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**Original Research Article** 

# To Determine the Prevalence of Subgroups of Blood Group A among the Blood Donors at Tertiary Care Hospital in Western Rajasthan, India

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**Conflict of interest: Nil** 

Abstract

**Background:** To determine the prevalence of the subgroup of blood group A among blood donors at a tertiary care hospital in Western Rajasthan, India

**Methods:** The current cross-sectional study conducted at the Department of Immunohematology and Transfusion Medicine, Sardar Patel Medical College, Bikaner, aimed to determine the prevalence of A subgroups (A1 and A2) among blood donors in Western Rajasthan, India. This investigation provides valuable insights into the regional distribution of these important blood subgroups, particularly relevant in transfusion medicine and organ transplantation practices.

**Results:** A total of 52,986 donors were included in the study, with blood grouping and subgrouping performed using a fully automated NEO IRIS analyzer, complemented by anti-A1 lectin for identification of A1 and A2 subgroups. The results showed that among 11,347 donors belonging to blood group A, the A₁ subgroup was found to be the most common, accounting for 10,691 donors (94.27%), while the A₂ subgroup was observed in 650 donors (5.73%).similarly among a total of 4716 donors belonging to the AB blood group, the A₁B subgroup was found to be predominant, accounting for 95.61% (≈4509 donors), while the A₂B subgroup was observed in 4.39% (≈209 donors).

Conclusion: The study confirms that the A1 subgroup is significantly more prevalent than the A2 subgroup among both A+ve and A-ve blood and AB+ve and AB-ve donors in Western Rajasthan. This trend mirrors findings in many Indian and nearby regional populations. Though A2 is less common, its detection is clinically important, as individuals with this subgroup may develop anti-A1 antibodies, leading to transfusion complications. The use of sensitive and automated systems like NEO IRIS enhances subgroup identification accuracy.

# **Keywords:** Blood, Donor, Subgroups.

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## Introduction

Blood transfusion is a life-saving procedure and a critical component of modern healthcare. The success of transfusion depends on the accurate determination and matching of blood groups to prevent transfusion reactions, which can range from mild to life-threatening. The ABO blood group system, first discovered by Karl Landsteiner in 1901, is the most important and clinically significant blood grouping system. It is determined by the presence or absence of antigens (A and B) on red blood cells and corresponding antibodies in plasma. The A blood group is one of the four major blood groups in the ABO system, and it is further classified into subgroups based on the quantity and nature of A antigen expressed on the surface of red

blood cells. [1-2] These subgroups can be distinguished from each other by reacting with Anti-A1 Lectin which occurs as a cold agglutinin and reacts with A1 cells. Anti-A1 lectin obtained from Dolichos Biflorus seeds is routinely used in blood banks to differentiate between A1 and A2 red blood cells. Persons having Blood Group-A2 and A2B may have anti-A1 antibody. Usually, anti-A1 exists as cold antibody which is naturally occurring, with a thermal amplitude of less than 25°C. However, cases of anti-A1 reacting at 37° C have also been reported in the literature. [3,4] The A1 and A2 subgroups are different from each other both qualitatively and quantitatively, with A1 red cells having 8.1 - 11.7×105 antigenic sites as

compared to 2.4–2.9×105 antigenic sites on A2 red cells. In routine testing, both A1 and A2 are strongly agglutinated by anti-A antiserum but problem arises when A2 blood group individuals develop anti-A1 antibody which can cause discrepancies in blood grouping. [5] When anti-A1 is active at body temperature, though rare, extensive destruction of A1 cells in vivo can occur and has been documented to cause severe hemolytic transfusion reaction. It is also important in cases of organ transplant. [6]

#### **Materials and Methods**

**Study Site:** Department of Immunohematology and Transfusion Medicine Sardar Patel Medical College and Associated Groups of Hospitals, Bikaner, Rajasthan, India.

**Study Design:** This study is a cross-sectional observational study conducted to determine the prevalence of subgroups of blood group A among blood donors at a tertiary care hospital in Western Rajasthan, India.

**Study Setting:** The study was conducted at the blood center department of a tertiary care hospital in Western Rajasthan, India.

**Study Period:** The study was carried out over a period of 15 months, from 1 January 2024 to 31 March 2025

**Study Population:** The study included all blood donors who donated blood at the tertiary care hospital during the study period.

**Sample Size:** The sample size consisted of all eligible blood donors who donated blood during the study period.

$$n = \frac{Z^2 X p X (1 - p)}{d^2}$$

- n = required sample size
- Z = Z-score (value from standard normal distribution corresponding to desired confidence level)
- Z = 1.96 for 95% confidence
- p = expected prevalence (as a proportion, i.e., 20% = 0.20)
- d = desired precision or margin of error (as a proportion, i.e., 5% = 0.05)

$$n = \frac{(1.96)^2 \cdot 0.20 \cdot (1 - 0.20)}{(0.05)^2} = \frac{3.8416 \cdot 0.20 \cdot 0.80}{0.0025} = \frac{0.614656}{0.0025} = 246$$

Although the minimum calculated sample size was 246, our study included all 52986 eligible blood donors over the study period. This approach was adopted to maximize statistical precision, reduce error margins, and ensure comprehensive evaluation of the pooled versus three-cell panel techniques. Inclusion of the entire donor population

not only enhances the reliability of prevalence estimates but also aligns with routine blood centre screening practices, making the study both feasible and highly representative, which offers several advantages:

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- 1. Increased Statistical Power: A larger sample enhances the reliability of results, reducing the margin of error and increasing confidence in the findings.
- Greater Representativeness: The large sample better reflects population diversity, allowing more accurate subgroup analyses and improving external validity.
- 3. Detection of Rare Events: With a larger cohort, it becomes feasible to identify and analyze less common occurrences or subgroups that may not be visible in smaller studies.
- 4. Stronger Basis for Conclusions: The scale of the data supports more robust, evidence-based conclusions that can potentially influence policy or clinical practice.

**Sampling Technique:** All whole blood donors donating at our Blood Centre in the duration of the study, meeting all inclusion criteria and not any of the exclusion criteria.

## **Inclusion Criteria:**

- 1. Whose age is 18-65 years for regular and 18-60 years for non-regular donors
- 2. Whose weight is >45 kg for collection of 350ml whole blood and >55kg for collection of 450ml whole blood
- 3. Hb >12.5gm/dl or equal to
- 4. There is no history of blood donation in last 3 months in males and 4 months in females
- 5. No complaints of any acute and chronic disease.
- 6. Not taking any medicine enlisted in causes for deferral from blood donation.
- 7. No history of any major surgery in the last 12 months
- 8. No history of vaccination enlisted in causes for deferral within deferral time limit for particular vaccine.
- 9. Non-pregnant and non-lactating female.
- 10. No history of blood transfusion in last 12 months
- 11. Following donor fitness criteria as per the revised guidelines of Drug and Cosmetic Act.

## **Exclusion Criteria:**

- Donors who did not provide informed consent.
- Donors who were ineligible according to the hospital's blood donation criteria.

# **Data Analysis:**

• The prevalence of each subgroup (A1 and A2) among blood group A donors was calculated.

- Descriptive statistics were used to summarize the data, including the number and percentage of each subgroup.
- The demographic characteristics of the donors (age, gender) were also analyzed.

# **Software Used:**

Data analysis was performed using SPSS version 23.0.

### **Outcome Measures:**

- Primary outcome: Prevalence of A1 and A2 subgroups among blood group A donors.
- Secondary outcome: Distribution of subgroups based on demographic characteristics.

#### **Observations**

In this study, the majority of donors belonged to the 21–30 years age group, comprising 50.04% (26,512 individuals) of the total 52,986 cases. This was followed by the 31–40 years age group with

29.56% (15,665 cases), and the 41–50 years group accounting for 10.32% (5,468 cases). The 18-20 years group represented 8.05% (4,264 cases), while individuals aged over 50 years made up the smallest proportion at 2.03% (1,077 cases). The distribution of donors according to gender showed a significant predominance of males, with 52,532 donors accounting for 99.14% of the total. In contrast, only 454 donors were females, making up just 0.86%. This indicates a highly male-dominated sample population out of the total 52,986 cases. The distribution of donors according to blood group revealed that the most common blood group was B+ve, found in 17,723 individuals (33.45%), and followed by O+ ve in 15,732 donors (29.69%). A+ve was present in 10,354 donors (19.54%). Other groups included AB+ve (8.13%), B-ve (3.37%), O-ve (3.19%), A-ve (1.86%), and the least common was AB-ve at 0.77%. These results indicate a predominance of B+ve and O+ve blood groups in the population studied.

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Table 1: Distribution of donors according to Sub Group in A+ve

Sub Group	Number	Percentage (%)
A1	9782	94.42
A2	572	5.52
Total	10360	100.00

In this study, among the 10,360 blood donors with the A positive (A+ve) blood group, the majority belonged to the A1 subgroup, which accounted for 9,782 individuals, making up 94.42% of the total A+ve donors. A smaller portion of donors, 572

individuals or 5.52%, were identified as belonging to the A2 subgroup. This result shows that the A1 subgroup is far more common than the A2 subgroup among people with A+ve blood in this population.

Table 2: Distribution of donors according to Sub Group in A -ve

Sub Group	Number	Percentage (%)
A1	909	92.10
A2	78	7.90
Total	987	100.00

In this study, out of 987 donors with the A negative (A-ve) blood group, the vast majority—909 individuals or 92.10%—belonged to the A1 subgroup. In contrast, only 78 donors, making up

7.90% of the A-ve group, were identified as A2 subgroup. This indicates that, similar to the A+ve group, the A1 subgroup is also predominant among individuals with A-ve blood.

Table 3: Distribution of donors according to Sub Group in AB+ve

Sub Group	Number	Percentage (%)
A1B	4177	97.01
A2B	129	2.99
Total	4306	100.00

In the AB+ve blood group, the A1B subgroup was overwhelmingly predominant, comprising 4177 donors (97.01%), while the A2B subgroup accounted for only 129 donors (2.99%). This shows that A1B is the most common subtype among AB+ve individuals.

Table 4: Distribution of donors according to Sub Group in AB-ve

Sub Group	Number	Percentage (%)
A1B	332	80.98
A2B	78	19.02
Total	410	100.00

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In individuals with the AB-negative blood group, the majority belonged to the A1B subgroup, comprising 332 donors (80.98%), while the A2B subgroup accounted for 78 donors (19.02%). This shows that A1B is the predominant subtype among AB-negative individuals in the study population.

Table 5: Distribution of donors according to Sub Group among the A GROUP

Sub Group	Number	Percentage (%)
A1	10691	94.27
A2	650	5.73
Total	11341	100.00

In the present study, among 11,341 donors belonging to blood group A, the A<sub>1</sub> subgroup was found to be the most common, accounting for 10,691 donors (94.27%), while the A<sub>2</sub> subgroup was observed in 650 donors (5.73%).

This finding indicates that the  $A_1$  phenotype is predominant among the A group population in this study. The lower frequency of  $A_2$  subgroup is consistent with the general population data, where  $A_2$  usually constitutes 4–8% of all A group individuals. The variation in the proportion of  $A_1$  and  $A_2$  subgroups between different studies may be

due to regional genetic differences, sample size, or methodology used (such as tube test or SPRCA). Identification of A<sub>1</sub> and A<sub>2</sub> subgroups is important in blood grouping because a small proportion of A<sub>2</sub> and A<sub>2</sub>B individuals may develop anti-A<sub>1</sub> antibody, which can cause ABO discrepancies or incompatible crossmatches if not properly recognized.

Thus, the present study emphasizes the need for routine A subgrouping, especially in donors and patients showing unexpected reactions during blood grouping.

Table 6: Distribution of donors according to Sub Group A among the AB GROUP

Sub Group	Number	Percentage (%)
A1B	4509	95.61
A2B	207	4.39
TOTAL	4716	100

In the present study, among a total of 4716 donors belonging to the AB blood group, the A1B subgroup was found to be predominant, accounting for 95.61% (≈4509 donors), while the A<sub>2</sub>B subgroup was observed in 4.39% (≈209 donors). This observation shows that A1B is the major subtype among AB group individuals, which is in agreement with findings from other studies conducted in different regions of India. The lower frequency of the A2B subgroup is expected, as it generally represents 2–5% of the AB group in most populations. The difference between A<sub>1</sub>B and A<sub>2</sub>B frequencies can be attributed to the genetic predominance of the A<sub>1</sub> allele and higher antigenic expression of A<sub>1</sub> antigen compared to A<sub>2</sub>. From a perspective, transfusion identifying subgroups is significant because some A2B individuals may produce anti-A1 antibody, which can lead to ABO grouping discrepancies or incompatible crossmatches if not properly identified. Therefore, routine determination of A subgroups, particularly among AB donors, is important to ensure accurate blood typing and safe transfusion practices.

## Discussion

The current cross-sectional study conducted at the Department of Immunohematology and Transfusion Medicine, Sardar Patel Medical College, Bikaner, aimed to determine the prevalence of A subgroups (A1 and A2) among blood donors in Western Rajasthan, India. This investigation provides valuable insights into the regional distribution of these important blood subgroups, particularly relevant in transfusion medicine and organ transplantation practices.

Table 6: Comparative table of various Indian studies of ABO Subgroup Distribution among blood donors

Author	Year	Place of Study	Sample Size	A1	A2	A1B	A2B
Meher et al [7]	2015	Mumbai	1036	96.8%	3.2%	98.3%	1.69%
Bhora et al [8]	2017	Jodhpur	1000	84.9%	15.1%	81.6%	18.4%
S. Giriyan [9]	2017	Karnataka	546	88.9%	11.1%	89.7%	10.3%
Yadav et al [10]	2019	Bhopal	720	91.0%	9.0%	84.37%	15.63%
Kalita et al [11]	2020	Guwahati	1354	93.94%	6.06%	91.04%	8.96%
Biswas et al [12]	2020	Eastern India	67954	82.8%	17.15%	84.93%	15.05%
Wasnik et al [13]	2024	Chhattisgarh	412	66.7%	33.3%	88.2%	11.8%
Present study	2025	SPMC, Bikaner	52,986	94.27%	5.73%	95.61%	4.39%

Several studies across India have reported varying distributions of ABO subgroup A among blood donors, reflecting genetic and ethnic diversity. Meher et al. (2016) conducted a study in Mumbai on 1036 donors and reported A1 at 96.8%, A2 at 3.2%, A1B at 98.3%, and A2B at 1.69%. Bhora et al. (2017) in Jodhpur found a comparatively lower A1 frequency (84.9%) and higher A2B percentage (18.4%) in 1000 donors.

In Karnataka, S. Giriyan et al. (2017) reported 88.9% A1, 11.1% A2, 89.7% A1B, and 10.3% A2B from 546 samples. Yadav et al. (2019) observed 91.0% A1 and 9.0% A2 among 720 donors in Bhopal, while Kalita et al. (2020) in Guwahati reported a notably lower A2B at 8.96% among 1354 donors. A large cohort by Biswas et al. (2020) from Eastern India (n=6794) recorded A2 at 17.15%, A2B at 15.05%, indicating significant A2 prevalence. Mangwana et al. (2021) in New Delhi and Wasnik et al. (2024) in Chhattisgarh also

highlighted increased A2 proportions at 15.3% and 33.3%, respectively, with A2B at 10.6% and 11.8%, pointing to regional variance. The current study (2025) from SPMC, Bikaner evaluated donors, identifying a predominant A1 subgroup (94.27%), with only 5.73% among A group while among AB donors A1B constituted 95.61% and A2B 4.39%.

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This data is clinically significant in transfusion medicine. Subgroups of A, particularly A2 and A2B, can occasionally develop anti-A1 antibodies, which may lead to ABO discrepancies in blood grouping or transfusion reactions if not identified properly. Recognizing the prevalence of these subgroups aids in better compatibility testing, donor selection, and transfusion safety, especially in organ transplantation and immunohematology workups. Regional differences also highlight the importance of local subgroup data in shaping transfusion policies and reference databases.

Table 7: Comparative table of various foreign studies of ABO Subgroup Distribution among blood donors

Author	Year	Place of Study	Sample Size	A1	A2	A1B	A2B
Olantunji et al [14]	2008	Egypt	200	94.4%	6.6%	91.7%	8.3%
Keokhamphou et al [7]	2012	Thailand	178	77.5%	4.5%	6.5%	1.5%
Parrajaramillo et al [15]	2014	Colombia	172	72.2%	4%	4.6%	1.7%
Omer et al [16]	2016	Sudan	200	93.4%	6.6%	91.6%	8.33%
Chiewsilp et al [17]	2017	Thailand	1302	83.6%	6.6%	91.7%	6.1%
Abubakar et al [18]	2020	Nigeria	37	90.9%	9.1%	0.0%	0.0%
Saboor et al [9]	2020	Pakistan	446	99.1%	0.4%	99.1%	0.4%
Abdelaziz et al [8]	2021	Fayoum, Egypt	10662	99.2%	0.4%	80.7%	0.4%
Abdelaziz et al [10]	2021	Egypt	2832	96.5%	1.05%	94.05%	1.05%
Present study	2025	SPMC, Bikaner	52986	94.27%	5.73%	95.61%	4.39%

The compiled data from various international and national studies provides a comparative overview of the distribution of ABO subgroups (A1, A2, A1B, and A2B) among blood donors across different geographic locations. It was observed that the A1 subgroup is the most predominant phenotype in nearly all regions, with frequencies ranging from approximately 72.2% to as high as 99.2%. This trend is especially prominent in countries such as Pakistan, Egypt, and India, where studies reported A1 prevalence above 96%. In contrast, lower A1 frequencies were reported in populations from Colombia and Thailand, suggesting regional or ethnic variability.

The subgroups A2 and A2B were consistently found to be less common, usually comprising less than 5% of the total population. In some instances, such as in the Nigerian study, A2 and A2B subgroups were entirely absent. This rarity of A2 and A2B highlights the importance of sensitive detection techniques, as their presence may have clinical implications. The A1B subgroup showed considerable variation in its distribution, from as

low as 4.6% to over 91%, which could reflect genetic diversity, population structure, or differences in testing methods.

The significance of these findings lies primarily in their implications for transfusion medicine and organ transplantation. Individuals with A2 or A2B subgroups may develop naturally occurring anti-A1 antibodies, which can sometimes be reactive at body temperature and cause transfusion reactions or ABO discrepancies if not properly identified. Furthermore, accurate determination of ABO subgroups is critical in organ transplantation, particularly in A2-incompatible grafts, where misclassification can lead to graft rejection. The marked differences in subgroup prevalence across studies underscore the influence of ethnicity and genetics on blood group distribution. This underlines the need for establishing local baseline data to guide safe transfusion practices and donor screening protocols. Moreover, this analysis contributes valuable epidemiological insights, reinforcing the necessity for routine ABO subgrouping, especially in high-volume transfusion

centers and transplantation units. Incorporating subgrouping into standard blood centre practices not only enhances patient safety but also supports more precise compatibility assessments, thereby improving overall transfusion outcomes.

## **Clinical Significance of Subgroup Identification**

The clinical implications of accurately identifying A1 and A2 subgroups are immense. A2 individuals may occasionally produce anti-A1 antibodies, which can lead to complications during transfusion or organ transplantation. For instance, Parra-Jaramillo (2018) noted hemolytic disease of the newborn and transplant rejections related to subgroup mismatches. These complications can be avoided by routine testing for A subgroups using anti-A1 lectin, especially in cases involving transfusion recipients and organ transplant donors/recipients.

Moreover, Sant'Anna Gomes et al. (2010) highlighted that A1 individuals may express higher levels of A antigens on platelets, which can be misinterpreted during platelet crossmatching and can influence transfusion strategies. They also emphasized the role of inherited patterns in A antigen expression, suggesting a hereditary basis for A1 and A2 subtypes.

The slightly higher frequency of A2 among A-ve donors (7.9%) as compared to A+ve donors (5.52%) in our study may warrant further genetic investigation, especially considering the limited sample size in the A-ve group. This could indicate either a real population trend or statistical variability due to a smaller denominator in the A-ve category.

## Conclusion

The study confirms that the A1 subgroup is significantly more prevalent than the A2 subgroup among both A+ve and A-ve blood donors in Western Rajasthan. This trend mirrors findings in many Indian and nearby regional populations. Though A2 is less common, its detection is clinically important, as individuals with this subgroup may develop anti-A1 antibodies, leading to transfusion complications. The use of sensitive and automated systems like NEO IRIS enhances subgroup identification accuracy. recommended that blood centers and transfusion centers routinely perform A subgrouping, particularly for A and AB group donors, to ensure safer transfusion practices and improve overall patient outcomes.

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