

Haemorrhological Properties of Blood in Phases of Menstrual Cycle in Female Undergraduate Students in Port Harcourt, Nigeria

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Abstract

Background: Blood flow pattern among various women in different location of the world had been noted to vary significantly with some anthropometric variations, geospatial locations, age, BMI and cardiovascular parameters. As a result, this study therefore investigated the flow properties of blood during menstrual cycle in female undergraduate students in Port Harcourt as a baseline study with a view of establishing local reference data for Port Harcourt, Nigeria.

Materials and Methods: 125 volunteers were recruited into the study using stratified random sampling techniques and were administered well-structured questionnaires. Their blood samples were collected and haemorrhological properties (Whole Blood Viscosity, Plasma Viscosity, ESR and haematocrits) were analysed against various phases of menstrual cycle namely: menstruation, follicular, ovulation and luteal phases among female undergraduate students in Port Harcourt.

Results and Discussions: Result of the study when compared with the WHO standard references showed a significant ($p \leq 0.05$) difference in ESR, PV, WBV and ESR for menstruation and follicular phases while HCT was not significant with ovulation and luteal phases. Flow properties was also significant ($p \leq 0.05$) with ages (16-23, 24-30 and ≥ 31) years as compared with the WHO Control. BMI and blood pressure was also significant ($p \leq 0.05$) with Haemorrhological properties of blood. **Conclusion:** The study therefore concluded that there is a significant ($p \leq 0.05$) correlation between flow properties of blood and phases of menstrual cycle among university students in Port Harcourt, Nigeria as compared with the WHO standard values.

Keywords: HCT, ESR, PV, WBV.

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INTRODUCTION

Background of the Study

Understanding menstrual cycle is paramount in women's health care and the flow properties of blood will help show the fluid dynamics of blood during the menstrual cycle.

Biorheology is the branch of medical sciences that studies the flow and deformation of biological materials under the influence of constraints applied to it. The part of biorheology focusing on blood is termed haemorrhology. Its purpose is therefore to study the flow of the blood in interaction with its surrounding environment in both macro- and micro circulation

(Karsheva, *et al.*, 2009). Blood is composed of both the cellular and liquid part. The cellular part includes red blood cells or erythrocytes, white blood cells or leukocytes and the platelets also known as thrombocytes. The liquid portion of blood is also called Plasma; it is a pale-yellow fluid that constitutes up to 55% of the blood's volume. It is made up of water, organic and inorganic substances. Cellular components contained in plasma, an aqueous solution including organic compounds, proteins, and salts make up whole blood, a two-phase liquid. The fluid properties of blood can be derived from understanding the volume, viscosity and specific gravity of blood and its constituents (El-sayed, *et al.*, 2012).

Appropriate tissue perfusion can only take place when bloods rheological characteristics fall within a certain range. Changes to these characteristics are crucial to disease processes (Baskurt and Meiselman, 2003). The haematocrit is one of the most important characteristics of blood as it shows the individual cellular volumes and properties as opposed to the total blood volume. It gives valuable information about the changes in the number and dimensions of red blood cells as well as the volume of plasma. Increase and decrease in the haematocrit value has significant meaning (Karsheva, *et al.*, 2009).

Blood viscosity is an important determinant of local flow characteristics, which exhibits shear thinning behaviour decreasing with increasing shear rates. Plasma viscosity, haematocrit (volume fraction of red blood cells, which make up 99% of the cellular constituents) and the mechanical characteristics of red blood cells all contribute to blood viscosity (Baskurt and Meiselman, 2003) The ability of RBC's to deform is crucial for blood flow in the microcirculation, but RBC deformability also influences blood flow in the microcirculation because it impacts blood viscosity.

Blood viscosity value is said to be between 3.5 and 5.5 cP to be normal. However, blood viscosity cannot be summarized in to one value and this is because of the shear thinning property of blood which is dependent on RBC's rheological properties causing the viscosity of the liquid to change depending on the hemodynamic conditions. This means blood viscosity is different in the large arteries, the veins and microcirculation where the shear rate varies from a few hundreds to thousands (El-sayed, *et al.*, 2012).

Blood viscosity depends on several factors: haematocrit, plasma viscosity, the ability of RBCs to deform under flow and the RBC aggregation-disaggregation properties. In fact, any one or more of these parameters may have varied effects on blood viscosity (Karsheva, *et al.*, 2009).

Findings had revealed a significant absence of local reference data on haemorrhological properties of blood in women in their respective menstrual phases. This absence had also significantly affected medical research on flow properties of blood in different menstrual phases and consequentially led to the current over-reliance on the global WHO standards.

The haemorheological values obtained in this study will provide information that will aid health practitioners properly diagnose early related causes of physiological changes, infections and diseases especially those related to the menstrual cycle in undergraduate females in Port Harcourt. It will also set a standard reference value of haemorheological data in Port Harcourt and as comparable to the other part of the world.

This research is aimed at investigating Haemorrhological properties of blood in phases of menstrual cycle in female undergraduate students in Port Harcourt, Nigeria. in other to achieve this, the following were determined; the effect of Haemorrhological properties on different phases of menstrual cycle, the statistical relationship between age variations and flow properties of blood, the possible effect of BMI on biorrheology of blood in females students, the effect of cardiovascular parameters (blood pressure and pulse rate) on haemorrhology of female students. Possible statistical relationship between nutrition and flow properties of blood were also determined and the findings from the study were compared with the global WHO Reference values.

MATERIALS AND METHODS

Study Area

Menstruating female students in the 3 universities in Rivers State namely University of Port Harcourt, Rivers State University and Ignatius Ajuru University of Education, all in Port Harcourt, Rivers State, Nigeria.

Bench Work/Testing: The Bench Work will be done in the Laboratory Unit of the Department of Human Physiology Department, Faculty of Basic Medical Sciences, College of Medical Sciences, Rivers State University, Port Harcourt.

Population of the Study

The Population of this study comprised of apparently healthy female students between the age of 16 to 45 years from three public universities in Rivers State, namely, University of Port Harcourt, Rivers State University and Ignatius Ajuru University of Education. Students below the age of 16 and those above the age of 45 were excluded from this study. Also sick students were also excluded from this study.

STUDY DESIGN

Table 1: Table showing summary of study design

Groups/ Variables	Sub-Divisions (N=120)	HCT (%)	ESR (mm/hr)	PV (mPa.s)	WBV
Group 1 (WHO Reference)	WHO Reference				
Group 2 (Phases of Menstrual Cycle)	Menstruation (Day 1-4)				
	Follicular (Day 1-13)				
	Ovulation (Day 14)				
	Luteal (Day 15-28)				

Groups/ Variables	Sub-Divisions (N=120)	HCT (%)	ESR (mm/hr)	PV (mPa.s)	WBV
Group 3 (Age variations)	16-23 years				
	24-30 years				
	≥31 years				
Group 4 (BMI)	≤18.4 (underweight)				
	18.5-24.9 (Normal)				
	25.0-29.9 (Pre-obessed)				
	30.0-34.9 (Obessed Class 1)				
	35.0-39.9 (Obessed Class 2)				
Group 5 (Blood Pressure in mmHg)	≥40 (morbid Obesity)				
	89-70/60-40 (Low BP)				
	120-90/80-61 (Normal)				
	130-121/90-81 (Elevated)				
	140-131/90 (Pre-hypertension)				
	≥141/≥91 (hypertension)				

Table 1 above is the summary of design on how the study will be carried out. A total of 120 volunteers as participants will be recruited into the study. They shall comprise of 30 non-menstruating healthy university females and 90 menstruating university female students. The study will be of 5 groups. Group 1 will be the control and having 30 non-menstruating healthy females. They will be used to compare with the test group. Group 2 to group 5 will be the experimental groups. Group 2 will be examining age differences and blood flow properties. Group 3 will be comparing different BMI with different flow properties of blood in healthy menstruating female students. Group 4 will be investigating possible relationship between those with high blood pressure and flow properties of blood in healthy menstruating female students while Group 5 will investigate possible effect of nutrition on flow properties of blood.

Sample and Sampling Techniques

The sample size comprise of 125 volunteers of which 29 were menstruating, 39 were in their follicular phase, 19 were ovulating and 39 luteal. The Control group was the WHO standard values. The experimental group was university girls comprising of both menstruating and non-menstruating and not sick of any pathos except a few who will be hypertensive. The sampling techniques deployed was stratified non-probability sampling techniques with consent of participants sought in accordance with the ethical provision of the College of Medical Sciences, Rivers State University

Ethical Consideration/Approval

The proposal of this research was submitted to the Ethical Committee of the Faculty of Basic Medical Sciences, Rivers State University for approval. After the

approval was granted, a Letter of Introduction was taken from my Head of Department, Human Physiology and taken to field. Consent of participants was sought after while presenting the letter of Introduction. All procedures were done in consonant with the ethical standards of the College of Medical Sciences, Rivers State University for use of human volunteers in research. The ethical clearance is presented at Appendix III on page 73 of this research.

Data/Statistical Analysis

The study utilized descriptive statistical test. The descriptive statistics was used to derive Means (\bar{x}) and Standard Error of Means (SEM) for all variables including the relationships between menstrual cycle group, Age Group, BMI, Blood Pressure, and nutrition of analysed variables of Haemorrhological Parameters. Result figures was expressed in Mean \pm SEM with N=125. The P-Value represents statistics that are significant upon comparison with the control which is also a primary data from the non-menstruating female students. The mean difference was significant when less than 0.05 ρ -level ($\rho < 0.05$). All data obtained from the study was analysed using Statistical Package for Social Sciences (SPSS) version 22.0.

RESULTS

Result and Analysis

The finding from this research is being represented in this chapter. The analysis includes the statistical representation of the flow properties of blood on menstrual cycle of female students in Port Harcourt. This chapter contains comparison with the WHO standards and significant discussions on the findings.

Table 2: Table of result for flow properties of blood on menstrual cycle against the WHO standards

Menstrual Cycle Phases	N (N=125)	HCT (%)	ESR (mm/hr)	PV (mPa.s)	WBV
WHO Female Reference		42.5 ± 5.5	10.00 ± 10.00	1.61±0.11	4.50 ± 1.00
Menstruation (Day 1-5)	29	34.55±3.22*	15.07±4.22	1.51±0.02*	3.10±0.10*
Follicular (Day 1-13)	39	35.61±4.32*	14.32±4.33	1.52±0.10*	3.21±0.00*
Ovulation (Day 14)	19	38.72±3.33	10.21±3.12	1.55±0.11*	3.70±0.22*
Luteal (Day 15-28)	38	40.67±4.77	8.37±3.22*	1.56±0.13*	3.78±0.14*

Result figures are expressed in Mean ± SEM, n = 125. The mean difference is significant (p) when less than 0.05 level, (p < 0.005)

Table 2 shows the statistical analysis of the flow properties of blood and stages of menstrual cycle relationship. Group 1 represents the WHO standard which is the control of this study while Group 2 represents the results from the experiments. Group 2 has been sub divided into four representing the various stage in the menstrual cycle the volunteers were in during collection of blood samples. The result of Group 2 is compared with that of Group1 (control).

From Table 2 Group 1 and the various parameters for the flow properties of blood which was studied using HCT, ESR, PV and WBV are 42.5 ± 5.5, 10.00 ± 10.00, 1.61±0.11, 4.50 ± 1.00 respectively.

Group 2 which was grouped into 4 representing the phases of menstrual cycle of the participants; The HCT, ESR, PV and WBV statistical value of those in menstruation phase is 34.55±3.22*,15.07±4.22, 1.51±0.02*, 3.10±0.10* respectively.

The subjects in follicular phase showed statistical value of 35.61±4.32, 14.32±4.33, 1.52±0.10*,3.21±0.00* for the flow properties parameter studied. The subjects in ovulation phase showed statistical value of 38.72±3.33, 10.21±3.12, 1.55±0.11*, 3.70±0.22*. The subjects for luteal phase showed statistical value of 3.70±0.22*, 3.70±0.22*, 1.56±0.13* and 3.78±0.14*

Table 3: Table of result for age difference and flow properties of blood on menstrual cycle

Age Groups	N (N = 125)	HCT (%)	ESR (mm/hr)	PV (mPa.s)	WBV
WHO Female Reference		42.5 ± 5.5	10.00 ± 10.00	1.61 ± 0.11	4.50 ± 1.00
16-23 years	45	39.23±4.55	14.32±3.55	1.61±0.04	3.70±1.02
24-30 years	43	40.11±4.33	12.81±4.11	1.65±0.03	3.75±1.70
31-45 Years	37	38.67±5.21	13.32±3.12	1.60±0.10	3.55±1.77*

Result figures are expressed in Mean ± SEM, n = 125. The mean difference is significant (p) when less than 0.05 level, (p < 0.005)

Table 3 shows the statistical result of the flow properties and the age of the volunteers' relationship. Group 1 represents the WHO standard which is the control of this study while Group 3 represents the results from the experiments. Group 2 has been sub divided into three age groups. The result of Group 2 is compared with that of Group1 (control).

From the Table 3 Group 1 and the various parameters for the flow properties of blood which was studied using HCT, ESR, PV and WBV are 42.5 ± 5.5, 10.00 ± 10.00, 1.61±0.11, 4.50 ± 1.00 respectively.

Group 3 which represents the age group of the subject has statistical values grouped into; The HCT, ESR, PV and WBV statistical value of those in 16-23years is 39.23±4.55, 14.32±3.55, 1.61±0.04 and 3.70±1.02 respectively. The subjects in 24-30 showed statistical value of 40.11±4.33, 12.81±4.11, 1.65±0.03 and 3.75±1.70 for the flow properties parameter studied.

The subjects in above 31 showed statistical value of 38.67±5.21, 13.32±3.12, 1.60±0.10 and 3.55±1.77* for the flow parameter studied respectively.

Table 4: Table of result for Body Mass Index and flow properties of blood on menstrual cycle

BMI Groups	N (N = 125)	HCT (%)	ESR (mm/hr)	PV (mPa.s)	WBV
WHO (Reference)		42.5 ± 5.5	10.00 ± 10.00	1.61 ± 0.11	4.50 ± 1.00
≤18.4 (underweight)	24	35.22±3.71*	11.37±4.47	1.58±0.10	3.87±1.65
18.5-24.9 (Normal)	28	39.41±4.56	9.71±6.34	1.61±0.05	3.80±1.70
25.0-29.9 (Pre-obese)	32	39.72±6.00	14.34±2.11	1.61±0.07	3.80±1.71
30.0-34.9 (Obese Class 1)	19	37.34±5.01	13.67±3.56	1.51.0.00	3.37±1.80
35.0-39.9 (Obese Class 2)	13	37.12±3.21	22.61±3.11*	1.49±0.00*	3.37±1.76
≥40 (Morbid Obesity)	9	36.76±4.01*	20.33±4.57*	1.34±0.17*	3.05±1.70*

Result figures are expressed in Mean ± SEM, n = 125. The mean difference is significant (p) when less than 0.05 level, (p < 0.005)

Table 4 shows the statistical result of the flow properties and body mass index of the subject's relationship. Group 1 represents the WHO standard which is the control of this study while the subjects are grouped based on their body mass index.

From the table the control group shows the various parameters for the flow properties of blood which was studied using HCT, ESR, PV and WBV are 42.5 ± 5.5 , 10.00 ± 10.00 , 1.61 ± 0.11 , 4.50 ± 1.00 respectively.

The statistical values for the underweight class include; $35.22 \pm 3.71^*$, 11.37 ± 4.47 , 1.58 ± 0.10 and

3.87 ± 1.65 . The statistical values for the normal weight class include; 39.41 ± 4.56 , 9.71 ± 6.34 , 1.61 ± 0.05 and 3.80 ± 1.70 . The statistical values for pre-obese class include; 39.72 ± 6.00 , 14.34 ± 2.11 , 1.61 ± 0.07 and 3.80 ± 1.71 . The statistical values for the Obese Class 1 include; 37.34 ± 5.01 , 13.67 ± 3.56 , 1.51 ± 0.00 and 3.37 ± 1.80 .

The statistical values for the Obese Class 2 include; 37.12 ± 3.21 , $22.61 \pm 3.11^*$, $1.49 \pm 0.00^*$ and 3.37 ± 1.76 . The statistical values for the morbid obesity class include; $36.76 \pm 4.01^*$, $20.33 \pm 4.57^*$, $1.34 \pm 0.17^*$ and $3.05 \pm 1.70^*$

Table 5: Table of result for Blood Pressure and flow properties of blood on menstrual cycle

Blood Pressure	N (N=125)	HCT (%)	ESR (mm/hr)	PV (mPa.s)	WBV
WHO Female Reference		42.5 ± 5.5	10.00 ± 10.00	1.61 ± 0.11	4.50 ± 1.00
89-70/60-40 (Low BP)	23	37.32 ± 6.77	13.56 ± 3.21	$1.52 \pm 0.40^*$	3.37 ± 1.74
120-90/80-61 (Normal)	56	39.33 ± 4.67	12.22 ± 2.12	1.69 ± 0.44	3.80 ± 1.60
130-121/90-81 (Elevated)	27	39.89 ± 3.33	9.89 ± 4.22	1.62 ± 0.32	3.80 ± 1.45
140-131/90 (Pre-hypertension)	12	$34.27 \pm 4.15^*$	$19.11 \pm 2.00^*$	$1.75 \pm 0.20^*$	$3.21 \pm 1.7^*0$
$\geq 141/\geq 91$ (hypertension)	7	$35.29 \pm 3.12^*$	$17.29 \pm 6.67^*$	1.69 ± 0.00	$3.30 \pm 1.70^*$

Result figures are expressed in Mean \pm SEM, n = 125. The mean difference is significant (ρ) when less than 0.05 level, ($\rho < 0.005$)

Table 5 shows the statistical result of the flow properties and the hypertension relationship. Group 1 represents the WHO standard (control) while the subjects are classified based on the level of their blood pressure.

From the table the control group being the WHO standard shows the various parameters for the flow properties of blood which was studied using HCT, ESR, PV and WBV are 42.5 ± 5.5 , 10.00 ± 10.00 , 1.61 ± 0.11 , 4.50 ± 1.00 respectively.

The subject samples were groups into four sub groups showing the different classes of blood pressure the subject is in.

The HCT, ESR, PV and WBV statistical value of those with low blood pressure is 37.32 ± 6.77 , 13.56 ± 3.21 , $1.52 \pm 0.40^*$, 3.37 ± 1.74 respectively. The subjects in 24-30 showed statistical value of 39.33 ± 4.67 , 12.22 ± 2.12 , 1.69 ± 0.44 , 3.80 ± 1.60 for the flow properties parameter studied.

The subjects in above 31 showed statistical value of $34.27 \pm 4.15^*$, $19.11 \pm 2.00^*$, $1.75 \pm 0.20^*$, $3.21 \pm 1.7^*0$ for the flow parameter studied respectively. The subjects in above 141/91 which is hypertension showed statistical value of $35.29 \pm 3.12^*$, $17.29 \pm 6.67^*$, 1.69 ± 0.00 , $3.30 \pm 1.70^*$ for the flow parameter studied respectively.

DISCUSSION OF FINDINGS

This study shows the flow properties of blood during menstrual cycle among female undergraduate

students. Using haematocrit, erythrocyte sedimentation rate, plasma viscosity and white blood count to ascertain the flow properties of blood and other parameters like blood pressure, body mass index and age as determinants. The significant differences showed in the statistical value will be used to study and understand further.

The internal friction of adjacent fluid layers moving past one another is a fundamental characteristic of fluids called viscosity. This internal friction contributes to the resistance to flow as described by Poiseuille's equation. The interactions between fluid layers are influenced by the fluid's chemical makeup and whether it is homogenous or heterogeneous in composition, having established that blood is a heterogeneous substance. That is, viscosity does not change with changes in velocity (Ofioritse, *et al.*, 2019).

Relationship between Phases of Menstrual Cycle and Haemorrhological Properties of Blood

The result obtained from Table 2 showed that compared result of the differences of the mean between the levels of menstrual cycle with the variables for flow properties which mean difference is significant at (ρ) 0.05 Level, showed that the WHO standard for the flow properties 42.5 ± 5.5 , 10.00 ± 10.00 , 1.61 ± 0.11 and 4.50 ± 1.00 was inconsistent throughout the menstrual cycle. The HCT variable shows significant increase as the cycle progresses from a low value of 34.55 ± 3.22 in the HCT value of those menstruating to 40.67 ± 4.77 for those in the luteal phase which is a closer value to the WHO standard value. The ESR reduced beyond the value of the

control in the luteal phase whereas the values in the menstruating, follicular and ovulation phase shows valuable increase than the value of the control group. The plasma viscosity and WBV values reducing below the control value throughout in all the phases of the menstrual cycle as it progresses.

This result suggest that flow properties is not the same throughout the cycle as the cycles involves physiological changes that may affect the flow properties differently at different intervals in the cycle.

Relationship between Age Variations and Haemorrhological Properties of Blood

The result obtained from Table 3 showed that compared result of the differences of the mean across age groups of the analytical variable of flow properties of blood parameters which mean difference is significant at (ρ) 0.05 Level, showed that $3.55 \pm 1.77^*$ revealed significant differences of the white blood volume proved insignificantly reduced. This result suggests that WBV increase with age, when compared between age group 16-23 and 24-30 years which are slight increased.

Effect of Body Mass Index on the Haemorrhological Properties of Blood

The result obtained from Table 4 showed the comparative difference between the body mass index group and analysed variables of the flow properties of blood. Comparing the result of the differences of the flow properties of the body mass index with that of the control it revealed significant differences in the HCT of the underweight and morbid obesity with analytical variable of 35.22 ± 3.71 and 36.76 ± 4.01 . There are also significant differences in the ESR and plasma viscosity of the obese class I. The flow properties of the morbid obesity class all showed interesting significant difference in all the flow properties of this class compared to the control group. This proved that the flow of blood can be affected especially the underweight and the morbid obesity class.

Relationship Cardiovascular Parameters on the Haemorrhological Properties of Blood

The result obtained from Table 5 showed that compared result of the differences of the mean between the levels of blood pressure with the variables for flow properties which mean difference is significant at (ρ) 0.05 Level, showed that the WHO standard for the flow properties 42.5 ± 5.5 , 10.00 ± 10.00 , 1.61 ± 0.11 and 4.50 ± 1.00 was not seen in any of the classes of blood pressure recorded, with significant differences in all the group's flow properties parameters. This result suggest that flow properties are has different parameters depending on the blood pressure.

CONCLUSION

After series of sample collection and issuing of questionnaire the blood samples were analysed using various methods of haemorrhological parameter procedures for their flow properties and the result obtained has been evaluated against the WHO standards, revealing significant difference in the flow parameters throughout the different levels of menstrual cycle.

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