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Study the role of inhibin b, TPO antibody, and thyroid hormones in patients with hyper and hypothyroidisim

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ABSTRACT: Hyper- and hypothyroidism have significant effects on the female reproductive system. However, little in the way of data is available on the relationship between ovarian paracrine control and thyroid function. The present study aimed to measure some of hormonal and biochemical parameters in women suffered from hyper and hypothyroidism, The experiment began on September 3, 2023, and continued until August 2, 2024, during which 90 cases of thyroid patients aged 15-90 years were obtained. All studied cases were examined by specialist doctors in the lobbies and laboratories of Tikrit General Hospital, relying on the necessary laboratory tests. The levels of Triiodothyronine (T3), Thyroxine (T4), and thyroid-stimulating hormone (TSH), were used as a criterion for diagnosing patients with hyperthyroidism and hypothyroidism. Hypothyroidism. The experiment samples were classified into three groups. The first is the **healthy group** consisted of 20 people, second group (patients), consisted of 35 sick people after confirming that they suffer from **hypothyroidism**. Third group of patients consisted of 35 sick people after it was confirmed that they had **hyperthyroidism**. The results of present study show levels of **TSH** had a significant differences (P>0.05) in the three different groups, for hypothyroid patients at a rate of 15.700 ± (5.270), in hyperthyroid patients at a rate of 0.275 ± (0.194) compared to healthy individuals at a rate of 2.402 ± (0.943), **T3** levels showed significant differences (P>0.05) in the three different groups, hypothyroidism patients at a rate of 15.354) ± (3.497), hyperthyroidism patients at a rate of 22.401) ± (1.959) compared to healthy people at a rate of 18.336) ± (1.011), The results of T4 showed significant differences (P>0.05) in the three different groups, hypothyroidism patients at a rate of 15.354) ± (3.497), hyperthyroidism patients at a rate of 22.401) ± (1.959) compared to healthy people at a rate of 18.336) ± (1.011, **inhibin b** showed significant differences (P>0.05) in the three different groups, hypothyroidism patients was 448.23 (± 29.33), for hyperthyroidism patients was 660.50 (± 38.50) compared to healthy subjects was 531.76 (± 22.75), **The Thyroperoxidase(TPO)** results showed significant differences (P>0.05) in the three different groups, for hypothyroidism patients was 3913 ± (454.7). for hyperthyroidism patients was 4907 ± (300.8) compared to healthy individuals was 2905 ± (214.0), The thyroglobulin (Tg) results showed significant differences (P>0.05) in the three different groups, for hypothyroidism patients was 4073.0 ± 318.0, for hyperthyroidism patients was 3337.3 ± 279.8 compared to healthy subjects was 2737.9 ± 211.0. In this study, negatively correlated between inhibin B with TSH, T4, T3, and Tg. TPO is positively correlated with hyperthyroidism, negatively correlated with TPO, Tg, and TSH, and positively correlated with T3 and T4 in hypothyroidism

Keywords: Hypothyroidism, hyperthyroidism, inhibin b

1. INTRODUCTION

The thyroid gland is an endocrine gland located in the neck under the Adam's apple. It takes the shape of a butterfly and produces thyroid hormones that regulate various vital activities in the body (1). It is considered the only gland that produces, stores and secretes thyroid hormones, which include

Triiodothyronine (T3) and Thyroxine (T4) (2, 3). Thyroid hormones play an important role in the processes of growth, differentiation, metabolism, and reproduction. The secretion of these hormones is regulated by thyroid-stimulating hormone (TSH), which is secreted from the anterior lobe of the pituitary gland (4). When the production of thyroid hormones increases, the pituitary gland produces what is known as hyperthyroidism. When the level of these hormones in the blood decreases, it results in what is known as hypothyroidism (5). Hyperthyroidism is an elevated level of thyroid hormones in the tissues. The primary causes of hyperthyroidism include Graves' disease, toxic multinodular goiter, and toxic adenoma. The predominant cause of increased negative secretion of thyroid hormones is painless thyroiditis (6, 7). Painless thyroiditis is the most prevalent cause of excessive negative secretion of thyroid hormones. This condition often occurs without any symptoms. Hypothyroidism is one of the disorders that appears most frequently all over the world. Hashimoto's disease is the most common cause of this condition (8).

On the other hand, hyperthyroidism can lead to weight gain. This is due to increased appetite, increased energy expenditure, and loss of muscle mass. Other common symptoms of hyperthyroidism include nervousness, irritability, excessive sweating, and heart palpitations. People with hypothyroidism may show a tendency to gain weight. This is partly due to a decreased basal metabolism, which results in a decreased calorie burn in the body. Common symptoms of hypothyroidism also include fatigue, constipation, dry skin, and increased sensitivity to cold (9). Many patients who have thyroid illness may not receive a diagnosis since the symptoms of the condition develop gradually and are not very specific. In spite of the fact that screening for thyroid disease seems to be suitable, universal screening has not yet been universally adopted due to a lack of clinical trials (10).

Early development of the thyroid antibody, Anti-Thyroperoxidase (anti-TPO), prior to the commencement of thyroid hormone imbalance; hence, the use of anti-TPO in conjunction with standard thyroid indicators, consisting of TSH and FT4, would help reduce long-term morbidity and the related health concerns (10). The thyroid gland is the primary organ responsible for the production of thyroglobulin (Tg), which is a homogenous glycoprotein. In addition to its role as a substrate for the production of thyroxine and triiodothyronine, it also plays a role in the storage of inactive forms of thyroxine and iodine. The endoplasmic reticulum is responsible for the secretion of thyroxine to its iodine site, which is followed by the manufacture of thyroxine in the follicular lumen (11). Since blood is the most affected body fluid in abnormal conditions that occur inside the body, the present study aimed to show the extent of the impact of evaluating the role of inhibin B with some antibodies on some blood parameters that represent the group of patients and compare them with healthy people who represent the control group.

2. MATERIALS AND METHODS

2.1 Experience Design

The experiment started on September 3, 2023, and continued until August 2, 2024, during which 90 cases of thyroid patients were obtained, ranging in age from 15-90 years. All the studied cases were examined by specialist doctors in the wards and laboratories of Tikrit General Hospital, based on the necessary laboratory tests, as the level of TSH, FT4, and FT3 were used as a standard for diagnosing patients with hyperthyroidism and hypothyroidism. The experiment samples were classified into three unequal groups; the first is the healthy group (Control Group) and represents the control group, as blood samples were taken from 20 people after ensuring that they were in good health and did not suffer from health problems. The second is the patient's group (Patients Group) and represents the infected group, as blood samples were taken from 35 sick people after ensuring that they were suffering from hypothyroidism (hypothyroidism). The third group is the Patients Group, which represents the group of patients. Blood samples were taken from 35 patients after confirming that they were suffering from hyperthyroidism, and specialist doctors diagnosed them in the hospital after conducting clinical and laboratory tests on them.

2.2 Collection of Blood Samples

Blood samples were drawn from the vein using a sterile medical syringe with a capacity of 5 ml, and the blood samples were deposited in gel tubes and maintained at room temperature for roughly thirty minutes until coagulation ensued. A centrifugation operation was conducted at 3000 rpm for 10 minutes to isolate the serum. The material was allocated into appendroff tubes and subsequently preserved at -20°C until required for the relevant testing.

2.3 Ethical approval

The study was carried out by the ethical principles outlined in the Declaration of Helsinki. The study was performed following the acquisition of both verbal and written consent from the patients before collecting the samples, this case-control study was approved by the Scientific Committee of the Department of Biology, College of Science, Tikrit University (3/7/5328 in 26-11-2023).

2.4 Measurement of Inhibin b, Triiodothyronine (T3), Thyroxine (T4), thyroid-stimulating hormone (TSH), thyroid-peroxidase (TPO), and thyroglobulin (Tg)

The levels of TSH, T3, T4, Inhibin b, TPO, and Tg were performed using the sandwich ELISA technique (Biotech, China). The plate was pre-coated with antibodies specific for human Inhibin b,

TSH, T3, T4, TPO, and Tg. The sample was supplemented with TSH, Inhibin b, T3, T4, TPO and Tg. The concentration of human Inhibin b, TSH, T3, T4, TPO, and Tg showed a positive correlation with the appearance of color in the substrate solution. Stopping the process was achieved by introducing an acidic stop solution, followed by quantification of the absorbance at a wavelength of 450 nm.

2.5 Statistical Analysis

The results of the study were statistically analyzed using the statistical program (Minitab ver. 17) to detect the studied variables (physiological blood parameters). The differences between the healthy and sick groups were measured using the Analysis of Variance (ANOVA) test. The arithmetic means were compared, and significant differences were determined according to Duncan's Multiple Range test at a probability level of ($P \leq 0.05$). The P-value and Pearson correlation coefficient were extracted and relied upon to find the extent of the relationship between the studied variables between the healthy and sick study groups at a significance level of ($P \leq 0.01$ and ($P \leq 0.05$) (12).

3. RESULTS AND DISCUSSION

Of the total number of men (90 people), the number of patients was 70. In contrast, the number of patients was divided equally into two groups: those suffering from hypothyroidism and hyperthyroidism and a healthy group of 20 who did not suffer from any disease.

TSH results showed significant differences ($P>0.05$) in the three different groups, as we noticed a significant increase ($P>0.05$) for hypothyroid patients at a rate of 15.700 ± (5.270). In contrast, the results showed a significant decrease ($P>0.05$) for hyperthyroid patients at a rate of 0.275 ± (0.194) compared to healthy individuals at a rate of 2.402 ± (0.943) Figure (3)

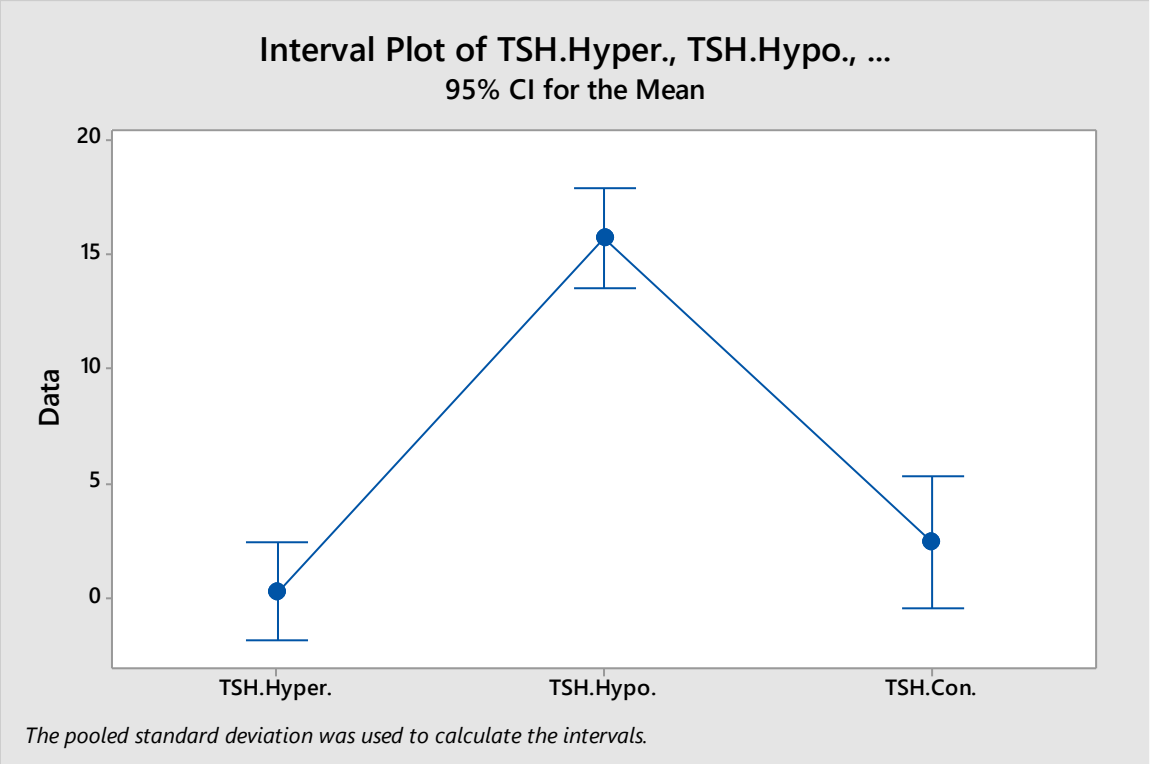

Figure (1) shows the concentrations of pg/ml TSH in the studied categories.

The results of T3 showed significant differences (P>0.05) in the three different groups, as we noticed a significant decrease (P>0.05) for hypothyroidism patients at a rate of 15.354) ± (3.497). In contrast, the results showed a significant increase (P>0.05) for hyperthyroidism patients at a rate of 22.401) ± (1.959) compared to healthy people at a rate of 18.336) ± (1.011) Figure (4)

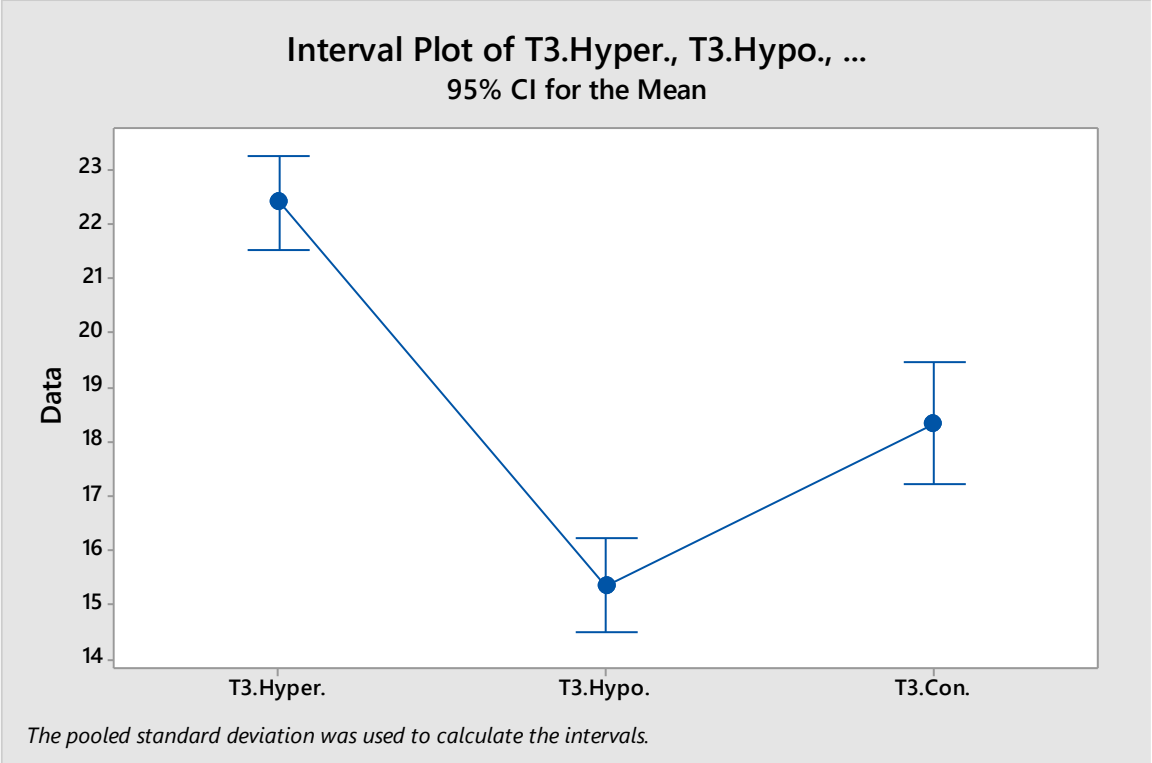

Figure (2) shows the concentrations of pg/ml T3 in the studied categories.

The results of T4 showed significant differences (P>0.05) in the three different groups, as we noticed a significant decrease (P>0.05) for hypothyroidism patients at a rate of 15.354) ± (3.497). In contrast, the results showed a significant increase (P>0.05) for hyperthyroidism patients at a rate of 22.401) ± (1.959) compared to healthy people at a rate of 18.336) ± (1.011 Figure (4)

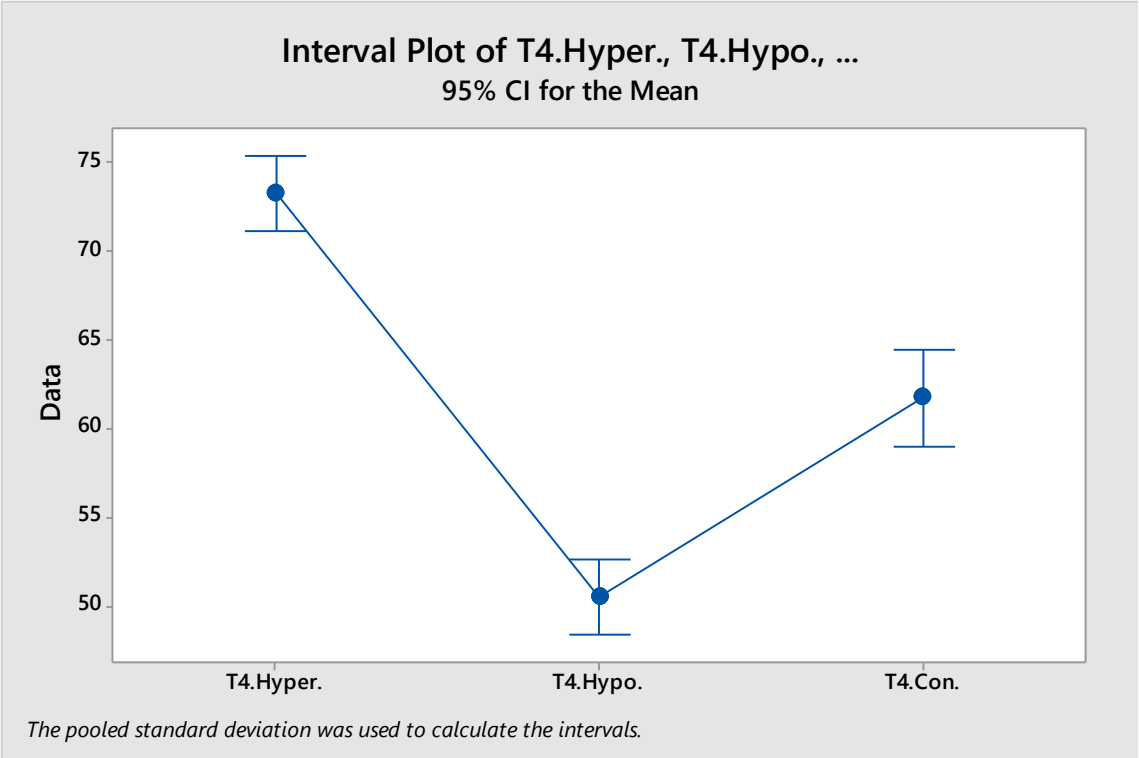

Figure (3) Shows the concentrations of pg/ml T4 in the studied categories.

Table (1) Average concentrations of TSH, T3, and T4 in the serum of men with thyroid diseases and healthy men.

Groups	TSH	T3	T4
Hyperthyroidism	0.194±0.275 b	1.959±22.401 a	7.840 ±73.260 a
Hypothyroidism	5.270±15.700 a	1.011±18.336 b	5.851 ±50.501 c
Healthy	0.943±2.402 b	3.497±15.354 c	2.470 ±61.732 b
p-valut	0.01**	0.01**	0.01**

The results of inhibin b showed significant differences (P>0.05) in the three different groups, as we noticed a significant decrease (P>0.05) for hypothyroidism patients at a rate of 448.23 (± 29.33). In contrast, the results showed a significant increase (P>0.05) for hyperthyroidism patients at a rate of 660.50 (± 38.50) compared to healthy subjects at a rate of 531.76 (± 22.75) in Figure 4 and Table 1.

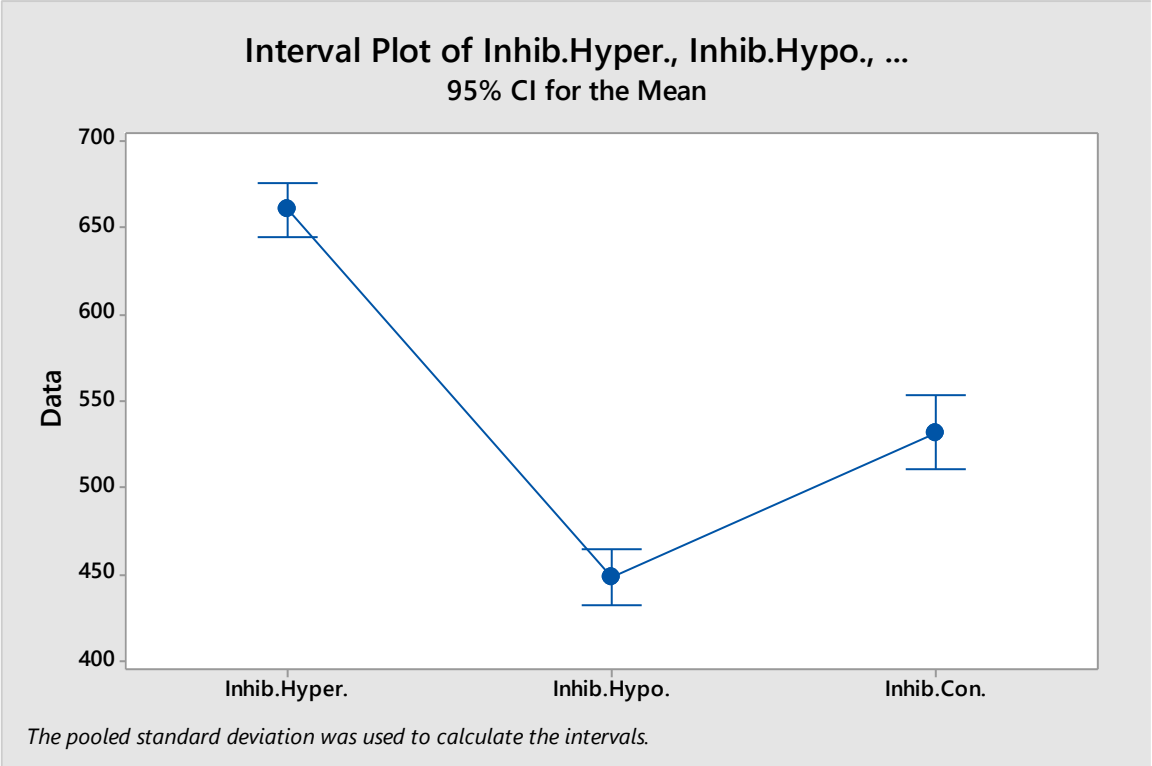

Figure (4) Shows the concentrations of pg/ml inhibin B in the studied categories.

The TPO results showed significant differences (P>0.05) in the three different groups, as we noticed a significant decrease (P>0.05) for hypothyroidism patients at a rate of 3913 ± (454.7). In contrast, the results showed a significant increase (P>0.05) for hyperthyroidism patients at a rate of 4907 ± (300.8) compared to healthy individuals at a rate of 2905 ± (214.0) in Figure 5 and Table 1.

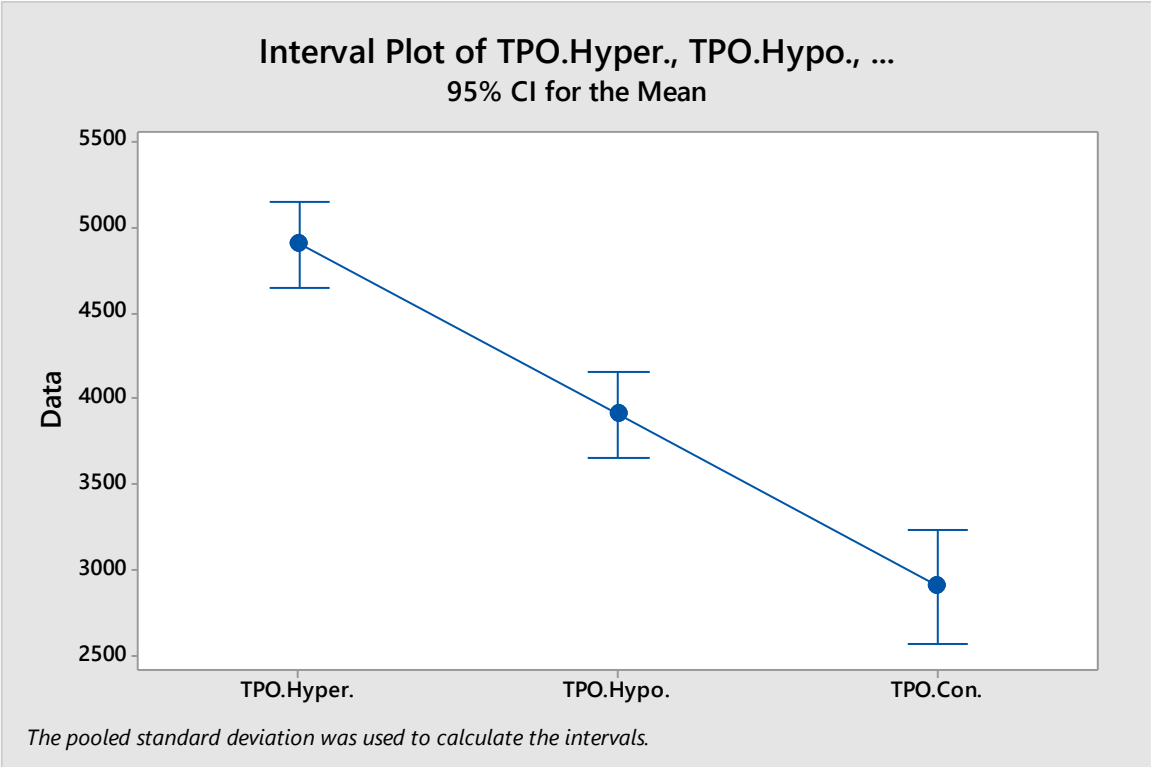

Figure (5) shows the concentrations of pg/ml TPO in the studied categories.

The Tg results showed significant differences (P>0.05) in the three different groups, as we notice a significant increase (P>0.05) for hypothyroidism patients with a rate of 4073.0 ± 318.0, while the results showed a significant decrease (P>0.05) for hyperthyroidism patients with a rate of 3337.3 ± 279.8 compared to healthy subjects with a rate of 2737.9 ± 211.0 in Figure 6 and Table 1.

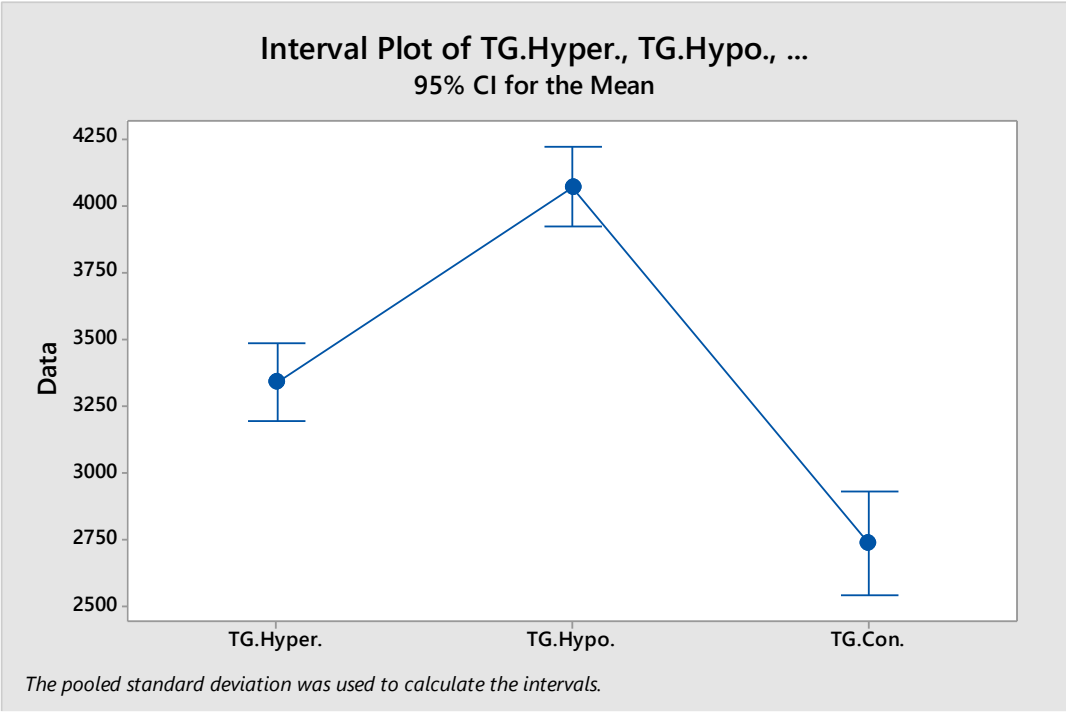

Figure (6) Shows the concentrations of pg/ml Tg in the studied categories.

Table (2) Average concentrations of Inhibin b, TPO, and Tg in the sera of men with thyroid diseases and healthy men.

Groups	Inhibin b	TPO	Tg
Hyperthyroidism	38.50 ±660.50 a	300.8 ±4907 a	279.8±3337.3 b
Hypothyroidism	29.33 ±448.23 c	454.7 ± 3913 b	318.0 ±4073.0 a
Healthy	22.75 ±531.76 b	214.0 ±2905 c	211.0±2737.9 c
p-valut	0.01**	0.01**	0.01**

These results agree with the TSH hormone. These results agree with (12, 13) that a high TSH hormone level in hypothyroidism patients will be a compensatory response to offset low levels of thyroid hormones in an effort to achieve a state of equilibrium. One possible cause of low thyroid hormones is the presence of certain disorders that have an effect on the gland and cause it to lose its efficiency. The hypothalamus is responsible for the production of thyrotropin-releasing hormone (TRH) as a consequence of this shortage. After that, it makes its way to the pituitary gland, where it stimulates the thyrotropin cells in the anterior lobe to boost the production of TSH. This is the reason why our study found a large rise in TSH in comparison to T3 and T4 levels.

These findings are in agreement with the findings of Hamasaeed *et al.*,(12), as well as AMMAR and his group in 2018, which demonstrated that a high TSH hormone level in hypothyroidism patients will be a compensatory response to offset low levels of thyroid hormones in an effort to achieve a state of equilibrium. One possible cause of low thyroid hormones is the presence of certain disorders that have an effect on the gland and cause it to lose its efficiency. The hypothalamus is responsible for the production of thyrotropin-releasing hormone (TRH) as a consequence of this shortage. After that, it makes its way to the pituitary gland, where it stimulates the thyrotropin cells in the anterior lobe to boost the production of TSH. This is the reason why our study found a large rise in TSH in comparison to T3 and T4 levels.

The results of inhibin b are in agreement with those of (14), Women with infertility and hypothyroidism exhibit diminished inhibin b levels and an increased propensity for obesity relative to healthy women. Inhibin b levels exhibit high sensitivity and specificity, warranting more frequent assessment in reproductive clinics.

Inhibin b levels did not appear to be significantly correlated with the patient's age or body mass index. This has already been seen in males with hypothyroidism, where it was found to be approximately half that of healthy males. These findings are consistent with our study, even if the

sex of the patients differed (15). Hypothyroidism and hyperthyroidism negatively affect the synthesis of reproductive hormones, follicle growth and development, and female reproductive fertility (16, 17). Polycystic ovary syndrome (PCOS) and thyroid diseases are intricately linked, and their concurrent presence may indicate patients at elevated reproductive and metabolic risk. Besides the established effects of thyroid-stimulating hormone receptor antibodies on fibroblasts in Graves' disease, the varied actions of thyroid antibodies align with disparities in the cellular localization of antigens, the specificity of circulating antibodies, the duration of antibody exposure, and the immune mechanisms involved in thyroid disease and Hashimoto's thyroiditis (18). The presence of antibodies to the enzyme thyroid peroxidase in the blood may indicate thyroid disease due to an immune system condition called Hashimoto's disease. In Hashimoto's disease, the immune system produces antibodies that mistakenly attack healthy tissue. The antibodies that attack the thyroid gland can cause swelling or inflammation of the gland (19). Overstimulation of the immune system may result in the inappropriate production of autoantibodies (20). Iodine sufficiency is associated with increased thyroid autoimmunity in iodine-sufficient areas. Some studies have reported that excessive iodine intake is a predisposing factor for thyroid dysfunction.

Studies from areas with high dietary iodine intake in Africa (21) and Japan (22) have reported increased TPO antibodies. Therefore, it is best to consume iodine under close control of the health care system and people's dietary habits, such as salt consumption (21). The presence of anti-TPO antibodies is associated with an increased risk of overt hypothyroidism (23, 24). Anti-TPO serves as a sensitive indicator of autoimmune thyroid illness. A study in Norway demonstrated a correlation between anti-TPO levels and both low and high TSH concentrations within the normal range (25). A significant correlation existed between thyroid illness and immunoglobulin E, the production of specific IgE against thyroid peroxidase (TPO), and blood IgE levels. IgE antibodies are commonly associated with autoimmune thyroid disease (26). Our study results agree with Can be elevated in people with Hashimoto's thyroiditis

Very low Tg values Thyroglobulin level may be an indicator of thyroid stability and successful thyroid treatment (27) Graves' disease is the predominant etiology of hyperthyroidism (28) With advancing age, the thyroid gland experiences increasing fibrosis and atrophy, leading to a reduction in its size, hence complicating palpation. The incidence of autoantibodies escalates with age and may partially account for the structural alterations in the thyroid gland (29, 30)

4. CONCLUSION

In this study, statistical significance was found. The correlation was made for the tests, and the correlation between them was reached, as inhibin B is negatively correlated with TSH, T4, T3, and Tg. TPO is positively correlated with hyperthyroidism, negatively correlated with TPO, Tg, and TSH, and positively correlated with T3 and T4 in hypothyroidism.

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