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A Study To Observe Bleeding Time, Clotting Time And Total Leucocyte Count And Neutrophil- Lymphocyte Ratio In Male And Female Young Adults Of Andhra Pradesh, India.

Kumar Sai Sailesh[1]*, Movva Swathi[2], Amarnath P[3], Sudhakar Reddy R[4], Tejaswani Santoshi Bandaru[5], and Suseelamma D[6].

[1]Department of Physiology, Vishnu Dental College, Bhimavaram, West Godavari District, Andhra Pradesh.
[2]Department of Physiology, Vishnu Dental College, Bhimavaram, West Godavari District, Andhra Pradesh.
[3]Department of Anatomy, Vishnu Dental College, Bhimavaram, West Godavari District, Andhra Pradesh.
[4]Department of Oral Medicine and Radiology, Vishnu Dental College, Bhimavaram, West Godavari District, Andhra Pradesh.
[5]BDS student, Vishnu Dental College, Bhimavaram, West Godavari District, Andhra Pradesh.
[6]Department of Anatomy, Vishnu Dental College, Bhimavaram, West Godavari District, Andhra Pradesh.

ABSTRACT

Haemostasis is defined as stagnation of the blood out of a cut blood vessel by natural mechanism. As bleeding and clotting time guidelines was written in nearly 30-35 years older, it is mandatory to refine the normal range and also gender and zonal differences. Although, the studies on male and females comparison are sparse, there may be difference among the genders. The present study was undertaken to compare the bleeding time, clotting time and total leucocyte count and neutrophil-lymphocyte ratio in male and female young adults of Andhra Pradesh. The present study was conducted at Department of Physiology, Vishnu Dental College, Bhimavaram, West Godavari District, Andhra Pradesh. A total of 50 first year dental students including both males (n=15) and females (n=35) were part of the study after obtaining written informed consent. All the tests were performed manually as per the standard guidelines specified in the literature. Significantly higher levels of clotting time and NLR were observed in males when compared with females. As NLR is associated with many physiological and pathological conditions, we recommend further detailed studies in this area to explore the diagnostic and prognostic role of NLR.

Keywords: Bleeding time, Clotting time, Total leucocyte count, Gender difference

Corresponding author

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INTRODUCTION

Haemostasis is defined as stagnation of the blood out of a cut blood vessel by natural mechanism. Bleeding time is performed to test the functioning of the platelets. It is the time duration between the onset and arrest of bleeding. The properties of the platelets will alter when there is a injury to the blood vessel. The cell membrane of platelets become more permeable to water and become more stick which favors the platelet plug formation. Earlier studies reported that bleeding time was higher in females when compared with males [1]. It was assumed that this higher bleeding time may be due to suppression of platelet functioning by female reproductive hormone estrogen [2]. In the process of blood coagulation, blood losses the fluidity and becomes gel like semi solid to decrease further loss of the blood. The factors involved in the process of coagulation are known as blood coagulation factors. All the coagulation factors responsible for blood coagulation were present in the blood itself.

However, they are kept in inactive state until there is any injury. These factors bring about blood coagulation through intrinsic or extrinsic mechanism. Determination of clotting time is a simple and cost effective method to assess the functioning of clotting factors. There was a contradiction regarding the values of clotting time as some studies reported that clotting time was higher in males [1] and some reported higher in females [2].

It was assumed that plasma fibrinogen levels are decreased in females due to action of estrogen [3]. As bleeding and clotting time guidelines was written in nearly 30-35 years older, it is mandatory to refine the normal range and also gender and zonal differences. White blood cells are nucleated type of blood cells which plays a key role in the immune functions. Although, the studies on male and females comparison are sparse, there may be difference among the genders. This is because of current day life style, which increased the stress levels. When the stress level alters, it has an impact on circulating blood cells [4] Further, it was reported that, stress causes greater increase in the cortisol response in females than males [5,6]. The present study was undertaken to compare the bleeding time, clotting time and total leucocyte count and neutrophil-lymphocyte ratio in male and female young adults of Andhra Pradesh.

MATERIALS AND METHODS

Study design: Cross sectional study

Study setting: The present study was conducted at Department of Physiology, Vishnu Dental College, Bhimavaram, West Godavari District, Andhra Pradesh.

Participants: A total of 50 first year dental students including both males (n=15) and females (n=35) were part of the study after obtaining written informed consent. The study was approved by institutional human ethics committee. The following criteria were used to recruit the participants.

Inclusion criteria:

1. Age group 17-25 years
2. Healthy and willing participants
3. Not under any kind of medication

Exclusion criteria:

1. Participants with any serious disorders or diseases
2. Under any therapy or treatment
3. Unwilling participants

Methods:

Determination of bleeding time: Bleeding time was determined by using Duke Method. The stop watch was set to zero and under aseptic conditions a puncture was made to a depth of about 2-3mm on the tip of the finger. As soon as the puncture was made, the stop watch was turned on. The oozing blood was blotted gently

with the filter paper after 30 seconds. The procedure was repeated in every 30 seconds using a fresh area on each occasion until there is arrest of bleeding. Each blood drop on the filter paper represents 30 sec flow of blood. The number of blood drops on the filter paper was counted and bleeding time was expressed in seconds [7, 8].

Determination of clotting time: Clotting time was determined by capillary glass tube method (Wright's method). Puncture was made to the fingertip under aseptic conditions and stop watch was started as soon as the puncture was made. The end of the glass capillary tube was introduced into the blood so that blood enters the tube by capillary action. The capillary tube was held in the palm of the hand to maintain body temperature. Every 30 sec, about 1 cm of the tube was broken and time was noted. This was continued till the first fibrin thread appears. Clotting time was expressed in seconds [7,8].

Determination of total leucocyte count (TLC) and differential leucocyte count (DLC): TLC and DLC was determined manually by standard methods specified in the literature [7,8]

Ethical consideration: The present study was approved by institutional human ethical committee of Vishnu Dental College.

Data analysis: Data was analyzed by SPSS 20.0. Student t test was applied to observe the significance of difference between the groups. P value less than 0.05 was considered as significant.

RESULTS

Results were presented in table no 1. There was no significant difference in the age and weight and BMI among males and females. Height was significantly ($P<0.01$) higher in males. Bleeding time was not significantly different among males and females. Clotting time was significantly ($P<0.01$) higher in males when compared with females. Total leucocyte count was not significantly different among males and females. NLR was significantly ($P<0.05$) higher in males when compared with females.

Table 1: Demographic data, bleeding time, clotting time and total leucocyte count and neutrophil-lymphocyte ratio in male and female young adults.

Parameter	Males	Females	P value
Age (years)	18.43±0.20	18.68±1.02	0.5380
Height (cm)	170.86±3.91	161.86±1.09	0.0038**
Weight (kg)	59.43±2.78	55.46±1.53	0.2479
BMI (kg/m^2)	20.42±0.90	21.14±0.49	0.5116
Bleeding time (sec)	98.57±5.53	82±4.97	0.1306
Clotting time (sec)	180±14.64	139.29±6.21	0.0078**
Total leucocyte count (cells/cu.mm of blood)	8829.29±315.47	8975.07±223.02	0.7611
Neutrophil- lymphocyte ratio (NLR)	2.20±0.15	1.86±0.07	0.0432*

(Data was presented as mean and SEM. *P<0.05 is significant, **P<0.01 is significant,***P<0.001 is significant).

DISCUSSION

Thrombocytes play a key role in the blood coagulation mechanism. Bleeding time is a simple and cost effective test to assess the functions of the platelets. The common factors that effects bleeding time includes thrombocytes number and functioning, temperature, race, diet. It was reported that there may be gender variation in the bleeding time [11,12]. There was conflict regarding the effect of age on bleeding time as few studies reported that no significant difference in bleeding time among different age groups [13]. In contrast, another study reported that in elderly population bleeding time was shorter [16]. Similarly, the gender difference of bleeding time and clotting time also a topic of interest. It was reported that clotting time and bleeding time was higher in females when compared with males [9, 14, 15]. However, in a study conducted by Mahapatra et al, they didn't observed any difference in bleeding and clotting time among males and females [10]. Prabhakar et al, reported that clotting time was higher in males compared to females [17]. BJ Jain etal

reported that the platelet count was higher in females when compared to males [18]. In the current study, there was no significant difference in bleeding time among males and females. However, clotting time was significantly higher in males when compared with females (P<0.001). Neutrophil count was found to be higher in males in earlier studies [18].

The studies comparing the total leucocyte count among the genders are very less. In the current study, total leucocyte count was not significantly different among males and females. Neutrophils and lymphocytes are The ratio of neutrophils and lymphocytes (NLR) is associated with several diseases [19] like coronary heart diseases [20]. Further, the ratio of neutrophil and lymphocytes was reported to be an excellent predictor for recurrence of the disease [21]. In the current study, there was significantly higher NLR in males when compared with females.

Limitations: The major limitation if the study was having fewer males than females.

CONCLUSION

Significantly higher levels of clotting time and NLR were observed in males when compared with females. As NLR is associated with many physiological and pathological conditions, we recommend further detailed studies in this area to explore the diagnostic and prognostic role of NLR.

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