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The role of anti-TPO as an additional analytical marker in thyroid disease patients in Erbil City

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Abstract

Background: There is a connection among anti-TPO and anti-TG antibodies and levels of thyroid hormone and both alone or in grouping have been utilized to expect perfection of hypo or hyperthyroidism.

Objective: Both alone or in grouping have been utilized to expect perfection of hypo or hyperthyroidism.

Patients and Methods: This is a cross-sectional study achieved in the laboratories of Rizgary Teaching Hospital and Erbil Teaching Hospital in Erbil, Iraq, within the period of May 2020 to April 2021. The study included 66 patients, both males and females included. These patients had clinical indications and were suspected of having a sort of thyroiditis. Anti-TPO electrochemiluminescence immunoassay analysis has been included by the analysts in conjunction with conventional markers immunoassay of thyroid TSH, T3, and Free T4 trusting that this would help in lessening morbidity and related wellbeing concerns.

Results: There was an increase in the level of anti-TSH antibodies in the hyperthyroidism group (60.6%) which was more than in those with hypothyroidism (36.4%), significant association exist (P≤0.049). Moreover, the level of Anti-TPO Abs was higher among hypothyroidism patients (63.6%) than among hyperthyroidism cases (33.3%), this association was statistically significant (P≤0.014).

Conclusion: Demonstrating the clinical importance of this antibody and the benefit of adding anti-TPO, in combination with TSH and FT4. Addition of one test could potentially save expenditure on long-term diseases such as overt thyroid disease and its attended morbidities.

Keywords: Thyroid autoantibodies, thyroid peroxidase, thyroglobulin, thyroid-stimulating hormone receptor

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Introduction

Thyroid organ has one of the foremost imperative functions in individual body because it regulates common physiological activities of the body. Thyroid hormones thyroxine (T4) and triiodothyronine (T3) are hormones delivered from thyroid gland and they have various activities including metabolism, development, protein synthesis, and the control of numerous other vital hormones. Dysfunction of the thyroid gland can influence the generation of thyroid hormones (T3 and T4) which can be

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connected to several pathologies throughout the body [1].

Thyroid dysfunction, which is described as a variety of conditions related to the thyroid gland has a significant negative impact on people's ability to prosper. In the United States, approximately 20 million people have thyroid illness in some form[1]. Functional thyroid dysfunction is often categorized as either hypothyroidism or hyperthyroidism, which are further split into overt and covert conditions[2].

Since symptoms of thyroid disease are vague and progress gradually, many sufferers go untreated. Evidence regarding the efficacy of treatment in patients found by screening to have subclinical thyroid dysfunction is inconclusive. No trials of treatment of subclinical hyperthyroidism have been done [3,4].

Antibodies against thyrocytes' transmembrane protein involved in the production of thyroid hormone, known as anti-thyroid peroxidase (TPO). The precursor to thyroid hormone thyroglobulin is attacked by anti-thyroglobulin (TG) antibodies. Since these antibodies are present in more than 90% of instances of Hashimoto's thyroiditis and more than 80% of cases of Graves' disease, anti-TPO antibodies, also known as anti-thyroid microsomal antibodies, and anti-TG antibodies are regarded as diagnostic for autoimmune thyroid diseases (AITDs) [5]. autoimmune thyroid diseases.

Because of their connection to thyroid stimulating hormone (TSH) levels, anti-TPO and anti-TG antibodies have both been employed separately or in combination to predict the beginning of hypo-/hyperthyroidism. Numerous studies have found a connection between changed levels of anti-thyroid antibodies and TSH and the development of hypothyroidism in euthyroid patients[6,7].Actually, a study linking anti-thyroid antibodies to the development of autoimmune thyroiditis came to the conclusion that cytotoxicity caused by complement was responsible [8].

As a result, their existence may really occur several years before the onset of obvious thyroid disease or abnormal tests of thyroid function. In anti-thyroid antibody persons, additional thyroid profile testing is very essential for making a timely diagnosis[9]. Studies on anti-thyroid antibodies in various patient populations, including SLE subsets, have varied (anti-TPO 54.76 percent) [10], patients with hepatitis C (anti-TPO 26.8%) [11], those who experience chronic urticaria (anti-TPO 57.4 percent and anti-TG 42.6 percent)[12]. However, the incidence of anti-thyroid antibodies and their relationship to the thyroid profile (TSH, T4, and T3) in the general population remain unknown. Building a correlation between anti-thyroid antibodies and thyroid profile testing may help identifying patients with exacerbated thyroid profiles who also need thyroid autoantibody testing to rule out a more serious autoimmune condition [13]. In light of this, we proposed the following hypothesis: Patients with an unbalanced thyroid profile had a higher prevalence of positive anti-thyroid antibodies[14].Therefore, their presence may actually precede progress of evident thyroid disease or disturbed thyroid function tests by several years.

Observing the results of lab tests of T3, free T4 hormone, TSH, anti- TPO, anti-TG, and

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anti-TSH, would identify those euthyroid subjects with potential risk of developing thyroid disease, prevent associated disease and long-term morbidity. Thus the researchers combined anti-TPO with conventional thyroid indicators. To lessen morbidity and associated health concerns.

Patients and Methods

Between May 2020 and April 2021, this cross-sectional study was carried out in the labs of Erbil Teaching Hospital and Rizgary Teaching Hospital in Erbil, Iraq. Sixty-six participants in age range 18-65 both males and females (16,50 respectively) participated in the trial; they all exhibited clinical symptoms and were thought to have some form of thyroid illness.

In individuals suspected of having thyroid-related illnesses, we looked at the results of thyroid hormone function, paying particular attention to TSH, T3, T4, anti-TG, anti-TSH, and thyroid peroxidase antibody (anti-TPO) titers. They were quantified using the (cobas e 411 analyzer), a completely automated analyzer that performs immunoassay analysis using patented electrochemiluminescence (ECL) technology.

The electrochemiluminescence immunoassay, or "ECLIA," developed by Roche to be used on the Electrosys and Cobas e immunoassay analyzers, was used to estimate TSH, T3, and T4. Range of measurement: 10.0-4000 IU/mL (defined by the lower detection limit and the maximum of the master curve).

The reported value for values below the lower detection limit is 10.0 IU/ml. Values that are higher than the measurement range are recorded as > 4000 IU/ml. The normal ranges for TSH, T4, and T3 are respectively 5.0 to 12.0 mU/dL, 80-220 ng/dl, and O.3-4.5 IU/ml.

Immunoassay for the in vitro quantitative measurement of antibodies to thyroglobulin in human serum and plasma is the intended usage of Elecsys Anti-TG. A tool for identifying autoimmune thyroid disorders is the anti TG determination. The Roche Elecsys and Cobas e immunoassay analyzers are intended for use with the electrochemiluminescence immunoassay, or "ECLIA."

Standard sampling tubes or tubes with separating gel, sodium heparin, K2 and K3 EDTA plasma are used to collect serum. Recovery within 85 to 115 percent of serum value, or slope 0.85 to 1.15 plus intercept within 0.95, is the criterion. Use of sodium citrate plasma or Li heparin is prohibited. 3 days at 28 °C and 1 month at -20 °C stable. range of 10.0-4000 IU/mL measurements (defined by the lower detection limit and the maximum of the master curve). Lower detection limit values are stated as 4000 IU/ml. anticipated values Studies using the Elecsys anti-TG assay were done at the 94th percentile level for Roche's Elecsys® Anti-TSHR, which is now the threshold value of 115 IU/mL.

The electrochemiluminescence immunoassay, or "ECLIA," is designed for use on Elecsys and Cobas e immunoassay analyzers by Roche. Elecsys Anti-TSHR (TRAK) is a fully automated test for the identification of autoantibodies to the TSH receptor. 0.8 to 40 (results below the LoD are given as 0.8 IU/L) is the measurement range. Limit: 1.75 IU/L.

The electrochemiluminescence immunoassay "ECLIA" is intended for use

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on (Elecsys) and (cobas e) immunoassay analyzers made by Roche. It is an immunoassay for the in vitro quantitative detection of antibodies to thyroid peroxidase in human serum and plasma (anti- TPO). Range of measurement: 5.00–600 IU/mL (defined by the lower detection limit and the maximum of the master curve). Anti-TPO has a normal range upper limit of >30 IU/ml [15].

A questioner regarding the age and weather they were taking any medications was included and filled by all patients. In this study we excluded the following groups; children, adolescents and pregnant women. The prevalence of unsuspected thyroid disease is lowest in men and highest in older women [4]. Another study should be done to assess the effects of different screening methods (and subsequent management) for thyroid dysfunction in pre-pregnancy and during pregnancy on maternal and infant outcomes. Every patient had this study explained to them, and their verbal consent—or that of their legal guardian—was obtained. Information privacy and secrecy were assured.

Statistical Analysis

The Factual Bundle for Social Sciences adaptation 26 was used to analyze the data (SPSS Inc., IBM Company, Chicago, Illinois, USA). Inferential results were contrasted between the individuals with different variables using a statistical significance level of 0.05, and when appropriate, Fisher's exact or Pearson Chi-square tests were used to examine the data. Descriptive analyses were expressed as frequencies and rates. The means and standard deviation of numerical data were examined.

Results

The total number of the studied sample was 66. The mean age ± SD of the studied sample was 39.05±13.73 years, mean ± SD of T3, T4, TSH, and Anti-TPO are 2.63±1.475, 118.48±79.105, 5.46±9.742, 91.35±121.108 respectively ranging from 47 to 65 years. The median was 44 years. Around two-thirds, 50 of the patients (62%) of the sample were females Table (1).

Table (10): Descriptive Statistics of the study variables

Variables	N	Range	Minimum	Maximum	Mean	Std. Deviation
Age	66	47	18	65	39.05	13.73
T3	66	5.41	1	6.41	2.63	1.47
T4	66	290.765	0.735	291.5	118.48	79.10
TSH	66	45.996	0.004	46	5.46	9.742
Anti-TPO	66	496	4	500	91.35	121.10

Around 40 patients (60.6%) had normal T3 levels while26 patients (39.4%) had abnormal T3 levels and among abnormal levels,14 patients(42.4%) had hypothyroidism while 12 patients (36.4%) had hyperthyroidism while 19 patients (57.6%) of those with normal T3 had hypothyroidism and 21 patients (63.6%)of normal T3 patients had hyperthyroidism with no significant difference and p-value was 0.614 Table (2).

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Table (2): T3 level among hyperthyroidism and hypothyroidism groups

T3	Groups		Total	Pearson Chi-square	
	Hypothyroidism	Hyperthyroidism			
abnormal	14 (42.4%)	12 (36.4%)	26 (39.4%)	p-value	0.614
normal	19 (57.6%)	21 (63.6%)	40 (60.6%)		
Total	33 (100%)	33 (100%)	66 (100%)		

Table (3) shows that 41 (62.1%) of patients with thyroid diseases had an abnormal level of T4 while 25 (37.9%)of patients had normal T4 levels and that 22 patients (66.7%) with hypothyroidism had abnormal T4 levels while 11(33.3%) of them had normal T4 level. Regarding those with hyperthyroidism 57.6% of them had abnormal T4 levels while 42.4% of them had normal T4 levels with no significant differences (p-value: 0.447).

Table (3): T4 level among hyperthyroidism and hypothyroidism groups

T4	Groups		Total	Pearson Chi-square	
	Hypothyroidism	Hyperthyroidism			
abnormal	22 (66.7%)	19 (57.6%)	41 (62.1%)	P-value	0.447
normal	11 (33.3%)	14 (42.4%)	25 (37.9%)		
Total	33 (100%)	33 (100%)	66 (100%)		

According to Table (4), 56 patients (84.8%) patients with thyroid diseases had abnormal TSH level 10 patients (15.2%) of patients had normal TSH level and 30 patients (90.9%) of those had hypothyroidism had abnormal TSH level and only 3 patients (9.1%) of them had normal level of TSH level and also 26 patients (78.7%) of those had hyperthyroidism had abnormal TSH level and 7 patients (21.2%) of them had normal TSH level with no significant difference (p-value: 0.170).

Table (4): TSH level among hyperthyroidism and hypothyroidism groups

TSH	Groups		Total	Pearson Chi-square	
	Hypothyroidism	Hyperthyroidism			
abnormal	30 (90.9%)	26 (78.8%)	56 (84.8%)	P-value	0.170
normal	3 (9.1%)	7 (21.2%)	10 (15.2%)		
Total	33 (100%)	33 (100%)	66 (100%)		

Table (5) revealed that although the level of ATG was higher in patients with hypothyroidism 9 patients (27.3%) in comparison with those with hyperthyroidism 8 patients (24.2%), but there was no significant statistical difference between ATG and study groups and p-value was 0.778.

Table (5): Distribution of ATG between study groups

ATG	Groups		Total	Pearson Chi-square	
	Hypothyroidism	Hyperthyroidism			
Positive	9 (27.3%)	8 (24.2%)	17 (25.8%)	P-value	p:0.778
Negative	24 (72.7%)	25 (75.8%)	49 (74.2%)		
Total	33 (100%)	33 (100%)	66 (100%)		

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Table (6) showed that the level of Anti-TSH was higher in those with hyperthyroidism 20 patients (60.6%) than those with hypothyroidism which was 12 patients(36.4%), this difference was statistically significant,chi-square test was done and the p-value was 0.049.

Table (6): Distribution of anti-TSH between study groups

Anti-TSH	Groups		Total	Pearson Chi-square	
	Hypothyroidism	Hyperthyroidism			
positive	12 (36.4%)	20 (60.6%)	32 (48.5%)	P-value	0.049
negative	21 (63.6%)	13 (39.4%)	34 (51.5%)		
Total	33 (100%)	33 (100%)	66 (100%)		

Table (7) showed that the level of Anti-TPO level was higher among hypothyroidism 21 patients (63.6%) than those with hyperthyroidism which was11 patients (33.3%), this difference association was statistically significant. Chi square test was done and p-value was 0.014.

Table (7): Distribution of anti-TPO between study groups

Anti-TPO	Groups		Total	Pearson Chi-square	
	Hypothyroidism	Hyperthyroidism			
positive	21 (63.6%)	11 (33.3%)	32 (48.5%)	P-value	p:0.014
negative	12 (36.4%)	22 (66.7%)	34 (51.5%)		
Total	33 (100%)	33 (100%)	66 (100%)		

Discussion

The most prevalent autoimmune diseases that affect the thyroid, known as autoimmune thyroid disorders (AITDs), are Hashimoto's thyroiditis and Graves' infection [16].

Despite the fact that only about 1% of people have AITDs, 15% of people with euthyroidism may have subclinical or focal thyroiditis and circulating antithyroid antibodies [17].

Direct assessment of blood concentration and Thyroid Stimulating Hormone (TSH) have been widely widely and appropriately used as a screening test. Unfortunately, the trend has been to rely on TSH measurements alone for the assessment of complicated thyroid disease [8].

In the current study, 62% of the participants were female, and the average age (± SD) of the sample under examination was 39.05±13.73 years. This is in agreement with a study by Meng *et al*. (2021), which found that women had a significantly higher overall incidence of hypothyroidism and hyperthyroidism than men did and that there was a significant tendency for the rate of hypothyroidism to increase with aging [19].

In the current investigation, we discovered that a normal T3 level was present in 60.6% of patients with autoimmune thyroiditis. In almost all cases of hyperthyroidism, the T3 rises and typically does so before the T4 increases. In order to confirm hyperthyroidism that a suppressed TSH has

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already shown, T3 levels are preferred, since it is a more sensitive sign of hyperthyroidism than total T4 levels. Additionally, T3 tests are useful for determining whether a patient has T3 thyrotoxicosis, a kind of hyperthyroidism that displays as extremely elevated T3 and suppressed TSH levels while maintaining (normal) T4 levels [20].

The cornerstone of thyroid function testing today is the serum thyroid stimulating hormone (TSH) concentration, which can be measured with an adequate sensitivity assay. For untreated groups of people that are at risk for primary thyroid dysfunction, a normal TSH concentration almost certainly rules out an anomaly. Nevertheless, serum TSH can provide a false signal of thyroid state in a number of crucial circumstances, most notably pituitary disorders and the early therapy of thyroid dysfunction [21].

In this study, the prevalence of anti-TPO was higher in hypothyroid subjects (63.6%) compared to hyperthyroid subjects (33.3%), with significant differences (p-value: 0.014). This finding was in agreement with a study by Bromiska et al They found that anti-TPO titers were above reference range values in individuals with hypothyroidism (60%) and euthyroid participants (31.4%) [22].

Despite the fact that only 1% of people have AITDs, 15% of people with normal thyroid function may have subclinical and localized thyroiditis and circulating antithyroid antibodies [23].

A transmembrane protein of thyrocytes involved in the manufacture of thyroid hormone, is the target of anti-thyroid peroxidase (TPO) antibodies. Antibodies to thyroglobulin, a precursor to thyroid hormone, are known as anti-thyroglobulin (TG) antibodies [24].

Since (anti-TPO antibodies) are present in more than 90% of cases of Hashimoto's thyroiditis and more than 80% of cases of Graves' infection, they are regarded as diagnostic of AITDs [25].

When compared to the combined control group, both subclinical/overt hypothyroidism and hyperthyroidism exhibited a significant number of participants who have anti-TPO before the onset of thyroid dysfunction [26 ,27].

Anti-TPO antibodies were more prevalent than anti-Tg antibodies in all outcomes, according to a study by Siriwardhane *et al*. They investigated whether these two indicators might be employed as standalone markers [28].

Microsomal antibody methods have been replaced by the TPO antibody radioimmunoassay, which is more sensitive and precise. Although anti-thyroglobulin assays are important for the accurate interpretation of serum thyroglobulin assays, they are less useful for the diagnosis of immunological thyroid disease [20, 30].

Anti-thyroid antibodies are more frequently present when TSH is out of range, according to research done by Mistry *et al*. These people should be tested for anti-thyroid antibodies. The fact that a considerable portion of older adults may have anti-thyroid antibodies makes screening even more crucial [31].

Conclusions

The current study verified the relationship between thyroid function test and anti-TPO antibody values, demonstrating the clinical importance of this antibody and

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recommending additional follow-up for those with high anti-TPO antibody titers in addition to a thorough and detailed clinical examination.

Recommendations

Our goal in doing this study was to demonstrate the advantage of using anti-TPO in addition to TSH and FT4 as a first-tier test. One additional test could potentially reduce costs associated with chronic conditions. Nevertheless, a large study should be performed especially on women of childbearing age, pregnancy, and cardiovascular diseases. In addition, involving other variables that can affect thyroid disease status such as current thyroid treatments and smoking habits with associated dysfunctions in reproductive health.

Source of funding: Kurdistan Higher Council of Medical Specialties.

Ethical clearance: This study is based on a cross-sectional analysis of identified laboratory data that had been got scientific and ethical approval from Kurdistan higher council of medical specialties. The data and materials in this manuscript have not been published elsewhere and are not under consideration by another journal.

Conflict of interest: The size of our sample dictates the amount of information we have and consequently, in part, determines the precision or level of confidence that we have in our sample estimates and therefore how valid and reliable our conclusions will be. The larger the sample size the more information we have and so our uncertainty reduces. Identifying and managing financial and non-financial conflicts is needed by the development of specific committees and potential empirical research will be important for defining which conflicts need to be better addressed and how to achieve this goal.

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دور مضادات بيروكسيداز الغدة الدرقية كعلامة تحليلية إضافية في مرضى الغدة الدرقية في مدينة اربيل

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الملخص

خلفية الدراسة: هناك علاقة بين الاجسام المضادة البيروكسيدا ز ومضادات ثايروغلوبيولين ومستويات هرمون المنبه الغدة الدرقية ، وقد تم استخدام كلاهما بمفرده أو في مجموعة لتوقعات في قصور الغدة الدرقية أو فرط نشاط الغدة الدرقية.

اهداف الدراسة: تم استخدام كلاهما بمفرده أو في مجموعة لتوقعات في قصور الغدة الدرقية أو فرط نشاط الغدة الدرقية.

المرضى والطرائق: هذه دراسة مقطعية تم أجرائها في مستشفى رزكاري التعليمي و مستشفى أربيل التعليمي في أربيل العراق. خلال الفترة من أيار ٢٠٢٠ الى نيسان ٢٠٢١.و شملت الدراسة ٦٦ مريضا من الذكور و الأناث. هؤلاء المرضى لديهم مؤشرات سريرية و يشتبه في أصابتهم بنوع من ألتهاب الغدة الدرقية.تم تضمين المقايسة المناعية المضادة للبيروكسيدايس من قبل المحللين بالتزامن مع المقايسة المناعية التقليدية للغدة الدرقية، هورمون المنبه للغدة الدرقية TSH مع هورمون الغدةالدرقية الحر T4,T3 و أن هذا من شأنه المساعدة في تقليل المرض و المخاوف المتعلقة بالرفاهية.

النتائج: كانت هناك زيادة في مستوى الأجسام المضادة لهورمون المنبه للغدة الدرقية في مجموعة فرط نشاط الغدة الدرقية (٦٠,٦%) والتي كانت أعلى من تلك الموجودة في مجموعة قصور الغدة الدرقية (٣٤,٤%) مع وجود دلالة أحصائية بفرق معنوي($P\leq0.049$) . بالأضافة ألى ذلك كان مستوى مضادات البيروكسيدايس أعلى بين مرضى قصور الغدة الدرقية٦٣,٦ % مقارنة بحالات فرط نشاط الغدة الدرقية و كان هذا الأرتباط ذو دلالة أحصائيةبفرق معنوي($P\leq0.014$) .

الاستنتاجات: أثبات الأهمية السريرية لهذا الجسم المضاد و فائدة أضافة مضاد البيروكسيدايس بالأشتراك مع هورمون الثايروويد الحر و يمكن أن يؤدي أضافة أختبار واحد الى توفير النفقات على الأمراض طويلة المدى مثل مرض الغدة الدرقية و الأمراض المصاحبة له.

الكلمات المفتاحية: الأجسام المضادة للغدة الدرقية ، بيروكسيداز الغدة الدرقية ، ثيروجلوبولين ،مستقبل الدرقية

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تاريخ قبول البحث: ٢١ آب ٢٠٢٢

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