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Study of the relationship between ferritin serum and thyroid function

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Abstract---The present study including study effect of thyroid function on ferritin. In Thi-Qar province, Iraq. The sample include 30 from patients and 10 samples was control in four age categories was (25≥, 26-35, 36-45, 46-55) during the period from August 2021 to January 2022. The current study show TSH level increased in women suffering from hypothyroidism, while it was lowest in women suffering from hyperthyroidism, As for the (T3,T4) Hormone the highest level was with hyperthyroidism, but women with hypothyroidism had lower levels when compared to the control group. The ferritin level decreased in the age group (46-55), while T3 and T4 was higher in the age group (26-35), but TSH increased in women less than 25 year. As for the Hb highest level was age category (26-35) while lowest in age category (46-55).

Keyword---hypothyroidism, hyperthyroidism, ferritin, TSH, T3, T4, Hb.

Introduction

Ferritin is an iron storing protein that can be found in nearly all bodily tissues. Thyroid illness patients' serum ferritin levels have also been reported to be changed. As a result, changes in ferritin concentrations in the blood reflect thyroid function.

Ferritin is the most common type of iron stored in the body. Small amounts of ferritin are secreted into the plasma of the body. In the absence of inflammation, the concentration of this plasma (or serum) ferritin is positively associated with the size of total body iron reserves. A low serum ferritin level indicates that iron stores have been depleted, but not necessarily the severity of the loss as time goes on. Ferritin concentrations fluctuate depending on age and gender. At birth,

concentrations are high, then climb for the first two months of life, then diminish throughout childhood (1). Concentrations begin to grow again around the age of one year and continue to rise until adulthood (2). Males, on the other hand, have greater values than females beginning in adolescence, and this trend continues until late adulthood. Men's values peak between the ages of 30 and 39, and then tend to stay steady until around the age of 70. Serum ferritin levels in women stay low until menopause, when they begin to rise (2). Unlike haemoglobin, body ferritin levels are unaffected by residence elevation above sea level or smoking habits. Ferritin, on the other hand, is a positive acute phase response protein, meaning that its concentration rises during inflammation and no longer reflects the size of the iron reserve. This complicates the evaluation of serum ferritin levels that are normal or high.

The thyroid gland is an important hormone gland that regulates metabolism, growth, and development in humans(5) (6). By continuously releasing a stable amount of thyroid hormones into the bloodstream, it aids in the regulation of numerous body functions. The thyroid gland generates extra hormones when the body need more energy in particular situations, such as when it is growing, cold, or pregnant (4).

The thyroid gland produce three hormone:

1-Triiodothyronine, also known as T3

2-Tetraiodothyronine, also called thyroxine or T4

3-Calcitonin

After diabetes mellitus, thyroid illness is the second most frequent disease (7) The thyroid gland's activity is linked to its two hormones, tetraiodothyroxine (T4) and triiodothyroxine (T3) (T3). TSH, which is produced by the pituitary gland, regulates the production of these two hormones (8). Thyroid hormone release is regulated by the amount of iodine in the body. Despite an increase in TSH, hypothyroidism occurs when there are insufficient iodine sources for T4 and T3 production. When high levels of iodine are present, the rate of T4 and T3 synthesis increases, but TSH levels decrease, resulting in a buildup of T4 and T3 in thyroid follicles and thyrotoxicosis, also known as hyperthyroidism (9).

Material and Methods

1-Ferritin test

Test principle

The method for measurement of Ferritin on the cobas® e601 is a sandwich principle with a total duration time of 18 minutes. The 1st incubation uses 10 µL of sample, a ferritin-specific antibody and a labeled ferritin-specific antibody to form a sandwich complex. The 2nd incubation occurs after the addition of microparticles that cause the complex to bind to the solid phase. The reaction mixture is aspirated into the measuring cell where the microparticles are magnetically captured onto the surface of the electrode. Unbound substances are then removed. Application of a voltage to the electrode then induces chemiluminescent emission which is measured by a photomultiplier. Results are determined via a calibration curve.

2- Thyroid Stimulating Hormone

Test principle

The TSH ELISA test is based on the principle of a solid phase enzyme-linked immunosorbent assay. The assay system utilizes a unique monoclonal antibody directed against a distinct antigenic determinant on the intact TSH molecule. Mouse monoclonal anti-TSH antibody is used for solid phase immobilization (on microtiter wells). A goat anti-TSH antibody is in the antibody-enzyme (horseradish peroxidase) conjugate solution. The test sample is allowed to react simultaneously with the two antibodies, resulting in the TSH molecules being sandwiched between the solid phase and enzyme-linked antibodies. After a 60-minute incubation at room temperature, the wells are washed with water to remove unbound labeled antibodies. A solution of TMB Reagent is added and incubated for 20 minutes, resulting in the development of a blue color. The color development is stopped with the addition of Stop Solution, changing the color to yellow. The concentration of TSH is directly proportional to the color intensity of the test sample. Absorbance is measured spectrophotometrically at 450 nm.

Reagents

Materials provided with the kit:

- ☐ Murine Monoclonal Anti-TSH-coated microtiter wells.
- ☐ Set of Reference Standards: 0, 0.5, 2, 5, 10 and 25 μ IU/ml, lyophilized.
- ☐ Enzyme Conjugate Reagent, 13 ml.
- ☐ TMB Reagent (One-Step), 11 ml.
- ☐ Stop Solution (1N HCl), 11 ml.

3- Thyroxine (T4)

Test principle

The Access Total T4 assay is a paramagnetic particle, chemiluminescent, competitive binding enzyme immunoassay for the quantitative determination of total thyroxine (T4) in human serum, using the Access Immunoassay System. A sample is added to a reaction vessel with anti-thyroxine antibody, thyroxine-alkaline phosphatase conjugate, and paramagnetic particles coated with goat anti-mouse capture antibody and a stripping agent to dissociate all T4 from serum-binding proteins. Thyroxine in the sample competes with the thyroxine-alkaline phosphatase conjugate for binding sites on a limited amount of specific anti-thyroxine antibody. Resulting antigen: antibody complexes bind to the capture antibody on the solid phase. Separation in a magnetic field and washing removes materials not bound to the solid phase. A chemiluminescent substrate, Lumi-Phos 530, is added to the reaction vessel and light generated by the reaction is measured with a luminometer. The photon production is inversely proportional to the concentration of T4 in the sample. The amount of analyte in the sample is determined by means of a stored calibration curve.

Total T4 measurements are used alone, or in conjunction with other thyroid tests, in the diagnosis and treatment of thyroid disorders. Elevated levels of T4 occur in Graves' disease, subacute thyroiditis, toxic nodule or secondary hyperthyroidism. Decreased levels occur in primary hypothyroid diseases such as Hashimoto's

thyroiditis and neonatal hypothyroidism due to defects at the hypothalamic-pituitary level.

4- Total Triiodothyronine, Total T3

Test principle

Access Total T3 Assay is a paramagnetic particle, chemiluminescent immunoassay for the quantitative determination of triiodothyronine levels in human serum and plasma using the Access Immunoassay Systems. The Total T3 Assay is a competitive binding immunoenzymatic assay. Sample is added to a reaction vessels with a stripping agent to dissociate T3 from the binding proteins. T3 in the sample competes with the T3 analogue coupled to biotin for anti-T3 alkaline phosphatase conjugate. Of the resulting antigen: antibody complexes, the T3 analogue: antibody complexes are bound to the streptavidin coated solid phase. Separation in a magnetic field and washing removes the sample T3; antibody complexes and other materials not bound to the solid phase. A chemiluminescent substrate, Lumi-Phos[®] 530, is added to the reaction vessel and light generated by the reaction is measured with a luminometer.

The light production is proportional to the amount of enzyme conjugate bound to the solid support. The amount of analyte in the sample is determined by means of a stored, multi-point calibration curve. Total T3 measurements are used in the diagnosis of thyroid disorders. Elevated concentrations of T3 can be found in Grave's disease, and most other classical causes of hyperthyroidism. Decreased concentrations occur in primary hypothyroid diseases such as Hashimoto's thyroiditis and neonatal hypothyroidism or secondary hypothyroidism due to defects at the hypothalamo-hypophyseal level.

Statistical analysis

The statistical analysis proceeded in all groups of study, descriptive statistics analyzed by using one way ANOVA (analysis of variations) test with LSD (least significant difference) were performed using mean and standard error (S.E) for continuous variables (p -value ≤ 0.05) was considered to be significant. All analyses were performed with statistical Package for the social sciences SPSS for Windows (version 23.0 SPSS Inc, Chicago, 111).

Result

It is noticed in table 1 ferritin level recorded highest in age category (26-35) while lowest in age category (46-55) Where there are significant differences between the two age groups, As for the Hb highest level was age category (26-35) while lowest in age category (46-55). (T3 and T4) highest level in age category (26-35) and lowest level age category (36-45, As for TSH level highest in age category (25>) while lowest in age category (36-45).

Table 1: levels TSH, T3, T4, Ferritin, Hb with Age categories in Women

Parameters	Age group	Mean	Std. Error	L.S.D
Ferritin	25 >	34.00 ^a	3.80	6.17
	26 - 35	34.07 ^a	4.25	
	36 - 45	30.57 ^a	5.65	
	46 - 55	15.33 ^b	2.84	
Hb	25 >	10.58 ^{ab}	0.45	0.72
	26 - 35	10.89 ^a	0.53	
	36 - 45	10.0 ^b	0.53	
	46 - 55	8.66 ^c	0.33	
T3	25 >	1.96 ^{ab}	0.26	0.52
	26 - 35	2.16 ^a	0.39	
	36 - 45	1.57 ^b	0.35	
	46 - 55	1.63 ^b	0.49	
T4	25 >	82.04 ^{ab}	15.27	14.83
	26 - 35	95.09 ^a	9.98	
	36 - 45	80.22 ^b	13.06	
	46 - 55	85.44 ^{ab}	28.86	
TSH	25 >	4.32 ^a	1.33	1.41
	26 - 35	3.04 ^{ab}	0.90	
	36 - 45	1.94 ^b	1.06	
	46 - 55	2.18 ^b	2.0	

*Results represent mean $\pm$ Standard Error(S.E)

* Means having different letters in the same column differed significantly (P $\leq$ 0.05).

(L.S.D) least significant difference.

Table 2 noticed T3 and T4 hormone level recorded the highest level in women with hyperthyroidism and lowest with hypothyroidism in comparing with the control group level. As for TSH hormone the highest level with hypothyroidism and lowest with hyperthyroidism in comparing with the control group level.

Table 2: level thyroid hormones in control hyperthyroidism and hypothyroidism in women

Parameters	Groups	Mean	Std. Error	L.S.D
T3	Control	1.72 ^b	0.12	0.53
	Hypo	1.22 ^b	0.19	
	Hyper	2.47 ^a	0.20	
T4	Control	79.72 ^b	8.94	10.01
	Hypo	56.95 ^c	5.84	
	Hyper	110.46 ^a	7.49	
TSH	Control	2.37 ^b	0.43	0.57
	Hypo	5.79 ^a	0.56	
	Hyper	0.19 ^c	0.05	

It is noticed Fig1 that Relationship between thyroid hormone, ferritin, Hb and categories age for age less than 25 years was T4 highest 82.04 while T3 was

lowest 1.96. while for age between (26-35) was T4 hormone highest 95.09 and lowest T3 hormone 2.16, but for women whose age were between (36-45) was T4 hormone highest 80.22 and lowest T3 hormone 1.57. while T4 hormone highest 85.44 and T3 hormone lowest 1.63 for age between (46-55). Ferritin level was lowest in age category (46-55) was 15.33. As Hb level was lowest in age category (46-55) was 8.66. T3 hormone was lowest in age category (46-55) was 1.63. age categories (36-45) lowest in T4 hormone level was 80.22. TSH hormone level recorded lowest in age between (36-45) was 1.94.

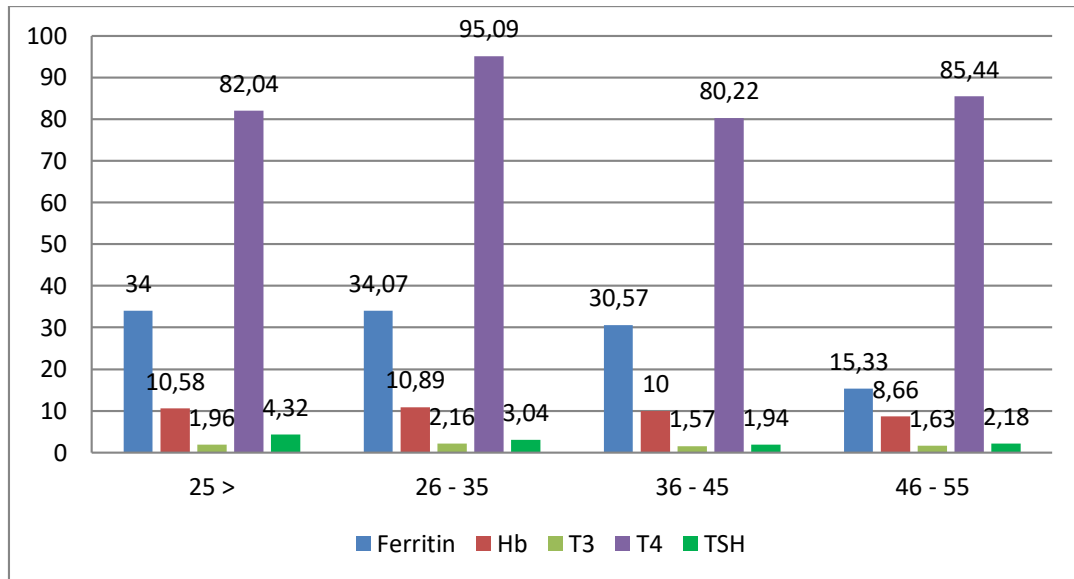


Fig 1: Relationship between thyroid hormone, ferritin, Hb and categories age

It can be observed that TSH level the highest with hypothyroidism was 5.79 and lowest hyperthyroidism 0.19 comparing with control group 2.37 (fig 2).

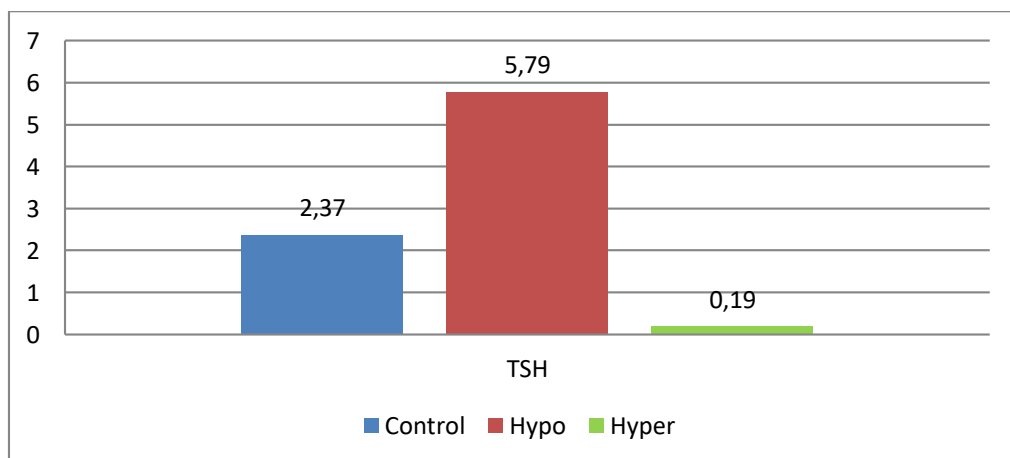


Fig 2: show TSH level in control hyperthyroidism and hypothyroidism in Pregnant and non-pregnant women

Show Fig 3 T3 level was the highest in pregnant 2.46 while in non pregnant was 1.48.

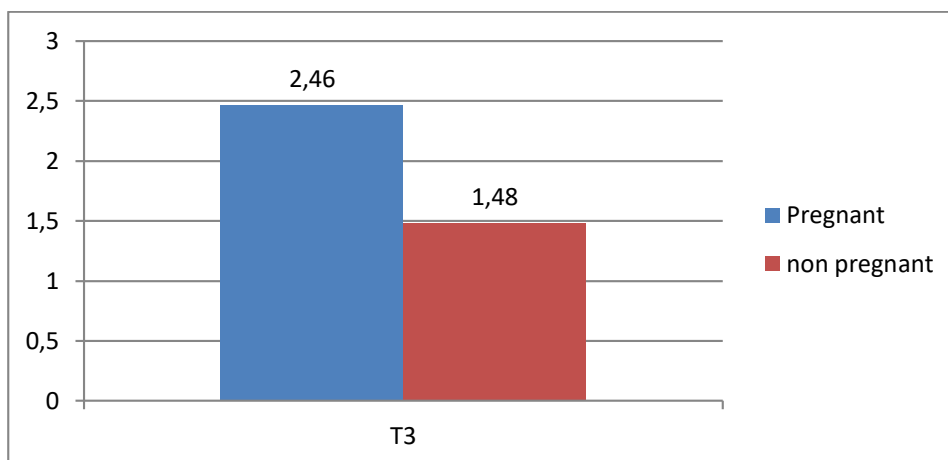


Fig 3: show T3 level in Pregnant and non-pregnant women

Show Fig 4 T4 level was the highest in pregnant 119.74 while in non pregnant was 65.69

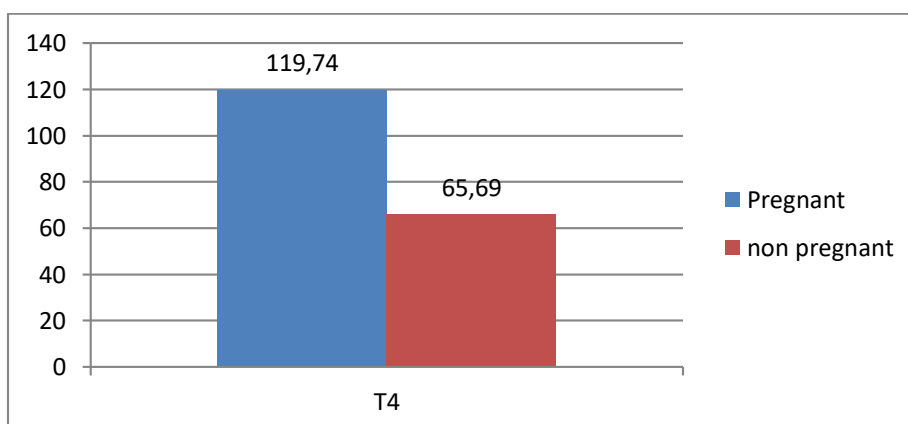


Fig 4: show T4 level in control hyperthyroidism and hypothyroidism in Pregnant and non-pregnant women

Discussion

Hypothyroidism (Hashimoto's thyroiditis) and hyperthyroidism (Graves' disease) are the most common thyroid dysfunction consequences (Swain et al, 2005) (10). Through the current study, the Patient of women with hypothyroidism with T3 and T4 is more than hyperthyroidism in relation to the study sample, while TSH level the highest with hyperthyroidism.

Also, it was found that women whose ages between (25≥) and (26-35) years were more likely to have thyroid disorders than the total of the sample, This contradicts with (Falgoos and Abdulredha, 2020) (11). The high prevalence of

thyroid disorders around the world, including in Iraq, can be linked to a lack of iodine (Al Shahrani et al, 2016)(12), in addition to other influencing elements such as pollution and hereditary abnormalities, which might be viewed as key influencing factors as well (Hamasaheed, 2019)(13).

When compared to the control group, the level of TSH increased in women suffering from hypothyroidism, while it was lowest in women suffering from hyperthyroidism, according to the findings of the current study. When it came to T4 and T3, women with hyperthyroidism had high levels, but women with hypothyroidism had lower levels when compared to the control group. The level of TSH rises when T4 and T3 levels fall. This result was in line with the previous one of Elzobir et al (2014)(14), study who found an increase in the level of TSH in patients with hypothyroidism. The high level of TSH and the decrease in the levels of T4 and T3 are the important indicators of thyroid tests that confirm a person has hypothyroidism (Sibia et al, 2018)(15). In the current study, the ferritin level decreased in the age group (46-55), while T3 and T4 was higher in the age group (26-35), but TSH increased in women less than 25 year, this Contrasting to study (Sachdeva et al, 2015)(16)

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