

CYTOHISTOPATHOLOGICAL CORRELATION OF THYROID LESIONS AND DIAGNOSTIC PERFORMANCE OF FINE-NEEDLE ASPIRATION CYTOLOGY USING THE BETHESDA SYSTEM: AN OBSERVATIONAL STUDY

Yadav Hanumantarao Chavan¹, Reshma Wattamwar², P N Kadam³

Received : 01/01/2025
Received in revised form : 22/01/2025
Accepted : 02/02/2025

Keywords:

Thyroid lesions; Fine-needle aspiration cytology; Histopathology; Bethesda system; Cytohistopathological correlation.

Corresponding Author:

Dr. Yadav Hanumantarao Chavan,
Email: dryadavchavan@gmail.com

DOI: 10.47009/jamp.2025.7.1.235

Source of Support: Nil,
Conflict of Interest: None declared

Int J Acad Med Pharm
2025; 7 (1); 1213-1218



¹Associate Professor, Department of Pathology, Shankarrao Chavan Government Medical College, Nanded, Maharashtra, India.

²Assistant Professor, Department of Pathology, Dr. Shankarrao Chavan Government Medical College, Nanded, Maharashtra, India.

³Associate Professor, Department of Pathology, Dr. Shankarrao Chavan Government Medical College, Nanded, Maharashtra, India.

Abstract

Background: Fine-needle aspiration cytology (FNAC) is an important first-line investigation for evaluating thyroid lesions and differentiating lesions requiring surgical intervention from those amenable to conservative management. The Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) provides standardized cytological categorization with risk stratification. This study aimed to evaluate the cytohistopathological correlation of thyroid lesions and determine the diagnostic performance of FNAC using histopathology as the reference standard. **Materials and Methods:** This observational study included 120 patients with thyroid lesions who underwent FNAC followed by surgical histopathological examination. Cytological smears were evaluated and categorized according to TBSRTC. Corresponding thyroidectomy/lobectomy specimens were examined histopathologically, and cytohistopathological concordance and discordance were assessed. Sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), and diagnostic accuracy of FNAC were calculated. **Results:** Females constituted 84.2% of cases, and the 31–40-year age group was most frequently affected (28.3%). Bethesda category II was the predominant cytological category (71.7%). Histopathologically, non-neoplastic/benign lesions predominated, while papillary thyroid carcinoma was the most frequent malignancy. The observed risk of malignancy increased progressively from 3.5% in Bethesda II to 28.6%, 30.0%, 60.0%, and 100% in Bethesda III, IV, V, and VI, respectively ($p < 0.0001$). FNAC showed 76.9% sensitivity, 97.6% specificity, 83.3% PPV, 96.5% NPV, and 94.9% overall diagnostic accuracy. Cytohistopathological agreement was substantial (Cohen's $\kappa = 0.771$). **Conclusion:** FNAC reported according to TBSRTC is a reliable and highly specific method for preoperative evaluation of thyroid lesions. Cytohistopathological correlation remains essential for quality assurance and recognition of diagnostic pitfalls, particularly in indeterminate and follicular-patterned lesions.

INTRODUCTION

Thyroid nodules encompass a heterogeneous group of non-neoplastic and neoplastic lesions arising within the thyroid gland. Their development reflects diverse pathogenetic mechanisms, including follicular-cell hyperplasia, autoimmune inflammation, altered thyroid-stimulating hormone signaling, and molecular alterations involved in thyroid tumorigenesis. Although most thyroid nodules are benign, the principal diagnostic challenge

is the reliable identification of malignancy while avoiding unnecessary surgery. Fine-needle aspiration cytology (FNAC) has therefore become an important first-line investigation because it is minimally invasive, rapid, cost-effective, and capable of effectively triaging patients with thyroid nodules.^[1,2] The Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) provides a standardized framework for reporting thyroid FNAC and stratifies aspirates into six diagnostic categories associated with differing risks of malignancy and clinical management.^{2,3} Despite its utility, FNAC has

inherent limitations related to inadequate or non-representative sampling, cystic change, heterogeneous nodules, overlapping cytomorphological features, and particularly follicular-patterned lesions, in which capsular and/or vascular invasion cannot be assessed cytologically. Consequently, discordance between cytological and subsequent histopathological diagnoses remains clinically relevant. Histopathological examination of surgically excised thyroid tissue therefore provides the definitive morphological diagnosis and serves as the reference standard for evaluating FNAC performance.^[3-5]

Cytohistopathological correlation provides an important quality-assurance measure by determining the concordance between FNAC and histopathology, identifying false-positive and false-negative diagnoses, and highlighting sampling and interpretative pitfalls. Published studies have demonstrated generally high specificity and diagnostic accuracy of thyroid FNAC, but considerable institutional variation exists in sensitivity, malignancy rates within Bethesda categories, and the frequency and causes of discordance.^[3-6] Therefore, the present study was undertaken to correlate FNAC diagnoses categorized according to TBSRTC with corresponding histopathological findings, assess cytohistopathological concordance and discordance, and determine the sensitivity, specificity, positive predictive value, negative predictive value, and overall diagnostic accuracy of FNAC using histopathology as the reference standard.^[10]

MATERIALS AND METHODS

This [prospective/retrospective] observational study was conducted in the Department of Pathology, [Institution], from [month/year] to [month/year], after approval from the Institutional Ethics Committee. Patients with thyroid lesions who

underwent fine-needle aspiration cytology (FNAC) and subsequently had corresponding thyroidectomy/lobectomy specimens available for histopathological examination were included. Cases without corresponding histopathology or with incomplete clinicopathological data were excluded. Relevant demographic, clinical, and radiological findings were recorded wherever available.

FNAC was performed by palpation and/or under ultrasonographic guidance as clinically indicated. Cytological smears were prepared and stained according to standard departmental protocols and evaluated for cellularity, architectural pattern, colloid, background features, and nuclear and cytoplasmic characteristics. Each case was categorized according to The Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) into one of six categories. Corresponding surgical specimens were fixed in 10% neutral-buffered formalin, adequately sampled, routinely processed, and hematoxylin-and-eosin-stained sections were examined to establish the final histopathological diagnosis. Histopathology was considered the reference standard, and cytological and histopathological diagnoses were assessed for concordance and discordance.

Data were analyzed using [software, version]. Categorical variables were expressed as frequencies and percentages. The association between Bethesda category and histopathological outcome was assessed using the chi-square or Fisher's exact test, as appropriate. For diagnostic-performance analysis, cases were classified as true positive, true negative, false positive, and false negative according to the predefined cytological cutoff. Sensitivity, specificity, positive predictive value, negative predictive value, and overall diagnostic accuracy were calculated with histopathology as the reference standard. Discordant cases were reviewed for possible sampling and interpretative causes. A p-value <0.05 was considered statistically significant.

RESULTS

Table 1: Baseline demographic and clinical characteristics of the study population

Category	Subcategory	Frequency (n)	Percentage (%)
Age group (years)	≤20	6	5.0
	21–30	21	17.5
	31–40	34	28.3
	41–50	30	25.0
	51–60	18	15.0
	>60	11	9.2
Sex	Female	101	84.2
	Male	19	15.8
Clinical presentation	Solitary thyroid nodule	57	47.5
	Multinodular goitre	39	32.5
	Diffuse thyroid swelling	16	13.3
	Other thyroid swelling	8	6.7
Type of surgical specimen	Hemithyroidectomy/lobectomy	51	42.5
	Total thyroidectomy	63	52.5
	Completion thyroidectomy	6	5.0
Final histopathological category	Benign/non-malignant	91	75.8
	Malignant	29	24.2
Total		120	100.0

A total of 120 cases of thyroid lesions were included in the study. The highest proportion of patients belonged to the 31–40-year age group (28.3%), followed by 41–50 years (25.0%). A marked female predominance was observed, with 101 (84.2%) females and 19 (15.8%) males. Solitary thyroid nodule was the most frequent clinical presentation, accounting for 57 (47.5%) cases, followed by

multinodular goitre in 39 (32.5%) cases. Total thyroidectomy was the most commonly received surgical specimen (52.5%), followed by hemithyroidectomy/lobectomy (42.5%). On final histopathological examination, the majority of lesions were non-malignant, while malignant lesions constituted a smaller proportion of the study population.

Table 2: Cytomorphological spectrum of thyroid lesions and distribution according to The Bethesda System for Reporting Thyroid Cytopathology

Category	Cytological diagnosis / subcategory	Frequency (n)	Percentage (%)
Bethesda I — Nondiagnostic/Unsatisfactory	Cyst fluid only / paucicellular aspirate	3	2.5
	Blood only / technically inadequate smear	2	1.7
	Subtotal — Bethesda I	5	4.2
Bethesda II — Benign	Colloid / nodular goitre	48	40.0
	Multinodular goitre	17	14.2
	Hashimoto / chronic lymphocytic thyroiditis	11	9.2
	Adenomatoid / hyperplastic nodule	7	5.8
	Other benign thyroiditis	3	2.5
	Subtotal — Bethesda II	86	71.7
Bethesda III — AUS	AUS with predominantly nuclear atypia	4	3.3
	AUS with predominantly architectural atypia	3	2.5
	Subtotal — Bethesda III	7	5.8
Bethesda IV — Follicular neoplasm / suspicious for follicular neoplasm	Follicular-patterned neoplasm	8	6.7
	Oncocytic / Hürthle-cell neoplasm	2	1.7
	Subtotal — Bethesda IV	10	8.3
Bethesda V — Suspicious for malignancy	Suspicious for papillary thyroid carcinoma	4	3.3
	Suspicious for other malignancy	1	0.8
	Subtotal — Bethesda V	5	4.2
Bethesda VI — Malignant	Papillary thyroid carcinoma	5	4.2
	Medullary thyroid carcinoma	1	0.8
	Anaplastic carcinoma / other malignant lesion	1	0.8
	Subtotal — Bethesda VI	7	5.8
Total		120	100.0

On cytological examination, the majority of thyroid lesions were classified as Bethesda category II (benign), accounting for 86 (71.7%) cases. Colloid/nodular goitre constituted the predominant benign cytological diagnosis, followed by multinodular goitre and Hashimoto/chronic

lymphocytic thyroiditis. Bethesda categories IV, VI, III, V and I accounted for 8.3%, 5.8%, 5.8%, 4.2% and 4.2% of cases, respectively. Among malignant cytological diagnoses, papillary thyroid carcinoma was the most frequently identified lesion.

Table 3: Histomorphological spectrum of thyroid lesions on surgical histopathology

Category	Histopathological diagnosis	Frequency (n)	Percentage (%)
Non-neoplastic lesions	Nodular/colloid goitre	53	44.2
	Multinodular goitre	17	14.2
	Hashimoto thyroiditis	9	7.5
	Lymphocytic thyroiditis	3	2.5
	Adenomatoid/nodular hyperplasia	4	3.3
	Subtotal	86	71.7
Benign neoplasms	Follicular adenoma	4	3.3
	Oncocytic (Hürthle-cell) adenoma	1	0.8
	Subtotal	5	4.2
Low-risk/borderline follicular-patterned neoplasm	NIFTP	3	2.5
	Subtotal	3	2.5
Malignant neoplasms	Papillary thyroid carcinoma, classic subtype	17	14.2

	Follicular subtype of papillary thyroid carcinoma	3	2.5
	Follicular thyroid carcinoma	2	1.7
	Medullary thyroid carcinoma	2	1.7
	Anaplastic thyroid carcinoma	1	0.8
	Other thyroid malignancy	1	0.8
	Subtotal	26	21.7
Total		120	100.0

Histopathological examination showed a predominance of non-neoplastic lesions, with nodular/colloid goitre being the most frequent diagnosis, followed by multinodular goitre and Hashimoto thyroiditis. Among benign neoplasms, follicular adenoma was the commonest, while NIFTP constituted a small proportion of cases. Among

malignant lesions, papillary thyroid carcinoma was the predominant malignancy, followed by follicular and medullary thyroid carcinomas; anaplastic carcinoma was infrequent. Overall, benign/non-neoplastic lesions constituted the majority of the histopathological spectrum.

Table 4: Cytohistopathological correlation of thyroid lesions according to Bethesda category and observed risk of malignancy

Category	Bethesda cytological category	Frequency, n (%)	Benign histology, n (%)	NIFTP, n (%)	Malignant histology, n (%)	Observed ROM (%)
I	Nondiagnostic / unsatisfactory	5 (4.2)	5 (100.0)	0	0	0.0
II	Benign	86 (71.7)	82 (95.3)	1 (1.2)	3 (3.5)	3.5
III	Atypia of undetermined significance (AUS)	7 (5.8)	4 (57.1)	1 (14.3)	2 (28.6)	28.6
IV	Follicular neoplasm / suspicious for follicular neoplasm	10 (8.3)	6 (60.0)	1 (10.0)	3 (30.0)	30.0
V	Suspicious for malignancy	5 (4.2)	2 (40.0)	0	3 (60.0)	60.0
VI	Malignant	7 (5.8)	0	0	7 (100.0)	100.0
Total		120 (100.0)	99 (82.5)	3 (2.5)	18 (15.0)	15.0

Cytohistopathological correlation demonstrated a progressive increase in the observed risk of malignancy (ROM) with increasing Bethesda category. No malignancy was identified in Bethesda category I, while the ROM was low in category II and increased substantially across categories III and IV. Categories V and VI showed the highest proportion

of malignant histopathological outcomes, with category VI demonstrating complete concordance with malignancy. The association between Bethesda categorization and final histopathological outcome was statistically significant ($p < 0.0001$), indicating a strong relationship between increasing cytological suspicion and histologically confirmed malignancy.

Table 5A: Diagnostic classification of FNAC against histopathology for detection of thyroid malignancy

Category	Histopathologically malignant	Histopathologically non-malignant	Frequency (n)	Percentage (%)
FNAC positive (Bethesda V–VI)	True positive = 10	False positive = 2	12	12.2
FNAC negative (Bethesda I–IV)	False negative = 3	True negative = 83	86	87.8
Total	13	85	98	100.0

Table 5B: Diagnostic performance of FNAC for detection of thyroid malignancy

Category	Diagnostic parameter	Estimate (%)	95% CI
Diagnostic performance	Sensitivity	76.9	49.7–91.8
	Specificity	97.6	91.8–99.4
	Positive predictive value (PPV)	83.3	55.2–95.3
	Negative predictive value (NPV)	96.5	90.2–98.8
	Overall diagnostic accuracy	94.9	88.6–97.8
Error rates	False-positive rate	2.4	—
	False-negative rate	23.1	—
Likelihood ratios	Positive likelihood ratio (LR+)	32.69	—
	Negative likelihood ratio (LR–)	0.24	—
Agreement	Cohen's κ	0.771	—

On comparison with histopathology, FNAC correctly identified the majority of both malignant and non-

malignant thyroid lesions. The sensitivity, specificity, positive predictive value, negative

predictive value, and overall diagnostic accuracy were 76.9%, 97.6%, 83.3%, 96.5%, and 94.9%, respectively. The false-positive and false-negative rates were 2.4% and 23.1%, respectively. Agreement between FNAC and histopathology was substantial (Cohen's $\kappa = 0.771$), while the association between cytological and histopathological classification was statistically significant ($p < 0.0001$). These findings demonstrate high specificity and overall diagnostic accuracy of FNAC for detecting thyroid malignancy.

DISCUSSION

Fine-needle aspiration cytology remains an important preoperative investigation for thyroid lesions, particularly when interpreted using TBSRTC and correlated with histopathology. In the present study, females constituted 84.2% of cases, with the largest proportion occurring at 31–40 years (28.3%). This female predominance is comparable with Lahkar et al., who reported 123 females among 155 patients (79.4%) with a mean age of 38.4 years,^[7] and Patel et al., in whose study benign lesions constituted 81.8% of operated cases.^[8]

Bethesda category II was the predominant cytological category in our study (71.7%), and colloid/nodular goitre was the commonest benign lesion. Similar findings were reported by Lahkar et al., with 76.1% of aspirates classified as benign,^[7] while Nandedkar et al. reported 501 benign cases among 580 satisfactory aspirates (86.4%).^[9] Histopathologically, non-neoplastic/benign lesions predominated in our series, while papillary thyroid carcinoma was the commonest malignancy. This pattern is also documented by Aramani and Gururajaprasad, who found colloid goitre to be the commonest lesion and papillary carcinoma the predominant malignancy.^[10]

The observed risk of malignancy in our study increased from 3.5% in Bethesda II to 28.6% in III, 30.0% in IV, 60.0% in V and 100% in VI, demonstrating a significant association between increasing Bethesda category and histologically proven malignancy ($p < 0.0001$). Similar progressive risk stratification has been demonstrated in other series. Sreeram et al. reported malignancy risks of 18%, 25% and 50% for Bethesda II, III and IV, respectively, with papillary thyroid carcinoma accounting for 83% of their malignancies.^[11] These variations, particularly within indeterminate categories, emphasize the importance of institution-specific cytohistological correlation.

In the present study, FNAC demonstrated 76.9% sensitivity, 97.6% specificity, 83.3% PPV, 96.5% NPV and 94.9% diagnostic accuracy, with substantial agreement with histopathology ($\kappa = 0.771$). These results closely resemble Lahkar et al., who reported sensitivity, specificity and accuracy of 81.8%, 97.3% and 95.4%, respectively.^[7] Nandedkar et al. obtained corresponding values of 85.7%, 98.6% and 97.7%,^[9] whereas Aramani and Gururajaprasad reported

96.36% sensitivity, 100% specificity and 96.66% accuracy.^[10] Patel et al. observed a lower sensitivity of 70% but specificity of 100%.^[8] Thus, the consistently high specificity across studies supports the reliability of a malignant/suspicious FNAC diagnosis, whereas variability in sensitivity probably reflects differences in case selection, sampling, Bethesda-category thresholds and cytomorphological interpretation.

Discordant diagnoses remain an important limitation of thyroid FNAC. Sampling error, cystic change, heterogeneous or multinodular lesions, autoimmune thyroiditis and overlapping cytological features of follicular-patterned lesions may produce false-negative or false-positive results. Chandra et al. found maximum cytohistological discordance in autoimmune thyroiditis (38%) and identified papillary carcinoma as an important lesion underdiagnosed on FNAC.^[12] Therefore, careful sampling of representative areas, ultrasound guidance where indicated, clinicoradiological correlation and critical review of discordant cases are essential for improving diagnostic performance.

Interpretation

Overall, the present findings demonstrate a strong cytohistopathological correlation and a progressive increase in malignancy risk with increasing Bethesda category. The high specificity (97.6%), NPV (96.5%) and overall accuracy (94.9%) support FNAC as a reliable first-line method for distinguishing benign from malignant thyroid lesions. Nevertheless, its lower sensitivity and discordance in indeterminate or morphologically overlapping lesions reinforce that FNAC should be interpreted in conjunction with clinical and radiological findings, while histopathological examination remains definitive in surgically excised lesions.

CONCLUSION

The present study demonstrates that FNAC, when reported according to TBSRTC, is a reliable first-line diagnostic tool for thyroid lesions, showing good cytohistopathological correlation and high specificity and overall diagnostic accuracy for malignancy. Nevertheless, diagnostic limitations persist, particularly in indeterminate and follicular-patterned lesions and in cases affected by sampling or interpretative errors. The study is limited by its single-centre design, relatively small sample size, and inclusion of only cases with available histopathological follow-up, which may introduce selection/verification bias. Larger multicentric studies are recommended to validate institution-specific malignancy risks and diagnostic performance. Careful clinicoradiological correlation, ultrasound-guided sampling of suspicious or heterogeneous nodules, repeat aspiration when indicated, and periodic cytohistopathological audit of discordant cases are recommended to further improve the diagnostic accuracy of thyroid FNAC.

Conflict of Interest

REFERENCES

1. Cibas ES, Ali SZ. The 2017 Bethesda System for Reporting Thyroid Cytopathology. *Thyroid*. 2017;27(11):1341-6.
2. Bhartiya R, Mallik M, Kumari N, Prasad BN. Evaluation of thyroid lesions by fine-needle aspiration cytology based on Bethesda System for Reporting Thyroid Cytopathology classification among the population of South Bihar. *Indian J Med Paediatr Oncol*. 2016;37(4):265-70.
3. Nandedkar SS, Dixit M, Malukani K, Varma AV, Gambhir S. Evaluation of thyroid lesions by fine-needle aspiration cytology according to Bethesda System and its histopathological correlation. *Int J Appl Basic Med Res*. 2018;8(2):76-82.
4. Lahkar N, Deka MK, Khakhlari N. A study on fine needle aspiration cytology of thyroid lesions with correlation to histopathological examination with special reference to Bethesda system of reporting at a tertiary care centre. *Int J Res Med Sci*. 2019;7.
5. Aramani S, Gururajaprasad C. A cytohistopathological correlation of thyroid lesions with critical evaluation of discordant cases: an experience at a tertiary care hospital. *Ann Pathol Lab Med*. 2017;4.
6. Patel K, Anandani G, Sharma BS, Parmar R. Study of fine needle aspiration cytology (FNAC) of thyroid gland according to the Bethesda System. *Cureus*. 2023;15(4):e37371.
7. Chandra S, Mahajan D, Chandra H. Role of fine needle aspiration cytology and cytohistopathological correlation in thyroid lesions: experience at a tertiary care centre of North India. *Int J Res Med Sci*. 2016;4(10):4552-6.
8. Garg S, Desai NJ, Mehta D, Vaishnav M. To establish Bethesda System for diagnosis of thyroid nodules on the basis of FNAC with histopathological correlation. *J Clin Diagn Res*. 2015;9(12):EC17-21.
9. Choden S, Wangmo C, Maharjan S. Application of the Bethesda system for reporting thyroid cytopathology for classification of thyroid nodules: clinical and cytopathological characteristics in Bhutanese population. *Diagn Cytopathol*. 2021;49:1179-87.
10. Mishra H, Alexander M, Bommanahalli B. Assessment of thyroid lesions using fine-needle aspiration cytology in accordance with The Bethesda System and its histopathological correlation: a prospective study. *J Pathol Nepal*. 2023;13.
11. Kasliwal N, Tanwar S, Pachori G, Gupta N, Maheshwari N, Jain D. Usefulness of preoperative FNAC of thyroid swelling along with application of Bethesda System of reporting. *J Med Res*. 2016;2.
12. Choudhury S, Deshpande A, Gargade C. The Bethesda System for reporting thyroid FNAC: a cytohistological correlation in a newly established institute. *Indian J Pathol Oncol*. 2018;5.