

Comparison of Hematological Profile of Athletes in Various Phases of Menstrual Cycle

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ABSTRACT

The purpose of the study was to compare the hematological parameters during the different phases of the menstrual cycle viz Menstrual, Follicular, Ovulation, and Luteal Phase. Thirty athletes were selected as subjects for the study who were involved in speed-endurance dominated sports. Blood samples were taken in each phase and analyzed for the following hematological variables - Red blood cells (RBCs), White blood cells (WBCs), Hemoglobin (Hb), and platelets. The results of the study showed variations in hematological parameters throughout the menstrual cycle. RBC and Hb levels significantly increased during the follicular phase (FP) but remain unchanged in the ovulation and luteal phase. WBC levels also increased during the FP, were almost similar in ovulation phase, and slightly decreased in the luteal phase. Platelet count followed a similar pattern as WBC. Platelet count increased in the FP but decreased in the luteal phase. Menstrual phase is characterized by hemorrhage which results in blood loss and decreased levels of hematological parameters. FP is seen as compensatory phase in which the hematological variables significantly increase. WBC and Platelet count decrease in the luteal phase in attributed to the decrease in the estrogen levels. The study concluded that variations in hematological parameters is observed during the different phases of the menstrual cycle.

Keywords: Follicular, Hematological, Luteal, Menstrual, Ovulation

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INTRODUCTION

Menstruation is the cyclical and rhythmic shift that occurs in a sexually mature female's reproductive life.^[1] When fertilization fails, it entails the deconstruction and loss of the endometrium that has been meticulously prepared for the growth of a fertilized ovum. Menstrual bleeding, also known as the ovarian cycle, is the regular outflow of blood from the vaginal canal because of the uterine lining being released.^[2] When fertilization does not occur, the endometrial lining is shed monthly in a mature female. The cycle lasts about 28 days on average.^[3] In normal women, this can take up to 45 days. Estrogen, which stimulates the follicular phase (FP), and progesterone, which promotes the luteal phase, are both involved in this process acts on the uterus to prepare it for the fertilized ovum's implantation. When fertilization does not occur, these hormones decrease, and the uterine lining becomes irritated. The lining then sloughs away, resulting in menstrual flow.^[4] The cycle is measured from the start day of one menstrual period to the 1st day of the next menstrual period. The menstrual cycle is characterized by cyclical changes in the levels of FSH, LH, estrogen, and progesterone.^[5]

The menstrual cycle is frequently described in terms of particular milestones and occurrences that denote different stages. The initiation of blood flow (menses), which also signals the start of the FP, is the 1st day of the ovarian cycle.^[5] The luteal phase begins after ovulation, which occurs around halfway through the menstrual cycle. These two stages are frequently differentiated into early, mid, and late follicular, as well as early, mid, and late luteal, based on estradiol and progesterone levels.^[6] These oscillations can have an impact on hematological parameters. Platelet function, for example, is altered on a regular basis during the ovarian cycle due to the influence of progesterone and estrogen on factor concentrations.^[7]

Data on Red blood cell (RBC), White blood cell (WBC), and Hemoglobin (Hb) in male and female cynomolgus macaques, capuchin macaques, and people show that females have

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significantly lower RBC, HCT, and Hb levels than males, which is thought to be due to menstruation-related blood loss.^[8] Prolonged and heavy bleeding results in increased blood loss (>80 ml), which may induce anemia, necessitating the estimation of RBCs, hemoglobin, and erythrocyte sedimentation rate.^[9] In nonhuman primates and women, blood loss during menstruation has been proposed as one of the reasons of iron deficiency anemia. Menstruation may affect RBC mass characteristics as well as serum total protein, albumin, and globulins in preclinical.^[10,11]

When it comes to menstruation and sports, some women experience a decline in physical capacity throughout the menstrual cycle, although olympic medal-winning performances have occurred at all stages of the menstrual cycle.^[12,13] Exercise has been reported to reduce period cramps due to better circulation to the uterus, but it has not been demonstrated to cure or worsen uncomfortable menstruation.^[14] Increased quantities of endorphins produced after extended physical activity, which may counteract pain, might possibly be linked to a reduction in menstrual cramps.^[15] Some women claim that exercising on a regular basis helped them feel better.^[16]

The biological relevance of oxygen transport by Hb is clearly demonstrated by anemia, in which lowered Hb also reduces exercise performance despite a compensatory increase in cardiac output^[17,18] and by enhanced aerobic performance when total Hb is increased.^[18] The quantity, size, and hemoglobin concentration of athletes' RBCs are of particular interest in hematology. This curiosity is unsurprising given the critical function in oxygen supply to tissues and, ultimately, exercise performance. However, as the body's defenders against infection, WBCs indirectly contribute to performance by keeping athletes well enough (infection-free) to continue with their training regimens.^[19]

WBC levels in top athletes have gotten little attention, despite the fact that this information is often collected at the same time and from the same blood sample as red cell information. Previous studies of WBCs in athlete populations only looked at overall WBCs.^[20,18] One of the kinds of WBCs from a single sport.^[21] The role of RBCs to buffering fluctuations in blood pH via CO₂ transport and H⁺ binding to Hb is most likely the most important. RBCs also absorb lactate, a chemical generated by skeletal muscle cells during high-intensity exercise. Metabolite uptake into RBCs lowers plasma concentrations. Finally, RBCs appear to be capable of lowering peripheral vascular resistance by producing the vasodilator NO^[22] as well as ATP, which increases endothelial NO production, resulting in arteriolar vasodilation and increased local blood flow.^[23] Platelets play a critical role in the development and progression of cardiovascular disorders.^[24] Two prior research on platelet function in female participants at rest found contradictory results regarding the influence of menstrual phases (MPs).^[25,26] Despite the fact that both investigations reported modest changes in platelet count, volume, and aggregation, one research discovered that plasma levels of 3-thromboglobulin and platelet factor 4 (indicators of platelet release response) rise during ovulation and menstruation.^[27,28]

Research conducted in the field of menstrual cycle focusing on the athletic population is not much and those conducted have not considered all hematological elements. Furthermore, numerous research have been conducted to investigate the changes in various types of blood cell counts and hormonal profile in the menstrual cycle, but the results have been inconsistent and conflicting. Researcher also noticed that studies have not focused on athletes who participate in sports dominated by speed endurance specifically. So, the purpose of the current study is to examine the hematological parameters during the menstrual cycle's follicular, ovulation, luteal, and menstruation phases focusing on athletes who participate in sports dominated by speed endurance.

MATERIALS AND METHODS

Site of Study

The study was conducted in the Health Center of Lakshmbai National Institute of Physical Education, Gwalior, Madhya Pradesh, India.

Study Duration

The study is carried out for 6 months, that is, from August 2021 to January 2022 in the LNIPE.

Selection of Subjects

The current study was conducted on 30 healthy young female speed and endurance athlete students from Lakshmbai National Institute of Physical Education, Gwalior, and Jiwaji University, Gwalior, aged 18–25 years, with a typical menstrual cycle of 28–30 days. Participant who took part in this study provided written informed consent. The research comprised women who had a normal menstrual cycle of 27–30 days in the past. Women having irregular menstrual cycles, those using any medicine or hormonal preparation, those suffering from any physical ailment, and those with a history of endocrine problems, bleeding disorders, or heavy bleeding during MP were all excluded from the research. The study's goal and procedures were properly described to all of them in order to get their full participation, and their calm mental state was required for suitable findings.

Data Collection

The following protocol was used for data collection - Participants were asked to record the Previous menstrual cycle by the help of Flo App. Data collection was done after the morning meal of the participants. After the participant reported at the pathology laboratory and they were instructed to rest for 30 min after which 35 mL of blood was collected from the forearm vein (medial cubital vein) which was analyzed for hematological parameters and platelet function baseline data. As an anticoagulant, sodium citrate was utilized. In a span of 3 months data was collected by the same measure. Subjects were instructed to visit the center during each phase of the menstrual cycle, i.e., 1st–2nd day of MP, 8th–10th day of FP, 14th–16th of Ovulation phase, and 21th–24th day of Luteal Phase. During each visit, the subjects' blood was collected and Analysed for various hematological parameters such as Hb, RBC count, WBC count, and platelet count.

Blood samples were drawn from the ante-cubital vein using a venepuncture under aseptic circumstances and placed in a specimen vial containing the anticoagulant Ethylene Diamine Tetraacetic Acid. The Swelab Alfa automated hematology analyzer was used to analyze blood samples within 1–2 h of collection. Electronic counters (Cell Dyn 100 and 400, Metertech) were used to determine the erythrocyte count, leukocyte count, platelet count, hematocrit, and hemoglobin concentration in venous blood.

Statistical Analysis

RBC, WBC, platelet, and Hb indices were measured during the menstrual cycle's Menstruation phase, FP, ovulation phase, and luteal phase, and statistical analysis was performed using the *t*-test $P < 0.05$ was considered significant.

RESULTS

Mean value and Standard deviation of Hematological variables in different phases of menstrual cycle is show in the Table 1.

Table 1: Mean and standard deviation of hematological variable in different phases of menstrual cycle

Variable	Menstrual	Follicular	Ovulation	Luteal
RBC	4.12±0.77	4.20±0.78	4.23±0.78	4.21±0.77
WBC	8.24±1.50	8.30±1.50	8.32±1.51	8.31±1.48
Platelets	252.3±71.38	265.56±72.58	263.46±60.42	258.7±71.25
Hemoglobin	11.32±0.96	11.69±0.91	12.48±0.90	12.47±0.95

As per the results shown in Table 2, there is a significant difference in the RBC level between Menstrual and FP ($P = 0.000$); while no significant difference exist between any other phases on the cycle ($P = 1.000$).

As per the results shown in Table 3, there is a significant difference in the WBC level between Menstrual and other three Phases ($P = 0.000$); likewise Significant difference was found in Ovulation and Luteal phase ($P = 0.000$); while no significant difference exists between Follicular and Ovulation ($P = 1.000$); and in Follicular and Luteal ($P = 1.000$).

As per the results shown in Table 4, there is significant difference in the Platelets level between Menstrual and Luteal Phases as $P < 0.05$ ($P = 0.000$); while no significant difference exists between any other phases on the cycle.

As per the results shown in Table 5, there is no significant difference in the Hb level between Menstrual and FPs ($P > 0.05$); while comparing the MP with Ovulation and Luteal phase Significant difference is found as the $P < 0.05$ ($P = 0.000$). Similarly, significant difference is found when comparing between Follicular with Ovulation and Luteal phase ($P = 0.000$); and ovulation and Luteal phase ($P = 0.000$).

Table 2: Pairwise comparison of RBC in different phases of menstrual cycle

Measure: RBC						
(I) Phases	(J) Phases	Mean Difference (I-J)	Std. Error	Sig.b	95% Confidence Interval for Difference ^b	
					Lower Bound	Upper Bound
1	2	-0.075*	0.002	0.000	-0.081	-0.068
	3	-0.101	0.193	1.000	-0.648	0.446
	4	-0.082	0.193	1.000	-0.630	0.466
2	1	0.075*	0.002	0.000	0.068	0.081
	3	-0.026	0.193	1.000	-0.573	0.520
	4	-0.008	0.193	1.000	-0.555	0.539
3	1	0.101	0.193	1.000	-0.446	0.648
	2	0.026	0.193	1.000	-0.520	0.573
	4	0.019	0.021	1.000	-0.041	0.078
4	1	0.082	0.193	1.000	-0.466	0.630
	2	0.008	0.193	1.000	-0.539	0.555
	3	-0.019	0.021	1.000	-0.078	0.041

Based on estimated marginal means. *The mean difference is significant at the 0.05 level. ^bAdjustment for multiple comparisons: Bonferroni.

Table 3: Pairwise comparison of WBC in different phases of the menstrual cycle

Measure: WBC						
(I) Phases	(J) Phases	Mean Difference (I-J)	Std. Error	Sig.b	95% Confidence Interval for Difference ^b	
					Lower Bound	Upper Bound
1	2	-0.060*	0.003	0.000	-0.069	-0.051
	3	-0.078*	0.016	0.000	-0.124	-0.032
	4	-0.066*	0.017	0.003	-0.115	-0.018
2	1	0.060*	0.003	0.000	0.051	0.069
	3	-0.018	0.017	1.000	-0.065	0.029
	4	-0.006	0.018	1.000	-0.057	0.045
3	1	0.078*	0.016	0.000	0.032	0.124
	2	0.018	0.017	1.000	-0.029	0.065
	4	0.012	0.009	1.000	-0.013	0.036
4	1	0.066*	0.017	0.003	0.018	0.115
	2	0.006	0.018	1.000	-0.045	0.057
	3	-0.012	0.009	1.000	-0.036	0.013

Based on estimated marginal means. *The mean difference is significant at the 0.05 level. ^bAdjustment for multiple comparisons: Bonferroni.

Table 4: Pairwise comparison of platelets in different phases of menstrual cycle

Measure: Platelets						
(I) Phases	(J) Phases	Mean Difference (I-J)	Std. Error	Sig.b	95% Confidence Interval for Difference ^b	
					Lower Bound	Upper Bound
1	2	-13.267	5.829	0.182	-29.771	3.237
	3	-11.167	9.534	1.000	-38.163	15.830
	4	-6.467*	0.398	0.000	-7.592	-5.341
2	1	13.267	5.829	0.182	-3.237	29.771
	3	2.100	7.392	1.000	-18.831	23.031
	4	6.800	5.640	1.000	-9.171	22.771
3	1	11.167	9.534	1.000	-15.830	38.163
	2	-2.100	7.392	1.000	-23.031	18.831
	4	4.700	9.412	1.000	-21.951	31.351
4	1	6.467*	0.398	0.000	5.341	7.592
	2	-6.800	5.640	1.000	-22.771	9.171
	3	-4.700	9.412	1.000	-31.351	21.951

Based on estimated marginal means. *The mean difference is significant at the 0.05 level. ^bAdjustment for multiple comparisons: Bonferroni.

Table 5: Pairwise comparison of hemoglobin in different phases of menstrual cycle

Measure: HB						
(I) Phases	(J) Phases	Mean Difference (I-J)	Std. Error	Sig.b	95% Confidence Interval for Differenceb	
					Lower Bound	Upper Bound
1	2	-0.367*	0.047	1.000	-0.499	-0.234
	3	-1.160*	0.105	0.000	-1.457	-0.863
	4	-1.160*	0.105	0.000	-1.457	-0.863
2	1	0.367*	0.047	1.000	0.234	0.499
	3	-0.793*	0.103	0.000	-1.084	-0.502
	4	-0.793*	0.103	0.000	-1.084	-0.502
3	1	1.160*	0.105	0.000	0.863	1.457
	2	0.793*	0.103	1.000	0.502	1.084
	4	0.000	0.000	0.000	0.000	0.000
4	1	1.160*	0.105	0.000	0.863	1.457
	2	0.793*	0.103	0.000	0.502	1.084
	3	0.000	0.000	1.000	0.000	0.000

Based on estimated marginal means. *The mean difference is significant at the 0.05 level. ^bAdjustment for multiple comparisons: Bonferroni.

DISCUSSION

Thirty healthy young female speed-endurance athletes between the ages of 21 and 25 who meet the inclusion requirements were chosen. Hematological indices such as Hb, RBC count, WBC count, and platelet count were measured and analyzed in subjects during various phases of the menstrual cycle.

When comparing RBCs in all four stages among the various phases, the findings revealed that there is significant difference in the RBC level between Menstrual and FP; while no significant difference exists between any other phases on the cycle. RBC count significantly increased in the FP. But there was no further increase in RBC levels during other three phases. Similar results were found in case of Hb and similar of Hb variation was observed during the whole menstrual cycle. The MP is characterized by huge amount of bleeding which leads to decrease in the RBC and Hb levels. This period of hemorrhage is followed by FP in which the body attempts to compensate the loss during the MP. As a result of this, there is increase in the RBC and Hb levels which is seen in FP, which almost remains unchanged during the ovulation phase but slightly decreases during the end the menstrual cycle.^[29]

Similarly, when WBCs were compared throughout menstrual cycles, it was discovered there is significant difference in the WBC level between Menstrual and other three Phases; likewise, significant difference was found in Ovulation and Luteal phase; while no significant difference exist between the Follicular and Ovulation, Similarly no significance was found in Follicular and Luteal. WBC levels increased during the FP and remain almost unchanged in the later phases of menstrual cycle. The reason for this increase is attributed to the increase in estrogen levels during the FP. Estrogen stimulated the increased production of granulocytes and release of neutrophils.^[30] These findings are consistent with the results of previously conducted studies.^[31]

On the basis of a statistical comparison of platelet counts throughout all stages. The result revealed that the platelet count was On the basis of a statistical comparison of platelet counts throughout all stages. The result revealed there is a significant difference in the Platelets level between Menstrual and Luteal Phases; while no significant difference exists between any other phases on the cycle. Platelet count increased in the FP and remains unchanged during the ovulation phase. The Luteal phase is marked with decrease in the platelet count. The cause for increase in platelet count in FP is due to increased megakaryocytes boosting by estrogen to promote platelet formation. Since level of estrogen

is found increased during the FP which in turn stimulates platelet production. But at the end of the menstrual cycle, when the ovum did not get fertilized, the estrogen levels again decrease which leads to decrease in platelet count.

CONCLUSION

The purpose of the present study was to compare the selected hematological variables i.e. RBCs, WBCs, Hb, and Platelets in different phases of menstrual cycle - menstrual, follicular, ovulation, and luteal phase. The findings of the study revealed that there is significant increase in all the hematological parameters in the FP. There is blood loss in the MP and it results in decrease in the hematological variables. RBC and Hb increase in the FP and remain almost unchanged in the later phases of the menstrual cycle. WBC and Platelet count also increase in the FP, remains constant in the ovulation phase but decrease at the end of the luteal phase.

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