

Fine Needle Aspiration Cytology of Thyroid Lesions in a Tertiary Care Hospital Catering Predominantly the Tribal Population: Special Reference to Autoimmune Thyroiditis

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ABSTRACT

Introduction: The most common organ specific autoimmune disorder resulting in thyroid dysfunction is Autoimmune Thyroiditis (AITD). Fine Needle Aspiration Cytology (FNAC) is the most sensitive and most specific test for diagnosis of all clinically detectable thyroid lesions. But estimation of serum level of thyroid autoantibodies is necessary for the confirmation of AITD. In India, there are only few numbers of studies related to FNAC of thyroid lesions. There is also lack of convincing data regarding AITD in tribal populations in rural Bengal.

Aim: To analyse FNAC of the thyroid lesions especially in respect to AITD among the tribal populations with biochemical and immunological parameters.

Materials and Methods: The prospective observational study was done from January 2017-December 2018, in Department of Cytopathology and Biochemistry of BSMC, Bankura, India. Total 927 cases of thyroid lesions were diagnosed and classified. They were further associated biochemically and immunologically for the diagnosis of AITD. Data has been analysed in Microsoft Excel 2013 and with the help of Statistical Package for Social Sciences (SPSS)

version 17. Agreement measure has been calculated using Cohen's kappa coefficient statistical test.

Results: Total 432 (46.6%) cases were diagnosed as colloid goitre, adenomatoid goitre was present in 132 (14.24%), malignancy was suspected in 84 (9%) patients, malignancy was present in 159 (17.15%) patients and lastly 120 (12.95%) cases were diagnosed as AITD cytologically. Among the cytologically diagnosed cases of AITD, number of female patients were 102 (85%), and maximum patients were hypothyroid. Among cytologically diagnosed cases of AITD, 113 (94%) patients were found biochemically positive for autoimmunity. The kappa values for FNAC vs anti-Thyroglobulin (TG) and FNAC vs anti-Thyropoxidase (TPO), analysed, came to 0.74 for both, (p-value <0.0001) denoting substantial agreement (0.61-0.82).

Conclusion: FNAC is a sensitive, specific, and accurate initial diagnostic test for evaluation of patients with thyroid swellings. AITD is the most common autoimmune disease affecting mainly the middle aged women. Antithyroid antibodies are good markers for the assessment of thyroid autoimmunity. Anti Thyroid Peroxidase (TPO) antibody is highly specific for AITD.

Keywords: Antithyroid antibody, Cytopathology, Thyroid peroxidase

INTRODUCTION

Thyroid gland is one of the most important endocrine organs secreting active hormones influencing diverse metabolic functions [1]. Goiter, enlargement of thyroid gland, may be diffuse or nodular due to impaired hormone synthesis [2]. AITD is the most common organ specific autoimmune disorder usually present clinically as thyroid dysfunction (hyperfunction, hypofunction or both) [3]. Both humoral and cellular mechanism is responsible for AITD. Circulating autoantibodies exist against thyroglobulin and other follicular cell antigens, notably thyrotropin (TSH) receptors (TSHR). Evidence suggests that AITD is under influence of multiple genes, is multifactorial [4].

In spite of widespread national salt iodisation programme, endemic goitre remains highly prevalent health problem [5]. The genetic predisposition of the Indian population may be the cause behind development of autoimmune goiter [6]. Autoimmunity could be induced by iodine itself in patients, which is evident in animal studies also [3]. Evidence shows iodine supplementation in iodine deficient areas increases prevalence of lymphocytic infiltration in thyroid by three-fold; with 40% increase in prevalence of serum antithyroid antibodies over 0.5-5 years [7].

Considerable lacunae in knowledge remain in clinical assessment of thyroid lesions by means of physical examination, thyroid scan, and Ultrasonography (USG). FNAC considered the most cost-effective,

safe, and initial diagnostic test in evaluation of thyroid lesions. An adequate thyroid aspirate is necessary for the interpretation of FNAC. FNAC primarily helps selecting patients who require surgery for a neoplastic disorder from functional or inflammatory abnormality and who can be followed-up clinically or treated medically [8]. FNAC is extremely useful in diagnosis and monitoring of AITD lesions, especially in clinically equivocal cases and in cases with normal or marginally abnormal biochemical and immunological parameters [9]. Biochemical test like Thyroid Function Test (TFT), anti TPO and anti-Thyroglobulin (TG) antibodies helps further in pinpointing the diagnosis. FNAC being the accurate diagnostic test in thyroid lesion also has certain limitations and diagnostic difficulties. The limitations are false negative and false positive results and a number of FNAC results that are not obviously benign or malignant and fall into the indeterminate or suspicious group [10]. The reported pitfalls are associated with specimen adequacy, sampling techniques, the skill of the physician performing the aspiration, the experience of the pathologist interpreting the aspirated material and the overlying cytological characteristics between some benign and malignant thyroid lesions [11].

The accuracy of cytological evaluation of thyroid lesion is mainly depends on the adequacy of aspirated material. Various radiological guidance mainly USG can be used to increase adequacy of material as well as the accuracy of diagnosis as it is one of the major causes of misdiagnosis. USG is safe and less expensive than other methods

of guidance. It not only helps in accurate localisation of needle tip but also identify the preferable solid area in a predominantly cystic thyroid lesion [12]. There is lack of convincing data regarding AITD in tribal populations in rural Bengal, we found no such study after extensive literature search.

In present study, the FNA of thyroid lesions had been studied in respect to AITD among the tribal populations with biochemical and immunological association.

MATERIALS AND METHODS

The prospective observational study was conducted for the time period of 23 months (January 2017 to December 2018), 18 months for data collection and five months for calculations, in Department of Cytopathology and Biochemistry of Bankura Sammilani Medical College and Hospital (BSMCH), West Bengal, India. After Institutional Ethical Committee approval (BSMC/Aca/3860).

Inclusion criteria: Patients coming for thyroid FNAC for palpable thyroid gland or very small, impalpable thyroid nodule or radiologically confirmed thyroid nodule, who are willing to undergo USG guided FNAC in the Department of Cytopathology, during the time period of the study, were included in this study.

Exclusion criteria: The subjects receiving either thyroxine or any other drug known to interfere with thyroid function at the time of evaluation, old cases of AITD or any other additional lesions observed in association with AITD diagnosed on cytology and patients without written consent were excluded from this study.

Study Procedure

Consecutive sampling method was used, 927 patients presented with thyroid swelling. Among them 120 patients were diagnosed as AITD. Patients attending Cytopathology Department for FNAC of thyroid lesion were approached. Plan of the study was explained in local language. All the patients giving written consent underwent conventional or USG guided FNAC. The estimation of serum TFT was done for all. Immunological tests were done in correlation with cytology to establish the diagnosis of AITD.

STATISTICAL ANALYSIS

Data has been analysed in Microsoft Excel 2013 and with the help of SPSS version 17. Charts and diagrams have been prepared using Microsoft Excel 2013. Sensitivity and specificity have been checked for the investigations done in this study population by using contingency table (2*2), where applicable. Agreement measure has been calculated using Cohen's Kappa coefficient statistical test. The kappa values for FNAC vs both anti-TG and anti-TPO denoting substantial agreement were considered as 0.61-0.82 [13].

RESULTS

In the present study, among 927 subjects, total 432 (46.6%) patients were diagnosed as colloid goitre, adenomatoid goitre was present in 132 (14.2%), malignancy was suspected in 84 (9%) patients, malignancy was present in 159 (17.15%) patients and lastly 120 (12.9%) patients were diagnosed as AITD, female population 102 (85%) were majority [Table/Fig-1]. The age wise sex distribution [Table/Fig-2] revealed young and middle-aged females were predominant among study population. The ethnic distribution revealed 71% of total study population were from tribal origin (n=658). Physical examination revealed 862 patients (93%) presented with diffuse enlargement of thyroid gland, which was noted on physical examination and rest 65 (7%) patients had nodular thyroid swelling.

For most patients, (92%) conventional FNAC (n=853) was sufficient very few patients 74 (8%) needed USG guidance for diagnosis.

| Thyroid lesion | No. (%) |
|-----------------------------------|-------------|
| Colloid goitre | 432 (46.6%) |
| Adenomatoid goitre | 132 (14.2%) |
| Autoimmune thyroiditis | 120 (12.9%) |
| Follicular neoplasm | 48 (5.1%) |
| Suspicious of papillary carcinoma | 36 (3.8%) |
| Papillary carcinoma | 133 (14.3%) |
| Medullary carcinoma | 24 (2.5%) |
| Anaplastic carcinoma | 2 (0.22%) |

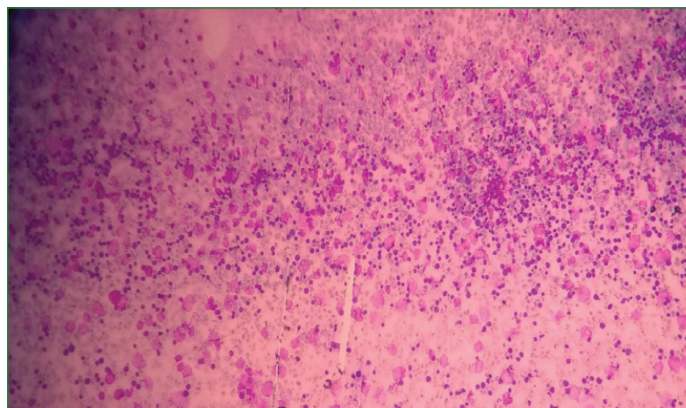
[Table/Fig-1]: Distribution of thyroid lesions (n=927).

| Age group (years) | Female | Male | Total |
|-------------------|--------|------|-------|
| 10-19 | 7 | | 7 |
| 20-29 | 39 | 01 | 40 |
| 30-39 | 53 | 10 | 63 |
| 40-50 | 03 | 07 | 10 |
| Total | 102 | 18 | 120 |
| Percentage | 85% | 15% | 100% |

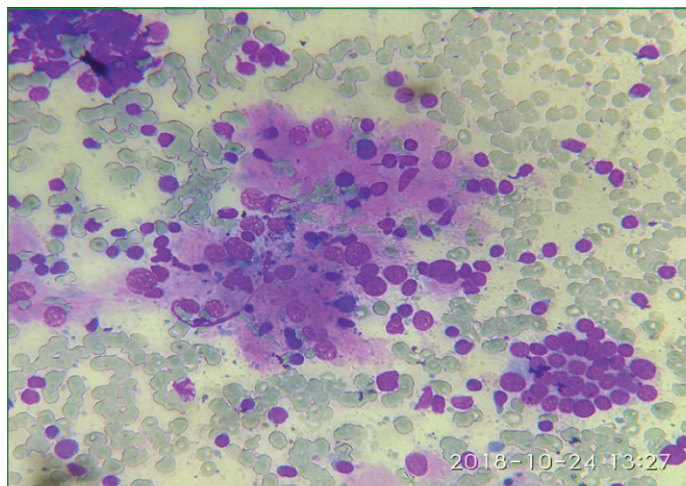
[Table/Fig-2]: Age specific distribution of AITD.

Among 120 cases of cytologically diagnosed AITD, 113 (94%) cases were true positive (biochemically proved).

Smears made from aspirated material showed destruction of thyroid follicles by the lymphocytic infiltrations [Table/Fig-3]. Sometimes there was also presence of plasma cell along with lymphocytes. Presence of hurthle cell directed the diagnosis to Hashimoto Thyroiditis (HT) [Table/Fig-4].



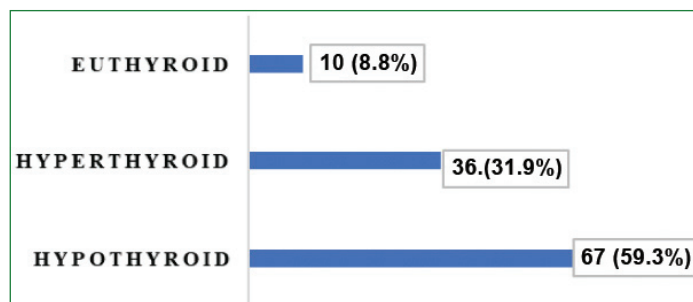
[Table/Fig-3]: Lymphocytic infiltration within thyroid follicular cell (Leishman-Giemsa stain, 10X).



[Table/Fig-4]: Hurthle cell changes and lymphocytic infiltration in Hashimoto's thyroiditis (Leishman-Giemsa stain, 40X).

Among the true positive cases i.e., 113 patients of AITD, maximum 67 (59.3%) patients presented with hypothyroidism [Table/Fig-5].

Now coming to biochemical parameters, all the cytologically diagnosed cases of AITD i.e., among 120 cases, 104 (87%) patients showed antithyroglobulin (Anti-TG) positivity and 113 (94%) patients were positive for anti-TPO.



[Table/Fig-5]: Thyroid status in AITD.

The kappa values for FNAC vs Anti-TG and FNAC vs anti-TPO has been analysed [Table/Fig-6,7], which came to 0.74 for both, (p -value <0.0001), denoting substantial agreement.

| FNAC | Anti TPO | |
|----------|----------|----------|
| | Positive | Negative |
| Positive | 113 | 07 |
| Negative | 26 | 106 |

[Table/Fig-6]: FNAC vs anti-TPO.

| FNAC | Anti TG | |
|----------|----------|----------|
| | Positive | Negative |
| Positive | 105 | 15 |
| Negative | 18 | 114 |

[Table/Fig-7]: FNAC vs anti-TG.

Statistical analysis showed that the false positive rate was 6% and false negative rate was 19%. The sensitivity and specificity were 81% and 94%, respectively. The positive predictive value was 94% and negative predictive value was 80%. Accuracy of FNAC in diagnosis of AITD was 87%.

DISCUSSION

Fine needle aspiration cytology is firmly established as the first line diagnostic test for the evaluation of thyroid lesions and classify and subclassify them [9,14]. USG findings in combination with FNAC of the lesion can increase the accuracy of results almost at par with histology, as claimed by different researchers [15]. In this study, the cytological diagnosis of AITD was correlated with their immunological status. A descriptive analysis of the data of the patients was included in the study.

The exact incidence of Hashimoto's thyroiditis is unknown, may be around 0.3-1.5 cases/1000 population per year. It is around 15-20 times more commonly seen in women as compared to men,

especially around 30-50 years of age. It can be seen in any age group, including children [16]. Most patients in present study with HT were female and belonged to the 30-39 years age group. Thomas T et al., Bhatia A et al. and Shetty A, also concluded same gender ratio with majority of patients were in 3rd and/or 4th decades [17-19]; But differences in disease distribution in specific age group has also been demonstrated in USA and Japan, that is in elderly individuals [20-22].

In this study, 93% of patients with AITD were having diffuse thyroid swelling, while only 8% patients presented with nodular enlargement. A study done by Thomas T et al., showed that among 100 cases of goitrous AITD, 68 patients presented with diffuse enlargement [17]. Mehanathan PB et al., in their study found out 47.9% having multinodular thyroid and 37.5% showed diffuse form both of which were histologically similar [23]. Shetty A, found out diffuse thyroid swelling in 77.4% and nodular thyroid swelling in 22.6% cases among 58 patients [19]. Iha et al., in their study showed diffuse swelling in 87% cases and nodular swelling in 12.9% cases [24].

A AITD is clinically characterised by an initial transient phase of hyperthyroidism followed by a chronic phase of hypothyroidism. Depending on the stage of the disease, the patients presented with features of hypo- or hyperthyroidism. Clinical presentation, hormonal status, and antithyroid antibody values might vary depending on the stage of the disease. Most patients in this study were hypothyroid and only 8.57% patients showed features of hyperthyroidism. These results were like previous studies from India and abroad [17-19,22,24-26].

The diagnosis of AITD is established by correlating clinical findings with cytological and serologic test results. In some cases, the authors found out the predominance of plasma cell which is like other study done by Anila KR et al., [22]. TPO, a transmembrane protein, synthesises thyroid hormones. Thyroid microsomal antibodies i.e., anti-TPO antibody is a thyroid autoantibody targeting TPOs [27].

Anti-thyroglobulin and anti-TPO levels were significantly higher in the AITD goiter group; however, anti-TPO is increased in greater number of cases in comparison to anti-thyroglobulin. Measuring circulating antibodies to thyroglobulin to detect autoimmune thyroid disease is uncommon than measurement of anti-TPO, that gives reliable information about autoimmune thyroid disease [19,24,26,28-30]. Antibody profiling was done in 27% patients in this study, 94% of them were anti-TPO positive and 87% were anti-TG positive. This is in concordance with the fact that TPO Ab is more specific than TG Ab. Comparison with similar studies depicted in [Table/Fig-8]. However, sero-negative cases of HT can be explained based on localised antibody production by intrathyroidal lymphocytes [29].

| Comparative studies | Place of study | No. of patients | Age (year) | Sex | Cytological diagnosis | Clinical presentation (%) | Anti-TPO (%) |
|-------------------------------|--|-----------------|------------|---------------|----------------------------------|------------------------------------|--------------|
| Anila KR et al., (2016) [22] | Regional Cancer Centre, Thiruvananthapuram | 60 | 5-74 | 55 F 5 M | 60 cases-Lymphocytic thyroiditis | Diffuse- 77% Nodular- 23% | 95 |
| Jayaram G et al., (1987) [26] | New Delhi, India | 40 | 40-50 | 40 F | 37 cases-Lymphocytic thyroiditis | - | 57.5 |
| Bhatia A et al., (2007) [18] | Postgraduate Institute of Medical Education and Research, Chandigarh | 76 | 6-60 | 70 F 6 M | 75 cases-Lymphocytic thyroiditis | Diffuse- 89.47% Nodular- 2.63% | 65.7 |
| Shetty A et al., (2019) [19] | Sapthagiri Institute of Medical Sciences, Bangalore | 110 | 30-40 | 55 F 3 M | 79 cases-Lymphocytic thyroiditis | Diffuse- 77.4% Nodular- 22.6% | - 72.13 |
| Iha et al., (2019) [24] | SGRRIM &HS, Dehradun, India | 31 | 13-66 | 30 F 1 M | 31 cases-Lymphocytic thyroiditis | Diffuse- 87.09% Nodular- 12.91% | 83.8 |
| Present study (2021) | BSMCH, West Bengal, India | 120 | 10-50 | 102 F 18 M | 120 cases | Diffuse- 93% Nodular- 7% | 94 |

[Table/Fig-8]: Comparison with similar studies.

Limitation(s)

The limitations of this study were poor generalisability, lack of follow-up and exclusion of patients presented with AITD along with any neoplastic lesion. Due to the special demographic distribution, resulted in only inclusion of tribal population-hence decreasing the result's generalisation. We failed to detect any neoplastic transformation in AITD patients too.

CONCLUSION(S)

To conclude, FNA cytology turned out to be a sensitive, specific, and accurate initial diagnostic test for evaluation of patients with thyroid swellings in this study. Minute search for various cytological features and accurate sampling can help in reducing the number of indeterminate, false-positive, and false-negative diagnoses. However, in difficult situations, an integrated approach will reduce potential pitfalls. In addition, US-FNA is an effective method for diagnosis of sub-centimeter nodules with a low non diagnostic rate in present study, when experienced operators and standard cytological preparation and evaluation were assumed. The most common circulating autoantibodies found to be anti-TPO and anti-TG, among them anti-TPO came out to be more specific than anti-TG in the diagnosis.

REFERENCES

- [1] Mullur R, Liu YY, Brent GA. Thyroid hormone regulation of metabolism. *Physiol Rev.* 2014;94(2):355-82.
- [2] Kumar V, Abbas AK, Aster JC. Robbins and Cotran Pathologic basis of disease. 9th ed. New Delhi: Reed Elsevier India Private Limited; 2015. Pp. 1086-88.
- [3] Franco JS, Amaya-Amaya J, Anaya JM. Thyroid disease and autoimmune diseases. In: Anaya JM, Shoenfeld Y, Rojas-Villarraga A, et al., editors. *Autoimmunity: From Bench to Bedside* [Internet]. Bogota (Colombia): El Rosario University Press; 2013 Jul 18. Chapter 30. Available from: <https://www.ncbi.nlm.nih.gov/books/NBK459466/>.
- [4] Rosai J. Thyroid Gland. In: Chan JKC, Arber DA, Brunning RD, Desmet VJ, Demasters BKK, Ordenez NG, Rosenblum MK, Tallini G, editors. *Rosai and Ackerman's Surgical Pathology*. 10th ed. St. Louis: Elsevier; 2011. Pp. 493.
- [5] Unnikrishnan AG, Menon UV. Thyroid disorders in India: An epidemiological perspective. *Indian J Endocr Metab.* 2011;15(Suppl. 2):78-81.
- [6] Zaletel K, Gaberšček S. Hashimoto's thyroiditis: From genes to the disease. *Curr Genomics.* 2011;12(8):576-88.
- [7] Pearce EN, Farwell AP, Braverman LE. Thyroiditis. *N Engl J Med.* 2003;348(26):2646-55.
- [8] Leopold GK. Koss' Diagnostic Cytology and Its Histologic Basis. 5th ed.: Lippincott William & Wilkins; 2006.
- [9] Jayaram G, Orell SR. Thyroid. In: Orell SR, Sterrett GF, editors. *Fine Needle Aspiration Cytology*. 5th ed. Gurgaon: Reed Elsevier India Private Limited; 2012. Pp. 118-55.
- [10] Zhu Y, Song Y, Xu G, Fan Z, Ren W. Causes of misdiagnoses by thyroid fine-needle aspiration cytology (FNAC): Our experience and a systematic review. *Diagn Pathol.* 2020;15(1):1. <https://doi.org/10.1186/s13000-019-0924-z>.
- [11] Bongiovanni M, Trimboli P, Rossi ED, Fadda G, Nobile A, Giovanella L. Diagnosis of endocrine disease: High-yield thyroid fine-needle aspiration cytology: An update focused on ancillary techniques improving its accuracy. *Eur J Endocrinol.* 2016;174(2):R53-63.
- [12] Kovacevic DO, Skurla MS. Sonographic diagnosis of thyroid nodules: Correlation with the results of sonographically guided fine-needle aspiration biopsy. *J Clin Ultrasound.* 2007;35(2):63-67.
- [13] McHugh ML. Interrater reliability: The kappa statistic. *Biochem Med (Zagreb).* 2012;22(3):276-82.
- [14] Sengupta A, Pal R, Kar S, Zaman FA, Sengupta S, Pal S. Fine needle aspiration cytology as the primary diagnostic tool in thyroid enlargement. *J Nat Sci Biol Med.* 2011;2(1):113-18.
- [15] Lee MJ, Hong SW, Chung WY, Kwak JY, Kim MJ, Kim EK. Cytological results of ultrasound-guided fine-needle aspiration cytology for thyroid nodules: Emphasis on correlation with sonographic findings. *Yonsei Med J.* 2011;52(5):838-44.
- [16] Akamizu T, Amino N. Hashimoto's Thyroiditis. [Updated 2017 Jul 17]. In: Feingold KR, Anawalt B, Boyce A, et al., editors. *Endotext* [Internet]. South Dartmouth (MA): MDText.com, Inc.; 2000-. Available from: <https://www.ncbi.nlm.nih.gov/books/NBK285557/>.
- [17] Thomas T, Sreedharan S, Khadilkar UN, Deviprasad D, Kamath MP, Bhojwani KM, et al. Clinical, biochemical & cytomorphologic study on Hashimoto's thyroiditis. *Indian J Med Res.* 2014;140(6):729-35.
- [18] Bhatia A, Rajwansi A, Dash RJ, Mittal BR, Saxena AK. Lymphocytic thyroiditis-is cytological grading significant? A correlation of grades with clinical, biochemical, ultrasonographic and radionuclide parameters. *Cytojournal.* 2007;4:10. Doi: 10.1186/1742-6413-4-10.
- [19] Shetty A, Chowdappa V. Cytomorphological Spectrum of Hashimoto's Thyroiditis and Its Correlation with Hormonal Profile and Hematological Parameters. *J Cytol.* 2019;36(3):137-41.
- [20] Hollowell JG, Staehling NW, Flanders WD, Hannon WH, Gunter EW, Spencer CA, et al. Serum TSH, T(4), and thyroid antibodies in the United States population (1988 to 1994): National Health and Nutrition Examination Survey (NHANES III). *J Clin Endocrinol Metab.* 2002;87(2):489-99.
- [21] Dong YH, Fu DG. Autoimmune thyroid disease: Mechanism, genetics and current knowledge. *Eur Rev Med Pharmacol Sci.* 2014;18(23):3611-18.
- [22] Anila KR, Nayak N, Jayasree K. Cytomorphologic spectrum of lymphocytic thyroiditis and correlation between cytological grading and biochemical parameters. *J Cytol.* 2016;33(3):145-49.
- [23] Mehanathan PB, Erusan RR, Shantaraman K, Kannan SM. Antithyroid peroxidase antibodies in multinodular hashimoto's thyroiditis indicate a variant etiology. *J Thyroid Res.* 2019;2019:4892329.
- [24] Iha, Bhardwaj A, Kumar R, Acharya S. Cytomorphologic study of lymphocytic thyroiditis: A correlation between cytological grade and biochemical parameters. *Journal of Clinical and Diagnostic Research.* 2019;13(6):EC05-09.
- [25] Jayaram G, Iyengar KR, Sthaneshwar P, Hayati JN. Hashimoto's thyroiditis-A Malaysian perspective. *J Cytol.* 2007;24(3):119-24.
- [26] Jayaram G, Marwaha RK, Gupta RK, Sharma SK. Cytomorphologic aspects of thyroiditis. A study of 51 cases with functional, immunologic and ultrasonographic data. *Acta Cytol.* 1987;31(6):687-93.
- [27] Czarnocka B, Ruf J, Ferrand M, Carayon P, Lissitzky S. Purification of the human thyroid peroxidase and its identification as the microsomal antigen involved in autoimmune thyroid diseases. *FEBS Lett.* 1985;190(1):147-52.
- [28] Gürkan Dumlu E, Kiyak G, Bozkurt B, Tokaç M, Polat B, Cakir B, et al. Correlation of thyroid fine-needle aspiration with final histopathology: A case series. *Minerva Chir.* 2013;68(2):191-97.
- [29] Rath M, Ahmad F, Budania SK, Awasthi S, Kumar A, Dutta S. Cytomorphological aspects of Hashimoto's Thyroiditis: Our experience at a tertiary center. *Clin Med Insights Pathol.* 2014;7:01-05. Doi: 10.4137/CPATH.S13580.
- [30] Kandi S, Rao P. Anti-thyroid peroxidase antibodies: Its effect on thyroid gland and breast tissue. *Ann Trop Med Public Health.* 2012;5:01-02.

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