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Original Research Article

Evaluation of Fine Needle Aspiration Cytology in Thyroid Lesions with Histopathological Correlation

Manal A. Khalaf[1, 2*], Omayma M. Osman[1]

[1]Pathology Department, Faculty of Medicine, Albaha University, Saudi Arabia
[2]Pathology Depaetment, Faculty of Medicine, Minia University, Egypt

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***Corresponding author:** Manal A. Khalaf
Pathology Department, Faculty of Medicine, Albaha University, Saudi Arabia

Abstract

***Background*:** Thyroid diseases are considered the commonest endocrine disorders worldwide. Fine needle aspiration cytology (FNAC) is the mosteasy, reliable screening test in diagnosing thyroid lesions. ***Objectives*:** To determine the frequency of Bethesda system diagnostic categories and cyto-histopathological correlation of FNAC results. ***Material and methods*:** This study comprised 488 thyroid FNA smears. cyto-histopathological correlation was conducted on surgically excised cases. ***Results*:** Out of 488 of thyroid FNAC smears, thyroidectomy was done for one hundred cases. Sensitivity, specificity, PPV, NPV and accuracy of FNAC were 82.8%, 94%, 88.9%, 90.4% and 89.9% respectively. ***Conclusion*:** FNAC is a highly sensitive and specific method in diagnosing different thyroid lesions.

Keywords: Thyroid diseases, Fine needle aspiration cytology (FNAC), diagnosing thyroid lesions.

1. INTRODUCTION

Thyroid diseases are the commonest endocrine disorders in the world [1]. Thyroid nodules prevalence is 3%–7% by palpation and increased to 19%–67% by ultrasonography [2, 3]. Majority of nodules are benign (90%), while thyroid cancer comprises (10%), with increasing incidence worldwide [4]. In Kingdom of Saudi Arabia (KSA), thyroid cancer accounted for about 11% of all newly diagnosed cancers, in 2008, in females [5-7].

Fine needle aspiration cytology (FNAC) is cost-effective, minimally invasive, and efficient method in identifying benign and malignant thyroid nodules and subsequently reducing unneeded surgery in benign lesions [8]. Yet, FNAC has some challenges due to suboptimal preparation of the smears, inadequate sampling, sampling errors and inter-observer variability. Additionally, lower accuracy is observed in follicular neoplasms and suspicious cytology [9, 10].

The Bethesda System for Reporting Thyroid Cytopathology (TBSRTC) has been adopted in 2007 for more consistency in reporting of FNAC and management plans [9, 11]. An updated version has been introduced in 2017 depending on the development in thyroid pathology field. In the new version of TBSRTC, the risk of malignancy (ROM) for each diagnostic category has been calculated when NIFTP is not included among malignancy, and when NIFTP is considered a malignancy [12].

The current study analyzed thyroid cytology based on Bethesda system, with cyto-histopathological correlation, aiming to determine the accuracy of FNAC in diagnosis of thyroid disorders.

2. PATIENTS AND METHODS

The current work comprised 488 thyroid FNA smears. They collected from Pathology Department, King Fahad hospital Al-Baha city, from March 2017 to February 2021.

Smears were fixed by air-dried methods and stained Papanicolaou stains. Smears were interpreted by two expert pathologists using TBSRTC system which includes six categories; non-diagnostic/unsatisfactory ND/US Bethesda system diagnostic categories1, (BDC I), benign (BDCII), atypia of undetermined significance/follicular lesion of undetermined significance AUS/FLUS (BDCIII), follicular neoplasm /suspicious for follicular neoplasm FN/SFN (BDCIV), suspicious for malignancy (SM) (BDC V), and malignant (BDC VI) [13].

One hundred patients underwent thyroidectomy. The specimens were processed and stained in automatic tissue processor and strainer. Two expert pathologists examined sections with correlation to cytological results.

Cases diagnosed of ND/US and AUS/ FLUS were excluded, as those two categories do not express definitive diagnosis of benign or malignant. Moreover, cases with histopathological diagnosis of incidental papillary carcinoma (PC) were excluded as they not aspirated by FNAC and the smears were from coexisting benign lesions. Then sensitivity, specificity, diagnostic accuracy, positive predictive value (PPV), and negative predictive value (NPV) were calculated to asses FNAC efficiency.

True-positive (TP) category included cases diagnosed as BDC IV, V, and VI and diagnosed neoplastic on histopathology, whilst cases that turned out to be non-neoplastic included in false-positive (FP) category. Cases with BDC II and confirmed histopathologically to be non-neoplastic were considered true-negative (TN). While false-negative (FN) category comprised those with neoplastic diagnosis.

The risk of malignancy was determined by correlating cytological results with histopathologic findings. Firstly, It was estimated when noninvasive follicular thyroid neoplasm with papillary like nuclear features (NIFTP) and well differentiated tumor of uncertain malignant potential (WDT-UMP) were considered as malignant, and recalculated when NIFTP and WDT-UMP were included in nonmalignant cases.

Statistical Analysis

Data were checked, coded, entered and analyzed by using SPSS (The Statistical Package for Social Sciences) version 21.0 software and MedCalc version 19.0 software. Descriptive data are presented as means ± standard deviation for continuous variables and percentages for categorical variables. Receiver Operating Characteristic (ROC) curve measures the accuracy that combines sensitivity and specificity with the estimation of the area under the curve of FNA as regarding the histopathologic findings of study group. Statistical significance was set at p-value < 0.05.

3. RESULTS

This study involved 488 of thyroid FNAC smears, those were taken from patients attending King Fahad Hospital in Al-Baha city from 2017 to 2021. The mean age was 42±11.6 years. The female to male ratio was about 7:1. More than half of biopsies were collected from the left lobe (50.4%) (Table 1).

Table-1: General characteristics of the studied thyroid biopsies

General characteristics	The studied thyroid biopsies No = 488	
	No	%
Age categories		
20 – 39 years	**174**	**35.6%**
40 -59 years	**242**	**49.6%**
60 -80 years	**72**	**14.8%**
Gender		
Female	**427**	**87.5%**
Male	**61**	**12.5%**
Year of specimen		
2017	**112**	**23%**
2018	**126**	**25.8%**
2019	**104**	**21.3%**
2020	**57**	**11.7%**
2021	**89**	**18.2%**
Site of the lesion in the thyroid		
Left lobe	**246**	**50.4%**
Right lobe	**178**	**36.5%**
Isthmus	**40**	**8.2%**
Bilateral	**24**	**4.9%**

Table 2 displayed cytological diagnoses of 488 of thyroid FNAC smears. ND/US category comprised 117 cases (23.9%), (Figure 1), benign category included 293 cases (60%) (Figure 2), AUS/FLUS category comprised 44 cases (9%) (Figure 3), FN/SFN included 12 cases (2.4%), Six cases (1.2%) were diagnosed as SM, finally malignant category included sixteen cases (3.3%) (Figure 4).

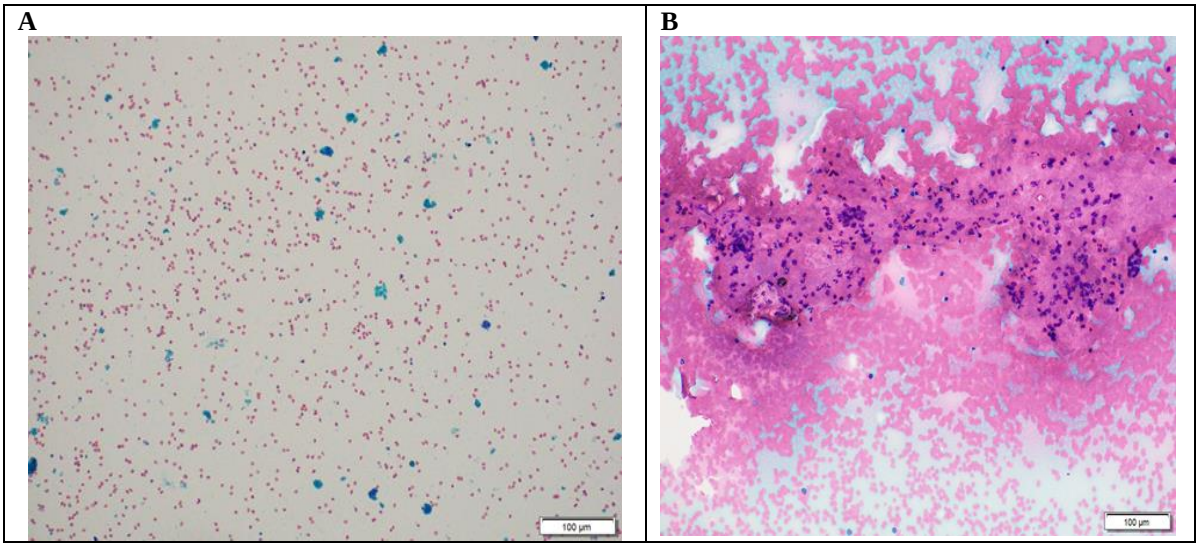


Figure-1: Non-diagnostic or Unsatisfactory;: **A,** Photomicrigraph showing a smear within adequate number of follicular cells(Smear, Papanicolaou stain,10 x magnification). **B,** Photomicrigraph displaying air dried smear (Smear, Papanicolaou stain, 20 x magnification).

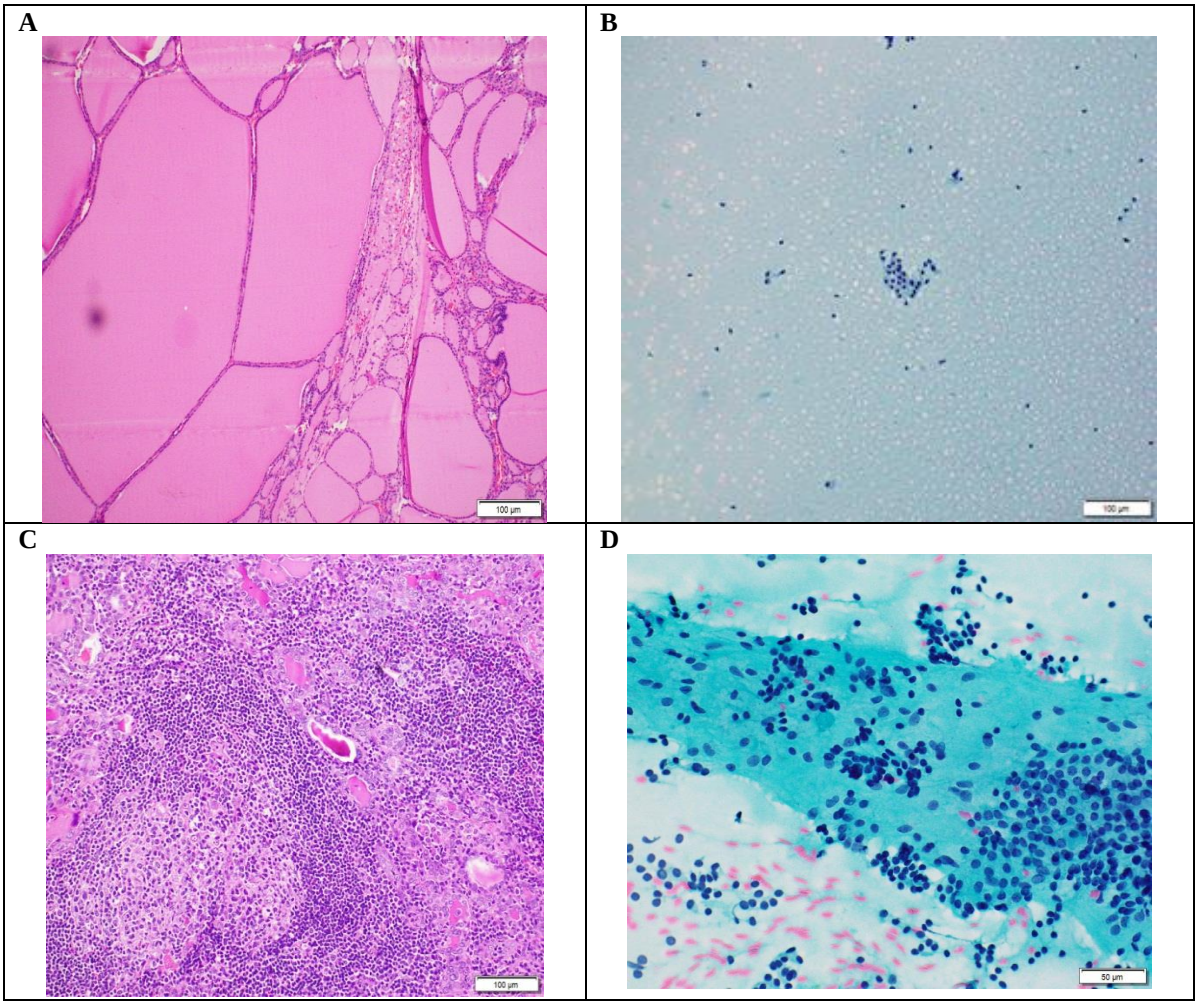


Figure-2: A, Colloid nodule: Photomicrograph displaying multiple nodules of dilated follicles lined by flattened epithelium and filled with colloid. (H & Estain, 20 x magnification). **B, Colloid nodule:** Photomicrigraph displaying sparsely cellular smears with abundant colloid and evenly spaced follicular cells (Smear, Papanicolaou stain, 20 x magnification). **C, lymphocytic thyroiditis**: Photomicrigraph displaying atrophic thyroid follicles with heavy lymphocytic infiltrate (H & E stain, 20 x magnification). **D, lymphocytic thyroiditis:** Photomicrigraph displaying benign appearing follicular cells and many lymphoid cells (Smear, Papanicolaou stain, 40 x magnification)

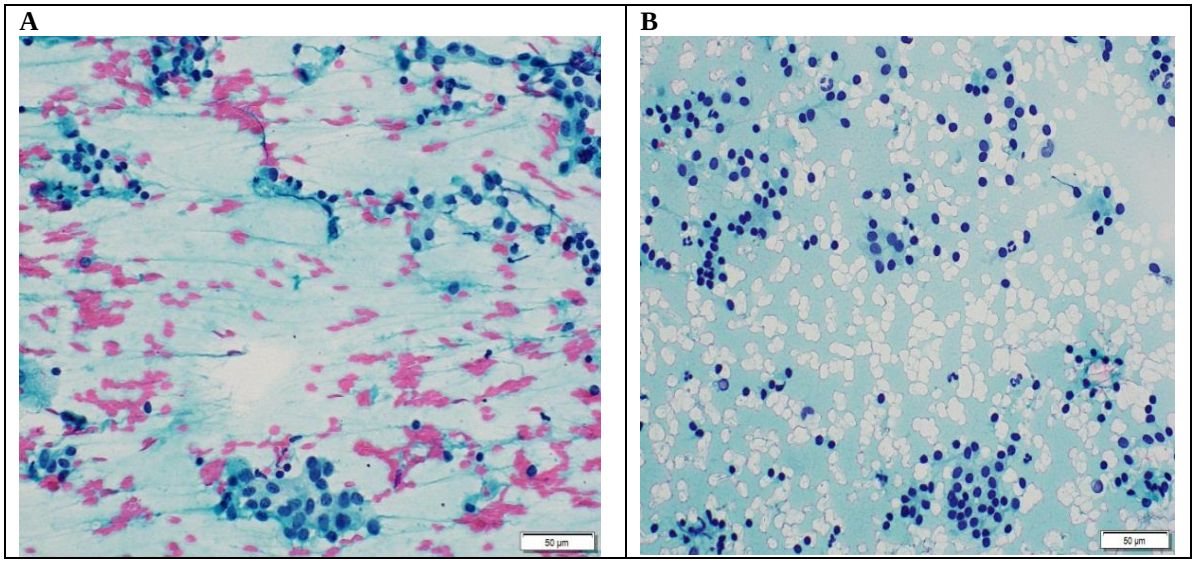


Figure (3): AUS/FLUS: A, Photomicrigraph displaying cells with focal nuclear enlargement with pale chromatin (Papanicolaou stain,40 x magnification). **B,** Photomicrigraph displaying moderately cellular smear with microfollicle formation (Smear, Papanicolaou stain, 40 x magnification)

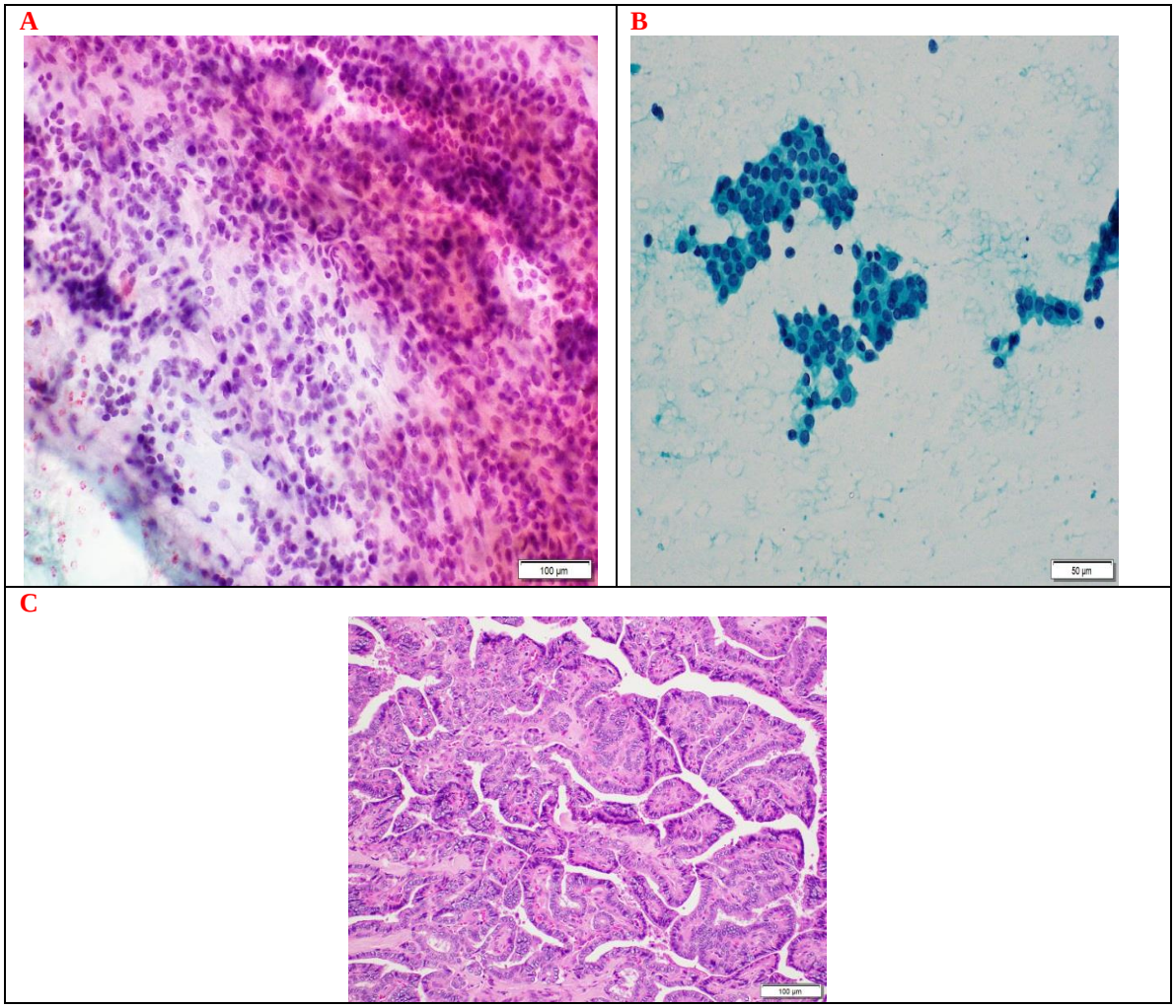
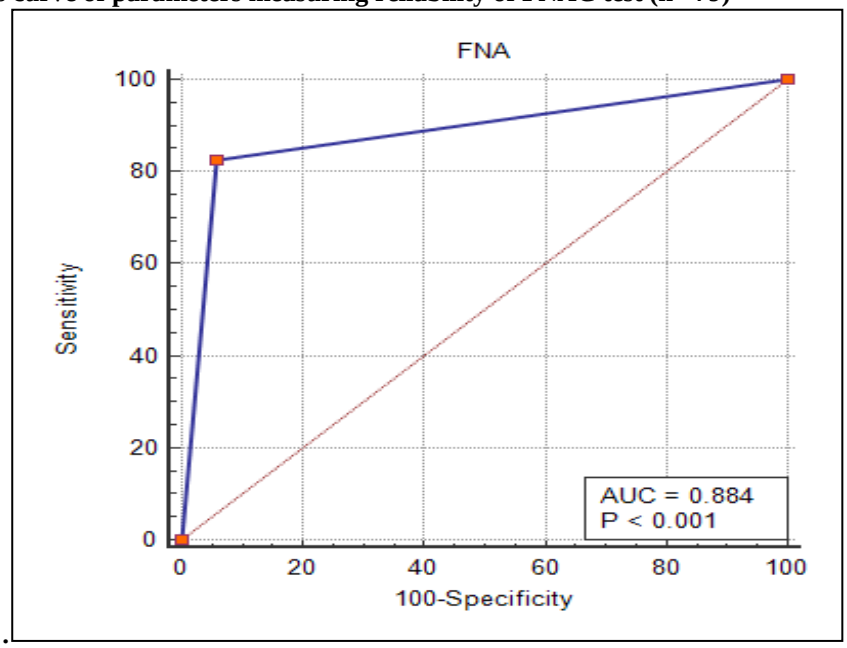


Figure 4: A, Suspicious for papillary carcinoma: Photomicrigraph displaying hypercellular smear with nuclear crowdening and focal clearing (Smear, Papanicolaou stain, 40 x magnification). **B, Papillary carcinoma**: Photomicrigraph displayingsheet of tumor cells with nuclear overlapping, enlargement and chromatin clearing (H&E stain, 40 x magnification). **C, Papillary carcinoma:** Photomicrigraph displaying papillae lined by cells with nuclear enlargement and crowding with many cells displaying nuclear grooves (Smear, Papanicolaou stain, 40 x magnification)

Figure 5: ROC plots curve of parameters measuring reliability of FNAC test (n =79)



AUC: Area under the curve, p-value <0.001

Table-2: Distribution of cytological diagnoses of 488 studied thyroid smears

Cytological diagnosis No (%)	The studied thyroid biopsies (No= 488)	
	Sub-categorical diagnosis	No. (%)
I. ND/US, 117 (23.9%)	**Paucicellular with colloid and RBCs**	**11 (2.3%)**
	Bloody smear	**6 (1.2%)**
	Paucicellular with mixed inflammatory cells	**4 (0.8%)**
	With few crushed follicular cells	**5 (1%)**
	Thick smear	**3 (0.6%)**
	No subcategory	**88 (18%)**
II. Benign, 293 (60%)	**-Benign follicular nodule**	**142 (29.1%)**
	Colloid nodule with cystic changes	**74 (15.3%)**
	Colloid nodule	**38 (7.8%)**
	Hashimoto thyroiditis	**24 (4.8%)**
	-Dominant adenomatoid nodule	**10 (2%)**
	-Benign follicular nodule with Hurthle cell changes	**5 (1%)**
III. AUS/FLUS, 44 (9%)	**No subcategory**	**44 (9%)**
IV. FN/SFN, 12 (2.4%)	**Hurthle cell neoplasms**	**3 (0.6%)**
	Favors carcinoma	**3 (0.6%)**
	No subcategory	**6 (1.2%)**
V. SM, 6 (1.2%)	**Favors (PC)**	**2 (0.4%)**
	No subcategory	**4 (0.8%)**
VI. Malignant, 16 (3.3%)	**Favor (PC)**	**10 (2.1%)**
	Favorsintracystic (PC)	**2 (0.4%)**
	Favor (FVPC)	**2 (0.4%)**
	Favors anaplastic carcinoma	**2 (0.4%)**

ND/US; non-diagnostic/unsatisfactory, AUS/FLUS;atypia of undetermined significance/follicular lesion of undetermined significance, FN/SFN follicular neoplasm /suspicious for follicular neoplasm, SM; suspicious for malignancy, PC; papillary thyroid carcinoma, FVPC; follicular variant papillary carcinoma

As shown in Table 3; one hundred cases underwent thyroidectomy. Histopathological examination revealed 59 (59%) cases were non neoplastic lesions. The neoplastic cases included 12 cases of follicular adenoma (FA), four cases of follicular carcinoma (FC), two cases were (WDT-UMP), and one case was (NIFTP). Papillary carcinoma comprised the most common cancer of 22 cases.

Table-3: Distribution of histological diagnoses in patients with thyroidectomy specimens

Histological diagnosis	No.	%	Subtype	No.
-Non-neoplastic	**59**	**59%**	**-Multinodulargoiter (MNG)** Simple MNG Toxic MNG MNG with focal adenomatous changes **-Hashimoto thyroiditis** **-Benign nodule with Hurthle cell changes**	**33** **10** **9** **2** **4** **1**
-Neoplastic				
Follicular adenoma	**12**	**12%**	**-**	**-**
Follicular carcinoma	**4**	**4%**	**Angioinvasive** **Minimally invasive** **Widely invasive**	**1** **1** **2**
WDT-UMP	**2**	**2%**	**-**	**-**
NIFTP	**1**	**1%**	**-**	**-**
Papillary carcinoma	**22**	**22%**	**Classic** **Intracystic** **Microscopic**	**18** **2** **2**

MNG; multinodular goiter, BFN; benign follicular nodule, WDT-UMP; well differentiated tumor of uncertain malignant potential; NIFTP; noninvasive follicular thyroid neoplasm with papillary like nuclear features,

Fifty four cases (14.2%) of benign category had undergone thyroidectomy. Forty seven cases of them were confirmed to benon-neoplasticon histopathology; therefore true negative FNAC findings were 47 cases (87%). While seven cases turned out to be neoplastic. After exclusion of two cases of incidental papillary microcarcinoma, the false negative results were 5(9.3%).

Surgery was done for nine cases (81.8%) of FN/SFN category. Seven cases were proved to be neoplastic on histopathology and those represent true positive results (77.8%).On the other hand, two cases were diagnosed MNG, and those considered false positive (22.2).

All three cases of SM category underwent surgical intervention. Two cases of them confirmed to be malignant (PC) and they represent true positive results (67%). While single case had thyroiditis and considered false positive finding (33%). Fifteen malignant cases (93.8%) were followed by surgical excision; all of them were proved to be neoplastic/malignant. Therefore true positive results were (100) in this category (Table 4).

Table-4: Correlation between cytotological and histopathological diagnosis in one hundred thyroid biopsies

Cytological diagnosis	Histological diagnosis of one hundred thyroid biopsies						
	Non-neoplastic N=59		Benign N=12	Malignant N=29			
	Goiter N=55 No (%)	Thyroiditis N=4 No (%)	Follicular adenoma N=12 No (%)	WDT-UMP N=2 No (%)	NIFTP N=1 No (%)	Follicular carcinoma N=4 No (%)	Papillary carcinoma N=22 No (%)
I. ND/US (N=6)	**5 (9.1%)**	**0 (0%)**	**0 (0%)**	**0 (0%)**	**0 (0%)**	**0 (0%)**	**1 (4.6%)**
II. Benign (N=54)	**44 (80%)**	**3 (75%)**	**2 (16.6%)**	**0 (0%)**	**1 (100%)**	**1 (25%)**	**3 (13.6%)**
III. AUS/FLUS (N=13)	**4 (7.3%)**	**0 (0%)**	**5 (41.7%)**	**0 (0%)**	**0 (0%)**	**2 (50%)**	**2 (9.1%)**
IV. FN/SFN (N=9)	**2 (3.6%)**	**0 (0%)**	**5 (41.7%)**	**1 (50%)**	**0 (0%)**	**1 (25%)**	**0 (0%)**
V. SM (N=3)	**0 (0%)**	**1 (25%)**	**0 (0%)**	**0 (0%)**	**0 (0%)**	**0 (0%)**	**2 (9.1%)**
VI. Malignant (N=15)	**0 (0%)**	**0 (0%)**	**0 (0%)**	**1 (50%)**	**0 (0%)**	**0 (0%)**	**14 (63.6%)**

ND/US; non-diagnostic/unsatisfactory, AUS/FLUS; atypia of undetermined significance/follicular lesion of undetermined significance, FN/SFN follicular neoplasm /suspicious for follicular neoplasm, SM; suspicious for malignancy, NIFTP; noninvasive follicular thyroid neoplasm with papillary like nuclear features, WDT-UMP; well differentiated tumor of uncertain malignant potential

The risk of malignancy was estimated regarding Bethesda system diagnostic category. When NIFTP and WDT-UMP were diagnosed malignant neoplasms, the ROMwas16.7% for BDC I, 11.1% for BDC II, 30.8% for BDC III, 22.2% for BDC IV, 66.7% for BDC V, and 100% for BDC VI. While, when NIFTP and WDT-UMP are considered non-malignant cases, the ROMs were 16.7%, 7.4%, 30.8%, 22.2%, 66.7%, and 93.3% respectively (Table 5).

Table-5: Risk of malignancy in each Bethesda system diagnostic category in relation to histological diagnosis in one hundred studied thyroid biopsies

Cytological diagnosis	Histological diagnosis	
	ROM when NIFTP and WDT-UMP considered malignant	ROM when NIFTP and WDT-UMP considered non-malignant
I. UD/UNS	16.7%	16.7%
II. Benign	11.1%	7.4%
III. AUS/FLUS	30.8%	30.8%
IV. FN/SFN	22.2%	11.1%
V. SM	66.7%	66.7%
VI. Malignant	100%	93.3%

NIFTP; noninvasive follicular thyroid neoplasm with papillary like nuclear features, WDT-UMP; well differentiated tumor of uncertain malignant potential, ROM; risk of malignancy

Based on cyto-histopathological comparison, the ROC plots curve calculated the sensitivity, specificity, PPV, NPV and accuracy of FNAC after exclusion of ND/US, AUS/FLUS categories. Furthermore, the two cases of incidental papillary microcarcinoma were excluded. The parameters were 82.8%, 94%, 88.9%, 90.4% and 89.9% respectively (Figure 5).

4. DISCUSSION

Thyroid diseases are misunderstood and are often overlooked and misdiagnosed [14]. Although FNAC is a rapid and easy procedure with few complications [15], it has limitations due to scanty sample, variation in sampling technique, skills of the technique performer, the experience of cytologist and vascularity of thyroid swelling. Those limitations impede reporting accurate diagnosis [16]. Thereby, our study was conducted to estimate the accuracy of FNAC test in identifying different thyroid lesions through correlation of FNA with histopathological diagnoses.

In the current work, FNAC interpreted 381 (60.7%) cases as benign. The most common benign diagnosis was benign follicular nodules (197) followed by colloid nodule with cystic changes (99). On histopathology, benign /non neoplastic lesions comprised the majority of cases 59 (59%), and MNG is the most common non neoplastic lesion (54 cases). Many studies reported close findings **[**10, 17, 18].

In the present study, PC comprised the most common malignancy 22 (22%), on histopathology, and the majority of them were of classic type. In line with our results, Gupta *et al*., [10] and Zarif *et al*., [18] demonstrated that PC was the most common malignancy, but Zarif *et al*., [18] reported that FVPC was the most common subtype. Nandedkar *et al*., [17] contradicted the previous findings and reported that FC was the common cancer in their study followed by PC.

In our study, ND/US category constituted 129 (20.9%) cases and this is comparable with Jo *et al*., [19] who reported (18.6%). Furthermore, in a meta-analysis of 8 studies by Bongiovanni *et al.*, [20], the range of incidence of this category was from 1.8% to 23.6%, and the average was 12.9%. Much Lower percentages of this category were mentioned in other studies [17, 18, 21], they reported 4.3%, 6.4% and 11% respectively. The incidence of this category is dependent on the aspirator's experience [22]. TBSRTC has recommended a reaspiration of cases with BDCI after a minimum interval of 3 months to prevent false positive interpretations due to reparative or reactive changes [23]. Repeating FNAC is diagnostic in 50%–88% of cases; while surgical excision should be done for cases with persistent inadequacy as the ROM is 10% [24].

Six (4.7%) cases of BDCI underwent surgical procedure, one case diagnosed PC, and the ROM was 16.7%. In accordance, Bongiovanni *et al.*, [20] reported 16.8% ROM, while higher ROM (34.6 %.) was reported by Zarif *et al*., [18].

FNAC results revealed 381 (60.7%) cases of benign BDC II. In accordance, many studies reported large percentages of this category from 59%, to 80% [20, 22, 25]. Abdulla *et al*., [21] reported slight lower rate of this category (55%) and they explained that because of selection bias.

Benign category are usually managed by clinical follow-up, Yet surgical intervention is performed for other concerns, cosmetic purposes or pressure symptoms [18]. In the current study, fifty four (14.2%) cases of BDCII had undergone thyroidectomy. Seven cases turned out to be neoplastic and the ROM was 11.1%, when NIFTP considered malignant and 7.4%, when NIFTP considered nonmalignant. Consistent with our findings, malignancy rates from 10.5%, to 15.6% were reported in other studies [18, 21, 25]. Lower malignancy rates were reported to be 0%–3% [9, 17]. Bongiovanni *et al.*, [20] displayed ROM range of 1%–10%.with a 3.7% mean. These discordant findings was yielded when NIFTP, WDUM, or occult papillary carcinoma are included or excluded from malignant results.

False negative results are those with benign/non neoplastic cytological diagnosis and turned

out to be neoplastic on histopathology. They represent a major source of limitation in using FNAC procedure, as many neoplastic cases would be missed without management. In the current cohort, after exclusion of the two cases of incidental papillary microcarcinoma, the false negative results were 5 (6.3%). Wide range of false negative rates was demonstrated by previous studies of 1.8-20% [10, 18, 26]. Coexistence of both benign and malignant lesions indicates the sampling from larger benign lesion not from adjacent malignant tissue as well as the overlap between low grade malignant and benign lesions, all those could yield false negative results [27]. Therefore, a negative FNAC diagnosis should never exclude malignancy in presence of obvious clinical suspicion, and cases with benign diagnosis should be followed up strictly [26].

AUS/FLUS diagnosis constitutes the most controversial category, as it yields inconclusive reporting. TBSRTC (2007) has recommended limitation in using BDC III to 7% or less [9]. While TBSRTC (2017) has extended the use of this diagnosis to 10% because of difficulty faced by many laboratories but ensured on using this diagnosis as the last option [17].

In the present study, AUS/FLUS category comprised 77 (12.5%) cases. In accordance, Abdullah *et al*., [21] and Chakravarthy *et al*., [28] showed (16.2%) and (12%) respectively. Our result was much lower than the percentage (79.5%) reported by Chirayath *et al*., [29]. Thirteencases (16.9%) within this category had a follow-up surgery and the ROM was 30.8%. In comparison, the malignancy rates of this category were ranged from15.9% to 29%, [20, 21, 30, 31]. Chakravarthy *et al*., [28], and Chirayath *et al*., [29] demonstrated much higher malignancy rates of 69% and 54.6% respectively. Hence, many studies recommended that those patients should be given strong consideration to surgery [28, 29, 32].

FN/SFN category identifies a lesion that might be a follicular carcinoma and expose it for surrgical excision [29]. The current work included 11 cases (1.8%) within this category. In comparison, the published studies reported a range of 1.7%- 20.3% [17, 18, 21, 22, 25, 33]. Malignancy risks of 15-30% and 25-40% were described by TBSRTC 2007 and TBSRTC 2017 respectively [9, 12]. In our study, surgery was done for 9 (81.8) cases with 22.2% ROM. Alshaikh *et al*., [25] showed the same our percentage (22.2%) and Abdullah *et al*., [21] reported close result (27.3%). On the contrary, higher malignancy rates were showed by Chirayath *et al*., [29] and Doodi *et al.,* [33], 72.4%, and 81% respectively.

Three cases (0.5%) were diagnosed as SM by FNAC; close result (1.15%) was reported by Nandedkar *et al*., [17]. Many previous studies showed higher percentages of 2.6%- 7.2% [18, 21, 22, 25, 26]. All 3 (100%) cases of SM category underwent surgical intervention with 66.7% ROM. Previous studies had close rates of 75.2%, 76.2%, and 72.7% respectively [20, 21, 25]. Higher malignancy rates of 100% and 95.7% were reported in other studies [17, 18].

In the current cohort, sixteen (2.6%) cases were within malignant category. This data is comparable with that reported in previous studies [17, 22, 25], as they reported rates of 1.98%, 2.2%, and 4.1%. Whilst Zarif *et al*., [18] and Sukumaran *et al*., [26] showed higher figures of 23.5% and 59.68% respectively.

Fifteen malignant (93.8%) cases were followed by surgical excision, all of them were confirmed histopathologically to be neoplastic/malignant (1 WDT-UMP and 14 PC). The malignancy rates were 100%, When WDT-UMP considered malignant and 93.3% When WDT-UMP considered nonmalignant. Many studies showed high reliability of FNAC procedure in identifying malignant lesions. As malignancy rates were reported to be 100**%** [17, 18, 25, 26] or 96%, and 98.6% [20, 21]

In our study, false positive results are those with cytologic diagnosis of FN/SFN, SM, or malignant but turned out to be non-neoplastic on histopathology. False positive finding comprised only 3 (3.8%) cases. In accordance, Nandedkar *et al*., [17] reported false positive rate of 1.2% and Zarif *et al*., [18] showed (4.3%). Gupta *et al*., [10] reported high percentage of 13.3%.

In this work sensitivity, specificity, PPV, NPV and accuracy of FNAC were 82.8%, 94%, 88.9%, 90.4% and 89.9% respectively. In comparison, several publications reported close results and proved that FNAC is highly sensitive and specific for diagnosis of thyroid diseases [17, 18, 21, 22, 34].

CONCLUSION

Based on aforementioned results, despite false-negative and false-positive results, FNAC is a highly sensitive and specific procedure in diagnosing different thyroid lesions. Thereby, it can help in decreasing number of surgeries in benign lesions.

Ethical Consideration: Ethical committee of Albaha University approved our study.

LIMITATIONS: Retrospective study.

Conflict of Interest: No conflict of interest.

Funding: Nil

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