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Original Research Article

Assessing the rate of malignancy in Bethesda category III thyroid nodules and its correlation with histopathology

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Abstract

Background: Fine-needle aspiration cytology (FNAC) is a key diagnostic tool for evaluating thyroid nodules. The Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) classifies thyroid nodules into six categories, with Category III (Atypia of Undetermined Significance - AUS) presenting diagnostic challenges due to its variable malignancy risk, reported between 13% and 30%. This study aims to assess the malignancy risk of Bethesda Category III nodules and analyse the role of cytological and radiological features in malignancy prediction.

Materials and Methods: Retrospective study was conducted on 20 patients diagnosed with Bethesda Category III thyroid nodules on FNAC who subsequently underwent surgical resection. FNAC slides were re-evaluated to assess cytomorphological features such as nuclear atypia, chromatin changes, microfollicular patterns, colloid presence, and lymphocytic infiltration. Histopathological findings were correlated with FNAC results to determine malignancy rates. Additionally, sonographic features such as echogenicity, vascularity, and the presence of calcifications were analysed to assess their predictive value for malignancy.

Results: Among the 20 cases, 35% (7/20) were malignant, while 65% (13/20) were benign. The malignant cases included follicular carcinoma (n=4) and the follicular variant of papillary carcinoma (n=3). The benign cases consisted of nodular goiter (n=6), follicular adenoma (n=6), and Hashimoto thyroiditis (n=1). Hypoechogenicity and irregular margins were observed in 5 malignant cases, and one case demonstrated microcalcifications. FNAC features such as nuclear grooves, chromatin pallor, and microfollicular patterns were significantly associated with malignancy.

Conclusion: Malignancy risk of Bethesda Category III thyroid nodules may be higher than traditionally estimated in the present study. A multidisciplinary approach, integrating cytological, radiological, and clinical findings, is crucial for guiding patient management, including repeat FNAC, molecular testing, or surgical intervention. Due to the limited sample size, further large-scale, multicenter studies are warranted to validate these findings and optimize diagnostic strategies.

Keywords: TBSRTC, AUS, FNAC, Histopathology, ROM.

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1. Introduction

The accurate diagnosis of thyroid nodule on Fine needle aspiration (FNA) poses a diagnostic challenge for the pathologist due to overlapping features. However it can guide the clinician by estimating the risk of malignancy and thereby helping in the management of the patients. It is also very cost effective and day care procedure with minimal risk of complication. The Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) is a valuable tool for improving the accuracy, consistency, and efficiency of thyroid FNA reporting, ultimately leading to better patient care and management.

TBSRTC 2023 categories thyroid FNA results into six categories each having different malignant risk. Among all, the categories III is the most confusing due to inter observer variability between pathologists. The term ‘‘follicular lesion of undetermined significance’’ used earlier was found to be ambiguous and thereby replaced by Atypia of undetermined significance, AUS. It is further sub classified into two types- Nuclear atypia and atypia others.[1-6] Nuclear atypia carries

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higher risk of malignancy than atypia others. The category III has been ascribed a malignancy rate of 13- 30%, however, malignancy rate varies between different institute and areas.[7-11] Therefore this study was conducted to determine the risk of malignancy in the category III thyroid nodule operated in GMC Kota which will help in tailoring the management of the patient.

2. Material and Methods

Retrospectively 20 cases were selected which were diagnosed with Bethesda Category III on FNAC and histopathology report was available post-surgery. All 20 patients were followed up for informations regarding clinical details, presenting complaints, radiological details.

To re- evaluate the cytological features of Category III thyroid nodules and the associations between cytological and histopathology report.

Association between sonography findings and malignant thyroid nodules.
To determine the malignancy rate of thyroid nodules classified as Category III
The following architectural features were studied:-

1. Formation of macrofollicle and/or microfollicle patterns
2. Presence of papillae or papilla-like groups
3. Discohesion, and the amount of colloid.
4. Nuclear changes such as coarsening, contour irregularity, elongation, clearing or coarse chromatin, presence of grooves, inclusions, equal distances between nuclei,or overlapping
5. Lymphocytosis/Plasmacytosis
6. Calcification and cystic changes

3. Results

Out of 20 cases, 7 cases were found to be malignant on histopathology (35%). 13 cases were benign (65%). Follicular adenoma (6 cases) and nodular goiter (6 cases) were the most common diagnosis among benign cases. One case was found to be Hashimottos thyroiditis. Follicular carcinoma was diagnosed in 4 cases whereas follicular variant of papillary carcinoma was the final diagnosis in 3 cases.(**Table 1**) Sonographic evaluation did not yield any conclusive results in malignant thyroid nodule. Hypoechogenic was seen in 5 cases and iso echogenic in 2 cases. Microcalcification was evident in one case. The peripheral flow was found in 4 cases, central flow in 2 cases and mixed flow in 2 cases during Doppler vascular flow study. Nodes were of sub centimeter in sizes in all the 7 cases.(**Table 2**)

Table 1: Correlation between cytology and Histopathological diagnosis

Histopathological Diagnosis	Number of cases	Percentage (n=20)
1. Benign	13	65%
Nodular goiter or adenomatoid	6	
Follicular Adenoma	6	
Hashimoto thyroiditis	1	
2. Maligant	7	35%
Follicular carcinoma	4	
Follicular Variant of papillary carcinoma	3	
Total no. of cases	20	

Table 2: Association between Sonographic characteristics and Histopathological diagnosis of malignancy

Sonographic Characteristics	No. of cases
1. Echogenicity	
Hypo echogenic	5
Iso echogenic	2
2. Doppler Vascular flow	
Peripheral flow	4
Mixed flow (Peripheral and central)	2
Central flow	1
3. Microcalcification	1
4. Node- sub centimeter in size	7

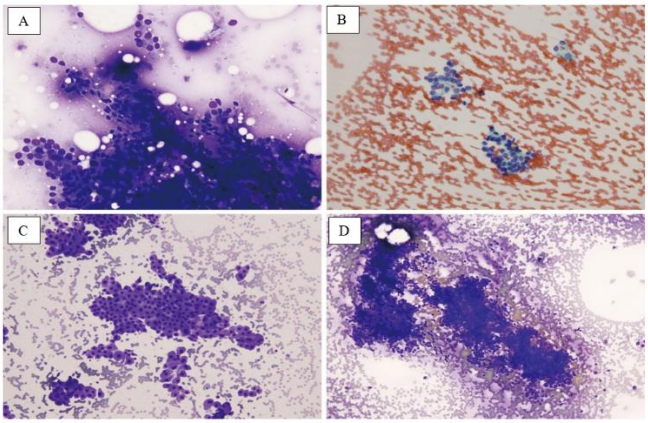

Figure 1: Atypia of undetermined significance with architectural and cytologic atypia. **A):** Follicular cells arranged as syncytial sheets/fragments with nuclear crowding/overlapping and loss of nuclear polarity. **B):** Sparsely cellular smear with a predominance of microfollicles. **C):** Follicular cells are arranged as large monolayer, honeycomb sheets/fragments with evenly distributed nuclei. **D):** Crowded three-dimensional configuration of follicular cells with nuclear overlapping

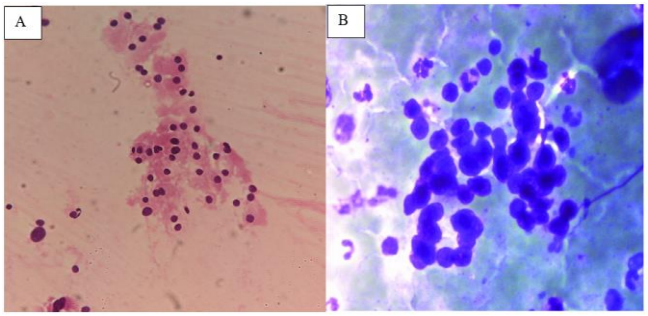

Figure 2: **A):** Scant cellular smear with smooth outlines and evenly spaced nuclei. Follicles contain colloid in back ground. **B):** Architectural atypia is manifested by crowed three- dimensional configuration of follicular cells. Cytology atypia is also evident with nuclear enlargement, slight chromatin pallor, nuclear crowding /overlapping

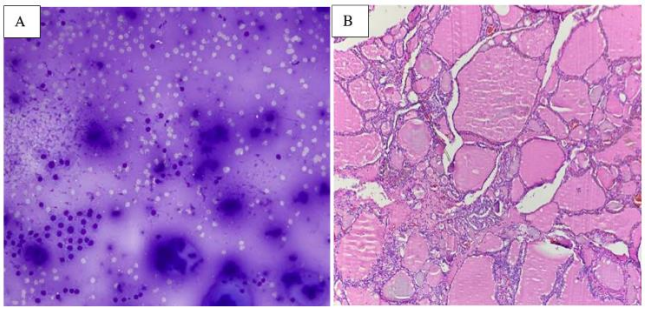

Figure 3: A): Cytology Colloid Goitre - 40x (MGG Stain) Follicular epithelial cells arranged in single cells, monolayer sheets, poorly cohesive clusters, **B):** 40X (H & E) Multinodular goitre- Variable sized dilated follicles with flattened to hyperplastic epithelium

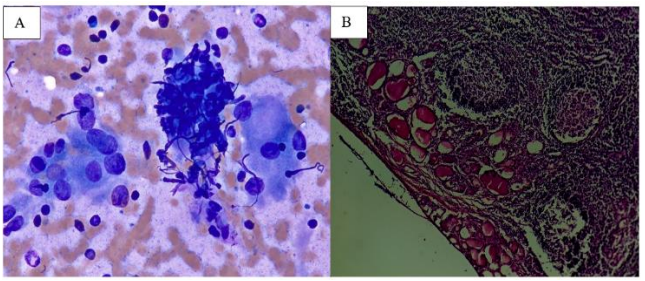

Figure 4: A): Focal cytologic atypia – Moderately cellular smear and cells have nuclear enlargement, pale chromatin, and irregular nuclear contours, lymphocytes infiltrating follicular epithelial cells. **B):** 40X (H & E) Hashimoto thyroiditis - Thyroid follicular destruction with diffuse infiltration of lymphocytes and lymphoid follicle formation with germinal centres

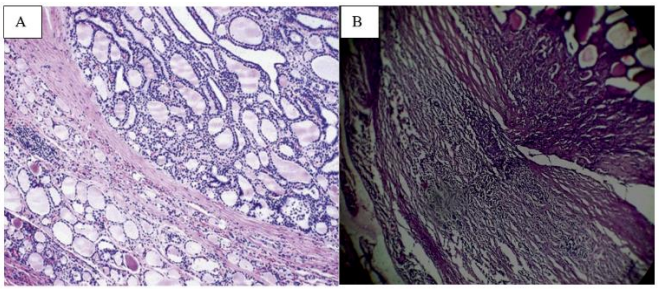

Figure 5: **A)** 40 X (H & E) Follicular adenoma -The tumour is separated from the adjacent thyroid by a complete fibrous capsule. No capsular or vascular invasion is present. **B):** 40X (H & E) Follicular carcinoma with capsular invasion

4. Discussion

The Bethesda system has simplified the cytology reporting of thyroid nodules and stratifies the risk of malignancy which guide the clinical management of the patients. The implied risk of malignancy associated with each category helps clinicians to make informed decisions regarding follow-up, further testing, and surgical management. The Category III is considered heterogeneous, encompassing a range of thyroid nodules with varying degrees of atypia, making it challenging to predict the risk of malignancy. It may require repeat FNA, imaging studies or surgical intervention.(**Figure 1** and **Figure 2**)

Among 20 cases of AUS nodules, malignancy was confirmed in 35% (07/20). 13 cases turned out be benign. Nodular goiter or adenomatoid nodule was found in 06 cases, 06 cases were follicular adenoma and 1 case was Hashimoto thyroiditis. Among the malignant cases follicular carcinoma and follicular variant of papillary carcinoma was the final diagnosis.(**Figure 3-Figure 5**)

The malignancy rate was found to be more than reported in the literature. However the rate of malignancy is variable among different studies. Bayrak et al[14] found the rates of malignancy were 25% for category III, whereas HO AS[15] reported 26.6-37.8% of AUS nodules harboring cancer. Alshalaan AM et al[9] reported 27.3% for patients with AUS on FNA but a higher rate of malignancy (69%) was evident in a study done by Park et al.[6] In his study, papillary carcinoma was confirmed in 84 cases (66.7%), follicular carcinoma in two cases (1.6%). He attributed this findings to patients willing to undergo surgery than repeat FNA and imaging. A few authors admits that AUS will always have inter observer variability due to overlapping features.[5]

A multi-institutional study found that when compared to the benign category, the AUS category exhibited the highest level of variability among cytopathologists. This variability is likely due to the different thresholds used by individual cytopathologists when applying diagnostic criteria.[16]

There are various approaches that can help reduce the overuse of the AUS category, which include, but are not restricted to.[7,8,12]

1. The cohesive sheets or fragments with a monolayer or honeycomb arrangement seen in benign follicular nodules should not be misinterpreted as syncytial sheets or fragments found in PTC and intact follicles or spherules as microfollicles.(**Figure 1**)
2. A few microfollicles can be seen in benign follicular nodular disease where predominant population of follicular cells are arranged in monolayer, honeycomb sheets and/or spherules with or without colloid.
3. Intranuclear grooves and pseudoinclusions are not specific for papillary carcinoma and can be seen in benign follicular disease.

In the current study, cytological features of FVPTC and follicular carcinoma that had been classified as category III on FNA were evaluated. Microfollicular pattern was observed predominantly along with macrofollicular pattern. Most of the cases has significant pallor, frequent grooves. Intra nuclear inclusion and nuclear enlargement were not evident. Nuclear atypia, oncocytic atypia and nuclear overcrowding were observed focally. Colloid was scant to moderate.

In 2023, the TBSRTC further classifies AUS into two subcategories: AUS with nuclear atypia (AUS-N) and AUS with other atypia (AUS-O), showing that the risk of malignancy (ROM) differs between these two subcategories. Bagis et al reviewed 1224 cases diagnosed as AUS and concluded that different types of atypia should be evaluated and reported according to stringent criteria. This may help in reducing the variability among pathologist and remodeling of TBSRTC.[17] On sonography, only one malignant case had microcalcifcations and 6 had poorly defined margins. Hypo echogenic was found in 5 cases. Some authors argue that hypo echogenicity, irregular margin and microcalcifcations, are the ultrasound findings most commonly associated with malignancy, whether occurring individually or together.[13] In the present study, there was no correlation found between sonography findings and malignant thyroid nodule.

Irrespective of the present study outcome, his classification assists clinicians predict the malignancy risk of the thyroid nodule and influences patient management, such as the need for repeat FNA, molecular testing, surgery, and the type of possible surgical intervention.

Presence of both nuclear and architectural atypia can occur together. It is recommended to report cases exhibiting architectural atypia, oncocytic atypia, atypical lymphocytic cells, and isolated psammomatous calcification as "AUS-other.

5. Limitation

There was no correlation found between Sonography findings and malignant thyroid nodule. ROM was found to be same between the nuclear atypia and atypia others. This may be due to small sample size, single-center design of the study, hypo cellularity and technical errors in a few cases.

6. Conclusion

Our findings suggest that the risks of malignancy and neoplasm in category III cases are slightly higher than recommended by TBSRTC. Separating Category III into subgroups revealed no significant differences in the present study, however nuclear atypia and others should be reported. The pathologist can communicate with the clinician and surgeon explaining the risk and help in the management of the patient. A multidisciplinary approach which includes endocrinologist, surgeon, cytologist and radiologist will help in improving the accuracy of diagnosis, reduced unnecessary procedures and improved patient outcomes.

7. Source of Funding

None.

8. Conflict of Interest

None.

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