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Original Article

Assessment of Iron Loss in Menstruating Female Students in a Nigerian University

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Abstract

Menstruation is a periodic physiological process of blood discharge in women of reproductive age. It is associated with normal adaptive variation in iron depletion, which could lead to pathological depletion of iron stores. This study assessed iron status before and after menstruation among menstruating female students. A longitudinal and paired comparative study was conducted in 42 female students. A structured questionnaire was administered, and blood samples were collected pre- and post-menstruation. A semi-automated analyzer (Chem 535) was used to measure serum iron (SI) and total iron-binding capacity (TIBC), while transferrin saturation (TSAT) was calculated. Data were analyzed according to age, duration of menstrual flow, and degree of menstrual flow, with $p < 0.05$ considered significant. Serum iron recorded a significant decrease from 19.41 ± μmol/L before menstruation to 16.14 μmol/L after menstruation ($p = 0.001$), while TIBC recorded a significant increase from 30.81 μmol/L pre-menstruation to 36.04 μmol/L post-menstruation ($p = 0.001$). TSAT declined significantly from 66.67% pre-menstruation to 48.44% post-menstruation ($p = 0.001$). Although all age groups showed reductions in serum iron and TSAT and increases in TIBC following menstruation, the differences between age groups were not statistically significant (serum iron, $p = 0.078$; TIBC, $p = 0.150$; TSAT, $p = 0.082$). Menstruation was associated with an acute reduction in serum iron and transferrin saturation, with increased total iron-binding capacity, indicating measurable acute iron loss among menstruating female students. The duration and degree of menstrual flow did not influence the extent of these changes.

Keywords: Menstruation, Serum Iron, Total Iron-Binding Capacity, Transferrin Saturation.

1. Introduction

Women of reproductive age are prone to physiological iron fluctuation because of menstrual blood loss (Munro et al., 2023). The body loses both blood and iron during menstruation, and the amount of iron lost varies greatly based on the length and volume of menstrual flow (Petraglia & Dolmans, 2022). Women typically lose 20-40 mL of blood during a typical monthly cycle, which is equivalent to 10-20 mg of iron. However, women who experience excessive menstrual bleeding may lose significantly more. Iron stores gradually run out when these losses are not sufficiently replenished by diet or supplements, which first causes iron deficiency and ultimately iron deficiency anaemia (MacLean et al., 2023).

Numerous physiological activities, such as DNA synthesis, oxygen transport, cellular respiration, and immunological function, depend on iron, an essential trace metal (Petraglia & Dolmans, 2022). About two-thirds of the iron in the body is stored as ferritin and hemosiderin or incorporated into enzymes and myoglobin; the remaining iron is mostly stored as haemoglobin in red blood cells (Munro, 2023). Maintaining iron balance mostly depends on sufficient dietary absorption to make up for daily iron losses because the human body has few systems for controlling iron excretion (Mirza et al., 2018).

Women and teenage girls are disproportionately affected by iron deficiency, which is still one of the most common nutritional problems in the world. Millions of women of reproductive age suffer from physiological iron fluctuations that could result in iron deficiency, which is the primary cause of anaemia worldwide (Petraglia & Dolmans, 2022). Inadequate dietary iron consumption, recurring illnesses, parasite infestations, and monthly blood loss all contribute to the high prevalence of iron insufficiency in low- and middle-income nations, such as Nigeria. Because women frequently face higher nutritional needs, erratic eating patterns, academic stress, and restricted access to balanced diets, all of which may predispose them to lower iron stores female university students are especially vulnerable.

Reliable biochemical indicators are necessary for the assessment of iron status. Although it is affected by dietary intake and daily fluctuations, serum iron represents the amount of circulating iron bound to transferrin (Munro, 2023). The highest amount of iron that transferrin can bind is measured by total iron-binding capacity (TIBC), which typically rises as body iron stores decrease (Fernandez-Jimenez et al., 2020). An indication of the percentage of transferrin occupied by iron is provided by transferrin saturation (TSAT), which is computed as the ratio of serum iron to TIBC reported as a percentage (Munro, 2023). The body loses both blood and iron during menstruation, and the amount of iron lost varies greatly based on the length and volume of menstrual flow (Fernandez-Jimenez et al., 2020). Prolonged or heavy menstrual bleeding has been associated with reduced serum iron concentrations, decreased transferrin saturation, elevated TIBC, and an increased risk of iron deficiency anaemia, according to several studies (Munro, 2023).

However, only a small number of studies have examined the acute biochemical changes that occur before and after menstruation in seemingly healthy female university students; the majority of studies that are now available have concentrated on women with clinically recognised heavy menstrual bleeding or established iron deficiency (Mansour et al., 2020). Additionally, nothing is known about menstrual iron loss among North-Central Nigerian female students.

In Nigeria, iron deficiency in women of childbearing age remains an important public health concern (Harika et al., 2017). Despite this, routine assessment of iron status in female university students is uncommon, and many cases of early iron depletion remain undetected until anaemia develops (Almasmoum et al., 2023). A study by Lugos et al. (2019) found that postmenstruating female students had lower packed cell values. Low dietary diversity was linked to lower haemoglobin concentrations, and poor dietary consumption raised the risk of iron deficiency and anaemia, according to Asare et al. (2024) in Ghana. According to Eric et al. (2024) among female students, haemoglobin levels were substantially correlated with nutritional status (BMI). Understanding the effect of menstruation on iron indices may facilitate early identification of students at risk and inform preventive strategies such as nutritional counselling, dietary modification, and appropriate iron supplementation (MacLean et al., 2023).

Therefore, this study was undertaken to assess iron loss among menstruating female students of the Department of Medical Laboratory Science, University of Jos, North-Central

Nigeria, by comparing serum iron, total iron-binding capacity, and transferrin saturation before and after menstruation. The study also evaluated the influence of age, duration of menstrual flow, and degree of menstrual flow on these iron status parameters and to further evaluated acute, short-term iron depletion rather than chronic iron deficiency.

2. Methods

2.1. Study Area

The study was carried out in the Department of Medical Laboratory Science, University of Jos, Plateau State. Jos-Plateau is a city in Nigeria's middle belt and is the administrative capital of Plateau State, between latitude 9˚30′N & 10˚10′N, and longitude 8˚15′E & 9˚15′E; and its topography comprises chains of highlands of variable heights coupled with flatter topography. With an altitude of 4062feet above sea level, it enjoys a more temperate climate (average monthly temperatures range from 20°C to 25°C and annual rainfall approximately ranges between 1500-2000mm)". The vegetation area is tropical wood-lands characterized by tall grasses, shrubs and stunted trees (Veenendaal et al., 2014). University of Jos is located in Jos North Local Government Area of Plateau State. It is one of the seventeen local governments of the state, and it shares boundaries with Jos East and Jos South. The people in the state area are mostly students, civil servants, businessmen and women.

2.2. Study Population

This study comprised female students of the Medical Laboratory Department, University of Jos, Plateau State.

2.3. Determination of Sample Size

The sample size was calculated using Thrusfield (2007)

$$n = \frac{1.96^2 \times P_{exp}(1 - P_{exp})}{d^2}$$

Where:
n= minimum sample size
P= Expected prevalence (2%) (Paul et al., 2016)
d= Desired absolute precision of 5% (0.05)

$$n = \frac{1.96^2 \times 0.02(1 - 0.02)}{0.05^2}$$

$$n = \frac{3.8416 \times 0.02\ (0.98)}{0.0025} = 30.11$$

$$n = 30$$

Minimum sample size = 30. Therefore, a sample size of 40 subjects was considered. And since samples were collected pre- and post-menstruation, a total of 80 samples were collected.

2.4. Study Design

This is a cohort study design with purposeful sampling. The study involved the Assessment of iron status amongst Females of the Medical Laboratory Science Department; University of Jos. Overnight fasting samples were collected between 8 am and 9 am on 2 days pre- and 2 days post-menstruation and were randomly collected among the study participants.

Serum Iron, total iron-binding capacity (TIBC), and Transferrin Saturation was carried out on the study participants.

2.5. Collection of Demographic Data

A structured questionnaire was administered to all eligible participants to enable them to fill in their demographic data, number of days in their cycle, Age of menarche, whether the flow was heavy or light, and use of iron supplements. Only females with a regular menstrual cycle who were not pregnant were included in the study, while those with an irregular menstrual cycle or who were pregnant were excluded.

2.6. Ethical Consideration

Ethical Clearance approval was obtained from the Ethics and Research Committee of the Jos University Teaching Hospital, Jos, Plateau State. Informed consent was obtained from all the participants.

2.7. Specimen Collection

Two (2ml) of venous blood was aseptically collected from each subject with the standard venepuncture technique into a plain bottle. The sample was centrifuged at 3000rpm for 5 minutes; serum was separated from the samples.

2.8. Laboratory Methods

2.8.1. Total serum iron

A semi-automated analyser (Chem-535) was used to assess serum samples from the subjects for Serum Iron.

A. Principle

Iron reacts with chromazurol-B and cetyltrimethyl-ammonium bromide (CTMA) to form a coloured ternary complex with an absorbance measured at 623nm spectrophotometrically. The intensity of the colour produced is directly proportional to the concentration of iron in the sample.

Table 1. Protocol for Serum Iron using Semi-Automated Analyser (Chem-535)

	Reagent Blank	Standard	Sample
Reagent	1.0ml	1.0ml	1.0ml
Standard		40µL	
Sample			40µL

The mixtures were thoroughly mixed and incubated at room temperature for 5minutes. The absorbance of the standard and sample was read against the reagent blank.

B. Calculation

$$\text{Iron concentration } (\mu g/dl) = \frac{\text{OD Sample}}{\text{OD Standard}} \times 200$$

SI unit: (µg/dl) x 0.1791 = µmol/L

Reference range = (6.6 - 28.0µmol/L)

2.8.2. Total Iron Binding Capacity (TIBC)

Semi-automated analyzer (Chem-535) was used to assessed serum samples from the subjects were assessed for Total Iron Binding Capacity using a semi-automated analyser (Chem-535)

A. Principle

Excess iron is added to the sample to give a saturation of the transferrin; the remaining iron will be absorbed mb magnesium carbonate. The bound iron in the supernatant is called TIBC, which is assayed in the same manner as the iron determination at 578nm spectrophotometrically.

B. Procedure

1) Preparation of the supernatant

Five hundred microliters (500µ) of reagent 1($FeCl_3$) were pipetted into a clean test tube. 250µL of the sample was added, mixed and allowed to stand at 25^{o}c for 30minutes. 1 full spatula of reagent 2 ($MgCO_3$) was added and allowed to stand for an additional 60 minutes, with periodic shaking 5 times within the 60 minutes. It was centrifuged for 10 minutes at 4000 rpm. A clear supernatant was obtained and used for the estimation of TIBC.

2) Determination of TIBC using Ferric S

Two (2) full spatulas of reagent 3 (Ascorbate) were dissolved in 50 mL of Ferren S buffer, mixed thoroughly and allowed to stand for 5 minutes to form the sample blank reagent. 1000µL of the above constituted solution (sample blank reagent) was pipetted into a clean test tube. 200µL of Feren S fluid (Chromogen) was added, mixed and allowed to stand for 10 minutes to form the reaction mixture. 50µL of the supernatant was added and incubated at room temperature for 10 minutes. It was analysed spectrophotometrically at 578nm. The standard was treated the same as the sample supernatant. The sample and standard blanks were prepared without the chromogen.

Table 2. Procedure for Serum Iron Using Semi-Automated Analyser (Chem-535)

	Sample	Sample blank	Standard	Standard blank
Clear supernatant	100µL	100µL		
Sample blank reagent		1000µL		1000µL
Reaction mixture	1000µL		1000µL	
Standard			100µL	100µL

C. Calculation

OD Sample

Iron concentration (µg/dl) =____________x 200

OD Standard

SI unit: (µg/dl) x 0.1791 = µmol/L

Reference range = (8.5 - 71.6µmol/L)

Transferrin Saturation (%TSAT)

The value of Transferrin saturation (%TSAT) for all the samples was calculated using the formula below.

Serum total iron

Transferrrin saturation (%TSAT) =____________________X 100

Total iron binding capacity

2.9. Statistical Analysis

Data were analysed using inferential statistics using the statistical software SPSS (statistically packaged for the social sciences, IBM Corporation) version 27.0.1. Results were expressed as Mean $\pm$ SD. T-test was used to compare mean of two parametric variables while

ANOVA was used to compare mean of three or more parametric variables Compare mean and one-way ANOVA. Statistical significance was set at $p<0.05$.

3. Results and Discussion

3.1. Research Results

Table 3 shows the 42 study participants' demographics, broken down by age group and level of menstrual flow. With 17 participants, or 40.5% of the sample, the highest age group represented is 26-30 years old. The age groups of 20-25 (16 individuals, 38.1%), 31-35 (5 participants, 11.9%), and 36-40 (4 participants, 9.5%) come next. In terms of menstrual flow, 24 women, or 57.1%, reported having a normal flow. 35.7% (15 participants) of the participants reported having a heavy flow, whereas 7.1% (3 participants) reported having a light flow. The subjects' menstrual flow lasted an average of four days, with a one-day standard deviation.

Table 3. Demographic Characteristics of the Participants (n=42)

Characteristics		N	%
Age Group (Years)			
	20 - 25	16	38.1
	26 - 30	17	40.5
	31 - 35	5	11.9
	36 - 40	4	9.5
Degree of flow			
	Normal	24	57.1
	Light	3	7.1
	Heavy	15	35.7
Duration of flow (Days), Mean (SD)		4(1)	4(1)

Table 4 presents the mean serum iron (SI), total iron-binding capacity (TIBC), and transferrin saturation (TSAT) for participants before and after menstruation, categorized by age groups. Before menstruation, participants aged 20-25 had a mean serum iron level of 17.81 µmol/L ±3.82, TIBC of 31.4 µmol/L ±5.0, and TSAT of 60% ±22. Those aged 26-30 had a slightly higher mean serum iron at 20.32 µmol/L ±3.68, with a TIBC of 31.1 µmol/L ±5.4, and a TSAT of 69% ±23. Participants aged 31-35 had the highest mean serum iron at 21.97 µmol/L ±2.28, the lowest TIBC at 26.3 µmol/L ± 5.2, and the highest TSAT at 86% ±18. The 36-40 age group had a mean serum iron of 18.74 µmol/L ±5.38, TIBC of 32.7 µmol/L ±6.9, and TSAT of 61% ±28.

Post-menstruation, all age groups showed a decrease in mean serum iron and TSAT, and an increase in TIBC. For the 20-25 age group, mean serum iron dropped to 14.66 µmol/L ±3.73, TIBC increased to 38.1 µmol/L ± 6.6, and TSAT decreased to 41% ± 18. The 26-30 age group had a post-menstruation mean serum iron of 17.13 µmol/L ± 3.48, TIBC of 35.0 µmol/L ±7.1, and TSAT of 52% ± 18. Participants aged 31-35 experienced a decrease in mean serum iron to 18.56 µmol/L ± 2.63, an increase in TIBC to 29.8 µmol/L ± 6.9, and a decrease in TSAT to 66% ± 21. The 36-40 age group had a mean serum iron of 14.89 µmol/L ±2.78, TIBC of 39.9 µmol/L ± 10.0, and TSAT of 40% ±17. The changes in these iron parameters highlight the physiological impact of menstruation across different age groups, though the exact statistical significance of these changes was not provided in the table.

Table 4. Effect of Age on Serum Iron (SI), Total Iron Binding Capacity (TIBC) and Transferrin Saturation (TSAT) of Pre- and Post-Menstruation

Age Group	Time	SI (µmol/L)	TIBC (µmol/L)	TSAT (%)
20-25	Pre	17.8±3.8	31.3±5.0	59.8±21.8
20-25	Post	14.7±3.7	38.1±6.6	41.1±17.7
26-30	Pre	20.3±3.7	31.2±5.4	68.7±22.6
26-30	Post	17.1±3.5	35.1±7.1	52.5±18.4
31-35	Pre	22.0±2.3	26.3	86.1±18.4
31-35	Post	18.6±2.6	29.8±6.9	65.9±20.9
36-40	Pre	18.7±5.4	32.7±6.9	61.0±28.2
36-40	Post	14.9±2.8	39.9±10.0	40.5±17.1
F value		2.457	1.874	2.411
P value		0.078	0.150	0.082

Table 5 presents the results of a paired sample t-test that analysed the changes in serum iron (SI), total iron-binding capacity (TIBC), and transferrin saturation (TSAT) before and after menstruation among 42 participants. The findings indicate significant variations in all three parameters due to menstruation. The mean serum iron level showed a notable decrease from 19.41 µmol/L ±SD 3.93 before menstruation to 16.14 µmol/L ± 3.63 after menstruation. This reduction, with a p-value of 0.001, highlights a statistically significant drop in serum iron levels post-menstruation.

Similarly, TIBC exhibited a significant increase, rising from a mean of 30.81 µmol/L ±5.46 pre-menstruation to 36.04 µmol/L ±7.46 post-menstruation. The p-value of 0.001 underscores the significance of this change, indicating an elevated iron-binding capacity after menstruation. Transferrin saturation also experienced a considerable decline, with the mean dropping from 66.67% ±23.11 before menstruation to 48.44% ±19.51 after menstruation. The p-value of 0.001 again signifies a statistically significant decrease in TSAT post-menstruation.

The study examined the impact of the duration of menstrual flow on Serum Iron (SI), Total Iron Binding Capacity (TIBC), and Transferrin Saturation (TSAT) by comparing pre- and post-intervention means across different flow durations (less than 2 days, 3-5 days, and 6 days & above).

Table 5. Effect of Menstruation on Pre- and Post-Serum Iron (SI), Total Iron Binding Capacity (TIBC), Transferrin Saturation (TSAT)

Variable	Pre Mean ± SD	Post Mean ± SD	T test	P value
SI µmol/L	19.41±3.93	16.14±3.63	10.092	0.001
TIBC µmol/L	30.81±5.46	36.04±7.46	8.333	0.001
TSAT%	66.67±23.11	48.44±19.51	11.671	0.001

Table 6 explains for Serum Iron (SI), participants with a menstrual flow of less than 2 days had pre- and post-intervention means of 22.33 µmol/L ±1.39 and 19.69 µmol/L ±1.37, respectively, with an F value of 7.319 and a P value of 0.035, indicating a significant decrease. Those with a flow of 3-5 days had means of 19.74 µmol/L ±3.94 pre-intervention and 16.01 µmol/L ±3.6 post-intervention, with an F value of 24.423 and a P value of 0.001, showing a highly significant decrease. Participants with a flow of 6 days & above had means of 18.48 µmol/L ±4.01 pre-intervention and 15.89 µmol/L ±3.79 post-intervention, with an F value of 6.610 and a P value of 0.013, indicating a significant decrease.

For Total Iron Binding Capacity (TIBC), participants with a flow of less than 2 days had means of 25.85 µmol/L ±1.91 pre-intervention and 28.3 µmol/L ±2.97 post-intervention, with an F value of 1.827 and a P value of 0.225, indicating no significant change. Those with a flow of 3-5 days had means of 30.76 µmol/L ±5.61 pre-intervention and 35.78 µmol/L ±8.02 post-intervention, with an F value of 13.108 and a P value of 0.001, showing a highly significant increase. Participants with a flow of 6 days & above had means of 31.57 µmol/L ±5.38 pre-intervention and 37.5 µmol/L ±6.42 post-intervention, with an F value of 30.220 and a P value of 0.001, indicating a highly significant increase.

For Transferrin Saturation (TSAT), participants with a flow of less than 2 days had means of 86.4%±0.99 pre-intervention and 70.22%±12.22 post-intervention, with an F value of 37.766 and a P value of 0.001, indicating a highly significant decrease. Those with a flow of 3-5 days had means of 67.95%±23.94 pre-intervention and 48.7%±20.26 post-intervention, with an F value of 18.494 and a P value of 0.001, showing a highly significant decrease. Participants with a flow of 6 days & above had means of 61.91%±22.32 pre-intervention and 45.1%±17.89 post-intervention, with an F value of 10.730 and a P value of 0.002, indicating a significant decrease.

Table 6. Effect of Duration of Flow on Pre and Post-Menstruation Serum Iron (SI), Total Iron Binding Capacity (TIBC), Transferrin Saturation (TSAT)

Variable	Duration of Flow (days)	Pre Mean	Post Mean	F value	P value
SI µmol/L	Less than 2	22.33±1.39	19.69±1.37	7.319	0.035
SI µmol/L	3-5	19.74±3.94	16.01±3.6	24.423	0.001
SI µmol/L	6 & above	18.48±4.01	15.89±3.79	6.610	0.013
TIBC µmol/L	Less than 2	25.85±1.91	28.30±2.97	1.827	0.225
TIBC µmol/L	3-5	30.76±5.61	35.78±8.02	13.108	0.001
TIBC µmol/L	6 & above	31.57±5.38	37.50±6.42	30.220	0.001
TSAT %	Less than 2	86.40±0.99	70.22±12.22	37.766	0.001
TSAT %	3-5	67.95±23.94	48.70±20.26	18.494	0.001
TSAT %	6 & above	61.91±22.32	45.10±17.89	10.730	0.002

Table 7 examines the correlation between the duration of menstrual flow and three key iron parameters: serum iron (SI), total iron-binding capacity (TIBC), and transferrin saturation (TSAT) among 42 participants. The correlations are expressed using Pearson's correlation coefficients along with their corresponding p-values. The Pearson correlation coefficient between the duration of flow and serum iron (SI) is -0.202, indicating a weak negative relationship. The p-value for this correlation is 0.199, which is above the conventional threshold for statistical significance ($p < 0.05$), suggesting that the observed correlation is not statistically significant.

For the relationship between the duration of flow and total iron-binding capacity (TIBC), the Pearson correlation coefficient is 0.234, indicating a weak positive relationship. The p-value for this correlation is 0.135, which also exceeds the threshold for statistical significance, indicating that this correlation is not statistically significant. The correlation between the duration of flow and transferrin saturation (TSAT) has a Pearson coefficient of -0.215, showing a weak negative relationship. The p-value here is 0.172, again higher than 0.05, implying that this correlation is not statistically significant.

In summary, the correlations between the duration of menstrual flow and the iron parameters (SI, TIBC, and TSAT) are weak and not statistically significant, as indicated by the respective p-values. This suggests that within this sample of 42 participants, the duration of menstrual flow does not have a significant impact on serum iron, total iron-binding capacity, or transferrin saturation.

Table 7. Correlation between Duration of Flow and Serum Iron (SI), Total Iron Binding Capacity (TIBC), Transferrin Saturation (TSAT)

	Duration of flow			Pearson Correlation	P value
	Less than 2	3-5	6 & above		
SIµmol/L	21.01±1.89	17.88±4.18	17.18±4.06	-0.202	0.199
TIBCµmol/L	26.08±2.48	33.27±7.31	34.54±6.55	0.234	0.135
TSAT%	78.31±11.72	58.33±24.01	53.50±21.64	0.215	0.172

Table 8 shows the effect of the degree of menstrual flow (normal, light, and heavy) on Serum Iron (SI), Total Iron Binding Capacity (TIBC), and Transferrin Saturation (TSAT) by comparing pre- and post-intervention means and analysing the statistical significance using F values and P values. For Serum Iron (SI), participants with a normal flow had pre- and post-intervention means of 19.81 µmol/L ±3.59 and 16.59 µmol/L ±3.61, respectively, with an F value of 19.201 and a P value of 0.001, indicating a significant decrease. Those with a light flow had means of 17.87 µmol/L ±1.53 pre-intervention and 16.37 µmol/L ±3.79 post-intervention, with an F value of 0.808 and a P value of 0.390, indicating no significant change. Participants with a heavy flow had means of 19.08 µmol/L ±4.77 pre-intervention and 15.38 µmol/L ±3.77 post-intervention, with an F value of 11.110 and a P value of 0.001, indicating a significant decrease.

For Total Iron Binding Capacity (TIBC), participants with a normal flow had means of 29.9 µmol/L ±5.31 pre-intervention and 36 µmol/L ±7.97 post-intervention, with an F value of 19.395 and a P value of 0.001, showing a significant increase. Those with a light flow had means of 33 µmol/L ±0.17 pre-intervention and 34.47 µmol/L ±1.68 post-intervention, with an F value of 4.608 and a P value of 0.057, indicating a trend towards significance. Participants with a heavy flow had means of 31.84 µmol/L ±6.13 pre-intervention and 36.41 µmol/L ±7.59 post-intervention, with an F value of 6.684 and a P value of 0.012, indicating a significant increase.

For Transferrin Saturation (TSAT), participants with a normal flow had means of 69.68%±22.23 pre-intervention and 50.29%±20.57 post-intervention, with an F value of 20.757 and a P value of 0.001, indicating a significant decrease. Those with a light flow had means of 54.17%±4.9 pre-intervention and 47.55%±11.36 post-intervention, with an F value of 1.479 and a P value of 0.252, indicating no significant change. Participants with a heavy flow had means of 64.37%±26.35 pre-intervention and 45.65%±19.66 post-intervention, with an F value of 9.033 and a P value of 0.004, indicating a significant decrease.

Table 8. Effect of Degree of Flow in Pre- and Post-Menstruation: Mean Serum Iron (SI), Total Iron Binding Capacity (TIBC), Transferrin Saturation (TSAT)

Variable	Degree of Flow	Pre Mean	Post Mean	F value	P value
SI µmol/L	Normal	19.81±3.59	16.59±3.61	19.201	0.001
SI µmol/L	Light	17.87±1.53	16.37±3.79	0.808	0.390
SI µmol/L	Heavy	19.08±4.77	15.38±3.77	11.110	0.001
TIBC µmol/L	Normal	29.90±5.31	36.00±7.97	19.395	0.001
TIBC µmol/L	Light	33.00±0.17	34.47±1.68	4.608	0.057
TIBC µmol/L	Heavy	31.84±6.13	36.41±7.59	6.684	0.012
TSAT %	Normal	69.68±22.23	50.29±20.57	20.757	0.001
TSAT %	Light	54.17±4.90	47.55±11.36	1.479	0.252
TSAT %	Heavy	64.37±26.35	45.65±19.66	9.033	0.004

Table 9 explores the correlation between the degree of menstrual flow and three key iron parameters: serum iron (SI), total iron-binding capacity (TIBC), and transferrin saturation (TSAT). The correlations are quantified using Pearson's correlation coefficients and the corresponding significance levels (Sig. 2-tailed), with a sample size of 84 participants. The Pearson correlation coefficient between the degree of flow and serum iron (SI) is -0.114, indicating a weak negative relationship. The significance level (p-value) for this correlation is 0.301, which is above the conventional threshold of 0.05, suggesting that this correlation is not statistically significant.

For the relationship between the degree of flow and total iron-binding capacity (TIBC), the Pearson correlation coefficient is 0.080, indicating a very weak positive relationship. The p-value for this correlation is 0.471, which is also higher than 0.05, indicating no statistically significant relationship. The correlation between the degree of flow and transferrin saturation (TSAT) has a Pearson coefficient of -0.106, showing a weak negative relationship. The p-value for this correlation is 0.336, again higher than 0.05, implying that this correlation is not statistically significant.

In summary, the correlations between the degree of menstrual flow and the iron parameters (SI, TIBC, and TSAT) are weak and not statistically significant, as indicated by the p-values. This suggests that within this sample of 84 participants, the degree of menstrual flow does not have a significant impact on serum iron, total iron-binding capacity, or transferrin saturation.

Table 9. Correlation between Degree of flow and Serum Iron (SI), Total Iron Binding Capacity (TIBC), Transferrin Saturation (TSAT)

	Degree of menstrual flow			Pearson	P
	Normal	**Light**	**Heavy**	**Correlation**	**value**
SIμmol/L	18.20±3.92	17.12±2.72	17.23±4.62	-0.114	0.301
TIBCμmol/L	32.95±7.38	33.73±1.34	34.13±7.17	0.080	0.471
TSAT%	59.98±23.34	50.86±8.62	55.01±24.74	-0.106	0.336

3.2. Discussion

As intestinal iron absorption rises to make up for menstrual blood loss, iron levels may experience a typical physiological recovery after menstruation. This process usually returns blood iron and transferrin saturation to pre-menstrual levels in healthy women with sufficient iron reserves without leading to clinical iron shortage. Therefore, unless they are severe, chronic, or accompanied by indications of iron insufficiency and depleted iron stores, short-term postmenstrual alterations in iron indices should be viewed as a typical physiological adaptation.

The present study assessed the effect of menstruation on iron status among female students of the Department of Medical Laboratory Science, University of Jos, North-Central Nigeria, using serum iron (SI), total iron-binding capacity (TIBC), and transferrin saturation (TSAT). The findings demonstrated a significant decline in serum iron and transferrin saturation after menstruation, accompanied by a significant increase in TIBC. These findings indicate that menstrual blood loss has a measurable impact on iron homeostasis even among apparently healthy young women.

The significant reduction in serum iron from 19.41 μmol/L before menstruation to 16.14 μmol/L after menstruation ($p = 0.001$) is consistent with the physiological loss of iron associated with menstrual bleeding. Menstrual blood contains considerable amounts of iron, and repeated monthly blood loss can gradually deplete circulating iron stores, especially when dietary iron intake is inadequate. A similar study has been reported by Munro et al. (2023), who observed that menstruating women generally have lower serum iron concentrations than non-menstruating women because of continuous menstrual iron losses. The observed reduction in serum iron in this study therefore reflects the immediate effect of menstrual blood loss on circulating iron availability.

The present study also found a significant increase in TIBC following menstruation (30.82 µmol/L versus 36.04 µmol/L, p = 0.001). TIBC is an indirect measure of transferrin concentration and typically increases when body iron stores decline in an attempt to maximise iron transport. This adaptive response has been widely reported among individuals with developing iron deficiency. Similar observations have been described by Manu et al. (2026), who noted that increased transferrin synthesis is one of the earliest biochemical responses to decreasing iron stores. The increased TIBC observed in the present study therefore suggests early compensatory mechanisms aimed at improving iron transport following menstrual blood loss.

Transferrin saturation reveals an acute decrease after menstruation, decreasing from 66.67% to 48.44% (p = 0.001). Because TSAT represents the proportion of transferrin occupied by iron, its reduction reflects decreased circulating iron after menstruation. This finding agrees with reports by Elamin et al. (2021), which indicate that declining transferrin saturation is one of the indicators of early iron depletion before iron deficiency anaemia develops.

Although all age groups demonstrated reductions in serum iron and transferrin saturation with corresponding increases in TIBC after menstruation, no statistically significant differences were observed among the age categories (serum iron, p = 0.078; TIBC, p = 0.150; TSAT, p = 0.082). This suggests that within the reproductive age range studied, age alone was not a major determinant of menstrual iron loss. The similar pattern observed across all age groups is expected because the physiological mechanism of menstrual blood loss remains relatively constant throughout reproductive life in healthy women (Habiba & Benagiano, 2023). Comparable findings have been reported in studies showing that menstrual characteristics rather than chronological age are stronger predictors of iron status among reproductive-aged women.

Duration of menstrual flow significantly influenced iron indices. Participants with menstrual periods lasting 3-5 days and those with menstrual flow lasting six days or more experienced significantly greater reductions in serum iron and TSAT, together with significant increases in TIBC. Although participants with menstrual duration of less than two days also showed reduced serum iron and TSAT, the increase in TIBC was not statistically significant (p = 0.225), probably because of the relatively smaller amount of blood lost. These findings agree with previous studies demonstrating that prolonged menstrual bleeding increases cumulative iron loss and predisposes women to iron deficiency. It was reported by Fraser colleagues that women with prolonged menstrual bleeding are more likely to develop depleted iron stores than those with shorter menstrual duration (Fraser et al., 2014).

The degree of menstrual flow also influenced biochemical markers of iron status. Women with normal and heavy menstrual flow experienced significant reductions in serum iron and TSAT and significant increases in TIBC after menstruation, whereas those with light menstrual flow showed no statistically significant changes. This finding supports the well-established relationship between menstrual blood volume and iron loss. Women with heavy menstrual bleeding lose substantially more iron each menstrual cycle, increasing their risk of iron depletion and eventually iron deficiency anaemia. Similar findings have been reported by Irving (2026) and Peuranpää et al. (2022), who demonstrated that heavy menstrual bleeding is one of the strongest predictors of reduced iron stores among women of reproductive age.

4. Conclusion

The findings of this study demonstrate significant decreases in SI and TSAT alongside increased TIBC levels in females before and after menstruation, with variation observed according to the duration and type of menstrual flow. These results offer insightful information about how menstruation affects certain iron parameters, which may have consequences for clinical iron depletion and future studies in the field of women's health.

This study is not without limitations. The small subgroup size in the light-flow category, comprising only three participants, may likely be responsible for the non-significance observed in that group. Additionally, the study was limited to one institution and department, which restricts the generalisation of findings; however, it provides useful information that could form a baseline for future studies.

Based on these findings, it is recommended that menstruating women considered at risk for iron shortage, particularly those with high or prolonged menstrual flow, have their iron status checked every two to three months. Women at risk of menstruation-related iron depletion should be closely monitored, with iron supplements administered where necessary.

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