Iron, TIBC and Ferritin Levels Correlation with Testosterone Deficient Males in Dhaka, Bangladesh

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Abstract
Low serum iron and ferritin levels with an elevated TIBC are diagnostic of iron deficiency. While a low serum ferritin is virtually diagnostic of iron deficiency, a normal serum ferritin can be seen in patients who are deficient in iron and have coexistent diseases (eg, hepatitis or anemia of chronic disorders). Men with low testosterone levels have lower hemoglobin and hematocrit levels, while men with anemia of unknown etiology. Testosterone directly regulates body iron levels through inhibition of the master regulator of iron metabolism, hepcidin. In our study we studied with 30 males. 80.0% males were observed with low iron level, 100.0% found with normal ferritin level, 66.7% with normal TIBC and finally we found 96.7% males with low serum testosterone level.

Keywords: Iron, TIBC, ferritin and testosterone.
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Introduction
Testosterone is the essential male sex hormone and an anabolic steroid. In male people, testosterone assumes a key job in the improvement of male regenerative tissues, for example, testicles and prostate, just as advancing optional sexual qualities, for example, expanded muscle and bone mass, and the development of body hair.[1] moreover, testosterone is engaged with health and well-being,[2] and the avoidance of osteoporosis.[3] Insufficient degrees of testosterone in men may prompt irregularities including feebleness and bone misfortune. Testosterone is a steroid from the androstane class containing a keto and hydroxyl bunches at the three and seventeen positions separately. It is biosynthesized in a few stages from cholesterol and is changed over in the liver to inert metabolites.[4] It applies its activity through authoritative to and enactment of the androgen receptor.[4] In people and most different vertebrates, testosterone is discharged fundamentally by the gonads of guys and, to a lesser degree, the ovaries of females. By and large, in grown-up guys, levels of testosterone are around 7 to multiple times as incredible as in grown-up females.[5] As the digestion of testosterone in guys is progressively articulated, the day by day generation is around multiple times more noteworthy in men.[6, 7] Females are likewise increasingly delicate to the hormone.[8]

Notwithstanding its job as a characteristic hormone, testosterone is utilized as a drug, for example in the treatment of low testosterone levels in men, transgender hormone treatment for transgender men, and bosom malignant growth in women. Since testosterone levels decline as men age, testosterone is now and again utilized in more established men to check this lack. It is likewise utilized unlawfully to improve physical make-up and execution, for example in athletes.[9]

Numerous examinations have announced a relationship between serum testosterone levels and body iron indices in people. For instance, men with low testosterone levels have lower hemoglobin and hematocrit levels, while men with weakness of obscure etiology are bound to have low testosterone levels contrasted with controls.[10-13] Also, men in the most minimal quartile of aggregate and free testosterone levels are fundamentally bound to create iron deficiency (5.4-and 13.1-overlay, individually) than those in the most noteworthy quartile.[14]

Increased hematocrit and hemoglobin are the most common side effects of exogenous testosterone therapy, with a 315% increased risk of erythrocytosis in men receiving testosterone.[15] This is conceivably helpful, however, as testosterone supplementation in hypogonadal men can diminish the commonness of iron deficiency from 30 to 10%.[16] Strikingly, preceding the standard utilization of erythropoiesis-animating operators, androgens were utilized to build hemoglobin fixations in dialysis patients.

While testosterone treatment is related with expanded hemoglobin levels, androgen deprivation therapy (ADT) has the contrary impact. ADT is every now and again used to treat metastatic prostate disease. With ADT, hemoglobin and hematocrit levels decline altogether, and up to 13% of treated men report symptomatic iron
deficiency.[17]

It is fascinating to take note of that the hours of the most elevated testosterone levels concur with the hours of the most elevated iron interest in people. Both serum testosterone and hemoglobin levels top during puberty.[18-21] This is related with a twofold to triple increment in iron prerequisite and a critical increment in the pace of dietary iron assimilation.[22, 23] Thus, testosterone levels in young men are low during childbirth however arrive at pubertal levels at 1–3 months of age before declining back to pattern levels by a half year of age. [24] Commencement of this "mini puberty" harmonizes with the nadir of baby hemoglobin fixations after which iron take-up and serum hemoglobin levels increment.[25]

Ferritin is a protein that adds to the iron-stockpiling primarily in our body and disseminates generally in all tissues, particularly in certain organs, for example, the liver, spleen and bone marrow,[26] and it is likewise a provocative serum biomarker.[27] Some past examinations have shown that degrees of ferritin were identified with a few ailment states. As we was already aware, it was related with metabolic disorder,[28] hematopoiesis,[29, 30] renal malady,[31, 32] liver illness,[33] autoimmunity,[34] disease,[35] the forecast of malignant growth [36] and the mortality of liver transplantation.[37]

It was accounted for that the two subunits of ferritin was incorporated under the influence of various qualities in chromosomes 11 and 19, separately.[38, 39] The degree of ferritin was influenced by the iron digestion in people body and cytokines, development factor just as oxidants added to the guideline.[40] Particularly, it ought to be noticed that the hormones additionally might be a member in the guideline, for instance the thyroid hormone.[40] At the point when it went to the sex hormones, they were notable as significant members in improving sexual organs, and furthermore identified with metabolic disorder,[41] iron deficiency,[13] irritation [42] and the mortality in the old.[43-45]

In perspective on regular infections and the guideline component of ferritin that were referenced above, sex hormones may conceivably partake in the guideline of ferritin, and afterward rise a few confirmations of connections among's them and serum ferritin levels. Be that as it may, scarcely any investigations described the connections between serum ferritin levels and sex hormones.[46, 47]

Materials and Methods

Study Design
A cross-sectional study.

Study Location
Popular diagnostic center, Narayanganj, Dhaka.

Study Duration
September-December, 2019.

Sample Size
Thirty (30) males.

Sample Collection
5 ml of blood was collected from the anticubital vein in fasting and postprandial state from all the subjects. The blood samples were collected in fluoride tubes, clot activated tubes and EDTA tubes for the estimation of serum iron, TIBC, ferritin and testosterone respectively.

Iron
Serum Iron is estimated by Ferrozine method using Biosystems kit.

TIBC
Serum Unsaturated Iron Binding Capacity (UIBC) was estimated by Ferrozine method using Biosystems kit. These parameters were analyzed on fully automated biochemistry analyzer BA 200 (Biosystems, Barcelona, Spain). Total Iron Binding Capacity (TIBC) was calculated by addition of serum Iron and UIBC.

Ferritin
Estimation of serum ferritin was done by using automated analyzer Biomeriux Minividas by Enzyme Linked Immunofluoroscent Assay (ELFA) method.

Testosterone
According to the manufacture, serum levels of Testosterone was measured by using TOSOH Bioscience automated immunoassay analyzer AIA-360. Competitive immunoenzymometric assay was used, testosterone in
the test sample compete with enzyme-labeled testosterone for limited number of binding sites on these hormones specific antibodies immobilized on magnetic beads, then magnetic beads were washed to remove unbound enzyme-labeled testosterone, and incubated with fluorogenic substrate 4-methylumbelliferyl phosphate (4 MUP). The rate of fluorescence produced by the reaction was measured fluorometrically at 365nm. The amount of testosterone that binds to the beads is inversely proportional to the testosterone concentration in the sample.

**Statistics**
Percentile distribution of the samples regarding different parameters such as age, iron, TIBC, ferritin and testosterone were analyzed by using SPSS.

**Results**
The total of 30 blood samples of male were collected from at 2019 (September-December) from Narayanganj, Dhaka, Bangladesh.

| Age Group | Frequency | Percent | Valid Percent | Cumulative Percent |
|-----------|-----------|---------|---------------|--------------------|
| >10-20    | 6         | 20.0    | 20.0          | 20.0               |
| >20-30    | 9         | 30.0    | 30.0          | 50.0               |
| >30-40    | 10        | 33.3    | 33.3          | 83.3               |
| >40-50    | 1         | 3.3     | 3.3           | 86.7               |
| >50-60    | 4         | 13.3    | 13.3          | 100.0              |
| Total     | 30        | 100.0   | 100.0         |                    |

In our study, table-1 showed the age distribution of subjects by their age groups. The highest of the study subjects underwent to the >30-40 years age group (10) and followed by >20-30 years age group (9), >10-20 years age group (6), >50-60 years age group (4) and <40-50 years age group (1) respectively.

| Iron (µg/dL) | Frequency | Percent | Valid Percent | Cumulative Percent |
|--------------|-----------|---------|---------------|--------------------|
| Under <65    | 24        | 80.0    | 80.0          | 100.0              |
| Normal 65-176| 6         | 20.0    | 20.0          | 100.0              |
| Above >176   | 0         | 0       | 0             | 100.0              |
| Total        | 30        | 100.0   | 100.0         |                    |

Among the subjects (n=30) 24 males (80.0%) were found with >65µg/dL, less than the standard level 65-176 µg/dL, serum iron and rest 6 subjects (20.0%) were found with standard serum iron level. No male subject was found with more serum iron level compare to the standard (Table-2).

| TIBC (µg/dL) | Frequency | Percent | Valid Percent | Cumulative Percent |
|--------------|-----------|---------|---------------|--------------------|
| Under <240   | 4         | 13.3    | 13.3          | 13.3               |
| Normal 240-450| 20       | 66.7    | 66.7          | 80.0               |
| Above >450   | 6         | 20.0    | 20.0          | 100.0              |
| Total        | 30        | 100.0   | 100.0         |                    |

Table-3 showed the distribution of TIBC level among 30 males. 13.3% (2) males found with below the normal level, 66.7% (20) males were within the normal level and 20% (6) males were diagnosed with above the normal blood TIBC level.

| Ferritin (12-300ng/dL) | Frequency | Percent | Valid Percent | Cumulative Percent |
|-------------------------|-----------|---------|---------------|--------------------|
| Under <12               | 0         | 0.0     | 0.0           | 0                  |
| Normal 12-300           | 30        | 100.0   | 100.0         | 100.0              |
| Above >300              | 0         | 0.0     | 0.0           | 100.0              |
| Total                   | 30        | 100.0   | 100.0         |                    |

Table-4 reflected the pattern of blood ferritin level among the target 30 males. 0.0% (0) male found with below the normal level, 100.0% (30) males were within the normal level and 0.0% (0) male was diagnosed with above the normal blood ferritin level.
Table-5: Blood Testosterone level among the study subjects (n=30).

| Testosterone Levels | Frequency | Percent | Valid Percent | Cumulative Percent |
|---------------------|-----------|---------|---------------|--------------------|
| Valid               | Under     | <9.2    | 29            | 96.7               | 96.7               |
|                     | Normal    | 9.2-31.8| 1             | 3.3                | 3.3                |
|                     | Above     | >31.8   | 0             | 0                  | 0                  |
| Total               |           |         | 30            | 100.0              | 100.0              |

Among the subjects (n=30) 29 males (96.7%) were found with <9.2 nmol/L testosterone in the blood, less than the standard level 9.2-31.8nmol/L and rest 1 subject (3.3%) was found with normal blood testosterone level. No male subject was found with more serum iron level compare to the standard (Table-5).

Discussion
Testosterone increases dietary iron absorption, providing a direct link between the two. As the body is unable to eliminate excess iron, a negative feedback mechanism allowing iron to inhibit testosterone production to maintain body iron homeostasis is proposed.[48]

A high TIBC, UIBC, or transferrin usually indicates iron deficiency, but they are also increased in pregnancy and with the use of oral contraceptives. A low TIBC, UIBC, or transferrin may also occur if someone has malnutrition, inflammation, liver disease, or nephrotic syndrome.[49]

If a ferritin test shows higher than normal levels, it could indicate a condition that causes body to store too much iron. It could also point to liver disease, rheumatoid arthritis, other inflammatory conditions or hyperthyroidism.[50] In our study we found normal ferritin level but low iron.

Different research results showed that low testosterone level due to low iron level. At low iron level TIBC level becomes high and at high ferritin level body stores more iron.

In our study we found low testosterone with low iron level; and found TIBC and ferritin level mostly normal.

Conclusion
In our study we studied with 30 males. 80.0% males were observed with low iron level, 100.0% found with normal ferritin level, 66.7% with normal TIBC and finally we found 96.7% males with low serum testosterone level.

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