ORIGINAL ARTICLE

# Bleeding Time in Different Blood Groups of the ABO System- A Descriptive Cross-Sectional Study

Bam S[1], Shrestha M[2], Jyoti S[2], Upreti A[2], Chaudhary S[1], Joshi AR[3]

**ABSTRACT**

**Introduction:** ABO blood grouping depends on the presence of specific antigens on red blood cell membranes: Type A, B, AB, and O have antigens a, b, both a and b, and neither a nor b, respectively. Bleeding time is the time between the puncture of vessels and to stoppage of bleeding. Certain disease seems more prevalent in specific ABO blood groups. Likewise, many studies found that blood group O and females exhibit a higher bleeding tendency. **Aims:** To study the bleeding time in different ABO blood groups and genders. **Method:** A descriptive cross-sectional study was conducted among 139 medical students of first and second year after Ethical approval from the Institutional Review Committee of Nepalgunj Medical College Teaching Hospital, during practical hours from June to October 2025. **Results:** Blood group O was the most common (n=63), followed by A and B (n=28 each), and AB (n=20). Among the ABO and Rhesus factors, O-positive was most prevalent (41%), followed by A-positive and B-positive (18% each), AB-positive (12.2%), O-negative (4.3%), and the least was A-negative, B-negative, and AB-negative (2.2%). We found more Rh-positive students (90.6%) than Rh-negative (9.4%). The mean bleeding time was 2.698 minutes, with a range of 1.5 to 6.5 minutes. Non-O blood group students (n=76) consisting of Type A, B, and AB, had a shorter bleeding time compared to the O blood group, which was statistically significant. Males (n=70, mean: 2.771±.8710) had a longer bleeding time than females (n=69, mean: 2.623±.8762). **Conclusion:** Blood group O was predominant among the ABO group, and with the Rhesus factor, O positive was predominant. Bleeding time was prolonged in the O blood group than in any other group. Bleeding time was prolonged among males than among females.

***Keywords:*** ABO grouping, Bleeding time (BT), Gender, Non-O blood group

**Authors:**

1. Mr. Sanjog Bam
2. Dr. Merina Shrestha
3. Dr. Sabita Jyoti
4. Dr. Arish Upreti
5. Mr. Shailesh Chaudhary
6. Mr. Abishek Raj Joshi

[1]Department of Physiology, Nepalgunj Medical College Teaching Hospital, Chisapani, Banke

[2]Department of Community Medicine, Nepalgunj Medical College Teaching Hospital, Kohalpur, Banke

[3]Department of Pharmacology, Nepalgunj Medical College Teaching Hospital, Chisapani, Banke

**Address for Correspondence:**

Mr. Sanjog Bam
Lecturer
Department of Physiology
Nepalgunj Medical College and Teaching Hospital
Chisapani, Banke
Email: bam.dsp@gmail.com

## INTRODUCTION

ABO blood grouping is based on the presence of an antigen on the surface of RBCs. Type A, B, AB, and O have the antigen a, b, both a and b, and neither of these, respectively, on the membrane; consequently, these typed individuals' plasma contains anti-B, anti-A, neither, or both anti-A and anti-B, respectively. [1-3] Bleeding time (BT) is the time from the onset of bleeding to the formation of a temporary hemostatic plug that stops bleeding. BT assesses the integrity of platelets.[4-6] Research consistently shows that Type O individuals exhibit a higher bleeding tendency due to significantly lower levels of von Willebrand factor (vWF).[7,8] Additionally, it has been noted that the absence of ABO antigens may expedite the clearance of vWF from plasma.[9] In contrast, a retrospective study done by B. Mahapatra and N. Mishra (2019) on 740 medical students found that BT was significantly higher in AB group than in persons with blood group O.[10] The relationship between blood groups and BT varies across populations, but it is clear that there is a notable lack of research on gender-based variations within blood groups, especially in the Nepalese context. The interaction between ABO group and gender regarding BT is still insufficiently explored. However, it is well-documented that gender differences in BT exist, with females typically exhibiting longer BT than males. [11-14]

Hence, this study was conducted to find out the predominant ABO blood group and its relation with BT, and also gender variation in BT.

## METHODS

The descriptive cross-sectional study was conducted in the Department of Physiology, Nepalgunj Medical College, from June to October 2025 on 139 students, including male and female medical students, between the age group of 18 and 26 years. The blood group and bleeding time of the students were determined. Comparison of bleeding time between males and females was performed. Moreover, any difference in bleeding time between O and non-O blood groups was determined, as well as effect of the Rh factor on bleeding time was assessed. Information regarding the history of bleeding disorder and drug intake (NSAIDS) or any recent trauma, surgery, or ongoing infection was obtained through a questionnaire to the students, and categorized as the exclusion criteria. All students participated, as none of them fell under the exclusion criteria. Inclusion criteria include the healthy physically and mentally fitted students of MBBS and BDS between 18-26 years old, irrespective of gender.

Formula used for the calculation of sample size:

n = N / [1 + N × e^2] Where,

n = the sample size

N = Target population = 210

e = the margin error in the calculation, i.e., 5%

All the data were collected after approval from NGMC IRC (ref. 68/081-082) and determined during practical time in the physiology laboratory. Blood samples were collected by finger prick with a sterile lancet after cleaning the puncture site with spirit. The sample blood was mixed with anti-A, anti-B, and anti-D serum. Blood groups were determined on the basis of the presence or absence of agglutination. Agglutination was confirmed by observing under a low-power objective of a compound microscope.[15]

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 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.[15]

**Statistical analysis**: One- way ANOVA and independent t-test were used in SPSS software to analyze the data.

## RESULTS

The sample size of a total of 139 medical students was analyzed. There was an almost homogeneous age group (17-26 years) as all belonged to the first and second year MBBS students and the BDS students. Out of 139 students, 70 were male, and 69 were female (Fig. 1, Table IV). We found that blood group O was predominant (n=63), followed by an equal number of A and B (n=28), and least in the AB blood group (n=20) among students. The percentage distribution of ABO blood group was in order of O (45.3%)> A (20.1%) = B (20.1%)> AB (14.4%) [Table V]. With Rh factor, in ABO blood group, we found O+ blood group in a more predominant number(41%), followed by A+(18%), B+ (18%), AB+(12.2%), O-(4.3%), A-(2.2%), B-(2.2%) and AB-(2.2%), displayed in Table I.

The mean value BT was 2.698 min, with 1.5 min and 6.5 min as the lowest and highest BT values in the study, respectively. In our study, there were more Rh-positive students (90.6%) than Rh-negative students (9.4%). Table I and II shows the distribution of ABO blood groups and the Rh blood groups. There were more non-O blood groups than the O blood group among students [Table III].

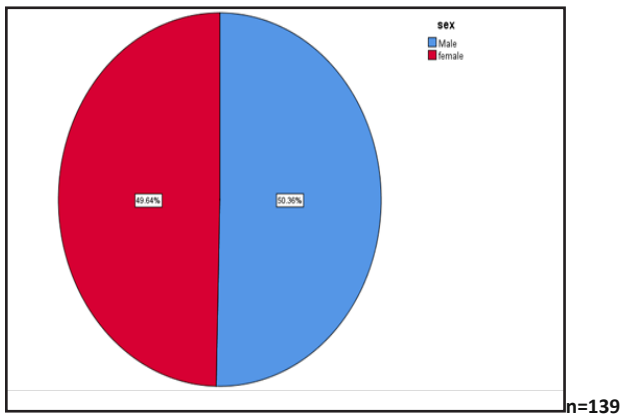


***Figure 1: Gender distribution among students***

| Blood group | Frequency | Percent |
|---|---|---|
| A+ | 25 | 18.0 |
| A- | 3 | 2.2 |
| B+ | 25 | 18.0 |
| B- | 3 | 2.2 |
| AB+ | 17 | 12.2 |
| AB- | 3 | 2.2 |
| O+ | 57 | 41.0 |
| O- | 6 | 4.3 |
| Total | 139 | 100.0 |

***Table I: Showing the distribution and frequency of blood group among students***

| Rh blood group | Frequency | Percent |
|---|---|---|
| Negative | 13 | 9.4 |
| Positive | 126 | 90.6 |
| Total | 139 | 100.0 |

***Table II: Showing distribution and frequency of Rh blood group among students***

| Blood group | Frequency | Percent |
|---|---|---|
| Non-O | 76 | 54.7 |
| O | 63 | 45.3 |
| Total | 139 | 100.0 |

***Table III: Showing distribution and frequency of blood groups among students***

| Sex | Bleeding time | | | | Significance |
|---|---|---|---|---|---|
| | ≥ 3.5 min | | < 3.5 min | | |
| | No. | % | No. | % | |
| Male | 15 | 10.8 | 55 | 39.6 | F=0.003 |
| Female | 11 | 7.9 | 58 | 41.7 | P=.319 |
| Total | 26 | 18.7 | 113 | 81.3 | |
| | Male | Female | Total | | |
| Number | 70 | 69 | 139 | | |
| Mean ± SD | 2.771± .8710 | 2.623± .8762 | | | |

***Table IV: Gender wise distribution of bleeding time with an independent t-test***

BT was prolonged more than or equal to 3.5 minutes among males (10.8 %) and among females (7.9%). Analyzing the data with an independent t-test did not show any significant difference BT of gender groups (p=.319) (Table IV).

| Blood group | Bleeding time | | | | Significance |
|---|---|---|---|---|---|
| | ≥ 3.5 min | | < 3.5 min | | |
| | No. | % | No. | % | |
| A | 4 | 2.9 | 24 | 17.3 | |
| B | 4 | 2.9 | 24 | 17.3 | |
| AB | 5 | 3.6 | 15 | 10.8 | |
| O | 13 | 9.4 | 50 | 36.0 | |
| Total | 26 | 18.7 | 113 | 81.3 | F=2.295 |
| Blood group | Number | Mean± SD | | | p =.081 |
| A | 28 | 2.482 ± .6869 | | | |
| B | 28 | 2.482 ± .8765 | | | |
| AB | 20 | 2.675 ± .6340 | | | |
| O | 63 | 2.897 ± .9762 | | | |
| Total | 139 | 2.698 ± .8736 | | | |

***Table V: Distribution of bleeding time on various blood groups with one-way ANOVA analysis***

BT prolonged more than or equal to 3.5 minutes among O (9.4%), followed by AB (3.6%), A (2.9%), and B (2.9%). Also from the table above, we found O-blood group students have prolonged BT than any other blood groups, whereas blood group A and B have equal but the least BT than others. Although there was a difference in BT of ABO blood groups but it was not found to be statistically significant (p=.081) (Table V).

| Blood group | Bleeding time | | | | Significance |
|---|---|---|---|---|---|
| | ≥ 3.5 min | | < 3.5 min | | |
| | No. | % | No. | % | |
| O | 13 | 9.4 | 50 | 36 | |
| Non-O | 13 | 9.4 | 63 | 45.3 | F=1.638 |
| Total | 26 | 18.7 | 113 | 81.3 | p = .014 |
| | O | Non-O | Total | | |
| number | 63 | 76 | 139 | | |
| Mean ±SD | 2.897± .9762 | 2.533± .7454 | | | |

***Table VI: Distribution of bleeding time on O and Non-O blood group with independent t-test analysis***

While considering O and Non-O blood groups, an equal number (9.4%) of both groups show BT prolonged more than or equal to 3.5 minutes. Analyzing with an independent t-test, data shows a statistically significant difference in BT of O and Non-O blood groups (p=.014) (Table VI).

In our study, we found that more than 90 percent of students were Rh positive; being disproportionate in size among Rh blood groups, it may be inappropriate to generalize the BT difference between the groups.

## DISCUSSION

In this study conducted on 139 students, the blood group O showed predominance (45.3%) in the percentage distribution of ABO blood groups, followed by A (20.1%), B (20.1%), and least AB (14.4%). Asian trend of prevalence of blood groups O>B>A>AB, with predominant O and least AB blood group, has been reported in many research studies, which is similar to our study.[10,16,17,18] Contrary to our study, a different trend of prevalence of blood groups B>O>A>AB, with the B blood group having a predominance, was observed in various research studies.[1, 19, 20] In our study, we found that there is a significant difference in mean BT between O and non-O blood groups, with a longer time in O than in non-O blood groups. Comparatively, we found a larger number of O blood group students having BT more than 3.5 minutes than any other blood group student. Similar to ours, many studies in the past have found BT prolongation in blood group O, and they also described it due to less expression of vWF.[13,21,22,23] Also in our study, we found O-blood group students have prolonged BT, followed by AB, and then blood group A and B having the least and equal BT, but the difference was not statistically significant.

In contrast, one study BT is found to be prolonged in blood group B, followed by blood group O.[24] Moreover, there is one retrospective study done by B. Mahapatra and N. Mishra in 2019 on 740 medical students, where clotting time was prolonged in blood group B and bleeding time was prolonged in blood group AB than in other blood groups, and the difference was statistically significant.[10]

vWF is important for hemostasis, which plays an important role in platelet adhesion and aggregation. Research indicates that the ABO gene locus, chromosome number 9, accounts for about 30% of the genetic factors affecting vWF levels, suggesting that the ABO blood group influences plasma vWF. And some studies suggest that the absence of ABO antigens may expedite the clearance of vWF from plasma. Both studies explain the lower levels of vWF in individuals with blood group O. Therefore, one can say that a lack of A and B antigens may lead to lower plasma vWF levels, explaining the increased bleeding tendencies associated with O blood groups.[9,25]

We found that in our study mean BT of males is greater than females, but it is not statistically significant; a larger number of male has BT more than 3.5 min compared to females. One study shows prolonged BT in males than females similar to us, but their difference was statistically significant, unlike ours.[24] Also, a similar result was found in a study done by Benjamin and Bagavad showing BT prolonged in males as compared to females.[26] Like ours, in some studies, no such significant difference in BT was observed between male and female.[2,10] It was reported in one study that testosterone inhibits platelet aggregation, and this effect was dependent on endothelial nitric oxide synthesis, which somehow favors our study.[27] In contrast, one study reported that thromboxane A2 synthesis increased by testosterone and this steroid hormone also facilitates platelet aggregation.[28] However, in some studies, it is well-documented that gender differences in bleeding time exist, with females typically exhibiting longer bleeding times than males, and it is thought to be due to the high estrogen, which may suppress platelet functions and also causes fibrinogen decrement in blood.[3, 11, 12, 13, 14, 29, 30]

## LIMITATIONS

The studied sample size was small; therefore, the findings may vary from those of other studies. A bigger multicentric study is suggested to verify the above-mentioned findings.

## CONCLUSION

In the present study population, the O blood group was predominant among other ABO blood groups. We found that bleeding time was prolonged in cases with blood group O than in other blood groups. Moreover, we found that the mean bleeding time of males is greater than females, and also a larger number of male has BT more than 3.5 compared to females, but the difference was not significant.

## REFERENCES


1. Roy B, Banerjee I, Sathian B, Mittal A, Baboo NS, Jha N. 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 J Epidemiol. 2011;1(4):135–40. doi:10.3126/nje.v1i4.5755. Available from: https://doi.org/10.3126/nje.v1i4.5755

2. Kaur M, Singh A, Bassi R, Kaur D. Blood group distribution and its relationship with bleeding time and clotting time. Natl J Physiol Pharm Pharmacol. 2015;5(3):253–57. doi:10.5455/njppp.2015.5.2609201433. Available from:https://doi.org/10.5455/njppp.2015.5.2609201433

3. Manandhar Adhikari S, Amatya A. Variation of bleeding time and clotting time in the ABO blood groups. J Physiol Soc Nepal. 2020;1(2):19–23. Available from: pdf

4. Pal GK, Pal P. Comprehensive textbook of medical physiology. Vol. 1. 1st ed. New Delhi: Jaypee Brothers Medical Publishers; 2017. ISBN:9789386056979. doi:10.5005/jp/books/12960. Available from:https://doi.org/10.5005/jp/books/12960

5. Pal GK, Pal P. Textbook of practical physiology. 2nd ed. Manipal: Universal Press; 2005. p.107–09.

6. Adhana R, Chaurasiya R, Verma A. Comparison of bleeding time and clotting time between males and females. Natl J Physiol Pharm Pharmacol. 2018;8(10):1388–90. doi:10.5455/ijmsph.2018.06201417062018. Available from: https://www.njppp.com/fulltext/28-1528542228.pdf

7. O'Donnell J, Laffan MA. The relationship between ABO histo-blood group, factor VIII and von Willebrand factor. Transfus Med. 2001;11:343–51. Available from: https://doi.org/10.1046/j.1365-3148.2001.00315.x

8. Franchini M, Mannucci PM. ABO blood group and thrombotic vascular disease. Thromb Haemost. 2014;112(6):1103–09.doi:10.1160/TH14-05-0457. Available from: https://doi.org/10.1160/TH14-05-0457

9. Gallinaro L, Cattini MG, Sztukowska M, Padrini R, Sartorello F, Pontara E, et al. A shorter von Willebrand factor survival in O blood group subjects explains how ABO determinants influence plasma von Willebrand factor. Blood. 2008;111(7):3540–45.doi:10.1182/blood-2007-11-122945.Available from:https://doi.org/10.1182/blood-2007-11-122945

10. Mahapatra B, Mishra N. Comparison of bleeding time and clotting time in different blood groups. Am J Infect Dis. 2009;5(2):106–08 Available from: ResearchGate

11. Kumar S, VK J, George J, Mukkadan J. Bleeding time and clotting time in healthy male and female college students of Karukutty Village, Kerala. Health Prospect. 2013;12(1):7–9. doi:10.3126/hprospect.v12i1.8720. Available from: Health Prospect

12. Verma A, Chaurasia R, Adhana R, Ballabh J, Kaur J. Interdependence of major blood group with bleeding time and clotting time. MedPulse Int J Physiol. 2019; 11:34-37.

13. Waghmare RV, Muniyappanavar NS. Influence of blood groups on bleeding and clotting time. Int Physiol. 2018;6(3):200–204. doi:10.21088/ip.2347.1506.6318.6. Available from: https://dx.doi.org/10.21088/ip.2347.1506.6318.6

14. Gupta SP, Dutta P, Anand S, Kanchan RK. Correlation of bleeding time and clotting time with ABO blood grouping among first year medical students. Natl J Physiol Pharm Pharmacol. 2021;11(5):525–529. doi:10.5455/njppp.2021.11.12371202018012021. Available from: http://dx.doi.org/10.5455/njppp.2021.11.12371202018012021

15. Pal GK, Pal P. Textbook of practical physiology. 3rd ed. New Delhi: Universities Press (India) Pvt Ltd; 2010. P.100–01.

16. Thenmozhi S, Neelambikai N, Aruna P. Comparison of bleeding time and clotting time in different ABO blood groups.

Natl J Physiol. 2013;1(1):19-24.

17. Kohli PG, Kaur H, Maini S. Relationship of bleeding time and clotting time with blood groups. Res J Pharm Biol Chem Sci. 2014 Jan 1;5(2):1780-3.Available from:https://www.rjpbcs.com/pdf/2014_5(2)/%5B210%5D.pdf

18. Sasekala M, Saikumar P. Relationship between bleeding time and clotting time among gender difference and varying blood groups in UG medical students. IOSR J Dent Med Sci. 2013;10(6):40–43. Available from: https://www.iosrjournals.org/iosr-jdms/papers/Vol10-issue6/I01064043.pdf

19. Abhishekh B, Mayadevi S, Meena D, Usha KC. Distribution of ABO and Rhesus-D blood groups in and around Thiruvananthapuram. Kerala Med J. 2011; 1:28–29. Available from: ResearchGate

20. Talib HV. Handbook of Medical Laboratory Technology. 2nd ed. New Delhi: CSB Publishers; 1991. P.205-10.

21. Jha RK, Kushwaha MS, Kushwaha DK, Tiwari S, Bhandari A, Nepal O. Blood group distribution and its relationship with bleeding time and clotting time in medical undergraduate students. Int J Res Rev [Internet]. 2017;4(9):10–15. Available from: IJRR003.pdf

22. Gavit S, Bhorania S. An observational study: association of blood groups with bleeding time and clotting time. Asian J Pharm Clin Res. 2022;15(7):119–22. doi:10.22159/ajpcr.2022.v15i7.44266 Available from: https://doi.org/10.22159/ajpcr.2022.v15i7.44266

23. Chinara A, Purohit P, Mahapatra B. No association of bleeding time and clotting time with four ABO blood groups in healthy young adults: an observational study. Natl J Physiol Pharm Pharmacol 2019;9(12):1193–97. doi:10.5455/njppp.2019.9.0931620092019. Available from: https://www.ejmanager.com/mnstemps/28/28-1568556156.pdf?t=1765625413

24. Adhikari B, Maharjan N. Blood group distribution and its association with bleeding time and clotting time among medical students. Asian J Med Sci 2024 Aug 1. doi:10.71152/ajms.v15i8.1389. Available from: https://doi.org/10.71152/ajms.v15i8.1389

25. Franchini M, Capra F, Targher G, Montagnana M, Lippi G. Relationship between ABO blood group and von Willebrand factor levels: from biology to clinical implications. Thromb J . 2007;5:14. doi:10.1186/1477-9560-5-14. Available from: https://doi.org/10.1186/1477-9560-5-14

26. Benjamin JJ, Geetha MB. Study of association of bleeding and clotting time with blood group among young adults. Indian J Clin Anat Physiol. 2020; 7(4):350-53. doi:10.18231/j.ijcap.2020.074. Available from:Indian Journal of Clinical Anatomy and Physiology

27. Borchgrevink CF. Platelet adhesion in vivo in patients with bleeding disorders. Acta Med Scand. 1961;170:231–43. doi:10.1111/j.0954-6820.1961.tb00234.x Available from: https://doi.org/10.1111/j.0954-6820.1961.tb00234.x

28. Campelo AE, Cutini PH, Massheimer VL. Testosterone modulates platelet aggregation and endothelial cell growth through nitric oxide pathway. J Endocrinol.2012;213(1):77–87.doi:10.1530/JOE-11-0441 Available from: https://doi.org/10.1530/JOE-11-0441

29. Adhana R, Chaurasia R, Verma A. Comparison of bleeding time and clotting time between males and females. Natl J Physiol Pharm Pharmacol.2018;8(10):1388-90.doi.org/10.5455/ijmsph.2018.06201417062018 Available from: https://doi.org/10.5455/ijmsph.2018.06201417062018

30. Aleem A, Wahid M. Correlation of blood groups, bleeding time and clotting time in male and female students: An observational study. Pak J Pharm Res. 2016;2(2):121-26.doi: 10.22200/pjpr.20162121-126 Available from: Researchgate