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Screening and Titration of Anti-A and Anti-B Haemolysins in Blood Donors: An Essential Test for Transfusion Immunosafety of Packed Red Blood Cells

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Abstract

The study aimed to examine the presence of anti-A and anti-B haemolysins in the serum of blood donors. **Methods**: A retrospective cross-sectional survey was conducted at the National Blood Transfusion Centre (CNTS) in Abidjan-Treichville, involving 1350 voluntary blood donors aged 18 to 65, belonging to blood groups A, B, and O. The immuno-haematology unit of the CNTS laboratory performed all biological analyses. Data collection forms from the CNTS Côte d'Ivoire computer system were used to collect socio-demographic data and haemolysin test results. Haemolysins were detected using a manual technique of direct haemagglutination of serum with A1 and B test red blood cells treated with papain, with titration performed through dilution series. Statistical analysis employed the Chi-square and Fisher tests at a 5% significance level. **Results**: Male donors constituted 86.3%, with a mean age of 31.8 ± 5.2 years, primarily in the 25-34 age group. The prevalence of haemolysins was 20.2%, with higher frequencies observed in males (73.3%, p=0.005), blood group O ((82.7%, p=0.000), and the 25-44 age group (49.8%, p<0.0001). Anti-B IgG was the most common haemolysin type (49.1%, p=0.0000). Anti-A IgG levels were notably higher in group O compared to group B (p=0.001) and in males compared to females (p=0.012), while anti-B IgG levels were higher in group O compared to group A (p<0.0001). Titration analysis indicated a prevalence of titration 2, with no high titres (≥ 64) detected. **Conclusion**: The study underscores that blood group O donors exhibited a higher likelihood of haemolysins. Despite generally low haemolysin titres, caution is advised during transfusions involving blood with these irregular antibodies due to their potential to cause severe complications in the recipient.

Keywords: Blood donors; Haemolysins; ABO blood-group system; Blood transfusion; Transfusion safety.

1. Introduction

Ensuring the immunological safety of blood transfusions is a fundamental aspect of preventing anti-erythrocyte alloimmunisation and minimising the risk of adverse immune reactions in patients [1]. The critical role of ABO blood groups is essential in ensuring this compatibility [1]. The ABO system is characterised by the constant presence of natural anti-A and/or anti-B antibodies in the plasma, corresponding to antigens that are not present on the surface of red blood cells. Typically, these natural antibodies belong to the IgM class and show optimal reactivity at a temperature of 4°C [2, 3]. In the context of the ABO system, it is recommended that isogroup transfusions be performed according to the principle of phenotypic identity [4]. If donors of identical blood groups are not available, non-isogroup transfusions may be considered based on the principle of phenocompatibility [4]. For example, group O blood could be given to a patient with group A, B or AB, or group A blood to a patient with group AB, or group B blood to a patient with group AB. In non-isogroup transfusions, the donor's natural antibodies pose a minimal risk to the recipient's red cells due to their low concentration and are rapidly diluted in the recipient's bloodstream [5, 6]. In addition to these natural antibodies, some people can develop anti-A and/or anti-B antibodies in response to various

immunological stimuli. These may be allogeneic, resulting from ABO blood group incompatibility in circumstances such as transfusion, pregnancy, or transplantation [4, 5], or heterogeneous, caused by substances related to blood group antigens as part of serotherapy, vaccination or infection [4, 5]. These immune antibodies, mainly of the IgG class, are called haemolysins because of their ability to induce haemolysis under standard conditions at 37°C [2, 4-7]. Anti-A and anti-B haemolysins can induce potentially fatal immune reactions in recipients who have the corresponding antigens [2, 5]. This concern is particularly relevant for whole blood transfusions. Currently, red cells concentrate contain a very small amount of residual plasma (≤ 25 ml), which reduces the associated risk. However, it is still essential to test for haemolysins because, although their concentrations are generally low, they can still cause haemolysis if transfused to an incompatible recipient, particularly in the case of massive transfusion [6]. This problem also applies to platelet concentrates, which contain about 30% plasma by volume. In addition, haemolysins are of major clinical interest in cases of foetal-maternal ABO incompatibility, as they can cross the placenta and cause haemolytic anaemia in the newborn [8, 9]. Detection of anti-A and anti-B haemolysins in blood donors is therefore a critical step in ensuring the safety of blood transfusion. Identification and appropriate management of these antibodies are essential measures to minimise the risk of incompatibility in non-isogroup transfusions. The aim of our study was to investigate the presence of anti-A and anti-B haemolysins in the serum of blood donors.

2. Methods

2.1. Study Population

Our study is a retrospective cross-sectional survey conducted at the National Blood Transfusion Centre (CNTS) in Abidjan-Treichville. It included a sample of 1350 voluntary blood donors, aged between 18 and 65 years, belonging to blood groups A, B and O. Group AB donors were not included in the study because transfusion of group AB red cell concentrates can only be performed between individuals of the same blood group (isogroup), unlike the other groups for which transfusion of compatible non-isogroups is possible. All biological analyses were carried out in the immuno-haematology unit of the CNTS laboratory.

2.2. Data Collection

Patient socio-demographic data and haemolysin test results were collected using data collection forms from the CNTS Côte d'Ivoire computer operating system.

2.3. Detection of Haemolysins

The method used was a manual technique of direct haemagglutination of serum using test red blood cells A1 and B, previously treated with papain.

2.3.1. Neutralisation of Natural Anti-A and/or Anti-B Antibodies

The serum to be tested is obtained after centrifugation of whole blood at 3000 rpm for 5 minutes. The procedure begins by diluting the serum in a haemolysis tube with saline (physiological water) in a 1:2 ratio. The purpose of this dilution is to reduce the concentration of antibodies, thus facilitating the neutralisation process. The diluted serum is then placed in a water bath at 70°C for 10 minutes. The aim of this step is to neutralise the natural anti-A and anti-B antibodies. After this 10-minute incubation, the tube is examined for an opalescent appearance, which would indicate successful neutralisation of the natural antibodies. To confirm this neutralisation, a Simonin test is performed using RH-negative test red cells A1 and B at a concentration of 10%. RH negative test red cells are used to avoid interference with any anti-D antibodies. If the Simonin reaction is positive, meaning that there are still natural antibodies present that have not been neutralised, the tube is reincubated in a water bath at 70°C for a further 10 minutes.

2.3.2. Preparing Papainised A1 and B Test Red Cells

RH-negative A1 red cells are washed three times with physiological water at a centrifugation speed of 3,000 rpm for 3 minutes to remove any residual plasma or antibody. The red cells were then incubated at 37°C for 7 minutes in the presence of papain in a 1:1 ratio. It should be noted that IgG, which has a low agglutinating capacity compared to normal red blood cells, becomes much more agglutinating when red blood cells are treated with proteolytic enzymes such as papain [2]. After this incubation, the red cells are washed three times with physiological water at 3,000 rpm for 2 minutes to remove any residual papain. The papain-treated erythrocytes are used to prepare a 5% erythrocyte suspension. The same process is repeated for B test red cells.

2.3.3. Detection of Anti-A and anti-B Antibodies (Anti-A and anti-B Haemolysins)

A drop of neutralised serum is placed in two separate haemolysis tubes. One drop of papainised A1 test erythrocyte suspension is added to the first tube and one drop of papainised B test erythrocyte suspension is added to the second tube. The two tubes are then incubated in a water bath at 37°C for 45 minutes. After this incubation period, the tubes were centrifuged at 1,000 rpm for 1 minute or 3,000 rpm for 20 seconds.

2.3.4. Reading and Interpretation of Results

The presence or absence of agglutination is observed with the naked eye by gently shaking the tubes. Positive agglutination indicates the presence of anti-A and/or anti-B haemolysins in the serum analysed. Absence of agglutination in both tubes indicates the absence of haemolysins in the serum.

2.3.5. Haemolysin Titration

Titration is carried out by making successive dilutions of the neutralised serum in a ratio of 2 (1/2, 1/4, 1/8 ... 1/128) to determine the maximum dilution at which agglutination can still be observed. The titre is then the reciprocal of this maximum dilution.

2.4. Data Analysis

Data were recorded, processed, and analysed using Microsoft Word and Excel 2007. Statistical analysis was performed using the Chi-square test and the Fisher test, with a significance level of α=5%.

2.5. Ethical Considerations

Throughout the study, we strictly adhered to ethical and confidentiality guidelines. This was reflected in the protection of the identity and privacy of participating donors.

3. Results

Of the 1350 blood donors included in the study, 1165 (86.3%) were male and 185 (13.7%) were female, giving a male/female sex ratio of 6.3. The mean age of donors was 31.8 ± 5.2 years, with extremes of 18 and 62 years. Donors aged between 25 and 34 years represented most of the group (44.7%), followed by those aged between 35 and 44 years (29%). The prevalence of haemolysins in all sera analysed was 20.2%. When the haemolysin cases were analysed according to sex, age, and blood group, we found a higher frequency in men (73.3%) than in women (26.7%) ($p=0.005$), as well as in individuals with blood group O (82.7%) ($p=0.000$) and in the 25-44 age group (49.8%) ($p<0.0001$). Anti-B IgG was the most common haemolysin type, accounting for 49.1% of cases ($p=0.0000$). Regarding the antigenic target, anti-A IgG levels were significantly higher in individuals from group O compared to group B ($p=0.001$) and in males compared to females ($p=0.012$). In addition, anti-B IgG levels were significantly higher in individuals from group O compared to group A ($p<0.0001$). It should be noted that although the proportions of these antibodies were higher in males, there was no significant difference in the prevalence of anti-B IgG and anti-A and anti-B IgG according to sex. Regarding haemolysin titres, titre 2 was the most common, followed by titres 4 and 8, irrespective of the variable considered. However, titer 4 was predominant in female donors and in individuals aged between 25 and 44 years. We also observed an anti-B titre of 16 in a female group O donor aged 25-34 years. It should be noted that no high titres (≥ 64) were recorded.

4. Discussion

The predominance of males, blood group O and young adults among blood donors is a common finding in the literature, as shown by several studies [6, 10-20]. The low proportion of female donors can be explained by several factors, including the selection criteria for blood donors, which exclude pregnant, lactating, and menstruating women. In addition, the relative under-representation of women in high schools and universities, which are the main sites for mobile blood drives, may also contribute to this disparity.

In our study, the overall prevalence of haemolysins among blood donors was 20.2%. Although this prevalence is relatively high, it remains lower than that found by several researchers in Nigeria, notably Kagu, *et al.* [11], Olawumi and Olatunji [13], Oyedeji, *et al.* [14], and Ugah, *et al.* [19], and in Burkina Faso [17], where prevalences ranging from 23.2% to 55.4% were observed. In contrast, much lower prevalence rates were found in Togo [15], Tunisia [6], and Nigeria **[20]** (0.5%, 6.6% and 10%, respectively). These differences between studies could be explained by differences in sample size, study duration or the susceptibility of populations to develop haemolysins due to their antigenic exposure [21]. With regard to the specificity of the haemolysins, anti-B IgG was the most common (49%), followed by anti-A IgG (30.7%) and the coexistence of anti-A and anti-B IgG (20.3%). This trend was also observed in the Fopa study in Cameroon [21] and in the Olawuni study in Nigeria [13]. However, in Burkina Faso [17], Togo [15] and certain regions of Nigeria [11], a predominance of the combination of anti-A and anti-B haemolysins predominated (14%). In Tunisia [6], Pakistan [18] and most studies in Nigeria [14, 19, 20], anti-A haemolysins predominated. Our study also showed that group O blood donors had the highest predisposition to develop haemolysins, with a prevalence of 82.7%. Group A donors followed with a rate of 11.4%, while group B donors accounted for 5.9% of cases. This observation explains why most studies on haemolysins have focused mainly on group O donors [10-18, 22]. The predominance of group O is explained by their high frequency in the world population and by the absence of A and B antigens on the surface of their red blood cells. As a result, their immune system is more exposed to these antigens, which are perceived as foreign. It should be noted that haemolysins are generally antibodies directed against antigens that are not present on the surface of the person's red blood cells. Group O is the only ABO group that can develop both anti-A and/or anti-B haemolysins. On the other hand, group A individuals develop mainly anti-B haemolysins, while group B individuals develop mainly anti-A haemolysins. Group AB individuals very rarely develop anti-A and anti-B haemolysins because they have both A and B antigens on their red blood cells, which reduces the likelihood of producing haemolysins directed against these specific antigens. Group O donors are generally considered to be universal donors, meaning that they can give blood to people of all blood groups. This is particularly useful in emergency situations where blood grouping could delay medical treatment or where there is a shortage of isogroup blood [1-5]. It is important to note, however, that for group Of individuals with ABO haemolysins, their blood must be reserved for isogroup transfusions because of the risk of incompatibility [1], hence their occasional nickname of "dangerous universal donors" in such circumstances

[22]. We also found differences in the prevalence of different haemolysin specificities according to blood group. In group O, 37.7% of donors had anti-B haemolysins, 24.9% had anti-A haemolysins and 20.1% had both anti-A and anti-B IgG. These prevalences were significantly higher than those observed for other blood groups. These results are consistent with those of Louati **[6]**, who also found significantly higher levels of haemolysins in group O donors than in non-O donors. In addition, donor age and sex influenced haemolysin specificity, with all specificities being predominant in males. These findings contrast with the Sawadogo study in Burkina Faso, where women had significantly more haemolysins than men (34% versus 48.3%).

In terms of haemolysin titres, titre 2 was the most common, followed by titres 4 and 8, regardless of the parameter considered. However, it should be noted that a titer of 16, of the anti-B type, was observed in a group O donor, female, aged between 25 and 34 years. Crucially, our study did not detect any high titres (≥ 64), which contrasts with the results of some studies by Uko in Nigeria [20], who obtained a 10% rate of haemolysins with a titre higher than 64, by Saidin in Malaysia, where titres ranged from 16 to 256 [16], and by Sharif in Pakistan [18], where 26.7% of the donors studied had a haemolysin titre of at least 128. Kagu, *et al.* [11], found results very similar to ours, with a low prevalence of lytic titres of 16 and 32 (1.1%). In general, a lytic antibody titer above 16 is often considered a threshold above which transfusion of incompatible blood becomes dangerous [8], while a titer of 32 or more is generally considered even more risky. Although the prevalence of anti-B haemolysins was higher than that of anti-A haemolysins, the titres of anti-A haemolysins were higher than those of anti-B haemolysins. This observation is consistent with the results of other studies [13, 17, 18, 21]. In fact, people of black African origin are generally more sensitive to B antigens (or B-like substances) than to A antigens (or A-like substances) [21]. However, it is also recognised that the haemolysing power of anti-A haemolysins is more pronounced than that of anti-B haemolysins. From a biochemical point of view, antigen A, whose immunodominant sugar is N-acetylgalactosamine, is considered more immunogenic than antigen B, whose immunodominant sugar is D-galactose, due to the presence of amino groups [21]. In general, proteins are more immunogenic than carbohydrates.

5. Conclusion

To ensure optimal blood transfusion and immunological safety, the use of isogroup blood is essential. In addition to standard immunohaematological tests, it is essential to screen blood donors for haemolysins. Like many similar studies, our aim was to determine the prevalence of haemolysins and our results showed a prevalence of 20.2% among blood donors in Côte d'Ivoire. Blood group O donors were most likely to have haemolysins. Although haemolysin titres were generally low, it is important to be particularly vigilant when transfusing blood containing these irregular antibodies, as they can lead to serious complications in the recipient.

Competing Interests

The authors declare no competing interest.

Authors' Contributions

All authors contributed to this work and read and approved the final version of the manuscript.

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Tables and Figures

Table I: Distribution of the 1350 donors by age group, sex, and ABO blood group
Table II: Distribution of anti-A and anti-B haemolysins by titre
Figure 1: Prevalence of haemolysins in all 1350 blood donors
Figure 2: Distribution of haemolysins by antigenic target
Figure 3: Distribution of haemolysins by ABO blood group donors
Figure 4: Distribution of haemolysins by sex of blood donors
Figure 5: Distribution of haemolysins by age range of blood donors

References

[1] Muller, J. Y., Chiaroni, J., and Garraud, O., 2015. "Sécurité immunologique des transfusions." *La Presse Médicale*, vol. 44, pp. 200-13. Available: https://doi.org/10.1016/j.lpm.2014.06.035

[2] Rouger, P. and Salmon, C., 1977. "Les allohémagglutinines du système ABO. Revue Française de Transfusion et Immuno-hématologie." vol. 20, pp. 627-55. Available: https://doi.org/10.1016/S0338-4535(77)80068-8.

[3] Système ABO (ISBT 001). Available: https://www.toutsurlatransfusion.com/immuno-hematologie/systemes/antigenes_ABO.php.Consultéle07-09-2023

[4] Lefrère, J. J. and Rouger, P., 2015. *Transfusion sanguine*. 5 ed. Elsevier Masson.

[5] Giraud, C., Korach, J. M., Andreu, G., Lacaze, C., Vaicle, M., and Schooneman, F., 2002. "Bases immunologiques de la transfusion." *Transfus Clin. Biol.*, vol. 9, pp. 163-7. Available: https://doi.org/10.1016/s1246-7820(02)00242-2

[6] Louati, N., Cherif, J., Ben, A. I., Rekik, H., and Gargouri, J., 2008. "Recherche des hémolysines chez les donneurs de sang. Tinisia." *J. Inform. Méd. Sfax.*, vol. 15, pp. 17-9.

[7] Flegel, W. A., 2015. "Pathogenesis and mechanisms of antibody-mediated hemolysis." *Transfusion* vol. 55, pp. S47–S58. Available: https://doi.org/10.1111/trf.13147

[8] Groupage-génotypage des femmes enceintes. Available: https://www.toutsurlatransfusion.com/immuno-hematologie/analyses-complementaires/ih-grossesse.php.Consultéle07-09-2023

[9] Sock, D. S., Kamdem, S. D., Boula, A., and Netongo, P. M., 2020. "Fréquence et titrage des hémolysines anti-A et anti-B chez les mères d'enfants ictériques à Yaoundé, Cameroun." *Pan. Afr. Med. J.*, vol. 35, p. 13. Available: https://doi.org/10.11604/pamj.2020.35.13.14770

[10] Fondoh, V. N., Ndzenjempuh, N., Stella, T., Fondoh, R. M., Awasom, C. N., and Enow-Tanjong, R., 2022. "Prevalence of alpha and beta haemolysin among blood group O donors in Bamenda, Cameroon." *Afr. J. Lab. Med.*, vol. 11, pp. 1-6. Available: http://dx.doi.org/10.4102/ajlm.v11i1.1432

[11] Kagu, M. B., Ahmed, S. G., Mohammed, A. A., Moshood, W. K., Malah, M. B., and Kehinde, J. M., 2011. "Anti-a and anti-b haemolysins amongst group "o" voluntary blood donors in northeastern Nigeria." *Journal of Blood Transfusion*, Available: https://doi.org/10.4061/2011/302406

[12] Khampanon, K., Chanprakop, T., Sriwanitchrak, P., Setthakarn, M., Oota, S., and Nathalang, O., 2012. "The characteristics of ABO antibodies in group O Thai blood donors." *J. Clin. Lab. Anal.*, vol. 26, pp. 223-6. Available: https://doi.org/10.1002/jcla.21499

[13] Olawumi, H. O. and Olatunji, P. O., 2001. "Prevalence and titre of alpha and beta haemolysins in blood group 'O' donors in Ilorin." *African Journal of Medicine and Medical Sciences*, vol. 30, pp. 319-21. Available: http://www.ncbi.nlm.nih.gov/pubmed/14510111

[14] Oyedeji, O. A., Adeyemo, T. A., Ogbenna, A. A., and Akanmu, A. S., 2015. "Prevalence of anti A and anti B hemolysis among blood group O donors in Lagos." *Niger J. Clin. Pract.*, vol. 18, pp. 328-32. Available: https://doi.org/10.4103/1119-3077.151760

[15] Padaro, E., Kueviakoe, I. M., Feteke, L., Agbetiafa, K., Magnang, H., and Alfa, T., 2015. "Prevalence des hemolysines chez les donneurs de sang de groupe O au centre national de transfusion sanguine (CNTS) de Lome." *Journal de la Recherche Scientifique de l'Université de Lomé,* vol. 17, pp. 371-77.

[16] Saidin, N. S., Noor, N. H. M., Yusoff, S. M., and Sauli, M. S., 2023. "Characteristics of ABO antibodies in Group O Malaysian blood donors." *Malays. J. Med. Sci.*, vol. 30, pp. 61-70. Available: https://doi.org/10.21315/mjms2023.30.4.6

[17] Sawadogo, S., Bationo, B. G., Nebie, K., Konseibo, A., Koanda, J. E., and Deneys, V., 2020. "Titre des hemolysines alpha et beta chez les donneurs de sang de groupe sanguin O et leur impact potentiel sur la sécurité des receveurs de produits sanguins au Burkina Faso." *Journal de la Recherche Scientifique de l'Université de Lomé,* vol. 22, pp. 281-91.

[18] Sharif, A., Raja, M. I., Hanif, W., Yazdani, M. S., Iqbal, H., and Anwar, N., 2022. "High titre anti-A and anti-B antibodies in group "O" blood donors - a tertiary care hospital blood bank experience." *Pakistan Journal of Pathology*, vol. 33, pp. 25-8. Available: https://doi.org/10.55629/pakjpathol.v33i1.700

[19] Ugah, U., Ibekailo, S., and Mbamagu, D., 2014. "Rate of haemolysins among blood donors in Abakaliki, Ebonyi State, Nigeria." *GJMR Stud*, vol. 1, pp. 61-5.

[20] Uko, E. K., Erhabor, O., Ahmed, H. M., Isaac, I. Z., Abdulrahaman, Y., and Wase, A., 2013. "Prevalence of high titre alpha and beta haemolysins among blood donors in Sokoto, Northwestern Nigeria." *Int. J. Med. Sci. Health Care*, vol. 1, pp. 1-8.

[21] Fopa, D., Tagny, C. T., Tebeu, P. M., Ndoumba, A., and Mbanya, D., 2013. "Recherche et titrage des hémolysines anti-A et anti-B chez les femmes en période du postpartum immédiat au Centre Hospitalier et Universitaire de Yaoundé." *Africa Sanguine*, vol. 16, pp. 4-8.

[22] Amita, R. and Vijayalakshmi, K., 2019. "Prevalence, and haemolytic significance of red cell antibodies among dangerous universal donors in a Tertiary Care Hospital in South India." *International Journal of Medical Laboratory*, vol. 6, pp. 234-40. Available: https://doi.org/10.18502/ijml.v6i4.1998

Table-1. Distribution of the 1350 donors by age range, sex, and ABO blood group

		NUMBER (n)	FREQUENCY (%)
Age range in years	18-24	219	16,2
	25-34	604	44,7
	35-44	392	29
	45-54	105	7,8
	55-62	30	2,2
Sex	Male	1165	86,3
	Female	185	13,7
ABO blood groups	Group O	757	56,1
	Group A	305	22,6
	Group B	288	21,3

Table-2. Distribution of anti-A and anti-B haemolysins by titre

HEMOLSIYNS TITRE						
	2	4	8	16	64	N (%)
IgG anti-A	87 (62,6%)	38 (27,3%)	13 (9,4%)	1 (0,7%)	0	139*
IgG anti-B	139 (73,5%)	37 (19,6%)	13 (6,9%)	0	0	189*
Group O	126 (55,8%)	82 (36,3%)	17 (7,5%)	1 (0,4%)	0	226 (100%)
Group A	25 (80,6%)	3 (9,7%)	3 (9,7%)	0	0	31 (100%)
Group B	13 (81,2%)	2 (12,5%)	1 (6,3%)	0	0	16 (100%)
Male	122 (61%)	59 (29,5%)	19 (9,5%)	0	0	200 (100%)
Female	34 (46,6%)	32 (43,8%)	6 (8,2%)	1 (1,4%)	0	73 (100%)
18-24 years	36 (70,6%)	15 (29,4%)	0	0	0	51 (100%)
25-34 years	42 (30,9%)	77 (56,6%)	16 (11,8%)	1 (0,7%)	0	136 (100%)
35-44 years	28 (54,9%)	15 (29,4%)	8 (15,7%)	0	0	51 (100%)
45-54 years	14 (70%)	6 (30%)	0	0	0	20 (100%)
55-62 years	14 (93,3%)	1 (6,7%)	0	0	0	15 (100%)
*Both anti-A and anti-B haemolysins were present in 55 patients						

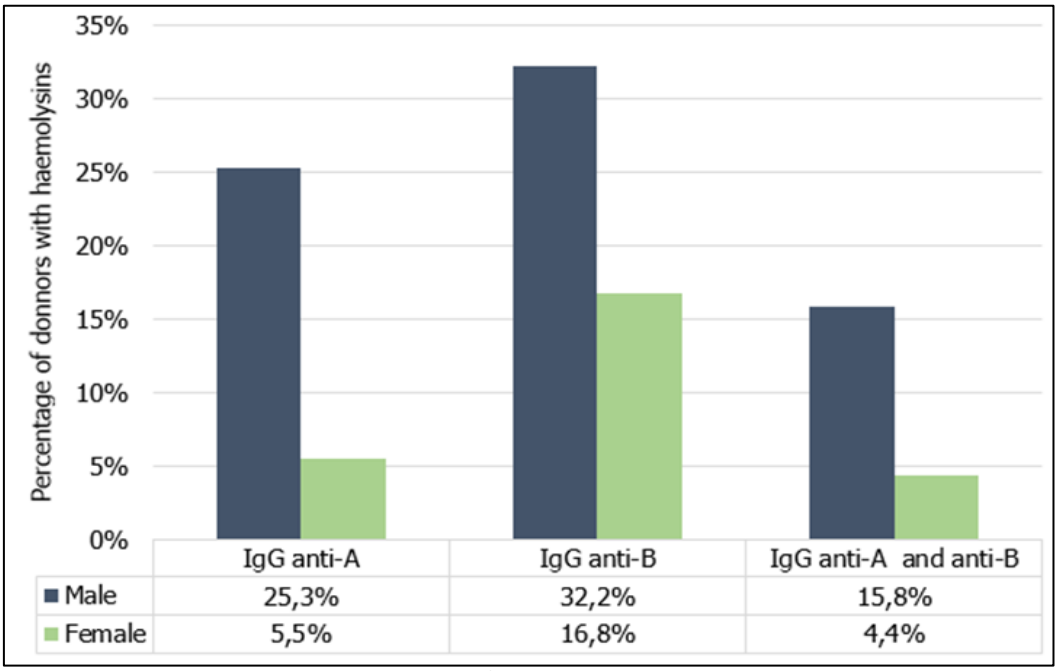

Figure-4. Distribution of haemolysins by sex blood donors

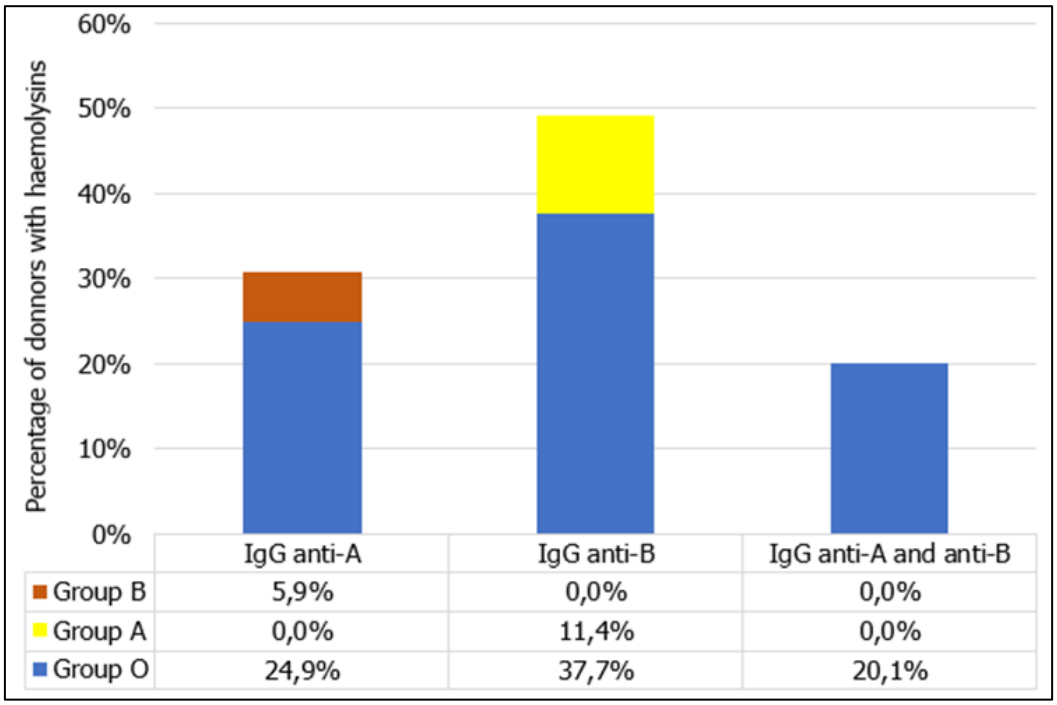

Figure-3. Distribution of haemolysins by ABO blood group donors

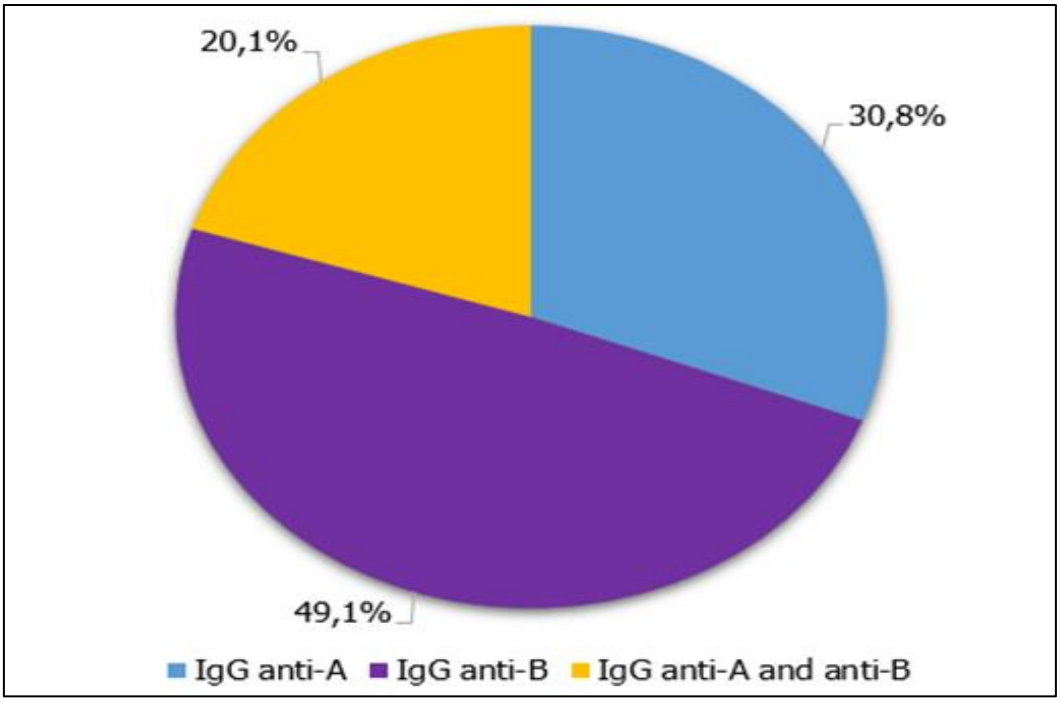

Figure-2. Distribution of haemolysins by antigenic target

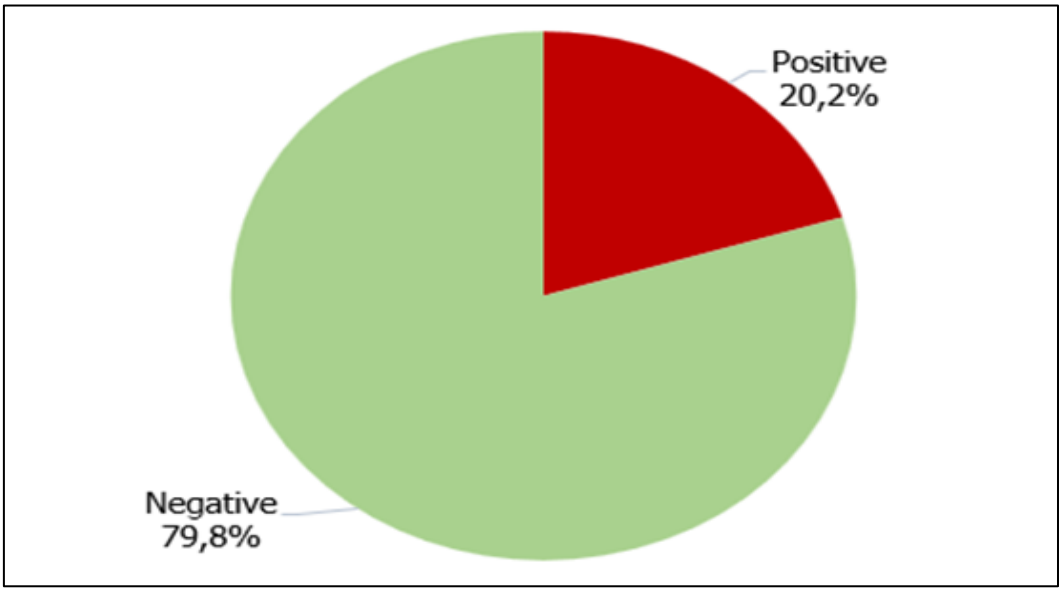

Figure-1. Prevalence of haemolysins in all 1350 blood donors

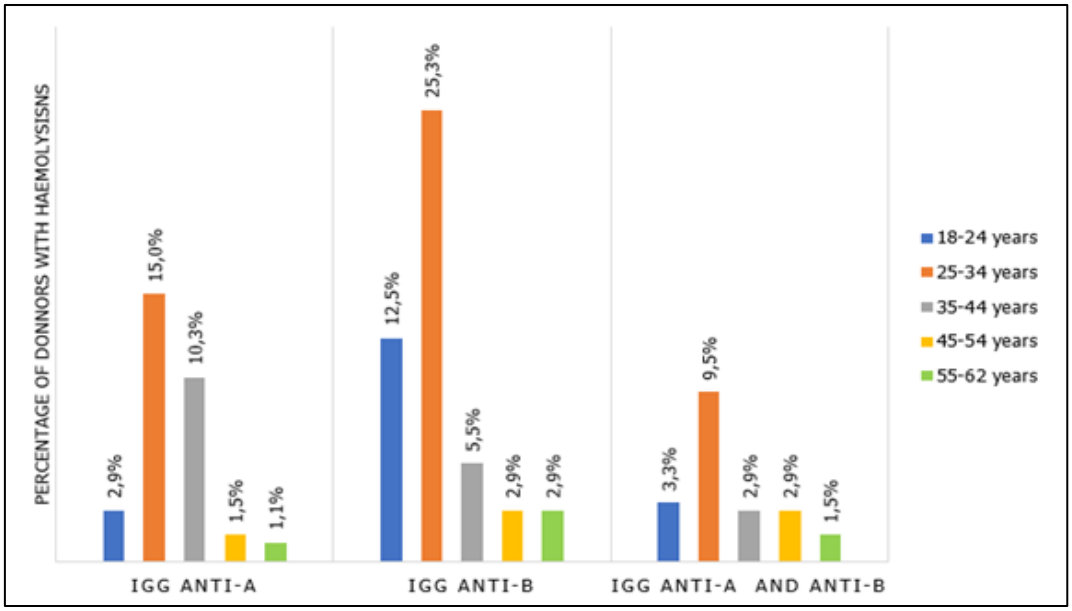

Figure-5. Distribution of haemolysins by age range of blood donors