

Diagnostic Value of Estimating the Ratio of Serum Free Triiodothyronine to Free Thyroxine in Differentiating Graves' Disease from Thyroiditis

Nesma A. Ibrahim^{1*} and Ahmed M. Hamam²

¹Department of Internal Medicine, Faculty of Medicine, Ain Shams University, Cairo, Egypt, and ²Department of Internal Medicine, Armed Forces College of Medicine (AFCM), Cairo, Egypt.

Abstract

Background: As the treatment approaches of Graves' disease (GD) and thyroiditis are different, the differential diagnosis is essential. **Aim:** To assess the diagnostic value of free triiodothyronine (FT₃) to free thyroxine (FT₄) ratio for differentiation between GD and destructive thyroiditis. **Subjects and Methods:** The present study was conducted on 100 newly diagnosed thyrotoxicosis patients; 61 with thyroiditis (group 1) and 39 with GD (group 2) and 20 healthy, age-matched subjects representing a control group (Group 3). All participants underwent medical history taking and clinical examination, serum levels of thyroid-stimulating hormone (TSH), FT₃, and FT₄ were measured, Technetium 99m thyroid Scan was done to all recruited patients. **Results:** The present results recorded significant increases in FT₃, FT₄, and decreases in TSH levels in Graves' disease group than in the thyroiditis group. There was a statistically significant difference between the study groups regarding FT₃/FT₄ ratio, which was lower in thyroiditis group than in Graves' disease group (0.21 ± 0.03 in group 1 vs 0.35 ± 0.18 in group 2, p -value < 0.01), while there was no significant difference between thyroiditis group and the control group. The FT₃/FT₄ ratio showed a predictive value, with an area under the ROC curve (AUC) of 0.936 (95%CI, 0.876-0.995) for Graves' disease vs. thyroiditis ($p < 0.001$). Based on the cutoff value of this ratio of more than 0.269 offered a sensitivity of 89.7% and specificity of 91.8%. **Conclusion:** The results of the present study demonstrated that free thyroid hormones and the FT₃/FT₄ ratio are useful to differentiate Graves' disease from destructive thyroiditis.

Keywords: Graves' disease, Thyroiditis, Free Thyroid Hormones, FT₃/FT₄ ratio

Introduction

Thyrotoxicosis is a hypermetabolic status characterized by excess of thyroid hormones serum levels. It may occur because of hyperthyroidism, the release of stored hormone or exogenous

thyroid hormone intake. However, hyperthyroidism is characterized by elevated thyroid hormone levels because of its excess synthesis and secretion from the thyroid gland. The most common causes are Graves' disease (GD), toxic multinodular goiter, and

*Corresponding Author: dr_nesmali@med.asu.edu.eg

autonomous toxic nodule^(1,2). Some studies reported that hyperthyroidism prevalence in iodine-sufficient areas ranges from 0.2% to 1.3%^(3,4). Overt and subclinical forms accounted for 0.5% and 0.7%, respectively⁽⁴⁾. The incidence rate was 51 cases per 100,000 per year⁽³⁾. The clinical presentation of thyrotoxicosis can provide clues for diagnosis, but not the etiology. It is crucial to identify the etiology of thyrotoxicosis, since symptomatic treatment is required in thyroiditis but extensive medical therapy as well as occasional thyroid ablation is needed in cases of GD⁽⁵⁾. Apart from typical clinical presentation, radioactive iodine (RAI) uptake, serum levels of TSH receptor antibody (TRAb), and evaluation of thyroidal blood flow on ultrasonography are commonly used⁽⁶⁾. Total T₃ to total T₄ ratio can also be useful in the assessment of the proper etiology of thyrotoxicosis when scintigraphy is contraindicated⁽⁴⁾. Measurement of FT₃ and FT₄ instead of total form is commonly performed, since free forms are less affected by thyroid hormone binding protein (TBP), FT₃/FT₄ ratio has been advocated for the differential diagnosis of GD and destructive thyroiditis⁽⁵⁾. Therefore, the present study evaluated the usefulness of FT₃/FT₄ ratio in predicting the etiology of thyrotoxicosis, and its role in differentiating patients with GD from patients with destructive thyroiditis, especially those with painless thyroiditis and subacute thyroiditis.

Subjects and Methods

Patients

This was a prospective descriptive study that was done on patients attending endocrine outpatient clinic in El-Maadi Armed Forces Hospital, with diagnosis of thyrotoxicosis at their first

visit and in whom thyroid scintigraphy could be done, in the period from July 2018 to February 2020. The diagnosis of thyrotoxicosis was made on clinical grounds (including heat intolerance, increased appetite, increased sweating, weight loss, muscle weakness, fatigue, tremors, and diffusely enlarged thyroid gland) along with values of thyroid function test results. The Institutional Ethics Committee approval was taken, and all participants gave informed consent before participating in the study.

Study design

The present study was conducted on 100 newly diagnosed thyrotoxicosis patients; 61 with thyroiditis (Group 1) and 39 with GD (Group 2) and 20 healthy, age-matched subjects representing a control group (Group 3). Diagnosis of subacute thyroiditis and painless thyroiditis based on the clinical presentations and low uptake values of thyroid scintigraphy. Graves' disease was diagnosed by clinical presentations and high uptake values of thyroid scintigraphy. Patients taking drugs that affect thyroid function or those who have already started antithyroid medication, patients below 15 years of age, pregnant patients, patients with palpable nodules and patients with single or multiple nodules discovered in thyroid scintigraphy were excluded.

Methods

Serum levels of TSH, FT₃ and FT₄, were measured, using automated chemiluminescent immunoassays (Architect i2000SR; Abbott Laboratories, Chicago, IL, USA). The laboratory reference ranges were (0.35–4.94 μIU/mL), (2.63–5.70 pmol/L) and (9.01–19.04 pmol/L) respectively. Technetium 99m Thyroid Scan was done at radiology centre in El-Maadi Armed Forces Hospital. Intravenous administration of 5 mCi dosage of

Tc 99m- per technetate was done and static image of thyroid gland after 20 minutes in multiple projections was taken. Quantification of total uptake of tracer by the gland was taken as percentage uptake of right and left lobe thus giving total uptake. Normal reference of total percentage uptake range was 0.5%-4%. Uptake values below 0.5% was diagnosed as thyroiditis and above 4% as GD.

Statistical analysis

Statistical evaluation was carried out by using SPSS program version 21.0 (SPSS Inc., Chicago, IL, USA). Numerical data were presented as mean±standard deviation (SD) and categorical data were presented as number and percentage of total. An ANOVA test was done to compare the three groups. Quantitative variables were compared in the two groups via post hoc test. Chi Square test (χ^2) was used to compare two qualitative variables. ROC curve was used to determine best cutoff, sen-

sitivity, and specificity of FT₃ / FT₄ ratio in predicting Graves' disease. Results were evaluated in 95% confidence interval and P value < 0.05 was considered statistically significant.

Results

A total of 120 subjects were included, and divided into three groups; as regard patients with destructive thyroiditis (subacute thyroiditis and painless thyroiditis) representing group 1, included 61 patients, their mean age were 38.03±8.92 yrs, of them 17 (27.87%) were males, and 44 (72.13%) were females. Group 2 included 39 patients with Graves' disease their mean±SD of age were 40.31±11.71 yrs, of them 12 (30.77%) were males, and 27 (69.23%) were females. Group 3 included 20 healthy, euthyroid subjects representing a control group, their mean±SD of age were 39.60±12.48 yrs, of them 7 (35%) were males, and 13 (65%) were females.

Table 1: Comparison between the study groups

	Group 1 (thyroiditis) (n=61)	Group 2 (GD) (n=39)	Group 3 (Control) (n=20)	X ² /F	P value
Male; n (%)	17 (27.87%)	12 (30.77%)	7 (35%)	0.827*	0.381*
Female; n (%)	44 (72.13%)	27 (69.23%)	13 (65%)		
Age (Years)	38.03±8.92	40.31±11.71	39.60 ±12.48	0.591	0.555
TSH (µIU/ml)	0.007±0.02 ^{a,b}	0.004±0.01 ^c	1.85±0.74	54.235	<0.01
FT ₃ (pmol/L)	5.52±0.61 ^{a,b}	8.60± 8.68 ^c	2.53±1.31	72.424	<0.01
FT ₄ (pmol/L)	19.59±2.38 ^{a,b}	24.31± 21.99 ^c	15.44±2.32	36.384	<0.01
FT ₃ /FT ₄ ratio	0.21± 0.03 ^a	0.35±0.18 ^c	0.19±0.09	60.523	<0.01
*= Chi Square test; a, b, c= significantly different at p<0.05, using post hoc test a=group 1 vs. group 2; b= group 1 vs. group 3; c= group 2 vs. group 3 TSH= thyroid stimulating hormone; FT ₃ = free triiodothyronine; FT ₄ =free thyroxine Age, TSH, FT ₃ , FT ₄ and FT ₃ /FT ₄ ratio are presented in mean±standard deviation					

There was no statistically significant difference between the study groups regarding age and gender (Table 1). Regarding thyroid function tests, FT₃ and

FT₄ levels were higher in Graves' disease group than in thyroiditis group, the difference was statistically significant; (FT₃, 8.60± 8.68 pmol/L in group

2 vs 5.52 ± 0.61 pmol/L in group 1, $p < 0.01$) and (FT4, 24.31 ± 21.99 pmol/L in group 2 vs 19.59 ± 2.38 pmol/L in group 1, $p < 0.01$). While TSH levels were lower in Graves' disease group than in thyroiditis group the difference was statistically non-significant (0.004 ± 0.01 μ U/ml in group 2 vs. 0.007 ± 0.02 μ U/ml in group 1, $p = 0.011$). FT3/FT4 ratio was lower in thyroiditis group than in Graves' disease group, the difference was statistically significant (0.21 ± 0.03

in group 1 vs 0.35 ± 0.18 in group 2, $p < 0.01$), while there was no significant difference between thyroiditis group and the control group (Table 1, Figure 1). The FT3/FT4 ratio showed a predictive value, with an area under the ROC curve (AUC) of 0.936 (95%CI, 0.876-0.995) for Graves' disease vs. thyroiditis ($P < 0.001$). Based on the cutoff value of this ratio of more than 0.269 offered sensitivity of 89.7% and specificity of 91.8% (Table 2, Figure 2).

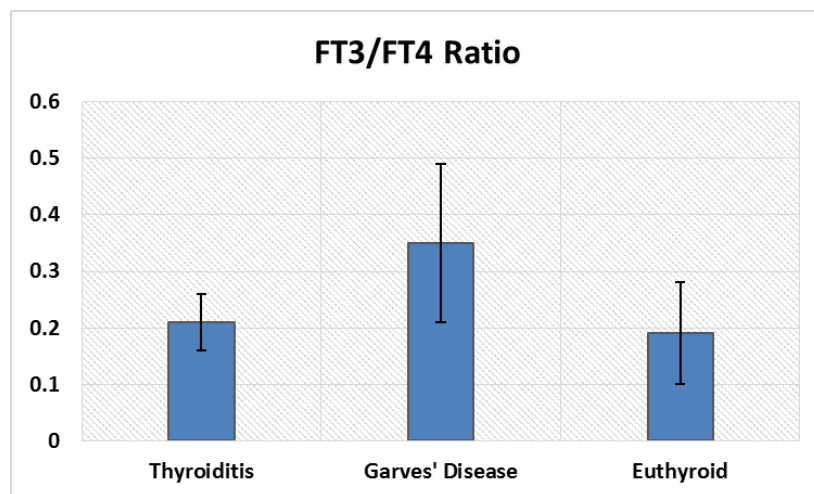


Figure 1: Comparison between the study groups regarding FT3/FT4 ratio. (High significant difference between the study groups)

Discussion

The initial assessment of thyrotoxicosis etiology is crucial to the appropriate clinical management. While the clinical diagnosis of patients is generally based on the physical examination and the laboratory results, it is difficult to differentiate GD from painless thyroiditis based on the physical manifestations. Apart from the typical presentation of patients with thyrotoxicosis, RAI uptake, measurement of serum levels of TRAb, as well as thyroidal blood flow on ultrasonography are commonly used⁽⁶⁾. Other markers that can assist the clinician in differentiating GD from subacute thyroiditis include; serum

calcitonin levels⁽⁷⁾, assessment of eosinophil to monocyte ratio⁽⁸⁾, stem cell factor⁽⁹⁾, total alkaline phosphatase activity⁽¹⁰⁾, RBCs zinc concentration⁽¹¹⁾, plasma fibrinogen levels⁽¹²⁾, and serial changes in liver function tests⁽¹³⁾. However, practically it is difficult to use such parameters. The use of ultrasonography for the differential diagnosis of thyrotoxicosis is less useful. However, when RAI uptake test is contraindicated (i.e. during pregnancy, lactation and in case of new iodine exposure), color flow Doppler ultrasonography may assist in diagnosing the etiology of thyrotoxicosis and to differentiate GD from destructive thyroiditis⁽¹⁴⁾. RAI uptake test is a valuable method in

differentiating the etiology of thyrotoxicosis; however, it is contradicted in some cases (i.e. during pregnancy and lactation). Moreover, exposure to large

amounts of iodine prior to undergoing RAI uptake test may lead to RAI uptake suppression, resulting in an inappropriate diagnosis⁽⁶⁾.

Table 2: Area under Curve (AUC) and best cutoff value of FT₃ / FT₄ ratio to predict Graves' disease

AUC	Cut-off	Sensitivity	Specificity	95% CI	
				Upper bound	Lower bound
0.936	0.269	89.7%	91.8%	0.876	0.995

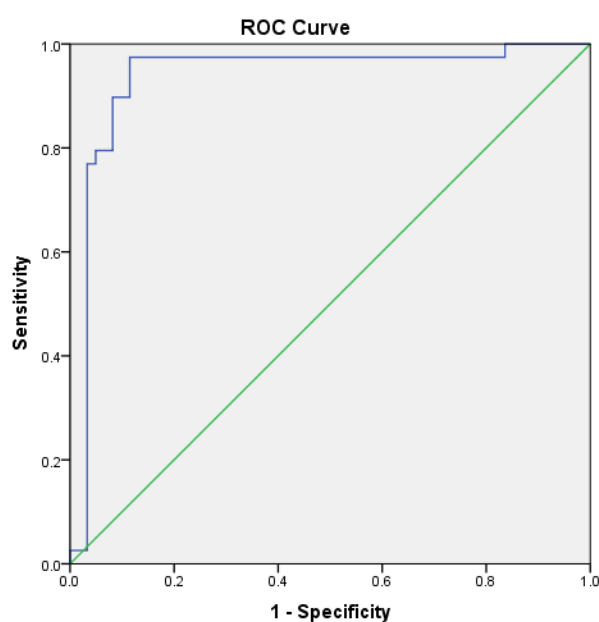


Figure 2: ROC curve to determine sensitivity and specificity of FT₃/FT₄ ratio in predicting Graves' disease

Measuring serum levels of TRAb has been commonly used in differentiating thyrotoxicosis etiology when RAI uptake is unavailable or contraindicated. However, patients with mild GD might have negative for TRAb⁽⁶⁾, TRAb may also be measured in patients with postpartum thyroiditis^(15,16). The ratio of total T₃ to total T₄ may also be useful in the assessment of thyrotoxicosis etiology when RAI uptake is contraindicated. The hyperactive gland produces more T₃ than T₄, resulting in elevation of T₃ above the upper limit of normal more than T₄ in thyrotoxicosis caused

by hyperthyroidism, whereas T₄ is elevated more than T₃ in thyrotoxicosis caused by thyroiditis⁽¹⁷⁾. Shigemasa et al. revealed that the ratio of total T₃ to total T₄ (ng/lg) was >20 in GD and toxic nodular goiter, and <20 for painless or postpartum thyroiditis⁽¹⁸⁾. American Thyroid Association/American Association of Clinical Endocrinologists (ATA/AACE) guidelines recommend TRAb measurement and total T₃/total T₄ ratio as an alternative method for the diagnosis of GD in the absence of thyroid scintigraphy and uptake or when these are contraindicated⁽¹⁵⁾.

However, TRAb measurement is not performed in many centers, it is expensive, and the results take time. Also, measurements of FT₃ and FT₄ are frequently ordered in the clinical practice⁽¹⁹⁾ rather than total T₄ and T₃ measurements, as the later are affected by serum TBG concentration which may be affected by many conditions in an unfavorable manner. So, a practical alternative was required for differentiating diagnosis in clinic. In the present study, we analyzed levels of FT₃, FT₄ and FT₃/FT₄ ratio in patients with GD and destructive thyroiditis (including subacute and painless thyroiditis). We found that the levels of FT₃, FT₄ and the FT₃/FT₄ ratio were higher in patients with GD than in those with thyroiditis. These results are consistent with the findings of previous literature. Yoshimura et al., reported that the FT₃/FT₄ ratio was useful in differentiating patients with GD and painless thyroiditis⁽⁵⁾. Moreover, Sriprapradang et al. revealed that the FT₃/FT₄ ratio might help in differentiating the etiology of thyrotoxicosis, with a higher FT₃/FT₄ ratio suggesting GD, whereas a very low ratio supported the diagnosis of thyroiditis⁽²⁰⁾. According to Izumi et al., the FT₃/FT₄ ratio was useful in differentiating GD and destructive thyroiditis⁽⁸⁾. However, Shigemasa et al. reported that the FT₃/FT₄ ratio was not useful in the differentiation between GD and painless thyroiditis⁽¹⁸⁾. T₃ is an active form of thyroid hormones that is resulted from the deiodinated T₄ by type 1 iodothyronine deiodinase (D₁) as well as type 2 (D₂). In humans, it was found that the thyroid gland expresses both D₁ and D₂⁽²¹⁾. In Graves' thyroid tissue, there is a high expression of D₂ which contribute to relatively high serum T₃ levels⁽²²⁾. On the other hand, there is leakage of thyroid hormone

from the breakdown thyroid follicle cell in destructive thyroiditis. In some cases of thyroiditis, the radioactive iodine uptake increases during the recovery phase and scintigraphy demonstrates increased activity leading to confusion with GD⁽²³⁾. So, patients in such phase were excluded from our study by asking their clinical history of interval decline in symptoms. According to our findings, FT₃/FT₄ ratio was useful in distinguishing the etiology of thyrotoxicosis. In the current era of economic crises, cost effectiveness is one of the most important health management concerns. Patients with mild GD or where thyroid scintigraphy were unavailable or contraindicated, the ratio of FT₃/FT₄ represents a simple and practical alternative to assess the cause of thyrotoxicosis. The finding of the present study is very helpful for both of patients and clinicians, in the settings of poor patient's compliance and limited country resources. However, FT₃/FT₄ ratio should be validated in other populations, and, due to the limited number of patients in our study, there is a need for studies with more patients. Selection of the recruited patients from a single center is another limitation of this study. Also, this ratio should not be applied to hospitalized patients who have the possibility of low T₃ levels from nonthyroidal illness.

Conclusion

FT₃/FT₄ ratio is useful for differentiating GD from destructive thyroiditis, with lower cost and hazards than thyroid scintigraphy. So, in the absence of thyroid scintigraphy and uptake or when these are contraindicated, the FT₃/FT₄ ratio may be used as an alternative method for distinguishing the etiology of thyrotoxicosis.

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Conflicts of Interest: None

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