - Chi-squared test.

preclude the risk of red cell alloimmunization, particularly in multiply-transfused patients such as those with SCD [2,19-20].

Jazan Province of Saudi Arabia has a unique population with a variation regarding the blood groups compared to the other ethnic backgrounds [13-18]. The present study used gel card technology to investigate the antigen prevalence of the three blood group systems (LE, LU, and P1) among Saudi blood donors in southwestern Saudi Arabia.

Regarding the LE blood group system, the prevalence of Le^a and Le^b antigens in Jazan Province were 24.6% and 58%, respectively. These findings were congruent

with a previous study on the African population, in which the Le^a and Le^b antigens were 23% and 55%, respectively [6]. Conversely, the only reported study in Saudi Arabia was in the Eastern Province, in which the Le^a and Le^b antigens accounted for 32% and 52%, respectively [21]. Furthermore, the prevalence of Le^a and Le^b antigens is inconsistent with this study, which was 18% and 63%, respectively.

Concerning the LU blood groups, the concurrence of the Lu^a antigen accounted for 4%. Interestingly, this is similar to the reported African population (5%) and double the findings reported in the Eastern Province of Saudi Arabia (2%) [6,21]. Indians have the highest

prevalence outcome of the Lu^a antigen with 8% [22]. The Lu^b antigen was observed in the entire sample population with 100%. Similar conclusions were reported on African [6], Caucasian [6], and Chinese populations [23]. The Eastern Region of Saudi Arabia reported 99% compared to 80% reported in the present study for the P1 antigen [21]. The results were relatively similar to the reported studies in Caucasians (79%) and the Eastern Region of Saudi Arabia (85%) [6,21]. However, other studies, including Chinese (38%), Indians (72%), and Africans (95%), were inconsistent [6,22-23]. The present study determined LE and LU phenotypes among Saudi blood donors in Jazan Province, Saudi Arabia (Tables 2 and 3). The most prevalent LE phenotype in Jazan Province was Le (a⁻b⁺) accounting for 58%. However, the reported occurrence of this phenotype was higher in Caucasian (72%) [6]. Additionally, the null phenotype Le (a⁻b⁻) in the Caucasian population was the lowest at 6% [6] compared to the Jazan, which was 17.3%. The highest frequency of the null LE phenotype was reported in the African population at 22% [6]. Notably, variations were seen between the Jazan and Chinese populations regarding the following Le (a⁺b⁻), Le (a⁻b⁺), and Le (a⁻b⁻) [23]. A statistically significant difference was found between the population in our study with Caucasian ($p < 0.05$), Chinese ($p < 0.05$), and African populations ($p < 0.01$) as indicated in Table 2.

This study only revealed two LU phenotypes, including Lu (a⁻b⁺) and Lu (a⁺b⁺). The most common phenotype in Jazan Province, as well as the other ethnicities, was Lu (a⁻b⁺). The Lu (a⁺b⁺) was presented at 4% in the present study, which was twice the Eastern Province of Saudi Arabia (2%), and lower than the Caucasian (7%) and African population (7%) [6,21].

In summary, the study results demonstrated the frequencies of LE, LU, and P1 antigens among the Saudi blood donors living in Jazan Province, Saudi Arabia. Additionally, LE and LU phenotypes were determined. Interestingly, the frequencies of the current study vary between the Eastern Province of Saudi Arabia and other ethnic populations globally. These outcomes may help the local blood banks to establish better blood group antigen screening and typing protocols for providing safe blood and better transfusion practices.

Acknowledgment:

The authors gratefully acknowledge Enago (<https://www.enago.com>) for English language editing.

Declaration of Interest:

The authors declare no potential conflicts of interest concerning this article's research, authorship, and/or publication.

References:

1. Alhamsan NA, Almazrou YY, Alswaidi FM, Choudhry AJ. Pre-marital screening for thalassemia and sickle cell disease in Saudi Arabia. *Genet Med* 2007;9(6):372-7. (PMID: 17575503)
2. Halawani AJ, Mobarki AA, Arjan AH, et al. Red cell alloimmunization and autoimmunization among sickle cell disease and thalassemia patients in Jazan province, Saudi Arabia. *Int J Gen Med* 2022 Apr 15;15:4093-100. (PMID: 35450032)
3. International Society of Blood Transfusion. Red cell immunogenetics and blood group terminology [Online]. 2023. Available at: <http://www.isbtweb.org/working-parties/red-cell-immunogenetics-and-blood-group-terminology/> [Accessed 26 May 2023]
4. Storry JR, Castilho L, Chen Q, et al. International Society of Blood Transfusion working party on red cell immunogenetics and terminology: report of the Seoul and London meetings. *ISBT Sci Ser* 2016 Aug;11(2):118-22. (PMID: 29093749)
5. Poole J, Daniels G. Blood group antibodies and their significance in transfusion medicine. *Transfus Med Rev* 2007 Jan;21(1):58-71. (PMID: 17174221)
6. Reid ME, Lomas-Francis C, Olsson ML. The blood group antigen facts book. 3rd ed. Boston: Academic Press; 2012. <https://www.elsevier.com/books/the-blood-group-antigen-facts-book/reid/978-0-12-415849-8>
7. Daniels G. Human blood groups. 3rd ed. Oxford: Wiley-Blackwell; 2013:182. <https://doi.org/10.1002/9781118493595>
8. Bashwari LA, Al-Mulhim AA, Ahmad MS, Ahmed MA. Frequency of ABO blood groups in the eastern region of Saudi Arabia. *Saudi Med J* 2001 Nov;22(11):1008-12. (PMID: 11744976)
9. Sarhan MA, Saleh KA, Bin-Dajem SM. Distribution of ABO blood groups and rhesus factor in Southwest Saudi Arabia. *Saudi Med J* 2009 Jan;30(1):116-9. (PMID: 19139784)
10. Abdullah SM. Frequency of ABO and Rh blood groups in the Jazan region of Saudi Arabia. *Pak J Med Sci* 2010 Oct;26(4):818-21. <https://www.pjms.com.pk/issues/octdec2010/abstract/article15.html>
11. Eweidah MH, Rahiman S, Ali MH, Al-Shamary AMD. Distribution of ABO and rhesus (RHD) blood groups in Al-Jouf Province of the Saudi Arabia. *Anthropologist* 2011 Apr;13(2):99-102. <https://www.tandfonline.com/doi/abs/10.1080/09720073.2011.11891182>
12. Alzahrani M, Jawdat D, Alaskar A, Cereb N, Hajeer AH. ABO and Rh blood group genotypes in a cohort of Saudi stem cell donors. *Int J Immunogenet* 2018 Apr;45(2):63-4. (PMID: 29441679)
13. Halawani AJ, Arjan AH. ABO, RH, and KEL1 antigens, phenotypes and haplotypes in Southwestern Saudi Arabia. *Clin Lab* 2021 Feb 1;67(2). (PMID: 33616335)
14. Halawani AJ, Saboor M, Abu-Tawil HI, Mahzari AA, Mansor AS, Bantun F. Prevalence of Duffy blood group antigens and phenotypes among Saudi blood donors in Southwestern Saudi Arabia. *Clin Lab* 2021 Jan;67(1). (PMID: 33491438)
15. Halawani AJ, Saboor M, Abu-Tawil HI, et al. The frequencies of Kidd blood group antigens and phenotypes among Saudi blood donors in Southwestern Saudi Arabia. *Saudi J Biol Sci* 2022 Jan; 29(1):251-4. (PMID: 35002415)

16. Halawani AJ, Habibullah MM, Dobie G, et al. Frequencies of MNS blood group antigens and phenotypes in southwestern Saudi Arabia. *Int J Gen Med* 2021 Dec;14:9315-9. (PMID: 34887679)
17. Halawani AJ, Mansor AS, Assaggaf HM, et al. Investigation of dombrock blood group alleles and genotypes among Saudi blood donors in Southwestern Saudi Arabia. *Genes* 2022 Jun;13(6):1079. (PMID: 35741842)
18. Saboor M, Zehra A, Hamali HA, et al. Prevalence of A2 and A2B subgroups and anti-A1 antibody in blood donors in Jazan, Saudi Arabia. *Int J Gen Med* 2020 Oct;13:787-90. (PMID: 33116770)
19. Yazdanbakhsh K, Ware RE, Noizat-Pirenne F. Red blood cell alloimmunization in sickle cell disease: pathophysiology, risk factors, and transfusion management. *Blood* 2012 Jul;120(3):528-37. (PMID: 22563085)
20. Nickel RS, Hendrickson JE, Fasano RM, et al. Impact of red blood cell alloimmunization on sickle cell disease mortality: a case series. *Transfusion* 2016 Jan;56(1):107-14. (PMID: 26509333)
21. Owaidah AY, Naffaa NM, Alumran A, Alzahrani F. Phenotype frequencies of major blood group systems (Rh, Kell, Kidd, Duffy, MNS, P, Lewis, and Lutheran) among blood donors in the eastern region of Saudi Arabia. *J Blood Med* 2020 Feb;11:59-65. (PMID: 32104128)
22. Thakral B, Saluja K, Sharma RR, Marwaha N. Phenotype frequencies of blood group systems (Rh, Kell, Kidd, Duffy, MNS, P, Lewis, and Lutheran) in north Indian blood donors. *Transfus Apher Sci* 2010 Aug;43(1):17-22. (PMID: 20558108)
23. Yu Y, Ma C, Sun X, et al. Frequencies of red blood cell major blood group antigens and phenotypes in the Chinese Han population from Mainland China. *Int J Immunogen* 2016 Aug;43(4):226-35. (PMID: 27320061)