

## BLOOD GROUPS AND ITS RELATIONSHIP WITH BLEEDING TIME AND CLOTTING TIME – AN CROSS SECTIONAL STUDY

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### Abstract

**Background:** In the field of transfusion medicine, the ABO blood group system plays an important role. ABO blood group system is determined by two antigens (A and B). These antigens are carbohydrate moieties located at the peripheral end of the carbohydrate chain of glycoprotein on the surface of red blood cells. The gene coding for these antigens are located on chromosome 9q34 which also exerts a major influence on the von Willebrand factor gene located on chromosome 12p12. Hence, ABO blood group system also determines the plasma levels of vWf. Some authors have also found 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 in them. The aim of this study is to assess the relationship between the Bleeding Time and Clotting Time in different blood groups. **Materials and Methods:** This is a cross sectional study which includes 150 subjects in the age group of 17-20 years. After obtaining informed consent, Bleeding Time and Clotting Time were determined by Duke's Filter Paper method and Wright's Capillary Tube method respectively. Statistical analysis was done by using One way ANOVA. The p value of <0.05 was taken as statistically significant. **Result:** Bleeding Time values reached up to 4 minutes in 69.5% subjects in AB blood groups and Clotting Time values reached up to 6 minutes in 13.9% subjects in B blood group but when the data was statistically analyzed by One way ANOVA test, the p value was not statistically significant for Bleeding Time(0.354) and Clotting Time (0.151). **Conclusion:** In our study, we found out that Bleeding Time and Clotting Time varies in different ABO blood groups, particularly the B and AB blood groups had a potential risk of bleeding.

## INTRODUCTION

In the field of Transfusion medicine, the ABO blood group system plays an important role. This blood group system was first discovered by Austrian Biologist and physician Karl Landsteiner. ABO blood group system consists of two antigens(A and B).<sup>[1,2]</sup> These antigens are carbohydrate moieties located at the peripheral end of the carbohydrate chain of glycoprotein on the surface of red blood cells. The gene coding for these antigens are located on chromosome 9 and 19.<sup>[3]</sup>

Recent research data shows that ABO blood groups are associated with various diseases, for example gastric cancer, periodontal diseases and venous thrombosis.<sup>[4,5]</sup> Some authors have found 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 in them.<sup>[6]</sup>

Von Willebrand factor(vWf) is a large multimeric blood glycoprotein involved in hemostasis, synthesis by Weibel - Palade bodies in the endothelial cells and the alpha granules of megakaryocytes. It helps in platelet adhesion and platelet aggregation. It also acts as a special carrier protein for the clotting factor VIII (Anti Hemophilic factor A). Thus, von Willebrand factor plays an important role in formation of temporary hemostatic plug and also the conversion of it into definite clot by activation of clotting mechanism. The gene codes for vWf is located on chromosome 12p12. Some research papers have proved that other genes like gene locus of ABO blood group on the chromosome 9q34 exert a major influence on the vWf gene.<sup>7</sup> Therefore, the ABO blood system influences the bleeding time and clotting time.

Although several studies have succeeded in correlating the association between bleeding disorders and clotting disorders with different ABO blood group, other workers could not find out any

association between bleeding tendencies and different ABO blood groups.<sup>[1,6]</sup> Hence, this study was conducted to find out existence of relationship between the different ABO blood groups with bleeding time and clotting time.

## MATERIALS AND METHODS

This cross sectional study was conducted after obtaining informed and written consent, one hundred and fifty(150) subjects in the age group of 17-20 years were selected. Personal details like H/O bleeding diathesis and H/O drug intake(NSAIDS) were obtained by administering a questionnaire to the subjects.<sup>[7,8]</sup> All the subjects were included in the study as none of them replied positive with regard to exclusion criteria.

Bleeding Time was determined by Duke's Filter paper method. A deep skin puncture was made and the length of the time required for bleeding to stop was recorded by blotting the drop of blood coming out of the incision every 30 seconds by using blotting paper. Bleeding Time was calculated by multiplying the number of drops on the filter paper and 30 seconds. The normal bleeding time by Duke's Filter paper method is usually in the range of 1-5 minutes.<sup>[9]</sup>

Clotting Time was determined by Capillary Tube method. A standard incision was made in the skin of the subjects and blood is taken into a capillary glass tube. The length of the time taken for the blood to clot can be calculated by breaking the capillary tube after 2 minutes, 1-2cm from one end every 30 seconds. The examiner looked for the appearance of fibrin thread. The normal clotting time estimated by this method falls in the range of 5-11 minutes.<sup>[9]</sup>

Blood Group was determined by using standard antisera. The sample blood was mixed with anti-A serum, anti-B serum and anti-D serum. Agglutination was confirmed by observing under low-power objective of the compound microscope.<sup>[9]</sup> These tests were done by the author with the help of the lab technician and the assistance of senior staff members. This study was conducted over a period of 15 days. The values of Bleeding Time and Clotting Time were cross checked thrice and the repeated value of BT and CT was taken as final value of BT and CT.

### Statistical Analysis

The data was expressed in frequency table and cross table in order to find out the sex ratio and to compare different ABO blood groups. Statistical analysis was done by using SPSS version 16. Data was analyzed using one way ANOVA.  $p < 0.05$  was considered to be statistically significant.

## RESULTS

Among the 150 subjects, 39.3% were males and 60.7% were females in the age group of 17-20 years.

The values of Bleeding Time and Clotting Time did not show any difference between males and females. In our study, 36% belonged to O blood group(O+ve) which was the dominant blood group and 64% belonged to non-O-blood groups (28.7% B+ve, 15.3% AB+ve, 17.3% A+ve and 2.7% A+ve blood group).

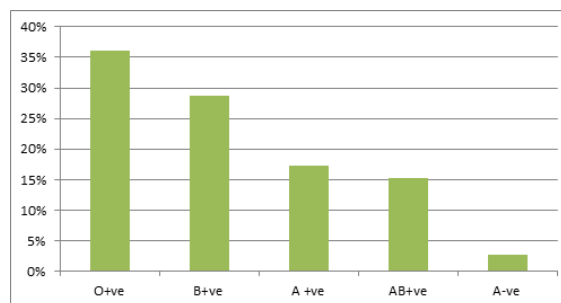


Figure 1: Percentage of different ABO blood groups.

Bleeding Time values reached up to 4 minutes in 69.5% of the subjects with AB blood group, 51.1% of the subjects with B blood group, 33.3% of the subjects with A blood group and 27.7% of the subjects with O blood group.

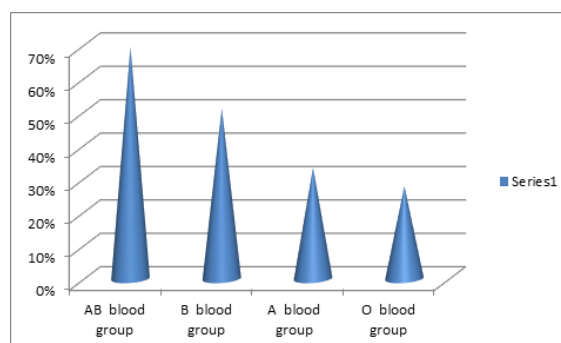


Figure 2: Percentage of different ABO blood groups with bleeding time value of 4 minutes.

One way ANOVA tests performed on the data did not show any statistically significant difference between the bleeding time of ABO groups( $p=0.354$ ).

Clotting Time values reached up to 6 minutes in 13.9% of the subjects with B blood group, 6.6% of the subjects with A blood group, 5.2% of the subjects with O blood group and 4.3% of the subjects with AB blood group.

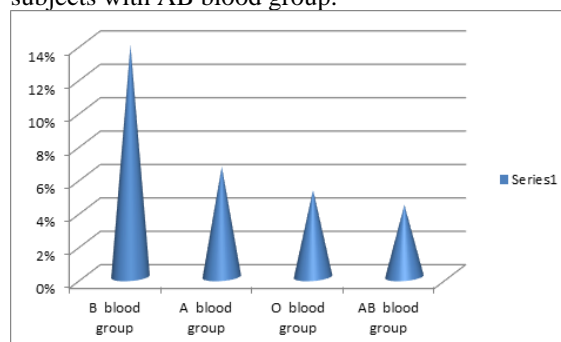


Figure 3: Percentage of different ABO blood groups with clotting time value of 6 minutes.

One way ANOVA tests performed on the data did not show any statistically significant differences

between the clotting times of ABO groups( $p=0.151$ ).

**Table 1: One way ANOVA analysis of bleeding time in different ABO blood group**

	Sum of squares	df	Mean square	F	Sig.
Between the groups	2.873	4	0.718	1.111	0.354
Within the groups	93.711	145	0.646		
Total	96.584	149			

**Table 2: One way ANOVA analysis of clotting time in different ABO blood groups.**

	Sum of squares	df	Mean square	F	Sig.
Between the groups	3.738	4	0.935	1.708	0.151
Within the groups	79.325	145	0.547		
Total	83.064	149			

## DISCUSSION

James Donnell et al have showed that the ABO blood group system plays an important role in determining the plasma level of Factor VIII – vWf complex. This is due to the influence of ABO gene over the genetic locus of vWf. The rate of synthesis of vWf from the endothelial cells, its secretion and plasma clearance are influenced by the ABO blood group system. Thus, the lower vWf:Ag level seen in O blood group may be due to rapid clearance which in turn will reduce the plasma half-life of vWf.<sup>[10-20]</sup>

Daniel et al reported that in Caucasian patients admitted with epistaxis mostly belongs to O blood group.<sup>[11]</sup> Ionescu et al observed that there is an increased risk for arterial thrombosis (eg. Coronary heart disease) in individuals of blood group A, B and AB.<sup>[12]</sup> Horwich et al observed that there is an increased risk for bleeding(eg. Cerebral hemorrhage and bleeding duodenal ulcer) in individuals of O blood group.<sup>[13,21-24]</sup>

Norman Cristian Souse et al observed an interesting finding that vWF:Ag and Factor VIII levels were lowered in A and B blood groups when compared to the AB group, but no difference was observed when compared with O blood group. Therefore, A and B blood group individuals are at potential risk of bleeding.<sup>[14,25]</sup>

Garcia et al reported that non-OO genotype carriers during treatment with vitamin-K antagonist have less risk of bleeding due to higher level of Factor VIII and vWf level when compared with O blood group individuals.<sup>[15]</sup>

Wiggins et al observed an increased risk of venous thrombosis and myocardial infarction with the A blood group, and increased risk of venous thrombosis and ischemic stroke with the B blood group.<sup>[16]</sup>

## CONCLUSION

In an aim to compare the Bleeding Time and Clotting Time in different ABO blood groups, we found that the values were in the normal limit, but most of the subjects in AB blood group bleeding time values were in higher level of normal limit when compare to other blood groups. Similarly in

most of the subjects in B blood group Clotting Time values were in higher level of normal limit when compare to other blood groups. This may be attributed to decreased expression of von Willebrand factor and Factor VIII levels in them. The statistically insignificant values may be due to the small sample size. Further research with larger sample size and conduct of multicentric studies are necessary to confirm this finding of different ABO blood groups with different bleeding tendencies and their association with vWf and Factor VIII.

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