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**Dr. Shaveta**  
 SGT Medical College Hospital  
 and Research Institute  
 Gurugram Haryana, India

**Dr. Sanjiv Kumar Bansal**  
 SGT Medical College Hospital  
 and Research Institute  
 Gurugram Haryana, India

## To study iron status in non anemic young female students

**Dr. Shaveta, Dr. Sanjiv Kumar Bansal**

### Abstract

**Introduction:** Iron deficiency (ID) is the one of the most prevalent nutritional deficiency worldwide. Therefore, maintaining body iron status is important in young female as they will be child bearers of future generation and early detection would call for early intervention. Therefore, main aim of our study was to assess the iron status in non anemic young female.

**Objective:** To assess the iron status of non anemic young female which included Mean corpuscular volume(MCV), Mean corpuscular hemoglobin(MCH), and mean corpuscular hemoglobin concentration(MCHC), Serum iron, total iron binding capacity- TIBC and serum Ferritin, Transferrin saturation

**Methods:** It was a cross-sectional study performed on two hundred students in SGT University, Gurugram. 18-25 years non pregnant female Hb>12mg/dl with normal RBC indices were included in this study. Blood samples were analyzed for MCV, MCH, MCHC, serum iron, TIBC and serum Ferritin by autoanalyser. Transferrin saturation was calculated from the data of serum iron and TIBC.

**Result:** The mean age of study population was 19.4 year (17 – 25 years) and BMI of 25.8. All the hematologic parameters like HCT, MCV, MCH, MCHC, and TLC were well within the reference range. Serum iron levels in all the study group students were also within the normal reference range of 50.25-149.92 µgm/dL. The Mean for TIBC 425.1 gm/dL, with 76/200 (26%) cases showing values above the reference range of 425 gm/dL. Mean ±SD for serum transferrin was 297.3±25.62 mg/ dL with 9/200(4.5%) were iron deficient. The Mean ±SD value of TSAT% and serum ferritin observed in this study was 20.7 ± 6.26 and 33.8± 21.1 µgm/L, showing 63/200 were ID.

**Conclusion:** Significant number of healthy women with normal hemoglobin was found to be iron deficient and therefore accurate analysis of iron status would help the clinicians in planning, treating and thus preventing the risk of adverse events due to iron deficiency.

**Keywords:** Iron deficiency, serum ferritin, serum iron

### Introduction

Iron deficiency is the single one most common nutritional deficiency worldwide. The World Health Organization (WHO) estimates that globally~293 million young children and 468 million non-pregnant women suffer from anemia, among which ~50% are estimated to be attributable to iron deficiency (ID) [1]. The effects of low iron in women can have broad global effects on their physical and cognitive capabilities [2, 3].

It is estimated that about 20%-40% of maternal deaths in India are due to anemia. India contributes to about 50% of global maternal deaths due to anemia [4]. In India, 74% of the children <5 year old and 52% of young women have anemia. Iron-deficiency also has important consequences for the future generations, as iron-deficiency anemia increases the risk for preterm labour, low birth weight, infant mortality and predicts iron-deficiency in infants after 4 months of age [5, 6].

Progression of iron deficiency can be divided into 3 stages [7].

1. The first stage (the negative iron balance) in which the demands for (or losses of) iron exceed the body's ability to absorb iron from the diet. Under these circumstances, iron deficiency is made up by mobilizing stores from reticulo endothelial storage sites. During this period iron stores- reflected by serum ferritin level or the appearance of stainable iron on bone marrow aspirations - decrease.
2. The second stage (iron deficient erythropoiesis) is when stores are depleted and serum iron begins to fall TIBC and red cell protoporphyrin increase. Marrow stores are absent when serum iron ferritin <15µg/L. As long as serum iron remains normal Hb is unaffected, once transferrin saturation falls to 15- 20% Hb synthesis is impaired

### Correspondence

**Dr. Shaveta**  
 SGT Medical College Hospital  
 and Research Institute  
 Gurugram Haryana, India

Peripheral blood smear reveals microcytic cells, hypochromic reticulocytes in circulation.

### 3. Iron deficiency anemia: Hb and haematocrit falls.

Signs relating to iron deficiency depend on severity and chronicity of anemia in addition to usual signs – fatigue, pallor, reduced exercise capacity. The serum iron level represents the amount of circulating iron bound to transferrin. TIBC is indirect measure of circulating transferrin.

Correction of Iron deficiency (Latent anaemia) & Iron deficiency anemia (IDA) have become critical goals all over the world because of their decreased immunity, increased morbidity and impaired cognitive performance. It is important to ensure that satisfactory iron status be maintained in young females before they go for pregnancy to prevent premature births, low birth weights and prenatal mortality [8, 9, 10]. ID through its effects on cognition and educational achievements among young students impairs productivity, work performance and learning skills. High prevalence rates highlight the need for new effective sustainable strategies to control ID.

Maintaining body iron status is very important in young female as they will be child bearers of future generation and early detection would call for early intervention. Therefore, main aim of our study is to assess the iron status of non anemic young female.

### Material and methods

The present study will be conducted in the Department of Biochemistry; SGT Medical College, Budhera, Gurugram.

### Type of study: Cross Sectional

**Selection of Cases:** Female students volunteering from SGT University conforming to the inclusion and exclusion criteria will be taken for study.

Volunteers will be subjected to Hemoglobin estimation by Sahli's method in samples collected by finger prick. Those with hemoglobin > 12mg% and fulfilling both inclusion and exclusion criteria will be taken for study.

**Sample size:** 200 subjects will constitute as the sample size. This sample size will be calculated according to the incidence of Non symptomatic Iron deficient population in the study. Screening will continue till the appropriate sample size for the study is attained.

### Inclusion criteria

1. Hb > 12mg/dl
2. Age 18-25 years with normal RBC indices
3. Non pregnant

### Exclusion criteria

1. Women on Iron therapy and OCP.
2. Women with chronic liver disease or any systemic illness.
3. Any acute or chronic infection.

### Collection of blood samples

Collection of blood samples  
About a total of 10 ml of venous blood will be drawn under aseptic precautions in the suitable vacutainers from selected subjects. The blood samples will be analyzed as follows:-

1. For Complete Hemogram – about 3ml sample In EDTA vial (WBC, RBC, Hb, HCT, MCV, MCHC, PLT Count,

MCH will be analyzed in SYSMEX KX 21 Autoanalyser.

2. 7ml of the sample will be drawn in plain vacutainer. Serum will be separated by centrifugation and will be used as analysis.
  - i. Serum Iron and TIBC will be estimated by Iron and TIBC kit on fully automated analyzer.
  - ii. Transferrin saturation will be calculated as Serum iron  $\times 100 / \text{TIBC}$ .
  - iii. Serum ferritin will estimated by Chemiluminescence Immunoassay.

### Statically analysis

The results were analyzed statistically using suitable software (SPSS) v 20.0. Mean and standard deviation of the study parameters was calculated. Pearson's correlation coefficient for finding correlation between markers pertaining to Iron status was calculated.

### Results

Study population comprised of young healthy female with the mean age of 19.4 year, and mean BMI 25.8.

**Table 1:** Study Group Characteristics

| Parameter   | Mean  | SD   | Range   |
|-------------|-------|------|---------|
| Age         | 90.4  | 1.96 | 18-25   |
| Body Weight | 153.2 | 6.87 | 141-175 |
| Height      | 162.8 | 6.15 | 150-173 |
| BMI         | 25.8  | 2.14 | 23-29   |

The haemoglobin, hematocrit and red cell indices of all the subjects were within the normal reference indicating no evidence of anemia. The total leucocyte count was also within the normal range ruling out any inflammation

**Table 2:** Hematologic Parameters of the study Group

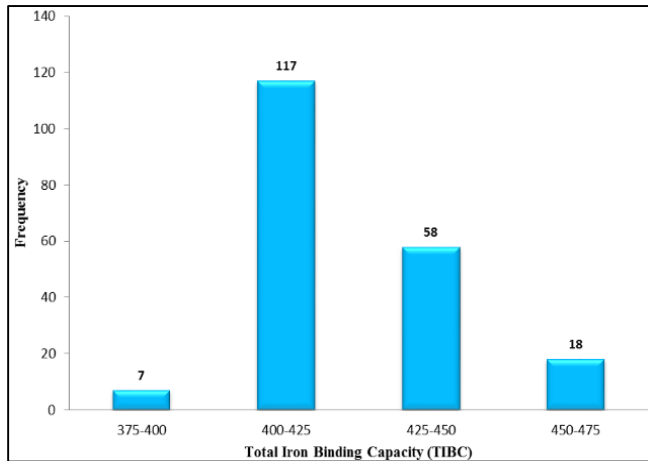
| Parameter           | Mean   | SD     | Range       |
|---------------------|--------|--------|-------------|
| Haemoglobin (gm/dL) | 13.1   | 0.69   | 12-15       |
| Haematocrit         | 40.1   | 2.11   | 36.7-45.7   |
| RBC Count           | 4.14   | 0.261  | 3.8-4.99    |
| MCHC                | 32.7   | 0.03   | 32.7-33.8   |
| MCH                 | 31.6   | 0.42   | 30-32.5     |
| MCV                 | 96.7   | 1.34   | 91.58-99.25 |
| TLC                 | 8028.5 | 648.34 | 5700-9100   |

Serum iron within the normal reference range in all the subjects, where as TIBC, Serum Transferrin, TSAT and Serum Ferritin were below the normal reference range in some of the subjects as seen in the frequency distribution bar graph.

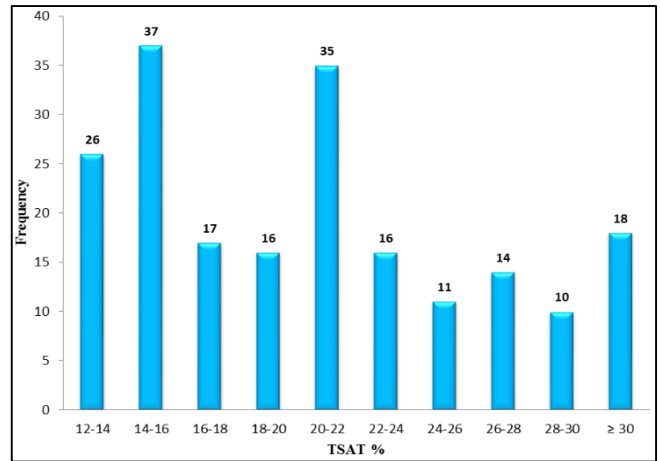
**Table 3:** Iron Status of the Study Group based on traditional Parameters

| Parameter                          | Mean  | SD    | Median | Range        |
|------------------------------------|-------|-------|--------|--------------|
| Serum Iron $\mu\text{gm/dL}$       | 87.4  | 23.59 | 86.7   | 50.25-149.92 |
| TIBC (gm/dL)                       | 425.1 | 16.38 | 420.4  | 386.6-460.9  |
| Serum Transferrin (mg/dL)          | 297.3 | 25.62 | 298.1  | 184.1-326.8  |
| TSAT (%)                           | 20.7  | 6.26  | 20.6   | 12.03-38.78  |
| Serum Ferritin ( $\mu\text{gm/}$ ) | 33.8  | 21.1  | 32.8   | 9.8-97.7     |

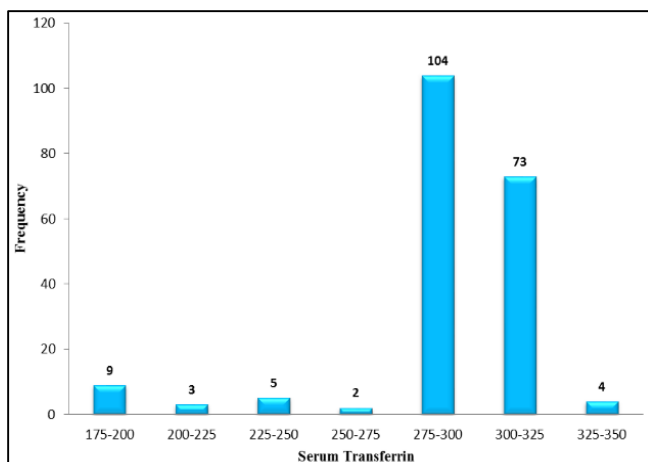
Frequency distribution bar graph was plotted for TIBC in the range of 375-475 gm/dl. The cut off value for ID was >425 gm/dl and is visible from the plot 76 students were above this value signifying Iron Deficiency



As the cut off s for serum transferrin for diagnosis ID was serum Transferrin <200mg/dl, 9 students were below this value.



In table 4 discrimination was made on the basis of Serum Ferritin with (Serum Ferritin ≤ 12µgm/L were taken as ID; Serum Ferritin 12 – 20 µgm/L were taken as equivocal; Serum Ferritin > 20µgm/L were taken as NID. Twenty three out of two hundred were diagnosed as ID accounting to 11.5%. Fifty out of two hundred were diagnosed as equivocal that is they could be included in ID or NID which accounted to 25%. NID accounted to 63.5% of the total population.



**Frequency distribution bar graph for Serum Ferritin µ gm/L**

The range of Serum Ferritin by Fig was between 8 – ≥20 µgm/L. The cut off s for Ferritin for diagnosing ID was Serum Ferritin ≤ 15µgm/L, fifty nine students were below normal [11].

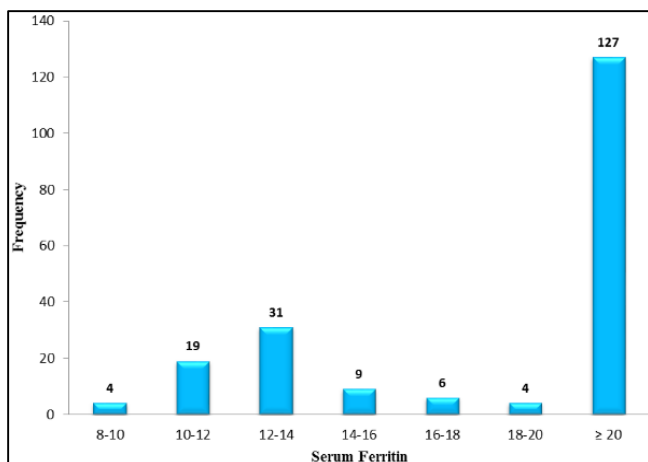
**Table:** Discrimination between ID and NID based on Serum Ferritin

| Parameter                    | Iron Deficient (ID)/ Non Iron Deficient (NID) | Total n = 200 |            |
|------------------------------|---|---------------|------------|
|                              |   | n             | Percentage |
| Serum Ferritin ≤ 12 µgm/L    | ID  | 23            | 11.5%      |
| Serum Ferritin 12 – 20 µgm/L | Equivocal                                     | 50            | 25%        |
| Serum Ferritin > 20 µgm/L    | NID   | 127           | 63.5%      |

Discrimination on the basis on Serum Transferrin, as per table 7, classifying ID as Serum Transferrin < 200 mg/ dL accounting for 4.5% of the total and NID being classified as Serum Transferrin ≥ 200 mg/ dL which included the rest 95.5% (191/200).

**Table:** Discrimination between ID and NID population based on Serum Transferrin

| Parameter                     | Iron Deficient (ID)/ Non Iron Deficient (NID) | Total n = 200 |            |
|-------------------------------|---|---------------|------------|
|                               |   | n             | Percentage |
| Serum Ferritin < 200 gm/dL    | ID  | 9             | 4.5%       |
| Serum Transferrin ≥ 200 mg/dL | NID   | 191           | 95.5%      |



**Frequency distribution bar graph for Transferrin Saturation %**

As the cut off s for TSAT% for diagnosing ID was TSAT ≤ 16%, sixty three students were below this value as visible from Fig 9. As visible from the frequency distribution plot, TSAT% range of values were from 12 – 36%.

Discrimination on the basis on TIBC, as per table 8, classifying ID as TIBC > 425mg/ dL accounting for 38% of the total and NID being classified as TIBC ≤ 425mg/ dL which included the rest 62% (124/200).

**Table:** Discrimination between ID and NID population based on TIBC

| Parameter       | Iron Deficient (ID)/ Non Iron Deficient (NID) | Total n = 200 |            |
|-----------------|---|---------------|------------|
|                 |   | n             | Percentage |
| TIBC > 425mg/dL | ID  | 76            | 38%        |
| TIBC ≤ 425mg/dL | NID   | 124           | 62%        |

As shown in Table 9 on differentiating between Iron Deficient and non Iron Deficient based on TSAT% and

Ferritin (TSAT  $\leq$  16% and Serum Ferritin  $\leq$  15 $\mu$ g/L were identified as ID; TSAT  $>$  16% and Serum Ferritin  $>$  15 $\mu$ g/L were identified as NID). Sixty three out of 200 were ID (31.5%), the rest 137/200 were NID (68.5%).

**Table:** Discrimination between ID and NID population based on TSAT% and Serum Ferritin

| Parameter         | Iron Deficient (ID)/ Non Iron Deficient (NID) | Total n = 200 |            |
|-------------------|---|---------------|------------|
|                   |   | n             | Percentage |
| TIBC $>$ 425mg/dL | ID  | 76            | 38%        |
| TIBC $>$ 425mg/dL | NID   | 124           | 62%        |

### Discussion

Iron deficiency anemia is the most common leading cause of disease in girls and women in developing countries. As per WHO iron deficiency anemia affects 1.3 billion people worldwide amongst which 43% are preschool children, 51% are pregnant women and 37% are school age children [12].

Our study population comprised of young university students with a mean age of 19.4 year and BMI of 25.8. 17-25 years. Study was intended for young healthy women, only those with Hb  $\geq$  12 gm /dl were included, mean being 13.1 and all the hematologic parameters like HCT, MCV, MCH, MCHC, TLC were well within the reference range. Serum iron levels in all the study group students were also within the normal reference range of 50.25-149.92  $\mu$ g/dL. Cook *et al* also reported that serum Iron was suitable only to detect advanced iron deficiency as it is subjected to variation in dietary intake, iron therapy and diurnal variation.

Mean value for TIBC 425.1 in this study, with 76/200 (26%) cases showing values above the reference range of 425 gm/dL. In another study conducted by on female medical students, Mean TIBC reported was 401.30 $\pm$ 89.06 [8] Mean  $\pm$ SD for serum transferrin was 297.3 $\pm$ 25.62 which was comparable to that of 280  $\pm$  0.6mg/dL in a study on the micronutrient status in university students [97]. The mean value of TSAT% observed in this study was 20.7  $\pm$  6.26, comparable to 20.27  $\pm$  8.20 observed in Modi s study in female medical students [80]. The Mean  $\pm$  SD for serum ferritin observed in this study was 33.8 $\pm$  21.1 20.64 $\pm$ 8 was the mean and SD observed for ferritin in Modi *et al.* study of iron status in medical students [13].

Ferritin is a intracellular 24 subunit protein that stores iron and releases it in a controlled fashion The ferritin levels have a direct correlation with the total amount of iron stored in the body. However, ferritin levels may be falsely high where ferritin is elevated in its capacity as an inflammatory acute phase protein and not as a marker for iron overload. The level of serum ferritin is affected by inflammation and food intake [14]. Iron deficiency with or without anemia during childhood, especially in infancy, has a negative impact on cognition, behavior, and motor skills of children [15]. Serum ferritin is a more sensitive marker as compared to serum iron which can be detected at the early stage of iron deficiency even without anemia.

### Conclusion

From the present study we may conclude that the prevalence of Iron deficiency in Young non anemic females in north India suggests the need for increasing awareness about nutritional diet, its effects and need to prevent it. High prevalence of Anemia suggests the need of screening the

vulnerable group of 15-49 years of females, so that iron deficiency anemia can be prevented.

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