Recent Advances in Pathology & Laboratory Medicine
Volume 10, Issue 3&4 - 2024, Pg. No. 9-15
Peer Reviewed & Open Access Journal

Research Article

Diagnostic Cytopathology Approach along with Malignant Risk Stratification in Thyroid Lesions

Saswati Das[1], Sonia Gon[2], Preeti R Chakarabarti[3], Debjani Mallick[4]

[1]Post-Graduate Trainee, [2]Head of the Department, [3,4]Professor, Department of Pathology ESI-PGIMSR & ESIC Medical College, Joka, Kolkata, India.
DOI: https://doi.org/10.24321/2454.8642.202406

I N F O

Corresponding Author:
Sonia Gon, Department of Pathology ESI-PGIMSR & ESIC Medical College, Joka, Kolkata, India.
E-mail Id:
dr.sonia.gon@esic.nic.in
Orcid Id:
https://orcid.org/0009-0006-1764-4699
How to cite this article:
Das S, Gon S, Chakarabarti P R, Mallick D. Diagnostic Cytopathology Approach along with Malignant Risk Stratification in Thyroid Lesions Rec Adv Path Lab Med. 2024;10(3&4):9-15.

Date of Submission: 2024-04-12
Date of Acceptance: 2024-05-15

A B S T R A C T

Objectives: Fine needle aspiration cytology (FNAC) plays a pivotal role in the initial diagnosis of thyroid swellings. The standardisation of cytomorphological classification by The Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) has simplified the treatment and malignancy risk analysis can help select the follow-up strategies. The aim of this study was to enumerate the cytomorphological features of thyroid swellings, classification in TBSRTC and corroboration of their radiological features along with illustration of diagnostic approaches in non-diagnostic cases and malignancy risk stratification in thyroid nodules.

Methods: FNAC were done in 153 cases of thyroid swellings and cytomorphological diagnoses were made as per TBSRTC. Repeat FNAC was done in non-diagnostic cases.

Results: Among 153 cases, benign lesions constituted 108 (70.6%) cases, malignancy constituted 9 (5.9%) cases, and 22 (14.4%) cases were non-diagnostic. Among benign cases, the majority (62, 57.4%) were cases of colloid and multinodular goitre. Five out of nine malignant cases (55.5%) showed a hypoechoic nodule in the ultrasound scan. Repeat FNAC succeeded in making a diagnosis in three cases (13.6%) of the non-diagnostic group. With guided FNAC, 10 cases (45.4%) were diagnosed and 2 cases (11.1%) were found to be malignant. Among the known risk factors, the number of nodules showed a significant causal association with malignancy (Odds ratio: 8.9; p value 0.04) and 121 cases (79%) were euthyroid irrespective of malignancy status.

Conclusion: FNAC is a sensitive, accessible, and inexpensive diagnostic tool and repeat FNAC in non-diagnostic cases is useful in diagnosing and analysing malignant risk in thyroid lesions.

Keywords: FNAC Thyroid, The Bethesda System, Malignancy Risk Stratification

Introduction

FNAC is widely utilised in the evaluation of space occupying lesions in various organs. It is valued for its simplicity, rapid results, and reduced risk compared to more invasive procedures, making it a valuable tool in the early detection and characterisation of palpable lesions.[1] It plays a pivotal role in the initial diagnosis of thyroid swellings and its accuracy is enhanced when combined with ultrasound imaging.[2] The Bethesda System for Reporting Thyroid Cytology (TBSRTC) is a standardised classification system developed to provide a consistent and uniform approach to reporting thyroid fine needle aspiration (FNA) cytology results.[3] Introduced in 2007 and last revised in 2023, TBSRTC categorises thyroid FNA results into six main diagnostic categories, ranging from benign to malignant, with each category associated with specific management recommendations.[4] This system helps streamline communication between pathologists, clinicians, and patients, facilitating a clearer understanding of the potential risks and appropriate follow-up for each diagnostic category.[5] TBSRTC has significantly improved the accuracy and consistency of thyroid FNA reporting, aiding in making decisions regarding patient management and treatment strategies based on the cytological findings.[6]

Though FNAC is one of the initial screening investigations of thyroid swelling, it has its own limitations, especially in non-diagnostic (Category I) cases where the material of aspiration is inadequate for evaluation.[4] It could be due to various reasons, commonly scanty material, haemorrhage obscuring cellular morphology or only aspiration of colloid or cystic fluid may yield unsatisfactory results.

This study aimed to enumerate the cytomorphological spectrum of various thyroid lesions and their subsequent classification in TBSRTC and to illustrate the diagnostic approaches in non-diagnostic cases. Analysing the known risk factors in palpable thyroid nodules was another objective of this study.

Materials & Methods

An observational, prospective institutional-based study was conducted for one year at a tertiary care hospital in the eastern region of India. The study population comprised patients referred to the FNAC clinic. The participants were selected through convenient sampling. Consenting patients of all age groups and of both genders who presented with thyroid swellings were included in our study. Exclusion criteria were non-consenting patients, patients with non-palpable thyroid lesions or with haemophilia or coagulation disorder. A total of 153 cases were studied. A thorough clinical history was taken along with the conduction of a local examination before performing FNAC. Ultrasound examination reports and reports of thyroid function tests were obtained. Informed written consents were taken. Using disposable hypodermic 22–27 gauge needles, materials were aspirated maintaining full aseptic precautions and smears were made. A few of the slides were air-dried for Leishman-Geimsa staining and the rest of the slides were fixed in 95% ethanol for staining with Hematoxylin-Eosin. In suspected cases, acid fast bacilli staining was also performed. Slides were examined and cytomorphological reports were given as per the TBSRTC.

Specimens showing at least 6 follicles comprising 10 follicular cells were accepted as adequate with consideration of colloid in the specimen.[3] In non-diagnostic cases, repeated FNACs and in a few cases, guided FNACs were done. Gross specimens for histopathological examination were received in a few for confirmation.

Data thus obtained were tabulated in Microsoft Excel Software and statistically analysed using SPSS software version 29. The thyroid lesions thus categorised in the Bethesda System were compared with clinic-pathological factors like age and gender and were also correlated with their respective ultrasound findings and results of thyroid function tests. In the non-diagnostic category, cases were followed up with either guided or non-guided FNACs and reports thus made were also statistically analysed to illustrate a practical diagnostic approach. Risk factor analysis was done in cases with palpable thyroid nodules with known risk factors and the Odds ratios with their respective p values were calculated and compiled.

Results

A total of 153 cases were included in this study. Using disposable hypodermic needles ranging from 22 to 27 gauge, samples were collected under strict aseptic conditions, and smears were prepared. Some slides were air-dried for Leishman-Giemsa staining, while others were fixed in 95% ethanol for Hematoxylin and Eosin staining. In cases where tuberculosis was suspected, acid-fast bacilli staining was also carried out. All slides were then examined, and cytomorphological findings were reported according to the Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) (Figure 1 and 2). Predominantly females presented with thyroid lesions (110, 72%) as compared to male patients (43, 28%). Maximum cases were seen in the age group of 31–40 years (49, 32%) followed by 41–50 years (43, 28%). Classification in The Bethesda System (Table 1) showed that a majority of cases were benign (108, 70.6%), malignancy was detected in 5.9% (9 cases) of patients, and 14.4% (22 cases) of cases were non-diagnostic on first attempt FNAC.[3]

ISSN: 2454-8642
DOI: https://doi.org/10.24321/2454.8642.202406

Regarding cytomorphological features associated with different Bethesda categories (Table 2), in benign (Category II) cases, maximum cases were of colloid goitre (36, 23.6%), followed by adenomatoid nodule (24, 15.6), multi-nodular goitre (17, 11.1%), lymphocytic thyroiditis (16, 10.4%) and granulomatous thyroiditis (15, 9.8%). In Category IV cases, follicular neoplasm was seen in 3.9% (6) of cases and hurthle cell neoplasm was seen in 3.2% (5) of cases. Among the malignant cases (Category IV), papillary thyroid carcinoma was predominant (8, 5.2%) with one case (0.6%) of medullary carcinoma of the thyroid.

On ultrasound findings, the majority of the thyroid lesions were hypoechoic (96, 62.7%) of which 74% (71 cases), on cytomorphology, showed benign features, and features of malignancy were seen in 5.2% (5) of cases. Thyroid lesions that were mixed in echogenesity, i.e. showing both solid and cystic components were 5.2% (8) of the total study population and 50% (4) of them turned out to be malignant in FNAC.

A total of 22 cases (14.4%) were categorised as non-diagnostic (Category I). Repeat FNAC was attempted with or without guidance and diagnosis was made by non-guided repeat FNAC in 13.6% (3) of cases, by USG-guided FNAC in 45.4% (10), and by histopathology in 22.7% (5) of cases, and 18.1% (4) of cases lost follow up (Figure 3). Among the total non-diagnostic cases, 88.8% (16) were benign but 11.1% (2) of cases turned out to be malignant.

Thyroid function tests were mostly normal (121, 79%), 18% (28) were hypothyroid and 3% (4) showed hyperthyroidism.

On risk factors analysis, the known risk factors i.e. the size of nodules (34% of > 4 cm vs 66% of < 4 cm), gender (28% male vs 72% female), age (35.6% of > 50 years vs 64.4% of < 50 years), consistency on palpation (64% firm to hard vs 36% soft cystic), echogenesity on ultrasound imaging (68% hypo and mixed echogenic vs 32% hypo and isoechogenic), ultrasound size of nodules (28.6% of > 4 cm vs 71.4% of < 4 cm), number of nodules (49.6% solitary nodules vs 50.3% multinodular) and thyroid function status (18.3% hypothyroidism vs 81.6% hyper and euthyroidism) were considered and univariate regression analysis was done.[7] Odds ratios were calculated for each risk factor and their respective p values were analysed to find out their causal association (Table 3).

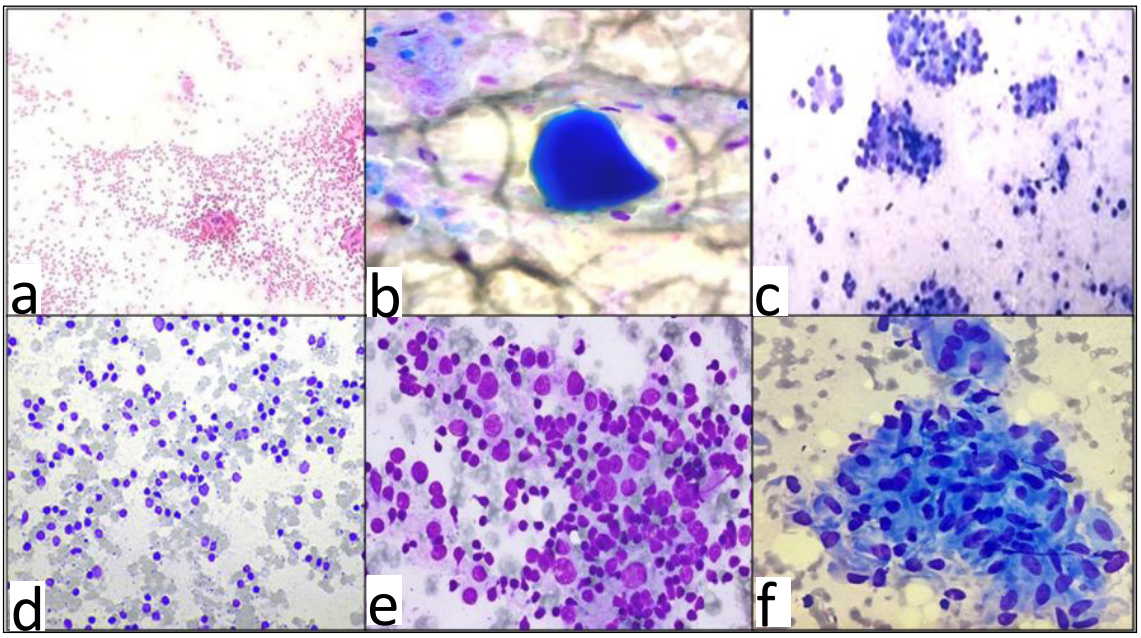

Figure 1.Non-Neoplastic Lesions of Thyroid. (a).H& E, 100X. Non-Diagnostic (Category I) Haemorrhagic Smear with Very Few Follicular Cells (b).Leishman-Giemsa, 400X. Colloid Goiter (Category II) Showing Thick Colloid (c). Leishman-Giemsa, 400X. Adenomatoid Nodule (Category II); Cellular Smear with Follicular Cells with No Atypia (d).Leishman-Giemsa,100X. Lymphocytic thyroiditis (Category II) (e).Leishman-Giemsa, 400X. Lymphocytic Impinging Inside Follicular Cell Clusters. (f).Leishman-Giemsa, 400X. Granulomatous Thyroiditis (Category II) - A Well-Formed Granuloma with Epithelioid Cells.

ISSN: 2454-8642
DOI: https://doi.org/10.24321/2454.8642.202406

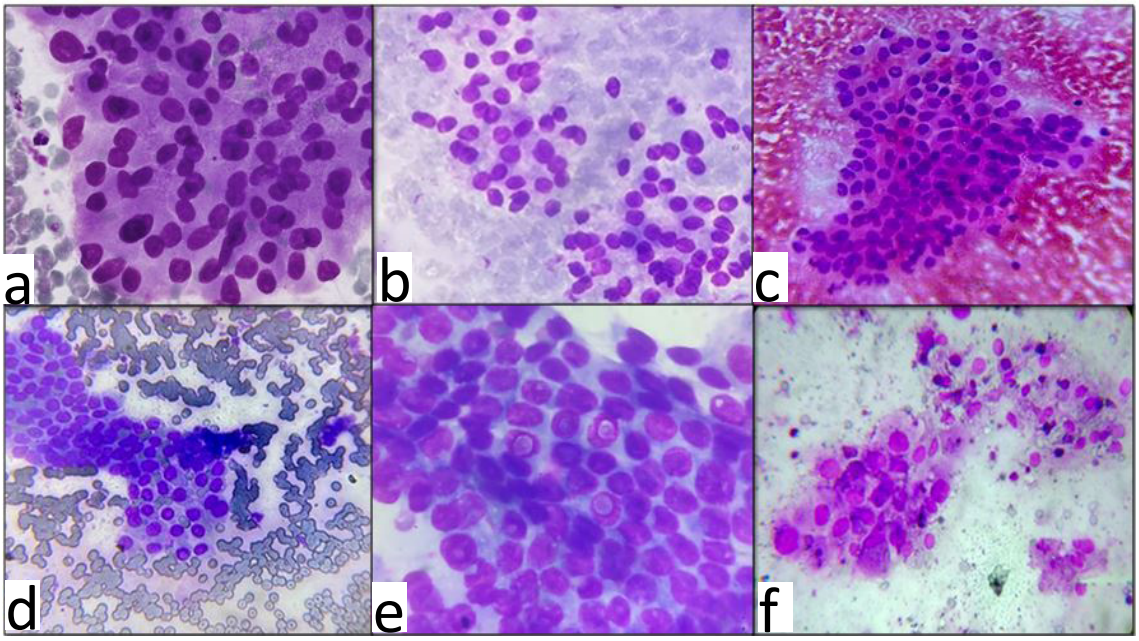

Figure 2.Neoplastic Lesions of Thyroid
(a) Leishman-Geimsa, 400X. Follicular Neoplasm (Category IV)(b).Leishman-Geimsa, 400X. Follicular Neoplasm (Category IV) (c). H & E, 400X. Papillary Thyroid Carcinoma (Category VI)-Formation of Papillae (d). Leishman-Geimsa, 400X. Papillary Thyroid Carcinoma (Category VI) (e).Leishman-Geimsa, 400X. Papillary Thyroid Carcinoma (Category VI) - Nuclear Pseudoinclusion (f).Leishman-Geimsa, 400X. Medullary Carcinoma of Thyroid.

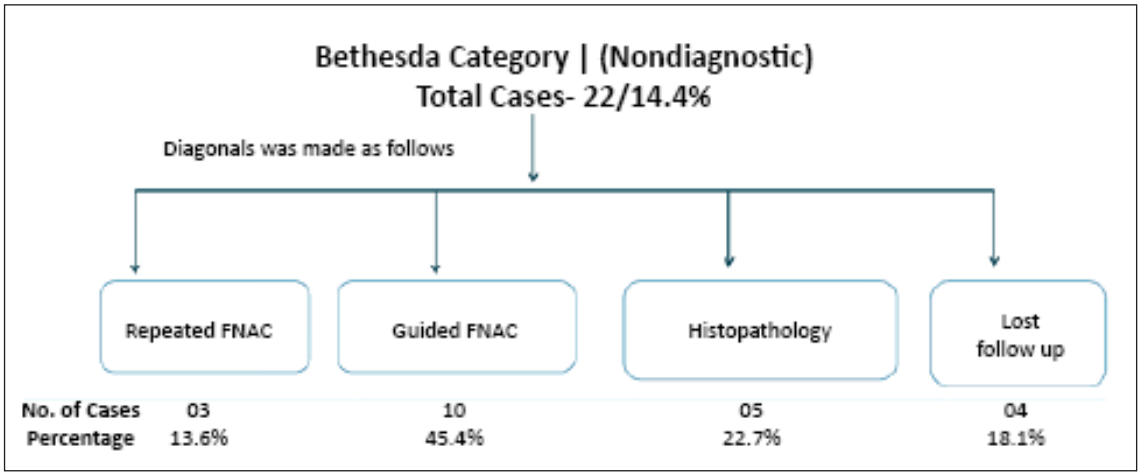

Figure 3.Distribution According to Bethesda Classification

Table 1.Distribution According to Bethesda Classification

Bethesda Category	I	II	III	IV	V	VI
Number of Cases	22	108	01	11	02	09
Percentage	14.4%	70.6%	0.6%	7.2%	1.3%	5.9%

ISSN: 2454-8642
DOI: https://doi.org/10.24321/2454.8642.202406

Table 2.Cytomorphological Spectrum

Bethesda Category	Cytomorphological Diagnosis	Number of Cases	Percentage
Non-neoplastic Lesions			
II	Colloid Goitre	36	23.6%
II	Multinodular Goitre	17	11.1%
II	Adenomatoid Nodule	24	15.6%
II	Lymphocytic Thyroiditis	16	10.4%
II	Granulomatous Thyroiditis	15	9.8%
III	Atypia of undetermined significance	01	0.6%
Neoplastic Lesions			
IV	Follicular Neoplasm	06	3.9%
IV	Hurthle cell Neoplasm	05	3.2%
V	Suspicious for Malignancy	02	1.3%
VI	Papillary Carcinoma	08	5.2%
VI	Medullary Carinoma	01	0.6%
Total		131	

Table 3.Risk Factors Stratification

Known Risk Factor[R]	Odds Ratio	95% of CI	P value
Size of Nodules (>4cm/<4cm)	2.8	0.325 to 11.267	0.54
Gender (Male/Female)	3.1	0.456 to 14.670	0.23
Age (>50 yrs/<50 yrs)	2.5	1.674 to 15.874	0.07
Consistency (Solid/Cystic)	1.5	0.217 to 7.098	0.09
USG Consistency (Hypoechoic/Hype rechoic)	1.2	0.261 to 8.542	0.10
TSH (Euthyroid/Hypothyroid)	0.9	0.4099 to 1.2903	0.27
Number of Nodules (Multiple/Single)	8.9	1.09-73.34	0.04
USG size of Nodules (>4cm/<4cm)	1.6	0.431 to 6.319	0.71

ISSN: 2454-8642
DOI: https://doi.org/10.24321/2454.8642.202406

Discussion

The present study showed that a majority of the cases were benign (Category II) as classified in The Bethesda System as comparable to a study done by Agarwal et al. and Akshatha et al.[8,9] Monappa and Kudva showed papillary thyroid carcinoma to be the commonest malignancy found in FNACs as found in the present study (88.8%).[10] Among the benign lesions, colloid goitre and adenomatoid nodule constituted the maximum number of cases (66.7%), comparable to a study done by Esmaili and Taghipour.[11] Nandedkar et al. showed non-diagnostic cases to be 4.29% after reaspiration, whereas the present study had 14.4% (22/153) of non-diagnostic cases after the first aspiration, out of which 59% (13/22) were diagnosed in follow-up repeat aspiration and 22.7% (5/22) required histopathological examination out of clinical suspicion for confirmation.[12] The reason behind those non-diagnostic cases was mainly the inadequate sample obtained on the first FNAC. Cystic thyroid SOL or nodule with mixed echogenesity presented maximally with inadequate sampling as the needle did not hit the solid tissue area accurately. Thus ultrasound guidance reduces this non-diagnostic rate significantly (45.4%) by improving the reach in the cystic or mixed thyroid nodules. A study done by Moon et al. also showed that among the non-diagnostic FNACs , ultrasound-guided repeat FNACs were useful in detecting 7.3% of cases to be malignant and 92.7% of cases to be benign.[13] Another cause of inadequate sampling was scanty follicular cells with obscuring haemorrhage or colloid. In such cases, repeat FNAC done with precision by an experienced hand yielded good results reducing the non-diagnostic rate by 13.6%. Similarly, Al Maqbali et al. and Yelave et al. both found that the rate of non-diagnostic cases was reduced significantly by repeat FNAC and ultrasound-guided FNAC.[14,15] In the study of Al Maqbali et al., among the 264 patients who received non-diagnostic (Category I) results in their initial FNAC, only 7% underwent surgery without a follow-up FNAC.[14] The majority of these surgeries were prompted by clinical suspicion, revealing a notable malignancy rate of 37% in this specific group. The present study showed that only 3.2% (5/153) of total cases underwent surgery for clinical suspicion without a follow-up FNAC after a non-diagnostic FNAC in the first attempt and 40% (2/5) of these operated cases showed malignancy in histopathological examination.

In the present study also, among the non-diagnostic cases (14.4%, 22 cases out of 153), repeated FNACs, ultrasound-guided and non-guided combined FNACs were successful in making the diagnosis in 59% of the total non-diagnostic cases (13 out of 22 cases) which is reasonably significant. As the procedure of FNAC is simple with minimal prerequisites, patient compliance was good and repeat tests conferred less diagnostic burden to the patients.

Most of the non-diagnostic cases turned out to be benign (88.8%) in the present study. Renshow also showed in his study that the chances of malignancy rate reduced in the non-diagnostic group.[16] According to him, the percentage of indeterminate aspirates in thyroid fine-needle aspiration (FNA) can vary significantly, ranging from approximately 5% to 30%. Various factors contribute to this variation, such as differences in the overall malignancy rate within a laboratory and variability in interpretations among cytopathologists.[16]

Though most of the malignancy cases showed hypoechogenic (55.5%) or mixed echogenesity (44.4%), echogenesity was proven to be a poor indicator of malignancy as 74% of the hypoechogenic nodule showed benign cytomorphology in FNAC. On the contrary, all the malignant thyroid lesions showed either a hypoechogenic or a mixed echogenic nodule on ultrasound. Hence regarding ultrasound echogenesity, it can be said that a hypoechogenic or mixed echogenic nodule with other clinically suspicious features possesses more chances of being malignant compared to a hyperechogenic or echogenic thyroid nodule.

Thyroid function tests were shown to be a poor determinant of benign or malignant state of thyroid swelling. The p value (2.6) reflected an insignificant causal association with the risk of malignancy comparable to the p value of 0.98 obtained by Al Maqbali et al. in their study.[14]

Regarding the risk analysis, among the known risk factors,[17,18,19] a number of nodules showed a potent causal association with malignancy with the Odds ratio being 8.9 and p value being significant (0.04). Other risk factors like size of nodules, gender, age, consistency on palpation, echogenesity on ultrasound imaging, ultrasound size of nodules and thyroid function status all showed no causal association. A study done by Al Maqbali et al. also showed a similar causal relationship with the p value being 0.006.[14] Multinodular lesions tend to be benign in most of the cases and FNAC should be done from more than one nodule with any suspicion clinically or as suggested in radiology. Solitary solid thyroid nodules with hypoechogenic or mixed echogenesity in ultrasound possess a substantial risk of malignancy as turned out in FNAC in the present study.

Conclusion

In thyroid lesions, FNAC serves as a potent screening tool as it is sensitive, inexpensive, and easy to perform and its efficacy enhances many folds when combined with ultrasound guidance.[20,21] The standardisation of reporting of cytomorphological features in TBSRTC provides valuable diagnostic information helping in treatment and follow-up. While the non-diagnostic cases (Category I) in TBSRTC continue to be an area of discussion, the present study has illustrated a practical diagnostic step-wise approach

ISSN: 2454-8642
DOI: https://doi.org/10.24321/2454.8642.202406

in those cases. In the non-diagnostic cases, repeat FNAC and guided FNAC yield better cytomorphological results helping in the diagnosis and analysis of the known risk factors will help in identifying the high-risk cases with the provision of appropriate follow-up.

Conflict of Interest: None

Source of Funding: None

References

1. Muratli A, Erdogan N, Sevim S, Unal I, Akyuz S. Diagnostic efficacy and importance of fine-needle aspiration cytology of thyroid nodules. J Cytol. 2014 Apr;31(2):73-8. [PubMed] [Google Scholar]
2. Hajmanoochehri F, Rabiee E. FNAC accuracy in diagnosis of thyroid neoplasms considering all diagnostic categories of the Bethesda reporting system: a single-institute experience. J Cytol. 2015 Oct-Dec;32(4):238-43. [PubMed] [Google Scholar]
3. Al Dawish MA, Robert AA, Muna A, Eyad A, Al Ghamdi A, Al Hajeri K, Thabet MA, Braham R. Bethesda system for reporting thyroid cytopathology: a three-year study at a tertiary care referral center in Saudi Arabia. World J Clin Oncol. 2017 Apr 10;8(2):151-7. [PubMed] [Google Scholar]
4. Ali SZ, Baloch ZW, Cochand-Priollet B, Schmitt FC, Vielh P, VanderLaan PA. The 2023 Bethesda system for reporting thyroid cytopathology. Thyroid. 2023 Sep;33(9):1039-44. [PubMed]
5. Anand B, Ramdas A, Ambroise MM, Kumar NP. The Bethesda system for reporting thyroid cytopathology: a cytohistological study. J Thyroid Res. 2020 Apr 16;2020:8095378. [PubMed] [Google Scholar]
6. VanderLaan PA, Marqusee E, Krane JF. Usefulness of diagnostic qualifiers for thyroid fine-needle aspirations with atypia of undetermined significance. Am J Clin Pathol. 2011 Oct;136(4):572-7. [PubMed] [Google Scholar]
7. Szumilas M. Explaining odds ratios. J Can Acad Child Adolesc Psychiatry. 2010 Aug;19(3):227-9. [PubMed] [Google Scholar]
8. Agarwal P, Dahiya S, Shams A. Cytomorphological spectrum of thyroid lesions using Bethesda system of reporting with histopathological correlation: a study of 87 cases. Indian J Pathol Oncol. 2019;6(3):454-9. [Google Scholar]
9. Akshatha N, Patil S, Bommanahalli BP. Clinical and cytological spectrum of thyroid lesions and the role of fine needle aspiration cytology in its diagnosis at a tertiary care hospital. Trop J Pathol Microbiol. 2019;5(8):523-8. [Google Scholar]
10. Monappa V, Kudva R. Cytomorphologic diversity of papillary thyroid carcinoma. J Cytol. 2017 Oct-Dec;34(4):183-7. [PubMed] [Google Scholar]
11. Esmaili HA, Taghipour H. Fine-needle aspiration in the diagnosis of thyroid diseases: an appraisal in our institution. Int Scholar Res Notic. 2012:912728. [Google Scholar]
12. 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 Apr-Jun;8(2):76-82. [PubMed] [Google Scholar]
13. Moon HJ, Kim EK, Yoon JH, Kwak JY. Malignancy risk stratification in thyroid nodules with nondiagnostic results at cytologic examination: combination of thyroid imaging reporting and data system and the Bethesda system. Radiology. 2015 Jan;274(1):287-95. [PubMed] [Google Scholar]
14. Al Maqbali T, Tedla M, Weickert MO, Mehanna H. Malignancy risk analysis in patients with inadequate fine needle aspiration cytology (FNAC) of the thyroid. PLoS One. 2012;7(11):e49078. [PubMed] [Google Scholar]
15. Yelave R, Mohite S, Mane V. Study of FNAC in diagnosis of thyroid lesions: a prospective study. Indian J Pathol Oncol. 2018;5(4):574-9. [Google Scholar]
16. Renshaw AA. Significance of repeatedly nondiagnostic thyroid fine-needle aspirations. Am J Clin Pathol. 2011 May;135(5):750-2. [PubMed] [Google Scholar]
17. Liu Y, Su L, Xiao H. Review of factors related to the thyroid cancer epidemic. Int J Endocrinol. 2017;2017:5308635. [PubMed] [Google Scholar]
18. Crnčić TB, Tomaš MI, Girotto N, Ivanković SG. Risk factors for thyroid cancer: what do we know so far? Acta Clin Croat. 2020 Jun;59(Suppl 1):66-72. [PubMed] [Google Scholar]
19. Rago T, Vitti P. Risk stratification of thyroid nodules: from ultrasound features to TIRADS. Cancers (Basel). 2022 Jan 30;14(3):717. [PubMed] [Google Scholar20. Borget I, Vielh P, Leboulleux S, Allyn M, Iacobelli S, Schlumberger M, de Pouvourville G. Assessment of the cost of fine-needle aspiration cytology as a diagnostic tool in patients with thyroid nodules. Am J Clin Pathol. 2008 May;129(5):763-71. [PubMed] [Google Scholar]21.Eloy C, Russ G, Suciu V, Johnson SJ, Rossi ED, Pantanowitz L, Vielh P. Preoperative diagnosis of thyroid nodules: an integrated multidisciplinary approach. Cancer Cytopathol. 2022;130(5):320-5. [PubMed] [Google Scholar]

ISSN: 2454-8642
DOI: https://doi.org/10.24321/2454.8642.202406