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Original Research Article

Comparative study of fine needle aspiration cytology and histopathology correlation of various thyroid lesions

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ABSTRACT

Background: Patients with anterior neck swelling constitute common problems encountered in the clinical practice. In all age groups, including children, thyroid cancer is the most prevalent endocrine system cancer. Initial diagnostic testing using fine needle aspiration cytology (FNAC) has been found to be quick and cost effective. Therefore, it is widely accepted and more thyroid cases are diagnosed at an early stage, but it is associated with pitfalls due to difference in correlations between FNAC and Histopathological Examination (HPE). The purpose of the study is to compare the results of FNAC with histopathological findings in order to evaluate the diagnostic efficacy of FNAC.

Materials and Methods: The present study was performed over a period of two years in a tertiary care center. Included in the study were 100 cases of thyroid nodule, which underwent FNAC and the post-operative specimens of whom were subjected to histopathological examination. The results obtained by FNAC were compared with histopathological findings.

Results: Out of the 100 cases taken for the purpose of the study, 92 were benign whereas 8 were malignant. The cytological and histopathological diagnosis correlated with each-other in 89 cases. A partial correlation between the cytological and histopathological diagnoses was found in 5 cases. In 6 cases the FNAC diagnosis was not correlating with histopathological diagnosis. FNAC revealed a sensitivity of 62.50%, a specificity of 97.83%, an accuracy of 95%, a positive predictive value of 71.43% and a negative predictive value of 96.77%.

Conclusions: FNAC of thyroid lesions is thus proved to be a simple, cost effective and accurate method to diagnose and to guide the management of palpable thyroid lesions. Thus, we can see that the diagnostic accuracy of FNAC procedure is very high making it a very useful investigation in the work-up of thyroid patients. However final diagnosis and treatment pattern should be based upon histopathology.

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1. Introduction

The thyroid is a butterfly-shaped gland located in the anterior part of the neck. Thyroid swellings are typically accompanied by neck swelling, which is a common clinical symptom. Thyroid cancer is most common cancer of endocrine system occurring in all age groups including

children and fastest growing cancer in both men and women.¹ The prevalence of thyroid nodules ranges from 4% to 10% in the general adult population and from 0.2% to 1.2% in children.² The majority of clinically diagnosed thyroid nodules are nonneoplastic; only 5%–30% are malignant and require surgical intervention.³ Many times these conditions cannot be diagnosed on the basis of clinical history and examination and require radiological and Fine

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Needle Aspiration Cytology testing for their diagnosis.⁴

Fine Needle Aspiration Cytology is a minimally invasive technique where cells are aspirated using a syringe and fine needle. It has been recommended by American Thyroid Association⁵ and National Comprehensive Cancer Network State as an initial diagnostic test because of its simplicity and cost-effectiveness. Worldwide FNAC has now been used increasingly to avoid unnecessary thyroid surgery.⁶ The primary goal of FNAC is to identify nodules that require surgery and those benign nodules that can be seen clinically, as well as to reduce the overall thyroidectomy rate in patients with benign diseases. This cuts down on unwanted hospital stay and operations and is cost-effective.⁷ The whole procedure from aspiration to processing of the smears can be completed with-in hours. Rapid diagnosis relieves patient's anxiety and saves precious time in planning a definitive treatment.

Not only has FNAC decreased the number of unnecessary thyroid surgeries, but also helped to diagnose many benign conditions infective, inflammatory and neoplastic conditions.⁸ It has increased the ratio of malignant to benign lesions resected. Though it cannot be a substitute to surgical histopathology, it is an extremely valuable diagnostic tool in the diagnosis and management of patients with thyroid lesions. Different ancillary tests like Radio-nucleotide ultrasonography, High resolution ultrasonography (USG) and thyroid antibodies are being used now-a-days in the diagnosis of thyroid swellings. However, FNAC of thyroid still is the simplest, cost-effective test with high sensitivity, specificity and diagnostic accuracy, especially for cystic lesions.⁹

Therefore, present study was undertaken to correlate the FNAC and histopathological findings of various thyroid lesions.

2. Aim

To correlate the FNAC and histopathological findings of various thyroid lesions

3. Objectives

1. To study the cytomorphological features in various thyroid lesions and to provide accurate diagnosis.
2. To correlate the cytological features with histopathological study of all surgically removed thyroid lesions.
3. To evaluate sensitivity, specificity and accuracy of FNAC as a procedure in diagnosing various thyroid lesions.

4. Materials and Methods

The present study was undertaken in a tertiary care hospital over a period of two years from December 2019 to November 2021. The study comprised of 100 patients who

presented with the history of swelling of thyroid (Figure 1) which were referred from the Departments of Surgery, Medicine & ENT.

Every patient underwent a complete clinical examination as per the proforma. To determine precisely where to aspirate thyroid tissue, the thyroid gland was palpated carefully. After a brief explanation of the procedure to the patient, the thyroid was aspirated with the patient supine or sitting with their necks extended, to make the thyroid swelling appear prominent. Under aseptic precautions, 21-gauge needle with syringe was inserted into the lesion and to and fro movements performed quickly. Under negative pressure, material got collected in the needle, after collection of material negative pressure gets released, needle with syringe holder was removed, the material spreads over a clean labelled slide and smears were prepared.

Smears were wet fixed in 95% ethyl alcohol and stained with Hematoxylin and Eosin (H & E). Whenever fluid was obtained, all the contents were aspirated and centrifuged. Smears were made from the sediment and stained by the stain described earlier. After surgery, the received specimens were fixed with 10% formalin and detailed gross examination was done and sections were taken from the representative areas for paraffin sections and stained by H & E. The sections were studied under light microscopy.

Cytological diagnosis was correlated with histopathology where ever possible and efficacy of FNAC was estimated by using the following methodology of Galen and Gambino.¹⁰

4.1. Terminology

1. True positive (TP) = the number of cases correctly identified as having thyroid neoplasm
2. False positive (FP) = the number of cases incorrectly identified as having thyroid neoplasm
3. True Negative (TN) = the number of cases correctly identified as not having thyroid neoplasm
4. False Negative (FN) = the number of cases incorrectly identified as not having thyroid neoplasm
5. Sensitivity measures the percentage of patient who are correctly identified as having thyroid neoplasm. Thus, sensitivity = $TP / (TP + FN)$
6. Specificity measures the percentage of patient who are correctly identified as not having thyroid neoplasm. Thus, specificity = $TN / (TN + FP)$
7. Accuracy measures ability of fine needle cytology to correctly identify the cases that having thyroid neoplasm and the cases that not having thyroid neoplasm. Thus, accuracy = $(TP + TN) / (TP + FP + TN + FN)$
8. Positive Predictive Value (PPV) is the proportion of positives that correspond to the presence of the thyroid neoplasm. Thus, PPV = $TP / (TP + FP)$

9. Negative Predictive Value (NPV) is the proportion of negatives that correspond to the absence of the thyroid neoplasm. Thus, $NPV = TN / (TN + FN)$

5. Results

The present study was performed over a period of two years in a tertiary care center. Included in the study were 100 cases of thyroid nodules, which underwent FNAC and their respective post-operative specimens were subjected to HPE. The results of the present study were as follows-lesions

5.1. Age

The age group of the patients included in the study was from 18 years to 77 years of age. Most of the cases were clustered around 18-40 years of age. The non-neoplastic lesions were concentrated in the age groups of 21-40 and 41-60 years. The neoplastic lesions have a bimodal peak at 21-40 years and then at 61-80 years of age.(Table 1)

5.2. Sex

Majority of the patients were females. Out of total of 100 patients, 79 were female and the rest 21 were male.(Table 2)

5.3. Benign and malignant lesions

Out of the 100 cases taken for the purpose of the study, 92 were benign whereas 8 were malignant.(Table 3) The benign lesions consisted majority of multinodular goitre-57 cases, (Figure 2) Hashimoto's cases-10, hyperplastic nodules-7 cases, follicular adenoma-19 cases and thyroglossal cyst- 2 cases. Among the 8 malignant lesions, 5 were papillary carcinoma of the thyroid gland, one was follicular carcinoma, one was medullary carcinoma and one was anaplastic thyroid carcinoma.

In this study among the 100 cases taken for study, in 89 cases the cytological and histopathological diagnosis correlated with each-other. There were 5 cases where the cytological diagnosis partially correlated to the histopathological diagnosis. These included cases where the FNAC slides showed goiter but missed the thyroiditis along with it. This is a well-known drawback of the FNAC procedure.

In six cases the FNAC diagnosis did not correlated with histopathological diagnosis.(Table 4) These included 3 cases where the FNAC found only cyst fluid but on histopathology they turned out to be multinodular goiter.(Figures 3 and 4) In one case of medullary carcinoma on FNAC, diagnosis of goiter was given. One case of goiter turned out to be adenomatoid nodule.(Figure 5) The FNAC also failed to diagnose follicular variant of papillary carcinoma.

Overall, present study shows 62.50% sensitivity, 97.83% specificity and 95% accuracy. Thus, we can see that the

diagnostic accuracy of FNAC procedure is very high making it a very useful investigation in the work-up of thyroid patient. In our study, negative predictive value was 71.43% and positive predictive value was 96.77%.

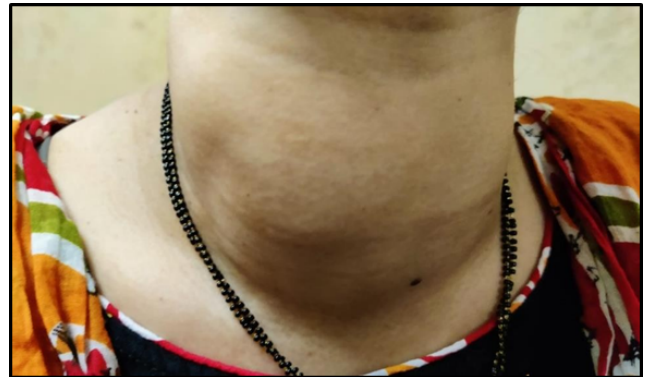


Fig. 1: Photograph of Anterior Neck swelling in patient

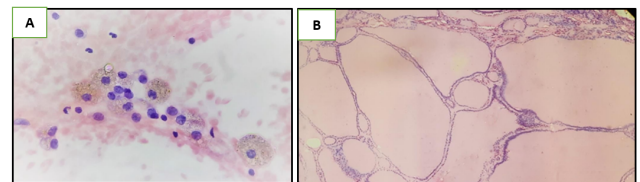


Fig. 2: **A):** Photomicrograph of Colloid Goiter with cystic change- (H & E stain, 40x); **B):** Photomicrograph showing multi-nodular goiter on histopathology- (H & E stain, 40x)

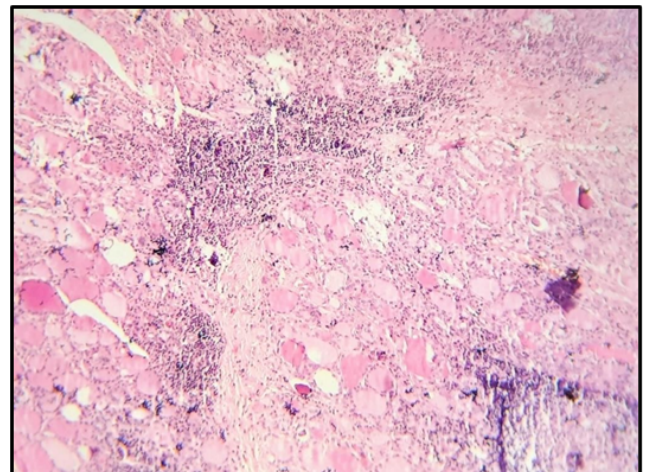


Fig. 3: Photomicrograph of smear showing only cyst macrophages-(H & E stain,40x)

6. Discussion

The current study examined the cytological characteristics of thyroid lesions and determined the management of

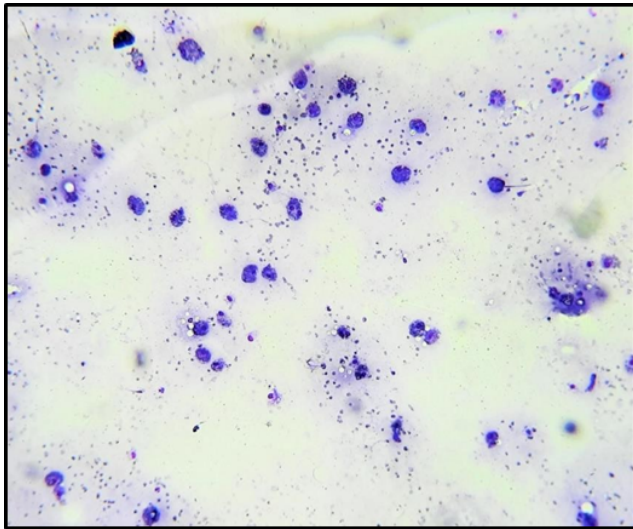


Fig. 4: Photomicrograph of smear showing only cyst macrophages-(MGG,40x)

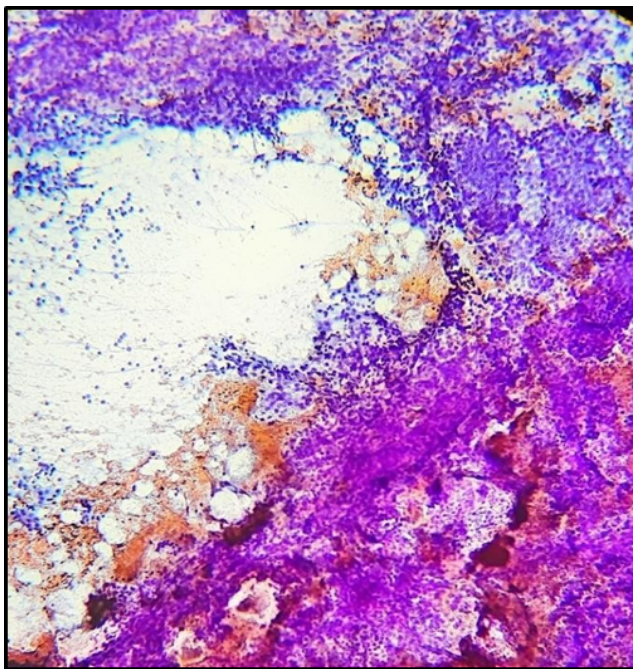


Fig. 5: Photomicrograph showing Adenomatoid Nodule on Histopathology-(H & E stain,10x)

Table 1: Distribution of patients according to age

Age in years	Non-neoplastic	Neoplastic	Total
0-20	7	2	9
21-40	39	4	43
41-60	36	0	36
61-80	10	2	12
			100

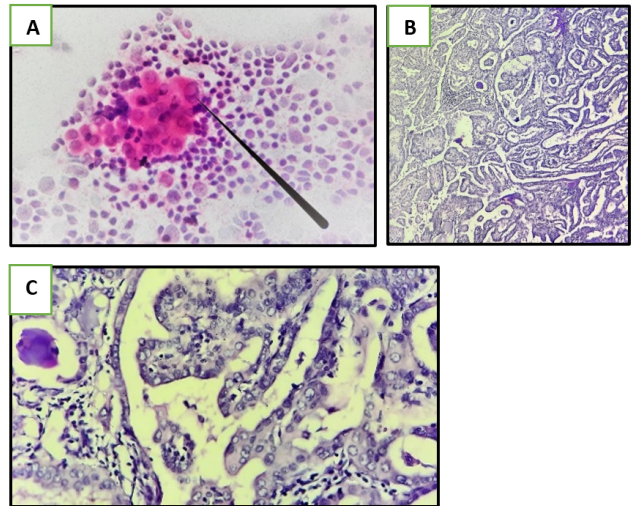


Fig. 6: **A):** Photomicrograph of Papillary carcinoma showing pseudo inclusions-(H & E stain, 40x); **B):** Photomicrograph showing Papillary Carcinoma Thyroid on histopathology-(H & E stain,10x); **C):** Photomicrograph of Papillary Carcinoma Thyroid showing Psammoma bodies and optically clear nuclei- (H & E stain, 40x)

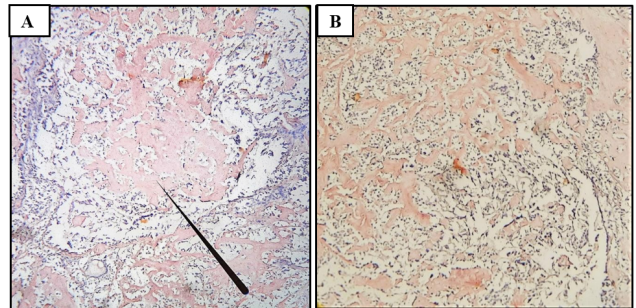


Fig. 7: **A):** Photomicrograph showing Medullary Carcinoma Thyroid-(H & E stain, 10x); **B):** Photomicrograph of Congo red stain in Medullary Carcinoma Thyroid showing amyloid-(10x)

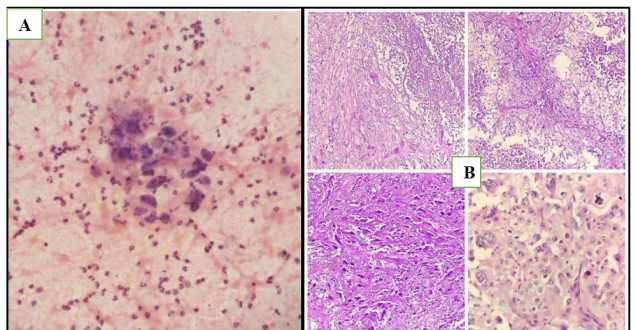


Fig. 8: **A):** Photomicrograph showing FNAC smear of anaplastic thyroid carcinoma – (H & E stain 40x); **B):** Photomicrograph showing histopathology of anaplastic thyroid carcinoma with squamoid, clear cells, spindle cells and many bizarre giant cells with mitosis- (H & E stain 40x)

Table 2: Distribution of patients according to gender

Gender	Frequency	Percent
Male	21	21%
Female	79	79%
Total	100	100%

Table 3: Distribution of benign and malignant thyroid lesions

Category	Frequency	Percent
Benign	92	92%
Malignant	8	8%
Total	100	100%

Table 4: Correlation of cytological and histopathological diagnosis of patient

Correlation	Frequency	Percent
Completely correlating	89	89%
Partially correlating	5	5%
Not correlating	6	6%
Total	100	100%

Table 5: Categorization of thyroid lesions according to the Bethesda category

Diagnostic Category	Frequency	Percent
I-Non diagnostic or unsatisfactory	4	4%
II-Benign	76	76%
III- Atypia of Undetermined Significance (AUS)/Follicular Lesion of Undetermined Significance (FLUS)	2	2%
IV-Follicular Neoplasm /Suspicious for a Follicular Neoplasm	14	14%
V-Suspicious for malignancy	1	1%
VI-Malignant	3	3%
Total	100	100%

Table 6: Correlation between FNAC and HPE

	Histopathology-Positive	Histopathology-Negative
Cytology/FNAC-Positive	5	2
Cytology/FNAC-Negative	3	90

these patients by categorizing them according to the Bethesda categories used for evaluating the thyroid lesions. The cytological findings were also correlated to the histopathological findings of these patients.

6.1. Age distribution

The present study included patients between the ages of 18–77 years of age. Majority of the cases are clustered in the age group of 20–40 years of age. The age distribution of present study was also compared to Roy PK et al¹¹ and Vargis RK et al¹² study. Babu SBK et al¹³ observed majority of patients

Table 7: Statistical values of FNAC-HPE

Statistic	Value	95% CI
Sensitivity	62.50%	24.49% to 91.48%
Specificity	97.83%	92.37% to 99.74%
Positive Likelihood Ratio	28.75	6.60 to 125.31
Negative Likelihood Ratio	0.38	0.16 to 0.94
Disease prevalence	8.00%	3.52% to 15.16%
Positive Predictive Value (PPV)	71.43%	36.45% to 91.59%
Negative Predictive Value (NPV)	96.77%	92.46% to 98.66%
Accuracy	95.00%	88.72% to 98.36%

belonged to age group between 30–45 years.(Table 1)

6.2. Sex distribution

Majority of the patients with thyroid lesions were females. In 100 cases, 79 were female and the rest 21 were male. This gives a male to female ratio of 1:3.7 in the present study. Similar findings were seen by Gupta M et al¹⁴ and Naveen Kumar C et al¹⁵ study. In a study by Rout K et al,¹⁶ 69.73% of patients were females, which was the comparable to finding in our study where 79% of the patients were females. (Table 2)

6.3. Comparative incidence of neoplastic and non-neoplastic lesions

Out of 100 cases, 92 were benign/ non-neoplastic in nature. Only 8 cases were malignant/ neoplastic in nature. Amongst the non-neoplastic cases, 57 were nodular colloid goiter, 10 Hashimoto thyroiditis, 2 thyroglossal cyst, 16 follicular adenomas and 7 hyperplastic nodules. Out of 8 neoplastic lesions, 5 were papillary carcinoma, 1 follicular neoplasm, 1 medullary carcinoma and 1 anaplastic thyroid neoplasm. Overall, the ratio of non-neoplastic to neoplastic conditions is 1:11.5 in the present study. The present study when compared with studies results was comparable with studies conducted by Kumar HC et al,¹⁷ Gangadhara KS et al¹⁸ and Handa U et al study.¹⁹ (Table 3)

6.4. Categorization of thyroid lesions according to the Bethesda category²⁰

The thyroid lesions were categorized based on FNAC findings in the six Bethesda categories of reporting the thyroid lesions. Using the Bethesda system for reporting the thyroid lesions is very effective as it provides uniform terms of reporting across all the hospitals. Also, it provides risk assessment for benign tumor turning into malignant.(Table 5)

6.5. Bethesda category-I (Non-diagnostic or unsatisfactory): 4%

In this study these were the patients of long-standing nodular goiter which showed cystic change. On FNA sampling we found only cyst macrophages in the fluid.

6.6. Bethesda category -II (Benign): 76%

This was major category comprised of multinodular goiter, Hashimoto's thyroiditis, adenomatoid goiter and follicular adenoma. Similar finding observed by Gangadhara KS¹⁸ study which showed 88.4% of cases belonged to category II.

6.7. Bethesda category III-(Atypia of undetermined significance/follicular lesion of undetermined significance): 2%

One of these patients was a case of nodular hyperplasia and the other was a case of long-standing multinodular goiter, which can sometimes show atypical cells in the smear. However, the atypia was not that significant to categorize it into the suspicious for malignancy –category 6. On histopathology these patients were found out to have benign lesions.

6.8. Bethesda category IV-(Follicular neoplasm or suspicious for follicular neoplasm): 14%

For diagnosis a follicular carcinoma we must show capsular and/ or vascular invasion. As these things cannot be visualized on the FNA smear, follicular carcinoma is a histopathological diagnosis.

6.9. Bethesda category V-(Suspicious for Malignancy): 1%

This was the patient of long-standing multinodular goiter and came with recent rapid enlargement of lesion. On gross examination, the swelling was hard to cut and showed variegated appearance. On the cut surface with areas showing necrosis, hemorrhage and calcification. The FNA smears showed atypical smears with squamoid appearance. Microscopy revealed some areas of squamous and clear cell differentiation along with squamoid, giant and spindle cells. Areas of necrosis and hemorrhage were also noted. On Immunohistochemistry (IHC) the tumor was positive for PAX8, CK19, P40, P 63 CD10, CK5/6, MIB1 and negative for Thyroglobulin, TTF1, PAX2 and Calcitonin. Based on the patient's history, FNAC smear findings, gross and microscopic findings and IHC markers, patient was diagnosed as anaplastic thyroid carcinoma.

6.10. Bethesda category VI-(Malignant):3%

In this study all the patients in this category were of papillary carcinoma thyroid.(Figure 6) On the cytology smears, thyroid follicular cells were seen in the papillary pattern. On the histopathology, we could appreciate the optically clear nucleus, psammoma bodies and pseudo inclusions in these cases. One case of papillary thyroid carcinoma also had lymph node metastasis.

6.11. Non-Neoplastic/ Benign lesions

Out of 100 cases, about 92 cases were benign in nature. As we can see the sensitivity of FNAC in providing the diagnosis in benign cases is 96%. The specificity is however only 71%. The major reason for missing out diagnosis is because in a cystic lesion, there might be focus of malignancy which will be missed if we don't do correct and accurate sampling of the patient.

In the present study a case of papillary carcinoma was diagnosed as colloid goiter on FNAC as sampling was only done from one nodule which showed goiter. Another case of papillary carcinoma with foci of Hurthle cell change was diagnosed as Hurthle cell neoplasm on FNAC.

6.12. Malignant/ Neoplastic lesions

Out of 100 cases 8 were malignant/ neoplastic. They comprised of 5 papillary carcinomas, 1 follicular carcinoma, 1 medullary carcinoma (Figure 7) and 1 anaplastic carcinoma.(Figure 8) The specificity of FNAC procedure for diagnosing malignant lesions is very high. However, in the present study the sensitivity of FNAC for finding out malignant cases is only 62.5%. These findings can be attributed to learning curve of the students doing FNAC procedure, the processing of the slides and patients coming to institute with long-standing thyroid lesions which have now showed secondary changes like cystic degeneration. In the present study there were 2 cases where the FNAC showed atypia of undetermined significance (AUS) and subsequent histopathology showed them to be benign in nature.

6.13. Cytology and histopathology diagnosis of papillary thyroid lesions

In the present study comprised of 5 cases of papillary carcinoma thyroid.(Table 6) Three (60%) of these cases were conventional papillary carcinoma and were correctly diagnosed on FNAC. One case had papillary carcinoma in a cystic lesion, the sampling of which was missed and lead to a false diagnosis of colloid goiter. One case of papillary carcinoma thyroid had focal hurtle cell change which led to a diagnosis of hurtle cell neoplasm. These findings were differed with Naveen Kumar C et al,¹⁵ Gagnetten CB et al²¹ and Gupta M.et al¹⁴ studies as the sample size of the study

were smaller for an accurate assessment.

6.14. Cytology and histopathology diagnosis of follicular thyroid lesions

Out of 2 cases of Follicular neoplasm, 1 case turned out to be follicular carcinoma on histopathology with a diagnostic accuracy of 50%. The diagnostic accuracy of FNAC in diagnosing Follicular neoplasm in the present study is comparable to the Silverman JF et al²² and Hall TL et al study.²³

6.15. Cytology and histopathology diagnosis of medullary and anaplastic thyroid lesions

The medullary carcinoma was diagnosed on histopathology and was reported as colloid goiter on FNAC. One case of anaplastic thyroid carcinoma was categorized as suspicious for malignancy Bethesda category IV on FNAC. dFNAC diagnosis of thyroid lesions.

The present study showed sensitivity of 62.50%, specificity of 97.83% and diagnostic accuracy of present study of 95%. (Table 7) Therefore, probability of having malignant disease following positive FNAC results for malignancy i.e., PPV was 71.43%. Therefore, probability of not having malignant disease following negative FNAC results for malignancy i.e., NPV was 96.77%.

On comparison with other studies the present study showed sensitivity of 62.50% which was comparable with study conducted by Ahmed MT et al study.²⁴ Same was demonstrated as 73.91% in a study by Reddy MR et al.²⁵ The highest sensitivity was reported in the study conducted by Handa U et al¹⁹ showing 97%. The present study showed specificity of 97.83% which was comparable with study conducted by Gangadhara KS et al.¹⁸ Our findings can be compared with observation of Kumar HC et al¹⁷ who reported specificity of FNAC was 87.5%. The highest specificity reported in the studies mentioned was in a study conducted by Babu SBK et al¹³ showing 100%. Srilekha Bodepudi et al²⁶ evaluated thyroid swelling in 93 cases and in their study FNAC demonstrated a sensitivity of 82.35%, and a specificity of 95.18%.

In our study the accuracy of FNAC in detection of the thyroid lesions was found to be 95%. It is well compared with the study done by Babu SBK,¹³ in which the accuracy was reported as 94%, and another study done by Abdullahi IM et al²⁷ the accuracy was 94.9%. The diagnostic accuracy of Correlation between FNAC diagnosis and final HPE diagnosis was 91.66% in a study by Gangadhara KS et al.¹⁸

The present study showed PPV of 71.43% which was accordance with studies conducted by Gangadhara KS et al¹⁸ and Srilekha Bodepudi et al.²⁶ In our study the NPV was found to be 96.77% which was similar of studies conducted by Gangadhara KS et al¹⁸ and Babu SBK et al.¹³ In a study by Reddy MR,²⁵ PPV and NPV of FNAC in the

diagnosis of thyroid lesions was evaluated to be 80% and 73%.

7. Conclusions

Fine Needle Aspiration Cytology of Thyroid lesions is thus proved to be a simple, cost effective and accurate method to diagnose and to guide the management of palpable Thyroid lesions. The method can accurately diagnose non-neoplastic Thyroid conditions and reduce the number of unnecessary Thyroid surgeries. In addition, it is also a simple procedure which does not require a long learning curve. The materials required for the test are easily available and hence this makes it a cost-effective test.

Accuracy of the results of the test depend on factors like skill of the person performing the test, nature of the swelling, preparation of the smears and the knowledge and expertise of the person interpreting the smears. The sensitivity, specificity and the diagnostic accuracy of the procedure is very good. This makes it a suitable first line investigation for the diagnosis of thyroid lesions and to guide the management of the patients with thyroid nodule.

Thus, we can very well say that though HPE is the final and confirmatory diagnostic procedure, FNAC remains the preliminary time-tested procedure of choice.

8. Source of Funding

No funding sources.

9. Conflict of Interest

None.

10. Ethical Approval

The study was approved by the Institutional Ethics Committee (IEC).

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