

# Anemia in Menstruating Women: Variation in Hematological Parameters in Different Phases of Menstrual Cycle

RB Kamal<sup>1</sup>, Abhilasha Mishra<sup>2,\*</sup>, Kavita Chawla<sup>1</sup>

## ABSTRACT

**Background and Aim:** Menstrual cycle is a physiological process that occurs in women. It is characterized by periodic vaginal bleeding due to shedding of uterine mucosa. This has its own effect on various systems and metabolisms. This study was attempted to understand the variations in hematological parameters in different phases of menstrual cycle. **Methods:** The present study was conducted on 55 healthy female volunteers within the age group of 18 to 24 years having regular menstrual cycle of  $28 \pm 3$  days from the female medical students of M. L. N. Medical College, Allahabad, U.P. We analyzed various hematological parameters on three different phases of menstrual cycle (Menstrual phase-2<sup>nd</sup> day, Proliferative phase -12<sup>th</sup> day and Secretory phase-22<sup>nd</sup> day). **Results:** Regarding hematological parameters, the mean TLC and PMN count increased with time i.e. highest in secretory phase followed by proliferative phase and least in menstrual phase. In contrast, mean Hb was highest in proliferative phase followed by secretory phase and menstrual phase. Conversely, mean eosinophils level was highest in menstrual phase followed by secretory phase and least in proliferative phase. **Conclusion:** This study was a moderate attempt to determine regular variations in some specific hematological parameters, during different phases of menstrual cycle in normal healthy females and evaluate various changes and their association with women's health. Eosinophil count increases in menstrual and premenstrual phase. In this phase exacerbation of menstrual asthma has also seen. **Key words:** Menstrual cycle, Hemoglobin, Anemia, Secretory phase.

## INTRODUCTION

Female reproductive phase comprises menstrual cycle which is an integral part of their lives. A normal menstrual cycle is dependent on the fluctuation in the ovarian hormones (estrogen and progesterone) during different phases of menstrual cycle—menstrual, proliferative and secretory phases. The characteristic rhythmic changes in the rate of secretion of ovarian hormones produce corresponding changes not only in the reproductive system but in other organ systems as well.

It has also been now recognized that important systemic as well as hematological changes are accompanying the various phases of menstrual cycle. It is suggested that stressful situations during ovulatory periods and menstruation may cause increased 17-hydroxy corticosterone levels with resulting eosinopenia.<sup>[1]</sup>

5–20% of women reporting severe dysmenorrhea may be associated with reproductive morbidities like infection. Thus, estimation of leucocyte count is an important tool. 9–14% of reproductive-aged women have blood loss that exceeds 80 mL<sup>2</sup> and prolonged and excessive bleeding may provoke or exacerbate anemia. Thus, there arises a need to estimate Hemoglobin, Red Blood Cell count and ESR during the menstrual cycle.

Ovarian hormones also alter the immune system like depression of the suppressor T cell activity). Human and animal studies suggest that there is a change in the distribution of immune cells during different phases of menstrual cycle. The ovulation phase of the menstrual cycle is the marker for fertilization and it may have unique hematological values. This study compares the hematologic values in the ovulation and luteal phases of the menstrual cycle. The aim of the present study is to find specific hematological parameters variability in different phases of menstrual cycle and to find any significant correlation between hematological parameters in reference to health of the menstruating women.

## MATERIALS AND METHODS

### Study Area

- The study was conducted in the Clinical Physiology lab of the Department of Physiology and Department of Pathology, Motilal Nehru Medical College, Allahabad over a period of one year (Aug 2014 to July 2015) after approval from institutional ethical committee.

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## Study Group

- This group included apparently healthy female medical students of Motilal Nehru Medical College, Allahabad between the ages of 18 to 24 years.

## Methodology

- The present study was carried in between (2014-2015) in healthy female undergraduate students' volunteers aged between 18-24 years having normal menstrual cycle of  $30 \pm 3$  days.
- The study protocol was explained to the subjects and oral and written informed consents were obtained.
- Prior to that all: age, height, weight were measured.
- A demonstration was given to the subject before performing the tests.
- The first sample was taken on 2<sup>nd</sup> day of onset of menstruation (Menstrual phase) Second sample during 12<sup>th</sup> day of Proliferative phase and the third sample was collected 22<sup>nd</sup> day during (Secretory Phase).
- All samples were taken at 10 am to avoid diurnal variations. The parameters analyzed were Hb, Total RBC count (Hayem's fluid) Total Leucocyte Count (Turk's fluid), Absolute Eosinophil Count (Pilot's fluid) and Differential Leucocyte Count (DLC) of neutrophils, Arneht Count by Tally bar method, eosinophils, basophils, lymphocytes, monocytes. Total Platelet count (1% ammonium oxalate) was done by hemocytometer. Bleeding time (Duke's method), clotting time (capillary glass tube method).

## Inclusion criteria

- This study included, healthy north Indian undergraduate Students population of age group 18-24 years having regular menstrual cycles.

## Exclusion criteria

- Contraceptive pills users, lactating women, H/O diabetes mellitus, Cases suffering from cardiovascular abnormalities, Psychiatric illness, Irregular periods, heavy Dysmenorrhoea, Oligomenorrhoea, Polymenorrhoea were excluded from the study.

## Statistical Analysis

Sample sizes were taken by Nonrandomized Judgmental Study and has been taken maximum possible number. Data collected from these tests were subjected to SSPS software analysis. Statistical analysis was done by Student's *t*-test. Comparison of data was done by using ANOVA. The *p* value of  $<0.05$  indicates statistically significant. The results were expressed as Mean  $\pm$  SEM (Standard error of mean).

Groups were compared by repeated measures one-way analysis of variance (ANOVA) and the significance of mean difference between the groups was done by Tukey *post hoc* test. A two-tailed ( $\alpha=2$ ) *p* value less than 0.05 ( $p<0.05$ ) was considered statistically significant. Analyses were performed on STATITICA (Windows version 6.0) software.

## RESULTS

### Hematological parameters changes

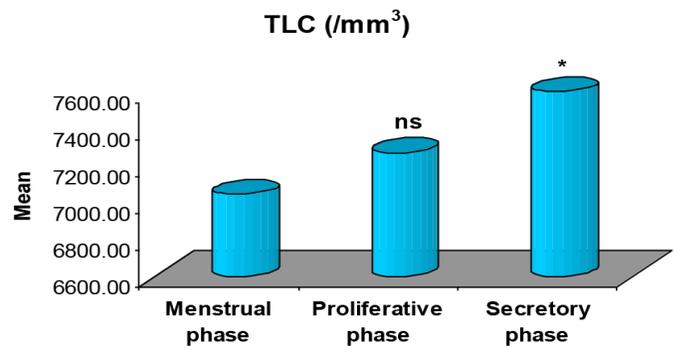
Like, pulmonary function test parameters, the mean levels of hematological parameters also varied between the periods. The mean TLC and PMN count as shown in Table 1 increase with time i.e. highest in secretory phase followed by proliferative phase and least in menstrual phase. Comparing the mean levels of each hematological parameter between the three groups, ANOVA revealed significantly ( $p<0.05$  or  $p<0.01$  or  $p<0.001$ ) different levels of all the parameters among the groups. The results are depicted in Table 2.

Further the mean TLC and PMN were significantly ( $p<0.05$  or  $p<0.01$  or  $p<0.001$ ) different and higher in secretory phase as compare to menstrual phase, shown in Figure 1, 2 and 3. In contrast, the mean Hb was significantly ( $p<0.001$ ) different and higher in proliferative phase as compared to both menstrual phase and secretory phase. Conversely, the mean Eosinophils was significantly ( $p<0.01$  or  $p<0.001$ ) different and higher in menstrual phase as compared to both proliferative phase and secretory phase and also higher significantly ( $p<0.05$ ) in secretory phase as compared to proliferative phase.

## DISCUSSION

Periodical change occurs in anatomical architect and hormonal fluctuation during menstrual cycle in females. It is also very much known to have a fluctuation in their hormone concentration. These hormones influence autonomic and metabolic activities.

In the present study mean levels of all the hematological parameters also varied between the periods. The mean TLC and PMN increase with time as suggested by Figure 1 and Figure 2 i.e. highest in secretory phase followed by proliferative phase and least in menstrual phase. Conversely, mean eosinophils level was highest in menstrual phase followed by secretory phase and least in proliferative phase



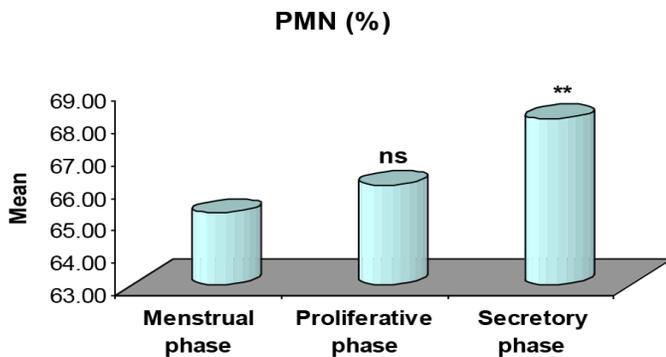
**Figur1:** Mean TLC at three different periods.  
<sup>ns</sup>*p*>0.05 or <sup>\*</sup>*p*<0.05- as compared to Menstrual phase.

**Table 1:** Hematological parameter levels (Mean  $\pm$  SD, *n*=55) at three different phases of menstrual cycle in undergraduate female students.

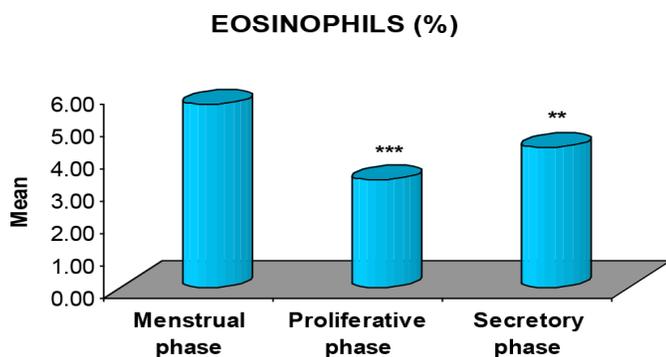
Parameters	Menstrual phase	Proliferative phase	Secretory phase	F value (2,162 DF)	<i>p</i> value
TLC (/mm <sup>3</sup> )	7045 $\pm$ 1026	7271 $\pm$ 1067	7600 $\pm$ 1333	3.23	0.042
PMN (%)	65.20 $\pm$ 3.83	66.07 $\pm$ 2.87	68.11 $\pm$ 7.29	4.84	0.009
Eosinophils (%)	5.64 $\pm$ 2.10	3.31 $\pm$ 1.67	4.31 $\pm$ 2.30	18.01	<0.001

**Table 2:** For each parameter, comparison ( $p$  value) of mean difference between the groups by Tukey test.

Comparisons	TLC	PMN	Eosinophils
Menstrual phase vs. Proliferative phase	0.559	0.634	<0.001
Menstrual phase vs. Secretory phase	0.031	0.007	0.002
Proliferative phase vs. Secretory phase	0.291	0.085	0.027

**Figure 2:** Mean PMN at three different periods.

<sup>ns</sup> $p > 0.05$  or <sup>\*\*</sup> $p < 0.01$  - as compared to Menstrual phase.

**Figure 3:** Mean Eosinophils at three different periods.

<sup>\*\*</sup> $p < 0.01$  or <sup>\*\*\*</sup> $p < 0.001$  - as compared to Menstrual phase.

In our present study there is an increase in neutrophil count percentage values during Secretory phase when compared to other phases, this changes is due to hormonal imbalance in ovary.<sup>[3]</sup> Some studies suggested that estrogen increases granular proliferation.<sup>[4]</sup> Increase in 17 beta estradiol concentration in circulatory pool of blood, in Secretory phase promotes granulopoiesis, which in turn increases the granulocyte numbers.

Several studies are in general agreement with the fact that the leucocyte count increased during the follicular phase and decreased during luteal

phase.<sup>[5]</sup> Although some exhibit a mild increase in total leucocyte count around follicular, yet no significant changes were observed. The changes in the circulating leucocyte count during the menstrual cycle is associated with the presumptive changes in blood estrogen and the possibility that they are influenced by blood gonadotrophic hormone at the time of ovulation, or by blood progesterone or body temperature during the latter half of the cycle, cannot be excluded. These observations are in contrary to some studies<sup>[6,7]</sup> in which they did not find any change in the number of circulating leucocytes in relation to the menstrual cycle.

## CONCLUSION

In the present study results, there is seen an increase in neutrophil count in secretory phase of menstrual cycle when compared to proliferative phase. It is also proved in few studies with increase in 17 beta estradiol concentrations in Secretory phase promotes granulopoiesis which in turn increases the WBC count. Eosinopenia during mid-cycle (14<sup>th</sup> day) is a result of physiologic stress and one might expect a simultaneous increase in levels of steroid hormones in blood.

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## CONFLICT OF INTEREST

The authors declares that there are no conflict of interest.

## ABBREVIATIONS

WBC: White blood cell; TLC: Total leucocyte count; PMN: Polymorphonuclear cells; DF: Degree of freedom.

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