Thyroid Science 5(9):1-5, 2010

www.ThyroidScience.com

Clinical and Laboratory Studies

Prevalence of Antithyroid Antibodies in Histologically Proven Autoimmune Thyroid Diseases and Correlation with Thyroid Dysfunction in South India

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Received: July 29, 2010
Accepted: August 27, 2010

***Abstract*. Objective**: The clinical diagnosis of autoimmune thyroid diseases (AITD) is usually confirmed by detection of various antibodies in the patient's blood or by histopathological evaluation. In this study, we evaluated the prevalence of serum antibodies in histopathologically proven AITD and assessed the correlation between antibodies and thyroid dysfunction. **Methods**: 100 consecutive patients with a histological diagnosis of AITD were included in the study and underwent detailed clinical examination and testing for thyroid function, thyroid antibodies, thyroid ultrasound scan, and thyroid gland fine-needle aspiration cytology. **Results**: Thyroid peroxidase antibodies (TPO Ab) tested positive in 89% of patients and negative in 11%. Antithyroglobulin antibody (ATG Ab) estimation was positive in 64 % of patients and negative in 36%. By thyroid function testing and serum antibody evaluation, of the 89 TPO-positive patients, 60.7% were hypothyroid, 6.7% hyperthyroid, and 32.6% euthyroid. Among euthyroid patients, 90% were TPO-Ab negative. In 64 ATG Ab-positive patients, 53.1% patients were hypothyroid, 4.7% hyperthyroid, and 42.2% euthyroid. But in the 36 ATG Ab-negative patients, 58.3% were hypothyroid. At the time of the first clinic visit, 55% of patients were hypothyroid, 6% hyperthyroid, and 39% euthyroid. **Conclusion**: In our study, TPO Ab was more sensitive than ATG Ab in predicting hypothyroidism. Similarly, TPO Ab was more sensitive than ATG Ab in autoimmune thyroiditis (98.1% vs 61.8%, p value < 0.005). Hypothyroidism was the most frequent thyroid dysfunction in patients with positive TPO and ATG antibodies . The absence of TPO usually is associated with no thyroid dysfunction, but the same cannot be said of ATG.

Keywords Antithyroid antibodies • Antithyroglobulin antibodies • Autoimmune thyroid disease • Hashimoto's thyroiditis • Hyperthyroidism • Hypothyroidism • Thyroid peroxidase antibodies

Introduction

Autoimmunity of the thyroid gland results in a spectrum of thyroid diseases in patients. The patients commonly present in outpatient endocrine clinics with goiter or thyroid dysfunction. Etiologically, patients with autoimmune thyroiditis, including Hashimoto's thyroiditis (chronic autoimmune thyroiditis) which has specific histopathological findings, are usually hypothyroid. Less commonly, patients are hyperthyroid.[1]

Autoimmune thyroiditis shows lymphocytic infiltration with large lymphoid follicles that have germinal centres. In addition, if large thyroid cells have an acidophilic staining character called "Hurthle" or "Askanazy" cells, the diagnosis is Hashimoto's thyroiditis. In this study, all patients with histological findings of autoimmune thyroiditis, with or without histopathological findings of Hashimoto's, are included under the title autoimmune thyroid disease.

The clinical diagnosis of autoimmune thyroid disease is usually confirmed by the detection of various antibodies in the patient's blood sample. Three types of antibodies are most commonly assayed: antithyroglobulin, thyroid peroxidase (previously termed antimicrosomal), and TSH receptor antibodies. TSH

receptor antibodies are either stimulating or blocking antibodies; respectfully, these are related to Grave's disease and myxoedema. We do not included in the present study, although their presence is classified as autoimmune thyroid disease. In autoimmune thyroiditis, high circulating blood titres of antithyroglobulin antibodies and antithyroperoxidase antibodies are usually present.

Table I. Classification of autoimmune thyroid disease.

Etiology	Clinical Presentation
Hashimoto's thyroiditis	Goitre, hypothyroidism (rarely hyperthyroidism)
Painless thyroiditis (silent/post partum)	Small goiter, transient hyper- or hypothyroidism
Atrophic/primary thyroiditis	Hypothyroidism/ thyroid atrophy
Grave's disease	Hyperthyroidism /goitre

The clinically pertinent classification of autoimmune thyroid disease, listed in Table 1, includes Hashimoto's thyroiditis and its variants, autoimmune atrophic thyroiditis (myxoedema) and Grave's disease.[2] Prevalence data from the American Thyroid Association state regional variations, but the generally accepted range of women worldwide developing autoimmune thyroiditis at some point in their lives is 5% to 7%. This percentage makes autoimmune thyroid disease a relatively common disorder.[3]

Routine thyroid antibody estimation in patients with thyroid enlargement has revealed an increased prevalence of autoimmune thyroiditis in Kerala.[4] In recent years, the hypothesis has gained ground that entities like puberty goitre in adolescents and multinodular goitre in adults are due to autoimmune thyroiditis. However, these conditions were not previously diagnosed because tests for antibody estimation were not available.[4]

To test this hypothesis, this study has two aims: (1) to find the regional prevalence of thyroid peroxidase and anti-thyroglobulin antibodies, and (2) to determine the relation of the antibody prevalence to thyroid dysfunction in patients with histological diagnoses of autoimmune thyroiditis and Hashimoto's thyroiditis.

Patients and Methods

Included in this study were 100 consecutive patients who attended the outpatient clinic and had a histological diagnosis of autoimmune thyroiditis or Hashimoto's thyroiditis with either symptoms of thyroid dysfunction and/or thyroid swelling. Besides a detailed clinical examination, each patient underwent the following investigations: blood tests for thyroid function (T_3, T_4, TSH), thyroid peroxidase antibodies, and antithyroglobulin antibodies. Each patient also had both a thyroid ultrasound scan and a fine-needle aspiration cytology of the thyroid gland. Thyroid function tests and antibodies were estimated by the chemiluminiscent method.[5]

Each patient's thyroid functional status (euthyroidism, hypothyroidism, or hyperthyroidism) was defined by TSH, free T_4, and free T_3 levels. Following are the values of the reference ranges for thyroid function and antibody tests in our laboratory: T_3 (1.0-to-2.8 nmol/L); T_4 (62-to-148 nmol/L); TSH (0.4-to-5.0 μIU/ml); antimicrosomal antibodies (< 9.0 IU/ml); and antithyroglobulin antibodies (< 60 IU/ml).

Table II. Age distribution of patients.

Age*	Male	Female	Total	%
< 21	1	3	4	4
21-30	4	23	27	27
31-40	2	31	33	33
41-50	2	23	25	25
51-60	0	9	9	9
> 60	0	2	2	2

*Age in years (Age range: 15-to-62 years)

Results

Of 100 patients studied, 91 were females and 9 were males. As stated previously, for analysis of data, both Hashimoto's thyroiditis and autoimmune thyroiditis were classified as autoimmune thyroid disease. Fifty-nine patients (59%) were classified as having Hashimoto's, and 41 patients (41%) were classified as having autoimmune thyroiditis.

The age of the studied patients ranged from 15 to 62 years. Four were less than 21 years of age and most were females. Patients aged less than 45 years made up 68% of the group (Table II). Many of the patients had long standing histories of thyroid dysfunction or goiter before presenting to us. Because of this, the exact ages at onset of thyroid disease was difficult to assess.

Antibody positivity and thyroid function. Thyroid peroxidase antibodies were estimated in all patients; 89 patients (89 %) tested positive and 11 (11%) tested negative. Antithyroglobulin antibody estimation was also done in all patients; 64 patients (64 %) tested

positive and 36 patients (36 %) tested negative (Table III).

Table III. Assay results of antibody levels.

Antibody	Positive	Negative	*p* value*
ATG**	64	36	< 0.005
TPO***	89	11	

* *p* value < 0.005 was considered significant.
** Antithyroglobulin Ab. *** Thyroid peroxidase Ab.

Of the 89 patients who were positive for thyroid peroxidase, 54 (60.7 %) were hypothyroid; 6 (6.7 %) were hyperthyroid. and 29 (32.6%) were euthyroid. However, all patients who were negative for thyroid peroxidase antibodies (100%) were euthyroid. This enhances the predictive value of positivity for thyroid peroxidase antibodies and thyroid dysfunction.

Of 64 patients who were positive for antithyroglobulin antibodies, 34 (53.1%) were hypothyroid; 3 (4.7%) were hyperthyroid, and 27 (42.2%) were euthyroid. Also, of the 36 patients who were negative for antithyroglobulin antibodies, 21 (58.3%) were hypothyroid, and 3 (0.08%) were hyperthyroidism. At the time of the first visit to our clinic, 55% of patients were hypothyroid, 6% were hyperthyroid, 39% were euthyroid. In predicting hypothyroidism (Table IV), thyroid peroxidase antibodies were much more sensitive than antithyroglobulin antibodies (98.1% vs 61.8%).

Ultrasound findings. Ultrasound testing show that 54 patients had multinodular goiter, 16 had diffuse goiter, 2 had cystic thyroid nodules, 3 had solitary nodules, and 25 had features of thyroiditis (i.e., diffuse or focal coarse echo texture in the thyroid gland and increased vascularity—generalized or focal thyroiditis). Of the 3 patients whose autoimmune thyroid disease included solitary nodules, 2 had histological evidence of papillary carcinoma (Table V).

Discussion

Historically, the first thyroid autoantibody discovered was antithyroglobulin antibody. That was in 1956. Antibodies to other antigens present in the cytoplasm of thyroid follicular cells (first termed "antimicrosomal antibodies") were detected in 1976. These "cytoplasmic" antigens were later found to be the enzyme thyroid peroxidase; hence today, these antibodies are more often called "thyroid peroxidase antibodies." Thyroid peroxidase antibodies appeared to be much more prevalent than antithyroglobulin antibodies.[6,7]

Table IV. Correlation between antibody levels and thyroid status.

Antibody assay	Status	Number of patients	Number and % of hypothyroid patients	Number and % of hyperthyroid patients	Number and % of euthyroid patients
ATG*	Positive	64	34 (53.1%)	3 (4.7%)	27(42.2%)
	Negative	36	21 (58.3%)	3 (8.3 %)	12 (33.3%)
TPO**	Positive	89	54 (60.7%)	6 (6.7%)	29 (32.6%)
	Negative	11	1 (9.09%)	0	10 (90.9%)

* Antithyroglobulin Ab. ** Thyroid peroxidase Ab.

The prevalence of autoimmune thyroiditis is increasing. We observed that about 70% of patients who present to outpatient clinics in our geographical region with thyroid problems have autoimmune thyroiditis. The prevalence of positive thyroid antibodies was 29% in pregnant patients in a region similar to ours.[8]

Hypothyroidism is the characteristic functional abnormality in autoimmune thyroid diseases. However, inflammation of the thyroid gland early in the disease may cause thyroid follicular destruction with thyroid hormonal release from the follicles. This results in transient hyperthyroidism.

These antibodies can even lyse the thyroid cells. B cells present the thyroid antigen to T cells. T cells secrete cytokines that activate a variety of other immune cells, and has a role in antibody production (Th2 cells) and apoptotic destruction of thyroid cells by activating cytotoxic T cells (Th1 cells).

Genetic and environmental factors such as toxins, bacterial and viral infections or iodine excess, appear to interact, leading to the appearance of auto antigens

and accumulation of antigen-presenting cells in the thyroid. Consequently, due to loss of immune tolerance, auto-reactive immune cells (T lymphocytes) activated by antigen-presenting cells invade the thyroid gland, interacting with the thyroid cells, and the apoptotic pathways are activated by certain cytokines produced locally by the T lymphocytes. It is likely that the regulation of apoptosis during this interaction between the invading lymphocytes and the defending thyroid cells, may play an important role in the clinical expression of autoimmune thyroid diseases.

Table V. Ultrasound scan findings.

Multinodular goitre	54
Cystic nodules	2
Thyroiditis features	25
Solitary thyroid nodule	3
Diffuse Goitre	16
Total	100

Of the patients in this study, 91% were female, and 84% of these were in the reproductive age group. Since thyroid dysfunction can lead to antenatal and neonatal complications, the diagnosis and correction of any thyroid disorder is very important in pregnant patients. Thyroid autoimmunity is a risk factor for pregnancy loss.[9] Some authors have reported that pregnant euthyroid females who are positive for thyroid peroxidase (antimicrosomal) antibodies and who undergo thyroid hormone therapy may improve their miscarriage rate by 75% and their premature deliveries rate by 69%.[10] A higher prevalence of thyroid antibodies also increases the risk of postpartum thyroiditis.[11]

Ultrasound scans revealed that a majority of our patients (54%) had thyroid gland multinodularity, and 16% were had diffuse goiters. Another 2% had papillary thyroid carcinoma.

In the latter 2% of patients, fine-needle aspiration cytology of thyroid gland from multiple sites was a must in view of the association of papillary carcinoma with autoimmune thyroid diseases. Here a pertechnetate thyroid scan can be used to assess functionality of the thyroid nodules before fine-needle aspiration cytology is considered. This procedure may identify the dominant and cold nodules which should not be missed.

Sixteen percent of the patients had no thyroid swelling but showed evidence of thyroiditis during ultrasound thyroid scans. In six patients with histologically proven autoimmune thyroiditis, no autoantibodies were detected in the blood; this suggests that autoimmune thyroiditis cannot be completely excluded in patients with no antibodies.

Positive antibodies were detected in the neonates of 4 patients who were positive for both thyroperoxidase and antithyrogobulin antibodies. Hence it appears prudent to test the offspring of women who are positive for both type of antithyroid antibodies, to diagnose any thyroid affliction in the neonates early. Since we have not done neonatal antibody screening routinely in all the patients postpartum, no conclusion could be drawn from this finding.

SUMMARY

Autoimmune thyroiditis is definitely not a rare disease, but it is under diagnosed. Each patients who has a swollen thyroid gland should be subjected to thyroid function tests and antibody estimation followed by fine-needle aspiration cytology. If all these tests favor of a diagnosis of autoimmune thyroiditis, surgery is not urgent unless the patient has symptoms of pressure, rapid thyroid gland enlargement, or suspicion of malignancy.

Hypothyroidism was the most frequent thyroid dysfunction in patients who were positive for thyroid peroxidase (60%) and antithyroglobulin antibodies (53.1%). It should be noted that only one subject who was negative for thyroid peroxidase was hypothyroid. However, 58.3% of patients who were negative for antithyroglobulin antibodies were hypothyroid. This shows that thyroid peroxidase antibodies are more sensitive than antithyroglobulin antibodies in predicting hypothyroidism.

Similarly, thyroid peroxidase antibodies were more sensitive than antithyroglobulin antibodies in autoimmune thyroiditis (98.1% vs 61.8%, $p < 0.005$). It is well recognized that autoimmune thyroid disease correlates with excess iodine intake; hence, the increased prevalence of thyroiditis may be related to the intake of iodine-rich salt by the majority of the sea-fish consuming population of Kerala (a southern coastal state in India where the study was conducted). However, the definite correlation of excess iodine intake and thyroiditis warrants study of the plasma and urinary iodide levels and thyroid iodine uptake levels in this population.

References

1. Roitt, I. M., Doniach, D., Campbell, P.N. et al.: Autoantibodies in Hashimoto's disease (lymph-

adenoid goitre). *Lancet*, 2: 820-821, 1956.
2. Fountoulakis, S. and Tsatsoulis, A.: Pathogenesis of autoimmune thyroiditis: an unifying answer. *Clin. Endocrinol*.,60(4):1,2004
3. The American Thyroid Association Public Health Committee: Study of maternal hypothyroidism during pregnancy and subsequent childhood neuropsychological development. *Thyroid*, 9:971-972, 1999.
4. Akkara, B.: Personal communication, February 2010.
5. Amino, N., Hagen, S.R., Yamada, N., et al.: Measurement of circulating thyroid microsomal antibodies by the tanned red cell heamagglutination technique: its usefulness in the diagnosis of autoimmune thyroid disease. *Clin. Endocrinol*., 5:115-125, 1976.
6. Libert, F., Ruel, J., Ludgate, M., et al.: Complete nucleotide sequence of the human thyroperoxidase-microsomal antigen cDNA. *Nucleic Acids Res*., 25;15(16):6735-6740, 1987.
7. McLachlan, S.M. and Rapoport, B.: The molecular biology of the thyroid peroxidise: cloning, expression and role as auto antigen in autoimmune thyroid disease. *Endocrine Rev.*, 13:192-206, 1992.
8. Vijayan, S., Paulose, J., Anitha, S., et al.: Thyroid Autoantibodies in Pregnancy and its implications. *Kerala Med. J*., 5:205-207, 2009
9. Glinoer, D.: Miscarriage in women with positive anti-TPO antibodies: Is thyroxine the answer? *J. Clin. Endocrinol. Metab*., 91:2500-2502, 2006.
10. Unnikrishnan, A.G.: Thyroid autoimmunity, pregnancy and finally, data from Kerala! *Kerala Med. J.*, 5:203-204, 2009.
11. Lazarus, J.H., Ammari, F. Oretti, R. et al.: Clinical aspects of recurrent postpartum thyroiditis. *Br. J. Gen. Pract.*, 47(418): 305–308, 1997.