

## A prospective study on cytology of thyroid lesions, its correlation with histopathology and evaluate diagnostic accuracy of FNAC in thyroid pathologies

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### Abstract

**Introduction:** Thyroid lesions are commonly seen in day to day clinical practice with an annual incidence rate of 4-7%. A preliminary and effective diagnostic tool is required to provide early diagnosis. FNAC is a handy technique and play an important role in early and differential diagnosis of benign and malignant lesions.

**Aims:** The aim of this study was to find out the spectrum of thyroid lesions with respect to age and sex, evaluate the diagnostic accuracy of FNAC in thyroid lesions by correlating the cytological finding with histopathology.

**Materials and Methods:** A prospective study was done during the period from 2012 to 2017. FNAC of all cases admitted to ENT and surgery ward with thyroid nodule was performed, irrespective of age and sex and all corresponding surgical specimens received in pathology department of thyroid lesions were collected. Thyroid lesions were classified according to Bethesda system of reporting. The statistical analysis was done.

**Results:** A total of 291 patients were evaluated in this study, FNAC was done on all cases. The thyroid lesions are more common in females (245 cases) than males (46 cases). Benign (240 cases) lesions are common than malignant lesions (27 cases). Benign lesions are more commonly seen in age group of 21-30 years (33.68%) and malignant ones in age group of 41-50 years (28.87%). Diagnostic accuracy in our study was 96.4%.

**Conclusion:** Thyroid lesions can be effectively categorized and reported by following the Bethesda system which minimizes the false positive cases. In this study FNAC of thyroid lesions has a high accuracy in differentiating between benign and malignant lesions.

**Keywords:** Diagnostic accuracy, FNAC, Histopathology, Sensitivity, Specificity, Bethesda.

### Introduction

Thyroid lesions are commonly encountered in day to day practice. Thyroid nodules show an incidence rate of 4-7%.<sup>1</sup> FNAC is an effective diagnostic tool in distinguishing benign and malignant lesions, provides an early diagnosis and avoid unnecessary surgery. FNAC has led to an appreciable decrease in the surgical treatment of the patients with thyroid nodular diseases and has increased the percentage of operated malignant nodules or suspicious of malignancy by 50%. FNAC is fast, safe, cost effective, minimally invasive, efficient and have high sensitivity and specificity.<sup>2</sup> However the major limitation of FNAC is its insensitivity in correctly diagnosing malignant follicular lesions and a proportion of cases cannot be evaluated due to scanty material. Despite its limitation, routine aspiration cytology is the most valuable and appropriate investigation in patients with thyroid lesions.<sup>3</sup> Against this background, in our study we have compared the cytological and histopathological diagnosis of patients who underwent thyroid surgery and aimed to evaluate the diagnostic accuracy of FNAC in terms of sensitivity, specificity and predictive values, which is over 90% in cases of malignancy.

### Materials and Methods

A prospective and interventional study was carried out in our hospital attached to a medical college. This study was conducted on approval from ethical

committee during a period of 5 years from 2012-2017. We included all the patients irrespective of age and sex referred for FNAC of thyroid nodule, who had also underwent surgery of thyroid gland. We excluded inadequate specimen and poorly preserved samples. 291 FNACs were done during this period. Thyroid nodule was defined as clinically palpable single or discrete lesion involving either lobes or the isthmus of the thyroid gland. Out of 291 patients, histopathological confirmation was available in 283 cases.

All the patient were evaluated in following order-history, clinical examination, thyroid function test, informed written consent, FNAC and histopathology.

Fine needle aspiration was done in position as per standard guidelines recommended by Svante R Orell.<sup>4</sup> The material was obtained by using a 2.5 cm long, 23-25 gauge needle attached to a 10cc disposable syringe by applying suction. Multiple smears were prepared and are equally air dried and wet fixed. Wet fixed smear were immediately fixed in ethanol taking care to avoid air dried artifact and were stained with papanicolaou stain. The air dried smears were stained with Giemsa stain.

Specimen was considered "unsuitable/inadequate" for cyto -diagnosis if consisted mainly of blood or if cellular material was absent. For a specimen to be considered adequate, 5-6 clusters of cells with more than 10 cells per cluster in at least two slides prepared from different aspirates.<sup>4</sup> However, adequacy also

depends upon lesion being aspirated. For example in case of colloid goiter FNAC many times yielded colloid with scanty cells, but it has been considered adequate. "Follicular pattern lesion" forms a grey zone in cytology and it is difficult to rule out malignancy in these lesions.<sup>5</sup> As the paper was specifically aimed to find out the efficacy of FNAC, and to keep our comparison clear, we included all cystic or follicular pattern lesion without atypia in benign category and those reported as suspicious in "malignant" category. All cystic lesion or follicular pattern lesions with atypia are classified in "suspicious category"

The cases were followed by postoperative specimen collected in 10% formalin and allowed to fix for 24 hours. Detailed gross examination was done, tissue were processed, sections were cut and evaluated after haematoxylin and eosin staining.<sup>6</sup>

Thyroid lesions were classified according to Bethesda system of reporting. The results of FNAC were compared with histopathology in 282 cases. The statistical analysis included sensitivity, specificity, false positive rate, false negative rate, positive predictive value, negative predictive value and accuracy.

## Results

A total of 291 patients were evaluated on which FNAC was done, but 9 out of which were excluded because of inadequacy. 282 patients having thyroid lesions form the study population, where both FNAC and subsequent histopathology examination was done. Table 1 show the age and sex distribution of all patients of thyroid lesions.

**Table 1: Age and sex distribution of patients**

Age group (years)	No. of cases			%
	Male	Female	Total	
<20	4	5	9	3.09 %
21-30	15	83	98	33.68%
31-40	10	64	74	25.43%
41-50	12	72	84	28.87%
51-60	5	17	22	7.56 %
>60	-	4	4	1.37 %
Total	46 (15.81%)	245 (84.19%)	291	100 %

Out of total 291 cases, 46(15.81%) were male patients and 245(84.19%) were female patients. The male to female ratio being 1:5.1. Females outnumbered males in all age groups. There was a significant association found between benign and malignant thyroid disease with gender and was higher towards the female side.

Following the Bethesda classification, in the present study Table 2 shows the distribution of thyroid lesions in which 240 cases (82.47%) out of 282 were diagnosed as benign, 27 cases (9.29%) were diagnosed

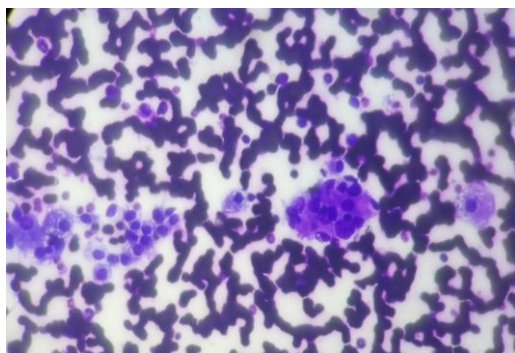
malignant and 9 out of 282 cases were in the category of suspicious of follicular neoplasm (2.06%) or suspicious of malignancy (1.03%) and 6 cases (2.06%) in atypical cell category. Benign lesions were more commonly encountered than malignant. Diagnosis of benign lesions were as follows- simple colloid goiter /colloid goiter with cystic change 150 cases (53%), adenomatous goiter 39 cases (13.40%), hyperplastic nodule 28 cases (9.62%), hashimotos thyroiditis 19 cases (6.52%), granulomatous thyroiditis 4 cases (1.37%).

**Table 2: Distribution of thyroid lesions based on Bethesda system (N=291)**

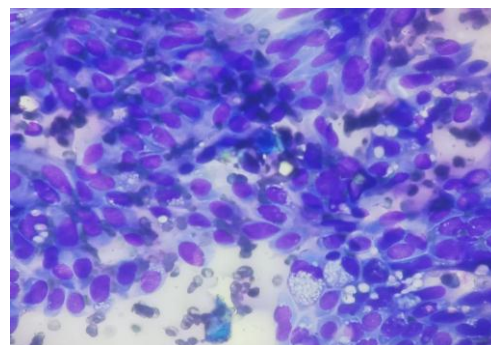
Category	Lesions	Cases
BI	Non Diagnostic/Unsatisfactory	9(3.09)%
BII	Benign- Colloid cyst/colloid goiter with cystic change Adenomatous goiter Hyperplastic nodule Hashimotos thyroiditis Granulomatous thyroiditis	240 (82.47%) 150(51.55%) 39(13.40%) 28(9.62%) 19(6.52%) 04(1.37%)
BIII	Atypia of Undetermined significance(AUS)	6(2.06)%
BIV	Follicular Neoplasm(FN)/ Suspicious of follicular Neoplasm(SFN)	6(2.06)%
BV	Suspicious of malignancy Papillary carcinoma(Follicular variant) Hurthel cell adenoma	3(1.03%) 2(0.69%) 1(0.34%)

BVI	Malignant	27(9.29%)
	Papillary	16(5.50 %)
	Medullary	6(0.02%)
	Anaplastic	5(0.02%)

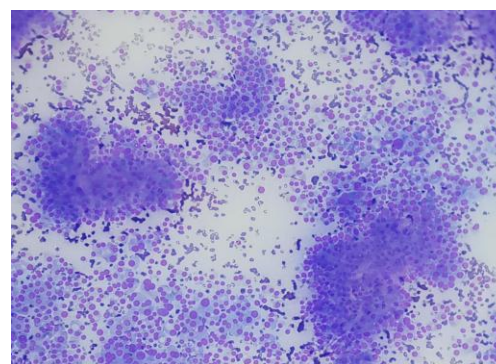
Among the benign lesions majority of them were simple colloid goiter/colloid goiter with cystic change. Fig. 1 the cytological examination of colloid goiter which revealed sheets of monolayered involutinal follicular cells, foamy macrophages in background of blood and thin colloid. Among the 27 malignancies, papillary carcinoma 16 cases (5.50%) was the commonest followed by medullary 6 cases (0.02%). Fig. 2 shows the cytological examination of papillary carcinoma which reveal high cellularity, minimal anisonucleosis of sheets of cells with focally nuclear crowding. Cells are columnar with oval pale nuclei and at places intranuclear inclusions. Out of 9 cases of suspicious category, majority of them were follicular neoplasm, as shown in figure 3 which revealed high cellularity with microfollicles, minimal anisonucleosis, nuclear overcrowding at places with absent colloid.



**Fig. 1: Colloid goiter with foamy macrophages and follicular epithelial cells(x40)**



**Fig. 2: Anisonucleosis, intranuclear inclusion in papillary carcinoma(x40)**



**Fig 3: Suspicious of follicular neoplasm on fine needle aspiration cytology(x40)**

Distribution of different lesions and its correlation with age and sex are shown in Table 3. Most of the patients were in the age group 21-30 year. Benign lesions in both males and females were more common in the age group of 21-30 year and malignant lesions in females were common in the age group of 31-50 years where as in males it is more common in the age group of 41-50 years.

**Table 3: Correlation of thyroid lesions with different age and sex (N=291)**

Lesions	11-20		21-30		31-40		41-50		51-60		>60	
	M	F	M	F	M	F	M	F	M	F	M	F
Non Diagnostic/Unsatisfactory	-	-	-	1	1	2	2	2			-	1
Colloid cyst/nodule	3	3	8	47	5	29	2	44	1	6	-	2
Benign follicular lesion	1	2	5	26	2	17	3	5	1	4	-	1
Hashimotos/lymphocytic/granulomatous thyroiditis	-	-	1	6	1	2	1	11	-	1	-	-
Atypia of Undetermined significance (AUS)	-	-	1	2	-	3	-	-	-	-	-	-
Follicular Neoplasm(FN)/Suspicious of	-	-	-	1	-	3	-	1	1	-	-	-

follicular Neoplasm(SFN)												
Suspicious of malignancy	-	-	-	-	-	-	1	1	1	-	-	-
Malignant	-	-	-	-	1	8	3	8	1	6	-	-

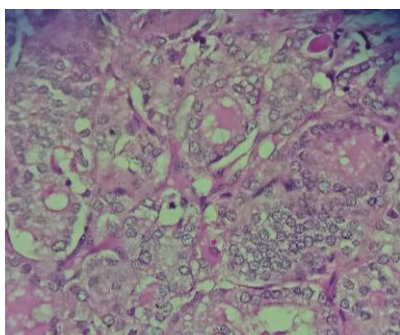
In the present study out of the total 291 cases of thyroid FNAC, cytohisto - correlation was done in 282 cases. Table 4 show the cyto-histopathological correlation of thyroid lesion and discordant cases. In this study we included all cystic or follicular pattern

lesion without atypia in benign category and those reported as suspicious in "malignant" category. All cystic lesion or follicular pattern lesions with atypia are classified in "suspicious category".

**Table 4: Cyto-histopathology correlation of cases of thyroid lesion and discordant cases**

Cytological diagnosis (N=282)	Histopathology diagnosis (N=282)	Discordant cases and its detail
Benign(N= 246 )	Benign(N= 243)	3(1-cystic PTC, 1-Papillary microcarcinoma, 1-follicular variant of papillary carcinoma)
Malignant(N= 27)	Malignant(N= 25)	2(nodular goitre with cystic change)
Suspicious(N= 9)	Malignant(N= 5)	Benign(N=4) (2-follicular adenoma, 1-adenomatous goiter, 1-Hyperplastic nodule with thyroiditis)

243/246 cases were confirmed as benign and 3/246 cases were discordant, which on histopathological examination proved to be malignant. Figure 4 shows histopathological examination of follicular variant of papillary carcinoma which is reported as benign on cytology and malignant on histopathology showing follicular architecture, individual cells show clear or ground-glass nuclei which is characteristic of papillary carcinoma. Out of 27 malignant cases in cytological examination, 25/27 cases were confirmed as malignant on histopathological examination and 2 cases were turned out to be benign. Rest 9 cases on cytology were of suspicious follicular neoplasm (6 cases) and suspicious of malignancy (3 cases), these on histopathological examination shows confirmed 5 cases as malignant, 4 cases were discordant and found to be benign.



**Fig. 4: Follicular variant of papillary carcinoma**

The diagnostic categories of thyroid lesions on cytology were compared with corresponding histopathological diagnosis. Among the benign lesions (246 cases), cyto-histo concordance were achieved in 243 cases and 3 were discordant. So false negative diagnosis in FNAC was given in 3 cases. Hence false

negative error rate was 9.4%. Out of 27 cases with FNAC diagnosis of malignancy, 25 proved to be the same on histopathological diagnosis and it differed in two cases. So the false positive rate was noted in 2.4 % cases of malignancy. Hence, the diagnostic accuracy of FNAC was found to be 94.3% in malignant cases. Out of nine cases suspicious of follicular neoplasm or suspicious of malignancy in cytology, five cases were confirmed same as malignancy, but four cases came out to be benign in histopathological examination on contrary to cytological diagnosis. Hence false positive error rate by FNAC was given in four cases as 2.5%.

Analysis of results yielded a sensitivity of 90.6%, specificity of 97.6%, positive predictive value of 80.6%, negative predictive value of 98.79%, false positive rate of 2.4%, false negative rate of 9.4% and diagnostic accuracy in differentiating benign and malignant thyroid lesion was 96.4%.

## Discussion

In the present study, a total of 282 cases of thyroid were included. The result of cytological findings and histopathological findings were compared and scrutinized for accuracy and other statistical values. Out of 282 cases 46(15.81%) were males and 245 cases (84.19%) were females, with a male to female ratio of 1:5.1. The findings are comparable to the studies of Hathila et al<sup>7</sup> who reported a ratio of 1:9 in a study of 60 cases, Al Rikabi et al<sup>8</sup> of 1:5.2 in 125 cases, Sirpal Y<sup>9</sup> of 1:1.4 in 1123 cases and Likhar KS et al<sup>10</sup> of 1:7 in 234 cases.

The highest number of cases were seen in the age group of 21-30 years(33.68%) followed by the age group 41-50 years (28.87%). This observation corresponds with the study conducted by Hathila R et al,<sup>7</sup> Savjani N et al.<sup>11</sup>

The main information to get from FNAC is to distinguish a malignant lesion from a benign one. This distinction has dramatically reduced the surgery rates in thyroid pathologies. However there are some limitations of FNAC which a pathologist must be aware of while reporting.<sup>2</sup>

There are various system for reporting thyroid cytology. The most widely used systems are the Bethesda system<sup>12</sup> and the Royal college of Pathologist system.<sup>13</sup> In the current study the cytological diagnosis of thyroid lesions follows the guidelines of Bethesda system and were categorized as unsatisfactory; benign; atypia of undetermined significance; suspicious of follicular neoplasm; suspicious of malignancy and malignant.

The present study reported 3.09% aspirates as nondiagnostic or unsatisfactory which correlates with studies of Yassa et al,<sup>14</sup> Nayar and Ivanovic<sup>15</sup> who reported 7% and 5% unsatisfactory smears in their studies, whereas Jo et al<sup>16</sup> reported much higher percentage(18.6%) of unsatisfactory smears. If smears are unsatisfactory or inadequate, then repeat FNAC was performed under ultrasound guidance. Therefore this rate was low in our study.

The present study shows that the benign lesions were (82.47%). These findings are consistent with study of Hathila et al (93.3%),<sup>7</sup> Kantasueb et al(87.5%).<sup>17</sup> The most common cytological diagnosis was colloid goiter (51.55%), similar findings were observed by Kantasueb.<sup>17</sup> Other diagnosis observed are adenomatous goiter(13.40%), hyperplastic nodule (9.62%), hashimotos thyroiditis (6.52%), granulomatous thyroiditis(1.37%). Among the lesions of category benign 243 out of 246 found to be concordant on the FNA and histopathology and 3 cases turned to be malignant reported as papillary carcinoma on final histopathology- false negative cases. These false negative cytology can occur due to sampling error in cases with coexistence of malignant and benign lesions, in which aspiration is obtained from benign lesions and adjacent small foci of malignant lesions can be missed. In our study false negative rate was 9.4% and all the three false negative cases were either papillary carcinoma or its variant. Most of the studies in the past also report a false negative rate in the range of 1-10%.<sup>3</sup> These errors can be minimized by repeating aspiration under ultrasound guidance or if there is a strong clinical suspicion then follow up of patients is to be done.

In lesions of category Atypia of undetermined significance/ Follicular lesions of undetermined significance(FLUS), out of 6 cases who underwent surgery, all cases were reported as colloid nodule with cellular area on histopathology. Literature shows 3-18% of thyroid lesions in category of FLUS and this diagnostic category should not be used indiscriminately and should be limited to less than 7% of all thyroid FNAs.<sup>14,15</sup> Our study shows 2.06% of thyroid lesions in

this category and is low as compared to the previous literature. This rate might be low in our study due to less use of this term and as the study was specifically aimed to find out the efficacy of FNAC and to keep our comparison clear, we considered all follicular patterned lesions without atypia in benign category. These follicular lesions form a grey zone in fine needle aspiration cytology. So attempt should be made to make specific diagnosis and if possible it should be correlate with clinical, pathological and radiological findings.

In this study, 6 cases were diagnosed as Follicular neoplasm /Suspicious of follicular neoplasm by FNA. This diagnosis were made if aspirate are cellular and composed of tightly cohesive follicular cells with significant cell crowding or microfollicle formation.<sup>4</sup> Of these 2 cases were diagnosed as malignant, mostly they are follicular variant of papillary carcinoma and rest of the 4 cases are follicular adenoma (2 cases), adenomatous goiter (1 case), and hyperplastic nodule (1 case). Literature also shows 27-68% of malignancies, in which diagnosis of follicular neoplasm was made on FNA and are reported as papillary carcinoma on histopathology.<sup>14</sup> This may be due to confusing cellular smears with follicular arrangement of cells, focal features of nuclear clearing and occasional nuclear grooves.

The present study shows that the suspicious lesions account for 3 cases (1.03%), which is in close relation with studies done by Moose et al (2.04%)<sup>18</sup> and Savjiani et al (2%)<sup>11</sup> and majority of the lesions on histopathology were papillary carcinoma. This diagnosis was made in cases which showed some features of malignancy but were not sufficient to make a conclusive diagnosis. In these cases some of the characteristic nuclear features of papillary carcinoma are present, on which diagnosis of malignancy cannot be made and are included in suspicious category. All the cases in our study of this category, on histopathology shows the nuclear features of papillary carcinoma which are focal.

In our study the malignant lesions account for 9.29%, which is in close relation with studies done by Moose et al(8.16%)<sup>18</sup> and Savjiani et al (12%)<sup>11</sup> and majority of the malignant lesions were papillary carcinoma (2.6%, 6.8%)<sup>11,18</sup> which were similar to our study which constitute (5.50%) of malignant lesions. Other malignant lesions were medullary carcinoma (0.02%) and anaplastic carcinoma (0.02%).

Analysis of FNAC results after the histopathological examination was done and the results are comparable with the results obtained by other studies. as shown in table 5. Results of various case series in previous years showed a wide range of the sensitivity of thyroid cytology from 66% to 100%. In the present study, sensitivity of thyroid cytology was 90.6%. On the other hand, specificity showed a narrow range in the previous case series varied from 90% to 100%. In the present study, specificity is 97.6% which

is very much similar to the study by Savjjiani et al<sup>11</sup> and Moose et al.<sup>18</sup> The accuracy of thyroid cytology in the

present study was 96.4% which is comparable to the other studies.

**Table 5: Correlation of statistical data**

Study	Sensitivity	Specificity	PPV	NPV	Accuracy
Ramteke DJ et al <sup>2</sup>	92.31%	97.01%	85.71%	98.48%	96.25%
Bagga et al <sup>3</sup>	66%	100%	100%	96%	96.2%
Hathila et al <sup>7</sup>	87.5%	96.15%	77.77%	98.03%	95%
Savjjiani N et al <sup>11</sup>	85.71%	97.67%	84.71%	96.67%	96%
Moose et al <sup>18</sup>	77.7%	97.67%	82.1%	94.6%	96.9%
Gulia S et al <sup>19</sup>	100%	90%	100%	97.67%	92.3%
Present study	90.6%	97.6%	80.6%	98.79%	96.4%

## Conclusions

Fine needle aspiration is a simple, reliable, cost effective and an effective diagnostic modality in the investigation of thyroid disease with high specificity and accuracy.

In recent trends, fine needle aspiration cytology has become an invaluable and minimal invasive procedure for screening or diagnosis of patients with thyroid nodule to guide the surgeons.

A benign fine needle aspiration cytology diagnosis should be viewed with caution, as false negative results do occur and so, the patient should be followed up with repeated aspirations to rule out missed neoplasm and to reduce the false negativity rate and false positivity rate.

The sensitivity, specificity and accuracy of FNA for malignant lesions is high, so the role of other diagnostic methods in detection of malignancy is low and role of FNA in determining the management of patient play an important part and it mostly replaces the need for histopathology which is a investigation of choice for confirmation of malignancy.

## Acknowledgements

The authors have no conflict of interest to declare and would like to acknowledge Professor and Head of Department of pathology and the Medical Director for their guidance and support provided.

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