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Research Article

Distribution And Comparative Analysis Of Anti- A And Anti- B Igm And Igg Antibody Titres Among Blood Donors At A Tertiary Care Hospital.

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Abstract: ***Introduction:*** Blood transfusion is one of the most vital and frequently performed medical procedures worldwide, saving millions of lives annually and playing a crucial role in the management of various haematological, surgical, obstetric, and emergency conditions. ***Aim:*** To estimate and compare Anti-A and Anti-B IgM and IgG antibody titres among blood donors across different ABO blood groups at a tertiary care hospital blood centre. ***Methodology:*** A descriptive cross-sectional study was conducted in 400, Group "ABO" blood donors in the Department of Immuno- Haematology and Transfusion Medicine, S.P. Medical College and Associated Group of P.B.M. Hospitals, Bikaner, fulfilling the laid down donor selection criteria from 1st December 2025 to 31st May 2026. ***Result:*** Group O donors showed substantially higher IgG titres, with elevated anti-A IgG in 61.6% and anti-B IgG in 55.8%; IgM titres were comparatively lower, with anti-A IgM more frequent than anti-B IgM. ***Conclusion:*** Simultaneous assessment of IgM and IgG using the NEO Iris platform helps identify high-titre ABO antibodies, particularly among group O donors, supporting method-specific screening to improve transfusion safety.

Keywords: ABO Blood Group, Haemagglutination, High-Titre Antibodies.

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INTRODUCTION

Blood transfusion is one of the most vital and frequently performed medical procedures worldwide, saving millions of lives annually and playing a crucial role in the management of various haematological, surgical, obstetric, and emergency conditions.1 Ensuring the safety and efficacy of blood transfusions is a complex process involving rigorous donor screening, compatibility testing, and post-transfusion monitoring. Among these, understanding the immunohaematological characteristics of blood donors, particularly the presence and titres of naturally occurring antibodies, is vital to prevent adverse transfusion reactions, especially haemolytic transfusion reactions (HTRs) which can be fatal. One of the major donor considerations in blood transfusion is to confirm whether the donation is safe for both donor and patient.2,3 Anti-Aand anti-B are naturally occurring antibodies of the immunoglobulin (Ig) M class and are present in practically all humans who lack the corresponding antigen.4 These antibodies will react immediately with a transfused antigen-positive cell and in most cases result in an intravascular haemolysis of the cell.

High titres of anti-A and anti-B seem to be one reason for haemolytic transfusion reactions (HTRs) and for ABO haemolytic disease in fetuses and neonates. Even though group specific packed red blood cells and component preparation have become widely used, haemolytic reactions are still occasionally recorded, albeit much less frequently5. Historically, the concept of antibody titres has been used predominantly in the context of Rh isoimmunisation and haemolytic disease of the fetus and newborn (HDFN). However, with the evolution of transfusion medicine, attention has extended to ABO isoagglutinins due to their immediate clinical significance. High anti-A or anti-B titres are associated with increased risk of HTRs, which can manifest as haemolysis, renal failure, disseminated intravascular coagulation6, or even death.

Levels of anti-A and anti-B antibodies appear to be influenced by environmental factors. Changes in the characteristics of these antibodies result from immunisation, pregnancy, or incompatible transfusions, especially in plasma incompatible platelet transfusions.Understanding the regional variations in anti-A and anti-B titres is important for establishing safe transfusion practices,7,8 especially considering the increasing demand for blood components and plasma-based therapies. It also aids in formulating protocols for donor screening, plasma donation, and managing transfusion reactions effectively.While the importance of antibody titres is well recognized, standardisation of measurement techniques and establishment of cut-off levels remain challenging. Different laboratories employ varying methodologies, including tube techniques, gel card methods, enzyme-enhanced assays, and flow cytometry, leading to inconsisten results. Variability also exists in defining 'high' titres, with thresholds ranging from 1:64 to 1:256 or higher, depending on institutional policies and regional practices.Moreover, the relationship between antibody titres and clinical outcomes is complex.

Some donors with high titres may never cause haemolytic reactions, whereas others with lower titres may do so under certain conditions.5 Factors such as antibody class (IgM vs. IgG), affinity, and recipient susceptibility also influence clinical risk.9 Therefore, establishing region-specific, evidence-based cut-off titres is essential for safe blood component management.The study was conducted in the Department of Immuno-Haematology & Transfusion Medicine, Sardar Patel Medical College & Associated Group of Hospitals, Bikaner, Rajasthan, India to assess the levels of anti-A and anti-B antibodies in blood donors to improve transfusion safety. This study aims to estimate the titres of anti-A and anti-B agglutinins in blood donors using the ABO blood grouping system at a tertiary care hospital in Western Rajasthan, with the overarching goal of enhancing transfusion safety.

AIM

To estimate and compare Anti-A and Anti-B IgM and IgG antibody titres among blood donors across different ABO blood groups at a tertiary care hospital blood centre.

METHODOLOGY

A descriptive cross-sectional study was conducted in 400, Group "ABO" blood donors in the Department of Immuno- Haematology and Transfusion Medicine, S.P. Medical College and Associated Group of P.B.M. Hospitals, Bikaner, fulfilling the laid down donor selection criteria from 1st December 2025 to 31st May 2026. Ethical Approval was obtained from the ethical committee of S.P. Medical College and Associated Group of P.B.M. Hospitals, Bikaner. The sample size was calculated based on a previous study40 that reported that, overall prevalence of ABO Anti-A and Anti-B Agglutinin Titers among Blood Donors in India 46% per 1000 populations. 95% level of confidence and Error rate set at 0.5 levels. Simple Random Sampling techniques were used. The age group to be included in the study is 18–60 years.Inclusion criteria included Donors who fulfilled the eligibility criteria as per the Drugs and Cosmetics Act, 1940 and Rules, 194550 will be included in the study after getting the written informed consent.Exclusion criteria involved AB blood group donors, donors with irregular red cell antibodies, those reactive for HIV, Hepatitis B, Hepatitis C, malaria, and syphilis, and those who had not given consent were excluded from the study.Informed Consent: Obtaining informed consent from donors prior to including them in the study. Confidentiality: Donor confidentiality was maintained throughout the study, adhering to relevant guidelines for data protection and anonymity.

RESULTS

Table 1: Age group and sex distribution

Age group (years)	Female n (%)	Male n (%)	Total n (%)
18-30	11 (6.8)	150 (93.2)	161 (40.2)
31-40	12 (10.2)	106 (89.8)	118 (29.5)
41-50	7 (8.3)	77 (91.7)	84 (21.0)
51-60	6 (16.2)	31 (83.8)	37 (9.2)
Total	**36 (9.0)**	**364 (91.0)**	**400 (100.0)**

The majority of donors belonged to the 18-30-year age group (161, 40.2%). Males predominated in the cohort, contributing 364 (91.0%) donors.

Table 2: ABO blood group and Rh type distribution

Blood group	Rh Negative n (%)	Rh Positive n (%)	Total n (%)
A	5 (5.8)	81 (94.2)	86 (21.5)
B	9 (6.3)	133 (93.7)	142 (35.5)
O	8 (4.7)	164 (95.3)	172 (43.0)
Total	**22 (5.5)**	**378 (94.5)**	**400 (100.0)**

Blood group O was the most frequent ABO group, followed by B and A. Rh-positive status was predominant across all ABO groups.

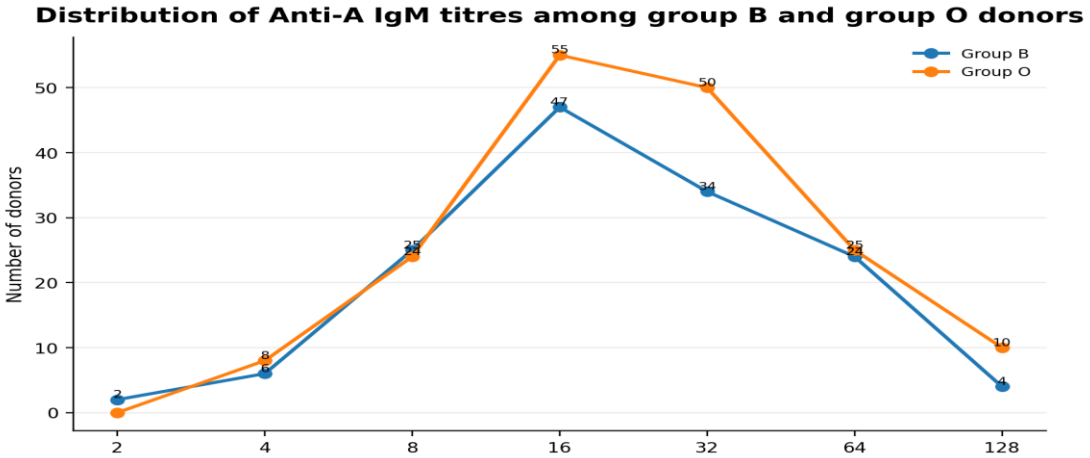


Figure 1: Distribution of Anti-A IgM titres among group B and group O donors

Anti-A IgM titres were applicable to group B and group O donors. The most frequent Anti-A IgM titre was 16, followed by 32.

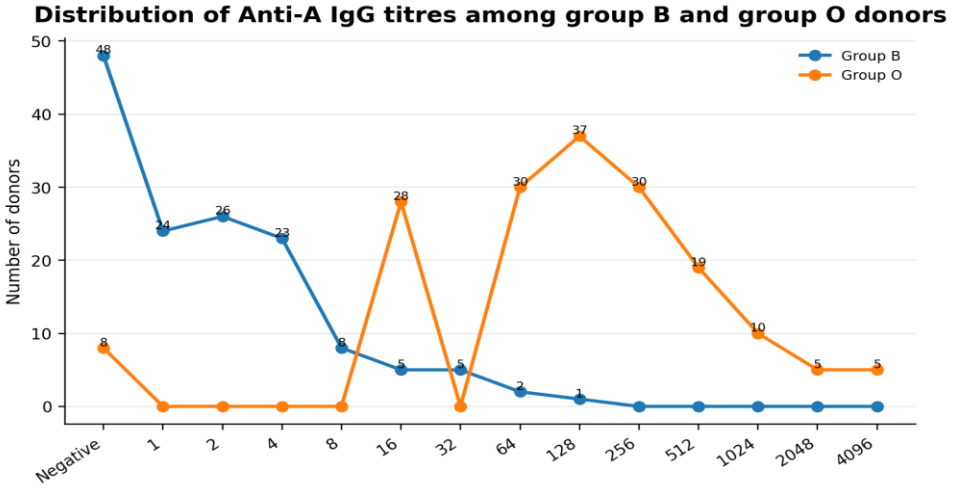


Figure 2:Distribution of Anti-A IgG titres among group B and group O donors

Negative results were coded as 0 for statistical analysis. Percentages are calculated within each column denominator; total denominator n=314. Anti-A IgG titres were markedly higher among group O donors than among group B donors. Group O donors showed titres extending up to 4096.

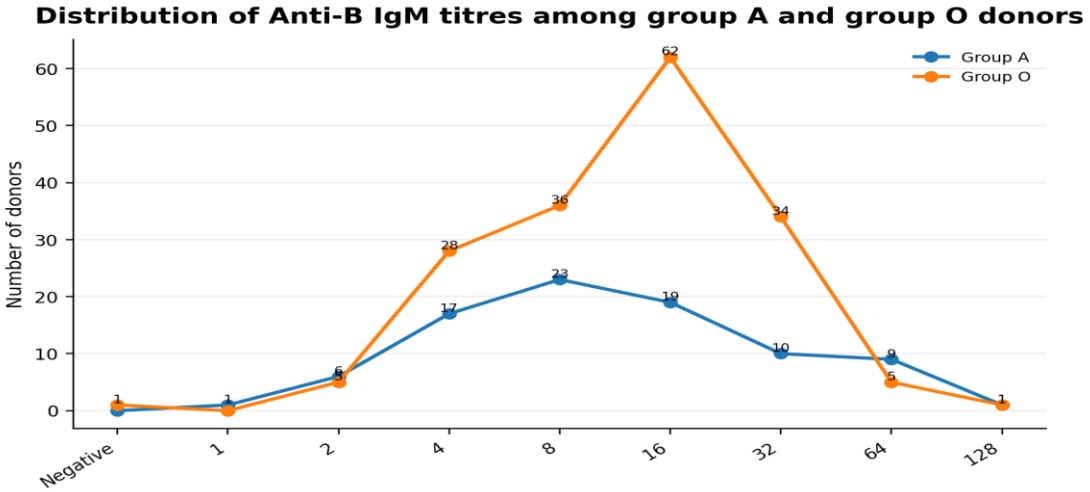


Figure 3: Distribution of Anti-B IgM titres among group A and group O donors

Percentages are calculated within each column denominator; total denominator n=258.

Anti-B IgM titres were applicable to group A and group O donors. The most frequent Anti-B IgM titre was 16.

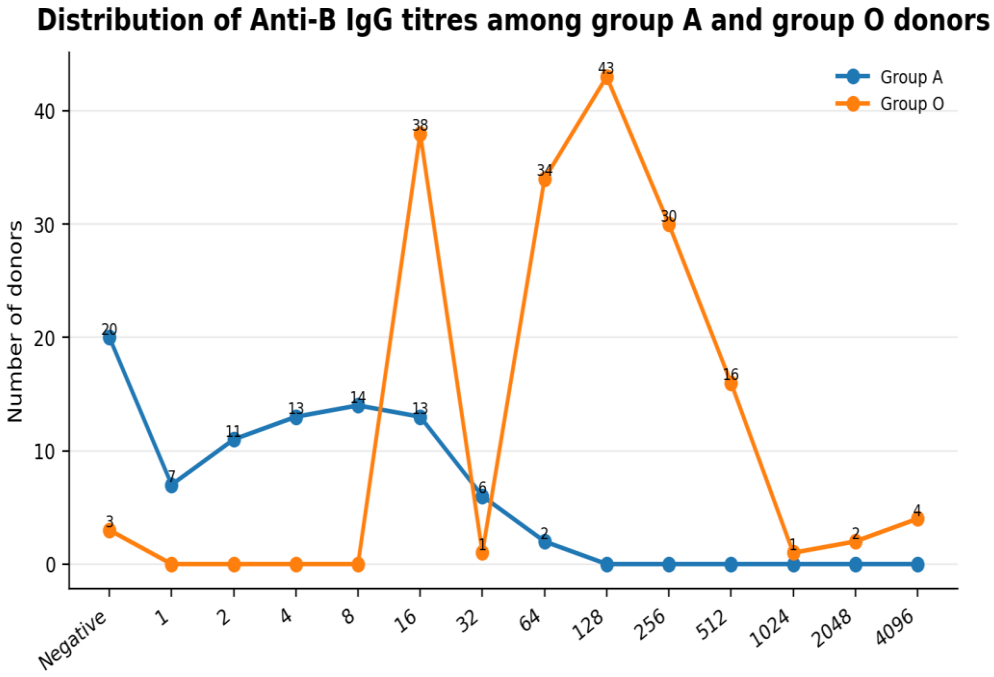


Figure 4: Distribution of Anti-B IgG titres among group A and group O donors

Negative results were coded as 0 for statistical analysis. Percentages are calculated within each column denominator; total denominator n=258. Anti-B IgG titres were substantially higher among group O donors than among group A donors. Group O donors showed a wider titre range and more frequent high titres.

Table 3: Median titre and interquartile range of ABO antibodies

Antibody	Blood group	n	Median (IQR)	Minimum	Maximum
Anti-A IgM	Group B	142	16 (16-32)	2	128
Anti-A IgM	Group O	172	16 (16-32)	4	128
Anti-A IgM	Overall	314	16 (16-32)	2	128
Anti-A IgG	Group B	142	1 (0-4)	Negative	128
Anti-A IgG	Group O	172	128 (64-256)	Negative	4096
Anti-A IgG	Overall	314	16 (1-128)	Negative	4096
Anti-B IgM	Group A	86	8 (4-16)	1	128
Anti-B IgM	Group O	172	16 (8-16)	Negative	128
Anti-B IgM	Overall	258	16 (8-16)	Negative	128
Anti-B IgG	Group A	86	4 (1-8)	Negative	64
Anti-B IgG	Group O	172	128 (64-256)	Negative	4096
Anti-B IgG	Overall	258	64 (8-128)	Negative	4096

For IgG analysis, Negative was coded as 0. IQR: interquartile range.

The median Anti-A and Anti-B IgG titres were substantially higher in group O donors than in non-O comparison groups. IgM titres showed comparatively narrower distributions.

Table 4: Blood-group-wise comparison of antibody titres

Antibody	Group 1	Median (IQR)	Group 2	Median (IQR)	p-value
Anti-A IgM	Group B	16 (16-32)	Group O	16 (16-32)	0.286
Anti-A IgG	Group B	1 (0-4)	Group O	128 (64-256)	<0.001
Anti-B IgM	Group A	8 (4-16)	Group O	16 (8-16)	0.178
Anti-B IgG	Group A	4 (1-8)	Group O	128 (64-256)	<0.001

Mann-Whitney U test was used because titre data were ordinal and non-normally distributed.

A statistically significant difference was observed for Anti-A IgG and Anti-B IgG titres between group O donors and corresponding non-O groups. IgM comparisons did not show statistically significant differences.

Table5: Correlation analysis among antibody titres in group O donors

Correlation pair	n	Spearman correlation coefficient	p-value
Anti-A IgM vs Anti-B IgM	172	0.357	<0.001
Anti-A IgG vs Anti-B IgG	172	0.222	0.003
Anti-A IgM vs Anti-A IgG	172	-0.015	0.844
Anti-B IgM vs Anti-B IgG	172	0.215	0.005

Spearman correlation was used because antibody titres are ordinal and showed a non-normal, dilution-based distribution. Negative results were coded as 0 for statistical analysis, and titres were plotted as log2(titre + 1) to accommodate the two-fold dilution pattern.

Among group O donors, Anti-A IgM and Anti-B IgM titres showed a statistically significant positive correlation (r=0.357, p<0.001), indicating that donors with higher Anti-A IgM titres tended to have higher Anti-B IgM titres. Anti-A IgG and Anti-B IgG titres also showed a weak but statistically significant positive correlation (r=0.222, p=0.003). No significant correlation was observed between Anti-A IgM and Anti-A IgG titres (r=-0.015, p=0.844), suggesting that Anti-A IgM and IgG titres varied independently. Anti-B IgM and Anti-B IgG titres showed a weak but statistically significant positive correlation (r=0.215, p=0.005). Overall, the correlations were weak to modest, indicating that each antibody titre should be interpreted separately.

DISCUSSION

The study differs from many earlier reports because both IgM and IgG were evaluated using the NEO Iris automated microplate platform. IgM titres were assessed by direct haemagglutination, while IgG titres were assessed by solid-phase red cell adherence using Capture-R indicator cells. Titre end-points are method-dependent and vary according to reagent cells, incubation conditions, reading platform and endpoint criteria; therefore, direct numerical comparison with tube or gel methods must be cautious.

The mean age of donors was 34.88 +/- 11.17 years, and the largest age group was 18-30 years (40.3%). Males constituted 91.0% of donors. This male predominance is consistent with most Indian blood-centre reports, where female donation is often limited by anaemia, low body weight, pregnancy-related deferral and sociocultural factors. Ravikanth et al.10 reported 98.5% males predominantly male donor population.

Group O was the commonest ABO group in the present study (43.0%), followed by group B (35.5%) and group A (21.5%). This order is comparable to to donor studies by Kannan et al.11 in which group O was common. The predominance of group O has practical importance because these donors may carry both anti-A and anti-B antibodies.

Using 1:64 as the descriptive cut-off for elevated IgM, elevated anti-B IgM was observed in 11.6% of group A donors, elevated anti-A IgM in 19.7% of group B donors, elevated anti-A IgM in 20.3% of group O donors and elevated anti-B IgM in 3.5% of group O donors. Therefore, elevated anti-A IgM was more frequent than elevated anti-B IgM among group O donors. A similar direction was reported by Ravikanth et al.10, Kannan et al11., Kumar et al.12 and França et al.13, although absolute rates differed. The median anti-A IgM titre was 1:16 in groups B and O, while the median anti-B IgM titre was 1:8 in group A and 1:16 in group O. Ravikanth et al.10 reported higher median titres in several corresponding groups, and Datta et al.14 also reported comparable or higher median titres depending on method. These differences reinforce the need for local method-specific reference data.

A major finding was the marked difference in IgG titres between group O and non-O donors. The median anti-A IgG titre was 1:1 among group B donors compared with 1:128 among group O donors. Similarly, the median anti-B IgG titre was 1:4 among group A donors compared with 1:128 among group O donors. At the descriptive threshold of 1:128, 61.6% of group O donors had elevated anti-A IgG and 55.8% had elevated anti-B IgG.4 This pattern is biologically plausible because group O donors may possess anti-A,B antibodies with a substantial IgG component. The finding adds value beyond IgM-only studies, including the JCDR article, and shows that screening only IgM may miss a subset of group O donors with clinically relevant IgG titres.

Correlation analysis among group O donors demonstrated a statistically significant positive relationship between anti-A IgM and anti-B IgM titres (Spearman r=0.357, p<0.001). A weak but significant positive correlation was also observed between anti-A IgG and anti-B IgG titres (r=0.222, p=0.003), while anti-B IgM and anti-B IgG titres showed a weak significant correlation (r=0.215, p=0.005). In contrast, anti-A IgM and anti-A IgG titres were not significantly correlated (r=-0.015, p=0.844). These findings suggest that some common immunological influences may affect paired anti-A and anti-B responses, but the generally weak correlations indicate that one antibody titre or class cannot be used as a reliable surrogate for another. Therefore, separate estimation of anti-A IgM, anti-B IgM, anti-A IgG and anti-B IgG remains important for identifying high-titre group O donors.

CONCLUSION

The study demonstrates that group O donors in Western Rajasthan have a substantial burden of elevated ABO agglutinin titres, particularly IgG anti-A and IgG anti-B. IgM titres were generally low to moderate, but IgG titres among group O donors were

markedly higher than those in corresponding non-O groups.The correlation findings further support separate assessment of IgM and IgG titres, as individual antibody titres cannot reliably predict the corresponding paired titre or antibody class.

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