Original Research Article

Blood Group Distribution and Its Relationship with Bleeding Time and Clotting Time in Medical Undergraduate Students

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ABSTRACT

Background: In 1901 an Austrian scientist Karl Landsteiner, discovered the ABO blood group in human. Blood group plays a vital role in the field of transfusion medicine. The previous studies found prolonged bleeding and clotting time in individuals with blood group O probably due to lower expression of von Willebrand factor, and elevated levels are risk for thrombosis.

Aims and Objectives: The objective of the study was to assess the relationship between bleeding time and clotting time among various blood groups and to identify gender difference between them.

Materials and methods: Our study included 100 medical undergraduates, aged between 17–22 years. The blood grouping was determined with the standard antisera and bleeding time and clotting time was estimated by duke method and capillary tube method respectively.

Results: Blood group O (33%) was more predominant among students followed by blood group A (32%), B (26%) and AB (9%). Bleeding time was found to be prolonged > 4 min in blood group O (21.21%), followed by blood group B (19.23%), A (18.75%) and AB (11.11%) but the difference was statistically insignificant (p = 0.867). Similarly clotting time was prolong > 6 min among blood group A (15.63%) followed by O (6.07%), B (3.84%) and AB (0%) but the difference was statistically insignificant (p = 0.192).Gender-wise bleeding time and clotting time were more prolonged in females than males.

Conclusions: In our study, blood group O was most predominant. Bleeding time was prolonged in blood group O whereas clotting time was prolonged in blood group A. Gender-wise bleeding and clotting time were higher in females than males.

Key words: Bleeding time, blood group, Clotting time, von Willebrand factor.

INTRODUCTION

In 1901 an Austrian scientist Karl Landsteiner, discovered the ABO blood group in human. The ABO blood group is determined by the presence or absence of A and B antigen on the surface of the red blood cells. Type A individuals have the antigen A, type B have antigen B, and type AB have both; type O have neither of these antigens. The antigens of the ABO system (A, B, and H) are polysaccharides which is located at the peripheral end of the carbohydrate chain of glycoproteins or glycosphingolipids. ^[1] The antigens are products of glycosyltransferases. The genes coding for these enzymes are located on chromosome 9 (A or B) and 19 (H). The A and B alleles encode slightly different glycosyltransferases that add Nacetylgalactosamine and D-galactose,

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respectively to a common precursor the H antigen and thus converting it into A or B antigens. The O alleles do not encode a functional enzyme and consequently OO carriers, who lack such transferase enzymes, express only the basic, unmodified, H antigen. ^[2] In addition to RBCs, these antigens are also expressed on the membranes of various cells including platelets, vascular endothelium, epithelial cells and von Willebrand factor (vWF). ^[3]

Von Willebrand factor (vWF) is a large adhesive glycoproteins, present in blood plasma synthesized and by endothelial cells and megakaryocytes (agranules of platelets). vWF participates in hemostasis ^[4] by binding with collagen, and promotes platelet adhesion as well as aggregation at sites of vessel injury. VWF also plays a major role in blood coagulation as they are the specific carrier of factor VIII in plasma and protects it from proteolytic degradation, thus prolongs its half-life in ^[5] Hereditary or acquired circulation. deficiency of vWF lead to bleeding diathesis of the skin and mucous membranes, causing menorrhagia, nosebleeds. and gastrointestinal bleeding while higher levels are risk for thrombosis.^[6]

The ABO blood group system influences the bleeding time (BT) and clotting time (CT). BT is the time interval between the skin puncture and spontaneous unassisted stoppage of bleeding. CT is the time interval between the puncture of blood vessels and formation of fibrin threads.^[7] Thus, relationships between BT, CT, and blood groups are important in certain clinical conditions such as epistaxis, surgery, thrombosis etc.

Several studies have documented the influence of ABO blood groups on plasma VWF levels. ^[7-9] Individuals with blood group O normally have significantly lower plasma levels of vWF and Factor VIII than non-O individuals. ^[10] Gill et al ^[11] conducted a study in 1117 healthy individuals and reported that blood group O subjects have lowest plasma von Willebrand factor levels and highest in group AB subjects. Reddy et al ^[12] demonstrated that epistaxis is commonly seen in people with O blood group when compared with other ABO blood groups, and they also observed that there is a lower expression of von Willebrand factor (vWF) in them. Thus the present study is undertaken to assess the frequency of blood groups among our students as well as their relationship with bleeding time and clotting time.

Aims and Objectives

- 1) To determine the blood group, bleeding time and clotting time of the subjects.
- 2) To assess the relationship between bleeding time and clotting time among various blood groups
- 3) To identify gender difference in bleeding time and clotting time.

MATERIALS AND METHODS

This cross-sectional study was conducted in the Department of Physiology, Kathmandu University School of Medical Sciences, Dhulikhel, Kavre. After obtaining consent, 100 undergraduate students in the age group of 17-22 years were selected. The study was approved by Institutional review committee. The exclusion criteria for selection of the students were history of bleeding disorders (hemophilia), and drug (nonsteroidal anti-inflammatory intake drugs).

Procedures

Blood group was determined during practical time in physiology laboratory by standard antisera. Blood samples were collected by finger prick with sterile lancet after cleaning the puncture site with spirit. The sample blood is mixed with anti-A, anti-B, and anti-D serum. Blood groups were determined on the basis of presence or absence of agglutination. Agglutination was confirmed by observing under low-power objective of a compound microscope. ^[7]

Bleeding time was determined by Duke's filter paper method. A deep skin puncture was made and the length of time required for bleeding to stop was recorded

by blotting the drop of blood coming out of the incision every 30 seconds using blotting paper. BT was calculated by multiplying the number of drops on the filter paper and 30 seconds. The normal BT by Duke's filter paper method is usually in the range of 1–5 min.^[7]

Clotting time was determined by capillary tube method. A standard incision was made in the skin and the blood was taken into a capillary glass tube. The length of time taken for the blood to clot was calculated by breaking the capillary tube after 2 min, 1-2 cm from one end then every 30 s till appearance of fibrin thread. The normal CT estimated by this method falls in the range of 5–11 min.^[7]

Statistical Analysis

The available data were entered in MS Excel then to software SPSS. Statistical analysis was carried out using SPSS, version 21 (SPSS, Inc., Chicago, IL). The Chi- square & one way analysis of variance (ANOVA) tests were applied to examine association between blood groups and bleeding time, clotting time. p-value of <0.05 was considered to be statistically significant.

RESULTS

Table 1. Showing gender percentage among students				
Gender	Frequency	Percent		
Male	50	50.0		
Female	50	50.0		
Total	100	100.0		

Table 2. showing distribution and frequency of blood group among students					
Blood group	Frequency	Percent			
A+ve	32	32.0			
B+ve	26	26.0			
AB+ve	9	9.0			
O+ve	33	33.0			
Total	100	100.0			

Table 3. Distribution of clotting time on various blood groups with one way ANOVA analysis

Blood group	Clot	ting tim	Significance (ANOVA)		
	$< 6 \min$ > 6 min			F = 1.729	
	No	%	No. %		p = 0.192
A+ve	27	29.3	5	62.5	
B+ve	25	27.2	1	12.5	
AB+ve	9	9.8	0	0	
O+ve	31	33.7	2	25	
Total	92	100	8	100	

Table 4. Distribution of bleeding time on various blood groups with one way ANOVA analysis

	Bleeding time					
Blood group					(ANOVA)	
	$< 4 \min$ > 4 min				F = 0.028	
	No	%	No. %		p = 0.867	
A+ve	26	32.1	6	31.6		
B+ve	21	25.9	5	26.3		
AB+ve	8	9.9	1	5.3		
O+ve	26	32.1	7	36.8		
Total	81	100	19	100		

Fable	5.	Gender-wise	distribution	of	clotting	time	with	one
way A	NO	VA analysis			-			

	Clott	ing tim	e	Significance	
Gender				(ANOVA)	
	<6 n	nin	>6 m	in	F = 0.536
	No	%	No.	%	p = 0.466
Male	47	51.1	3	37.5	
Female	45	48.9	5	62.5	
Total	92	100	8	100	

 Table 6. Gender-wise distribution of bleeding time with one way ANOVA analysis

Blee	ding tin	ne	Significance (ANOVA)	
<4 n	nin	>4 m	in	F = 1.618
No	%	No.	%	p = 0.206
43	53.1	7	36.8	
38	46.9	12	63.2	
81	100	19	100	
	Blee <4 m 43 38 81	Bleeding tin <4 min	Bleeding time <4 min	Bleeding time <4 min >4 min No % No. % 43 53.1 7 36.8 38 46.9 12 63.2 81 100 19 100

The available data of 100 students were analyzed. The age group was homogeneous in our study population (17-22 years) as all of them belonged to the first year and second year undergraduates. Of 100 students, 50 were males and 50 were females (Table 1). Our results showed that blood group O was more predominant, followed by blood group A, B and AB. The percentage distribution of ABO blood groups was in order of O (33%) > A(32%) > B (26%) > AB (9%).

Table 2 shows that the blood group O (33%) was more predominant among students followed by blood group A (32%), B (26%) and AB (9%). The distribution of clotting time and bleeding time according to blood groups is shown in tables 3 and 4 respectively. Table 3 shows that CT prolonged more than 6 min among blood groupA (15.63%) followed by O (6.07%), B (3.84%) and AB (0%). One way ANOVA tests performed on the data did not show any statically significant difference between the CT of ABO groups (p = 0.192).

Table 4 shows BT more than 4 min in a greater number among blood group O

(21.21%), followed by blood group B (19.23%), A (18.75%) and AB (11.11%). One way ANOVA tests performed on the data did not show any statically significant difference between the BT of ABO groups (p = 0.867).

While considering the role of gender, CT and BT were more prolonged in female than male as shown in Table 5 and 6 respectively. Table 5 shows that the CT is more than 6 min in female (10%) as compared to 6% in male students. The difference was not statistically significant (p=0.466%). Table 6 shows that 24% female students had more than 4 min of BT compared to 14 % male students, the difference was again not statistically significant (p=0.206).

DISCUSSION

In this study conducted on 100 P students, the percentage distribution of blood groups showed predominance of blood group O (33%) followed by blood group A (32%), B (26%) and AB (9%). Similar results were obtained from a study conducted on 322 medical students by Pramanik T, Saikia TC and Bandopadhyya M, ^[13] which reported blood group O as the most prevalent one (32%), followed by A (29%), B (26%) and AB (13%); another study on 1310 hospital patients by Pramanik T and Adhikari P, ^[14] observed that frequencies of distribution of blood groups O, A, B and AB were found to be 35.5%, 28.5%, 27.3% and 8.7%. Study conducted by Bedanta and their colligue ^[15] also reveals that the Nepalese students had predominant blood group O (35.2%), followed by A (30.5%), B (28.9%) and AB (5.5%). Contrary to our study. the prevalence of blood group A was most prevalent followed by blood group O, B and AB has been reported by many research studies. [16,17]

Several studies have been carried out to correlate the association between blood groups and bleeding time and clotting time. Individuals with blood group O have significantly lower plasma levels of vWF

and Factor VIII as compared to non-O group individuals. Thus the non-O group individuals can have an increased risk of [10-12] [12] Reddy et thrombosis. al demonstrated that epistaxis is commonly seen in people with O blood group when compared with other ABO blood groups, and they also observed that there is a lower expression of von Willebrand factor (vWF) in them. Franchini et al ^[18] stated that the ABO group can affect the vWF catabolism, which means the plasma vWF levels may depend on blood group of the individual. This concept was accepted by other studies10 who observed that vWF is 25% more in non-O group individual compared to group O individuals. This means that CT & BT will be elevated among the O group individual compared to the other groups. In our study we also found that BT was more prolong in blood group O followed by A, B and AB but the difference was not statistically significant. Similar nonsignificant prolong BT was seen in blood group O in other studies. ^[19,20] In contrast to our study BT was found more prolonged in the blood group AB compared to other group, which was statistically significant. [21]

In our study CT was raised in blood group A followed by O, B and AB which was not statistically significant (p<0.05). Similar non-significant raised CT was seen in Roy et. al. ^[15] however significant prolonged CT was found in B group in other studies. ^[21,22]

Regarding gender-wise distribution of BT & CT, both BT and CT were prolonged in female as compared to males but it was not statistically significant. Similar findings of raised BT and CT were seen in females compared to males but were statistically significant. ^[15,22] There was no such difference in BT and CT in genderwise in study carried by Mahapatra and Mishra. ^[21] This is probably due to presence of estrogen in female, estrogen is found to decrease the level of fibrinogen in the plasma and increase the clotting time. ^[23]

CONCLUSIONS

Blood group O is the most common blood group among the students and AB is the least common blood group. Clotting time was prolonged >6 min among blood group A followed by O, B, and AB. Bleeding time was prolonged >4 min in blood group O followed by A, B, and AB. Also there was statistically no significant relation between blood groups, BT, and CT. Gender-wise BT and CT were higher in females than males, but the difference was statistically insignificant. The statistically insignificant values may be due to the small sample size. Further research with larger sample size and conduct of multi-centric studies are necessary to confirm this finding of different ABO blood groups with different bleeding tendencies and their association with vWF and factor VIII so that preventive measures could be adopted before the onset of such disorders.

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REFERENCES

- 1. Bijlani RL and Manjunatha S. Understanding Medical Physiology.4th edition. Jaypee Brothers Medical Publishers (P) Ltd.2011;88-89
- Yamamoto F. Molecular genetics of ABO. VoxSanguinis. 2000;78 (suppl.2):91–103
- Oriol R, Mollicone P, Coullin A, Dalix M, Candelier JJ. Genetic regulation of the expression of ABH and Lewis antigens in tissues. APMIS. 1991;100(27):28–38
- Sarode R, Goldstein J, Sussman II, Nagel RL, Tsai HM. Role of A and B blood group antigens in the expression of adhesive activity of von Willebrand factor. British Journal Hematology. 2000;109(4): 857–64
- 5. Sadler JE. "Biochemistry and genetics of von Willebrand factor". Annual

Review of Biochemistry. 1998;(67):395–424.

- Roldan V, Francisco M, Begona M, Torregrossa JM, Hernandez D, Valdez M. et al. Plasma von Willebrand factor levels are an independent risk factor for adverse events including mortality and major bleeding in anticoagulated atrial fibrillation patients. Journal of the American College of Cardiology. 2011;57(25): 2496–2504.
- Ghai CL. A Textbook of Practical Physiology. 5th edition. Jaypee Brothers Medical Publishers (P) Ltd. 1999;84– 101.
- Gastineau DA, Moore SB: How important are ABO-related variations in coagulation factor levels? Transfusion. 2001;41:4-5.
- Viel K, Machiah D, Warren D, Khachidze M, Buil A, Fernstrom K. A sequence variation scan of the coagulation factor VIII (FVIII) structural gene and associations with plasma FVIII activity levels. Blood. 2007; 109(9): 3713–3724.
- 10. O'Donnell J and Laffan MA. The relationship between ABO histo-blood group, factor VIII and von Willebrand factor. Transfusion Medicine. 2001; 11(4): 343–51
- 11. Gill JC, Endres-Brooks J, Bauer PJ, Marks WJ, Montgomery RR. The effect of ABO blood group on the diagnosis of von Willebrand disease. Blood. 1987;69:1691-1695.
- 12. Reddy VM, Daniel M, Bright E, Broad SR, Moir AA. Is there an association between blood group O and epistaxis? J Laryngol Otol. 2008;122(4):366–8.
- Pramanik T, Saikia TC, Bandopadhyya M. Prelimilary report on the trend of blood group distribution among Nepalese and Indian medical students. J Nepal Med Assoc. 2001;41:258-61.
- 14. Pramanik T, Adhikari P. Trend of blood group distribution among the different ethnic groups of Kathmandu valley. Nepal Med Coll J. 2006;8(4):248-9.
- 15. Roy B, Banerjee I, Sathian B, Mondal M, Saha CG. Blood group distribution and its relationship with bleeding time and clotting time: A medical school based observational study among Nepali, Indian and Sri Lankan students.

Nepal journal of Epidemiology. 2011;1(4):135-140.

- Singh R. Distribution of ABO blood group in Nepal. J Nepal Med Assoc. 1985;23:1.87-1.92
- 17. Pramanik T, Pramanik S. Distribution of ABO and Rh blood groups in Nepalese medical students: a report. East Mediterr Health J. 2000 Jan;6(1):156-8.
- Franchini M, Franco C, Giovanni T, Martina M, Giuseppe L. Relationship between ABO blood group and von Willebrand factor levels: from biology to clinical implications. Thromb J. 2007;5:14.
- Patil SV, Gaikwad PB, Vaidya SR, Patil US, Kittad SD. To study the blood group distribution and its relationship with bleeding and clotting time in dental students. Asian J Medical Pharmaceutical Sci. 2013;1(1):1–4.

- Adhikari P, Pramanik T, Pokhael R, Khanal S. Relationship between blood group and epitaxis among Nepalese. Nepal Med Coll J. 2008;10(4):264–5.
- 21. Mahapatra B, Mishra N. Comparison of bleeding time and clotting time in different blood groups. American J Infectious Dis. 2009; 5(2):113–15.
- Sasekala M, Saikumar P. Relationship between bleeding time and clotting time among gender difference and varying blood groups in UG medical students. IOSR J Dental Medical Sci. 2013;10(6): 40–3.
- 23. Derham RJ, Buchan PC. Haemorheological consequences of estrogen and progestogen therapy. Eur J ObstetGynecolReprod Biol. 1989;32: 109-14

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