

## Research article

**Grading of Hashimoto's thyroiditis in cytology and its association with ultrasonographic and biochemical parameters**

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Corresponding author: **Cryslle Saldanha**. Email: cryslesaldanha@gmail.com**ABSTRACT**

**Introduction and Aim:** In the post-iodization period, Hashimoto's thyroiditis (HT) is most likely the most prevalent cause of goitre and hypothyroidism, particularly in women of reproductive years. This study was undertaken to grade HT on Fine Needle Aspiration Cytology smears and determine their usefulness in predicting thyroid damage in association with clinical history, radiological and biochemical findings.

**Materials and Methods:** Total of 151 cases were diagnosed cytologically in duration of over 2 years as Hashimoto's thyroiditis/lymphocytic thyroiditis. Cytology scoring technique was used to analyse smears and were correlated with radiological, biochemical findings and were evaluated statistically by ANOVA and Kruskal Wallis tests.

**Results:** In the current study, the most frequent group of individuals with Hashimoto's thyroiditis were between the ages of 40 and 50. Females (145 cases, 96%) were commonly affected and the commonest presentation was a diffuse (125 cases, 82.8%) thyroid enlargement. Cytological grading of Hashimoto's thyroiditis was as follows, 54 (35.8%) patients were Grade I, 81 (53.6%) patients were Grade II thyroiditis and 16 (10.6%) patients were Grade III thyroiditis. Biochemically, 78 (56.5%) of the cases were hypothyroid, 51 (37%), euthyroid, and 9 (6.5%) were hyperthyroid. Ultrasonography revealed 76 (50.3%) patients with hypoechoic micronodules, 71 (47%) with echogenic septae, 101 (66.9%) with increased vascularity, and 6 (4%) with normal findings. The grades of lymphocytic thyroiditis and the biochemical data were found to be statistically significant ( $p=0.001$ ). Furthermore, there was a strong correlation between the grades and specific ultrasound findings, such as echogenic septae ( $p=0.042$ ) and diffuse hypoechogenicity ( $p=0.046$ ).

**Conclusion:** Cytological scoring system in Hashimoto's thyroiditis can predict thyroid functional status and aid in determining the severity of the disease.

**Keywords:** Hashimoto's thyroiditis; cytology; AntiTPO.

**INTRODUCTION**

**H**ashimoto's thyroiditis (HT), is the most common cause of goitre and hypothyroidism in the post-iodization era, particularly in women of reproductive age (1,2). Iodine can cause autoimmune thyroiditis through the production of anti-thyroglobulin (Tg) antibodies and activating the immune system's defences, culminating in an exponential increase in thyroid lymphocytic infiltration (3).

Clinically, these individuals present as diffuse or nodular swelling in front of the neck which is with associated euthyroidism, hypothyroidism, or occasionally hyperthyroidism (1). Hashimoto thyroiditis typically has a diffusely enlarged gland, hypoechoic parenchyma, hypervascularity, and a micronodular pattern on sonography (4). Thyroid peroxidase is the most significant antibody aimed towards thyroid tissue (1). Fine needle cytology (FNAC) is the most often used method for HT diagnosis because antibody production in the early stages of HT may be limited to intrathyroidal cells or specific locations (5). HT on smears is determined by the presence of epithelial oxyphilic (Hurthle) cells, lymphocytes and plasma cells infiltrating follicles, as well as a considerable amount of these cells in the

background and either little or no colloid (6). The goal of the present research is to assess the severity of Hashimoto's thyroiditis using FNAC smears to determine their usefulness to predict thyroid injury. The present study was undertaken to evaluate and grade fine-needle aspiration cytology (FNAC) smears from patients with chronic lymphocytic thyroiditis and to establish their importance in predicting thyroid injury utilising cytology grading and comparison with clinical, biochemical, and ultrasound data.

**MATERIALS AND METHODS**

A two-year cross-sectional study was done in a tertiary care hospital on 151 patients diagnosed cytologically as Hashimoto's thyroiditis/lymphocytic thyroiditis (LT).

**Sample size calculation**

Formula for calculation of sample size (6):

$$\text{Estimate sensitivity: } n \geq \frac{Z_{1-\frac{\alpha}{2}}^2 \times p(1-p)}{d^2}$$

Alpha ( $\alpha$ ) -0.05

Estimated proportion (p)-0.61

Estimation error (d)-0.1

Minimum total sample size needed: 61

The inclusion criteria were met by newly identified cases of chronic lymphocytic thyroiditis. Subjects, who were using thyroxine or another drug that was thought to be interfering with thyroid function at the time of evaluation, as well as any additional lesions identified in conjunction with chronic lymphocytic thyroiditis observed on cytology, were excluded from the study.

Several factors were documented, including the patient's clinical presentation, thyroid function tests (TSH, Anti-TPO, T3, T4), and radiological results. Smears with a diagnosis of chronic lymphocytic thyroiditis were assessed using the cytology grading system developed by Alka et al., (6). The Grades were correlated with the above parameters using ANOVA and Kruskal Wallis tests.

The following grading method was used in the current study (6), Grade 1: Mild follicular lymphocytic infiltration or a disproportionately high number of background lymphocytes. Grade 2: Moderate degree of lymphocytic infiltration of the follicles or presence of anisonucleosis/hurthle cells/giant cells/ along with mild follicular lymphocytic infiltration. Grade 3: Characterised by development of germinal centres and lack of the majority of follicular cells as a result of severe lymphocyte infiltration.

**RESULTS**

Hashimoto's thyroiditis was observed among 151 patients over a two-year period. The most common age group for Hashimoto's thyroiditis in the current study was (Table 1) 40-50 years. Females (145 cases, 96%) were commonly affected and the commonest presentation was a diffuse (125 cases, 82.8%) thyroid enlargement. Remaining 26 (17.21%) patients exhibited a nodular thyroid enlargement. According to the standards established by Alka et al., (6) (Table 2), thyroiditis was graded.

**Table 1:** The distribution of age and gender in individuals with Hashimoto's Thyroiditis

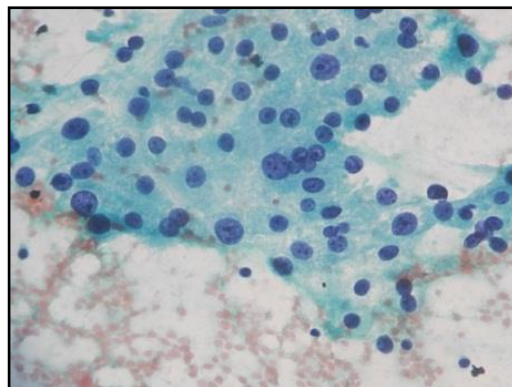
		No. of cases	Percentage %
Age	<=20	7	4.6%
	21-30	24	15.9%
	31-40	42	27.8%
	41-50	47	31.1%
	51-60	22	14.6%
	>60	9	6.0%
Sex	Female	145	96.0%
	Male	6	4.0%

**Table 2:** Grading of thyroiditis

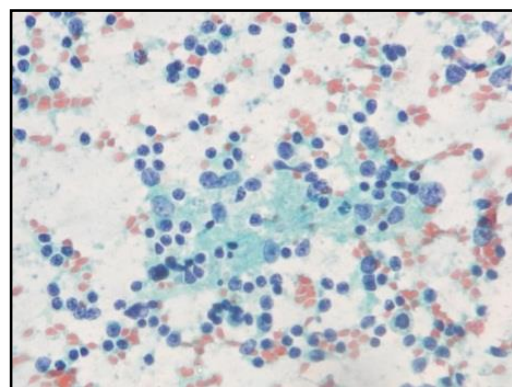
Grading of thyroiditis	Frequency (Percentage %)
Grade I	54 (35.8)
Grade II	81(53.6)
Grade III	16 (10.6)
Total	151(100.0)

Hashimoto's thyroiditis cytology scoring method demonstrated various degrees of severity. Grade I

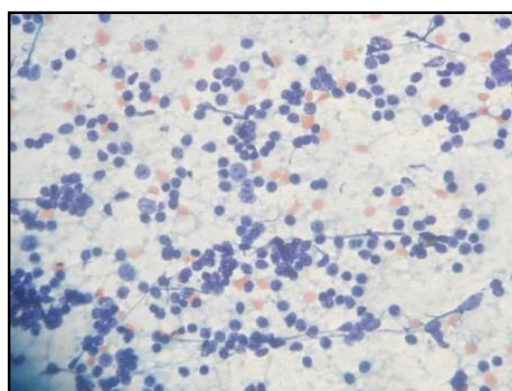
thyroiditis affected 54 (35.6%) patients, showing mild lymphocytic infiltration of thyroid follicular cells (Fig. 1), while Grade II thyroiditis (Fig.2) included 81 (53.6%) patients, exhibited a significant lymphocytic infiltrate and destruction of thyroid follicles and 16 (10.6%) individuals had Grade III thyroiditis (Fig. 3), which is indicated by extensive lymphocytic infiltrates with germinal centres.



**Fig. 1:** Microphotograph showing Grade I thyroiditis with few lymphocytes infiltrating the thyroid follicular cells (PAP, 40X).



**Fig. 2:** Microphotograph showing Grade II thyroiditis with significant lymphocytic inflammation (PAP, 40X).



**Fig. 3:** Microphotograph showing Grade III thyroiditis with marked lymphocytic inflammation (PAP, 40X)

Data on biochemical parameters was available in 138 patients. 78 (56.5%) of the 138 patients in the current study had biochemically low thyroid function, 51

**Table 3:** Contrast of severity of Hashimoto's thyroiditis with the hormonal status

Biochemical status	Mild thyroiditis		Moderate thyroiditis		Severe thyroiditis	
	No. of cases	Percentage %	No. of cases	Percentage %	No. of cases	Percentage %
Euthyroid	30	63.8%	19	25.0%	2	13.3%
Hyperthyroid	5	10.6%	4	5.3%	0	.0%
Hypothyroid	12	25.5%	53	69.7%	13	86.7%
Total	47	100.0%	76	100.0%	15	100.0%

**Table 4:** Comparison of severity of Hashimoto's thyroiditis with the results of ultrasonography

Ultrasonographic findings		Mild thyroiditis		Moderate thyroiditis		Severe thyroiditis	
		No. of cases	Percentage %	No. of cases	Percentage %	No. of cases	Percentage (%)
Hypoechoic micronodule	Present	26	48.1%	40	49.4%	10	62.5%
	Absent	28	51.9%	41	50.6%	6	37.5%
	Total	54	100.0%	81	100.0%	16	100.0%
Diffuse hypoechoic	Absent	38	70.4%	42	51.9%	12	75.0%
	Present	16	29.6%	39	48.1%	4	25.0%
	Total	54	100.0%	81	100.0%	16	100.0%
Vascularity	Normal	17	31.5%	30	37.0%	3	18.8%
	Increased	37	68.5%	51	63.0%	13	81.3%
	Total	54	100.0%	81	100.0%	16	100.0%
Echogenic septae	Present	22	40.7%	45	55.6%	4	25.0%
	Absent	32	59.3%	36	44.4%	12	75.0%
	Total	54	100.0%	81	100.0%	16	100.0%
Normal study		5	83.3%	1	16.7%	0	0
	Total	6	100.0%	6	100.0%	6	100.0%

(37%) had euthyroid function, and 9 (6.5%) had hyperthyroid function (Table 3). The degree of lymphocytic thyroiditis and the biochemical results were statistically significant (p 0.001). In 60 cases, the levels of AntiTPO were known. Only 3 (5%) cases had a normal anti-TPO result, while the value was elevated in 57 (95%) cases. The severity of thyroiditis assessed on smears did not correlate with the levels of AntiTPO in the serum (p= 0.575).

In 151 cases, ultrasonography was performed, and a predetermined set of sonographic characteristics were seen (Table 4). Ultrasonography revealed hypoechoic micronodules in 76 (50.3%) cases, echogenic septae in 71 (47%) cases, enhanced vascularity in 101 (66.9%) cases, and a normal study in 6 (4%) cases. In the current investigation, there was a statistically significant relationship between grades and two ultrasonography findings: diffuse hypoechoic (p- 0.046) and echogenic septae (p- 0.042).

**DISCUSSION**

The age range for onset of Hashimoto's thyroiditis in the current study was 14 to 70 years. The most prevalent age range was between 40 and 50 years old, similar to as observed in literature studies (7,8). Hashimoto's thyroiditis was common among females which was similar to other studies (6,9). The significant female prevalence of Hashimoto's thyroiditis may be due to the numerous sex- and immune-related genes found on the X chromosome, which are essential for the maintenance of immunological tolerance (10,11). Based on the results of the hormonal test, hypothyroidism was found in 78 cases (56.5%) of the

patients in the current study. The prevalence of subclinical hypothyroidism in our study was 26 cases (17.2%), which was comparable to the findings of Singh *et al.*, (1,11) 19.3% and Bagchi *et al.*, (12,13) 8-17%, respectively.

The most frequent appearance was diffuse goitre (125 cases, 82.8%), while nodular thyroid enlargement was seen in 26 patients (17.21%) of the remaining patients. Rathi *et al.*, (12) and Bijwe *et al.*, (10,12) conducted research in which nodules were found in 29(19%) and 16(32%) of the cases, respectively. Individuals have a nodular appearance during the early stages of the disease before clinical and hormonal changes are evident (10-13). In the present study, hypoechoic micronodules were often discovered in 76 (50.3%) of the cases. These results are comparable to the findings of earlier investigations by Alka *et al.*, (6) and Uma (9). The presence of echogenic septae, a sign of fibrosis and a more severe disease, was observed in 71 (47%) of the cases. The presence of hypothyroidism or its potential development in the future is strongly predicted by a diffuse hypoechoic pattern, which has been linked to infiltration of the gland by lymphocyte and fibrous septae (13,14). In the current study, 59 individuals (39.07%) had diffuse hypoechoic, which was equivalent to other studies' findings (9,14).

The criteria established by Alka *et al.*, (6) were used to classify 151 cases of lymphocytic thyroiditis into Grade I, Grade II, and Grade III thyroiditis in 35.8%, 53.6%, and 10.6% of cases, respectively. Asymptomatic patients (63.8%) were found to make up a large portion of Grade I thyroiditis patients. In thyroiditis of Grades II and III, a high prevalence of hypothyroid patients was

seen. Lesions of Grade I and II only exhibited hyperthyroid features. Clinical characteristics and lymphocytic thyroiditis grades were observed to significantly correlate statistically ( $p = 0.003$ ). The majority of Grade III thyroiditis (86.7%) cases were hypothyroid. Biochemical hypothyroid cases were found to have a strong correlation with thyroiditis of Grades II and III. In cases of biochemical euthyroidism (63.8%), Grade I lesions were the most common type. Statistically, there was a significant relationship between the biochemical characteristics and lymphocytic thyroiditis grades ( $p 0.001$ ).

Ultrasonography revealed hypoechoic micronodules in the majority (62.5%) of individuals with Grade III thyroiditis. In (55.6%) of Grade II lesions, echogenic septa were observed. Six cases had normal ultrasonography results, of which five (83.3%) had Grade I thyroiditis and one (16.7%) had Grade II thyroiditis. According to the results of the current study, there was a statistically significant correlation between the severity of lymphocytic thyroiditis and specific ultrasonographic abnormalities, namely echogenic septae ( $p -0.042$ ), diffuse hypo-echogenicity ( $p - 0.046$ ).

A study on the cytological grading of Hashimoto's thyroiditis was carried out by Uma *et al.*, (9). Patients with Grades I and II lymphocytic thyroiditis were asymptomatic and had subclinical hypothyroidism, in contrast to Grade II and Grade III cases, which were hypothyroid. Hypoechoic associated with Grade I and Grade II lymphocytic thyroiditis on cytology followed by hypoechoic micronodules were the most often observed sonographic findings.

Kumar *et al.*, (14) attempted the semiquantitative classification of infiltration of lymphocytes as mild and moderate to heavy infiltration in 55 patients of autoimmune thyroiditis. 61.9% of the cases exhibited mild lymphocytic infiltrate, compared to 38.1% of cases with moderate to high lymphocytic infiltration. They discovered an association between the patient's hormone levels and lymphocytic infiltration that was statistically significant ( $p=0.02$ ).

According to Alka *et al.*, (6) there is no statistically significant relationship between the severity of lymphocytic thyroiditis and any clinical, biochemical, or ultrasonographic indicators. It is critical to highlight that their study's relatively small sample size of only 76 cases may be the cause of this lack of a significant correlation. The present study found a strong correlation between the clinical, biochemical, and ultrasonographic criteria for grading lymphocytic thyroiditis, such as echogenic septa and widespread hypoechoic.

## CONCLUSION

The severity of the condition can be determined by the cytological classification of lymphocytic thyroiditis, which additionally predicts thyroid function.

According to cytology findings, asymptomatic and subclinical hypothyroidism are both linked to Grades I and II lymphocytic thyroiditis, while overt cases are linked to Grades II and III. Hypoechoic was the most frequent finding on USG results associated with Grade I as well as Grade II lymphocytic thyroiditis, while the second most frequent feature was hypoechoic micronodules. Ultrasonography, biochemical levels, and grading of lymphocytic thyroiditis paired with each other can identify subclinical hypothyroidism and serve as a therapeutic guide.

## CONFLICT OF INTEREST

The authors declare no conflicts of interest.

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