Original article:
A study on evaluation of solitary nodular thyroid lesions by FNAC and its histopathological correlation

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Abstract:
Background: Solitary nodular enlargement of thyroid is one of the common indications for surgical excision. Since most of the nodules are benign, symptomless, normal thyroid hormone profile, they do not require excision. This study was done to evaluate the utility of FNAC as a rapid diagnostic method in various thyroid lesions and guide the clinician for further treatment.

Objective: This main objective of the study to evaluate the cyto-histopathological findings, and the accuracy of fine needle aspiration cytology (FNAC) procedure in solitary nodular thyroid lesions. Materials and Methods: This prospective study comprises total 160 cases with thyroid lesions referred to cytopathology laboratory and gland morphology studied by ultrasonography (USG). Based on ultrasonography report Fine Needle Aspiration were done manually or guided and aspirated material fixed on fixative and stained to examine. Results: Age incidence varied from 10-80 yrs. The commonest age group affected was 21-40 yrs. The ratio of male to female was 1:2.2. The duration of symptoms ranged from 10 days to 15 years. Maximum number of cases presented with midline swelling of the neck. On cytology 52 (32.5%) cases were diagnosed as colloid goiter, 46 (28.75%) as colloid goiter with cystic change, 16(10%) as thyroiditis - among them 6(3.75%) as granulomatous thyroiditis, 8(5%) as lymphocytic thyroiditis, 2(1.25%) as dequervain thyroiditis, 10 (6.25%) as follicular neoplasm, 2(1.25%) as hurthle cell adenoma, 5(3.3%) as papillary carcinoma, 2 (1.25%) as medullary carcinoma, 1(0.33%) anaplastic carcinoma and 1(0.33%) NHL as suspicious smear. Histopathological examination was possible in 60 cases. The overall sensitivity was 90.2%, specificity was 98.2% and accuracy was 97.1%. Conclusion: FNAC is a rapid, efficient, cost-effective, relatively painless procedure and produces a early result with a high diagnostic accuracy. It has high rates of sensitivity and specificity to diagnose the solitary thyroid lesions and thereby it is an important diagnostic tool and further management of patients with thyroid lesions.

Keywords: FNAC; Solitary nodule; ultrasonography (USG); cytopathology

Introduction
Thyroid disorders are the most common endocrine diseases and most of them are curable to medical and surgical management. Thyroid lesions are challenging tasks to modern clinician in judging the nature, and thereby, advocating precise and adequate management. Thyroid lesions become vulnerable particularly in countries where iodine intake in diet is low. Fine needle aspiration cytology (FNAC) is a useful evaluation method in assessment of palpable and non-palpable thyroid masses. FNAC is a quick, safe, inexpensive outpatient procedure with minimum morbidity and without mortality. It is a rapid result oriented procedure, which helps in the evaluation of thyroid lesions helping the clinician in deciding the further line of treatment. Now a day it is the first line screening in the diagnosis of thyroid lesions. Usually the thyroid cancers are presented as palpable

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nODULES OR INCREASE IN SIZE OF A PRE-EXISTING NODULE. IT BECOMES IMPORTANT FOR A SURGEON TO INVESTIGATE THESE CHANGES CORRECTLY AND TIMELY. IT HAS BEEN ESTIMATED THAT THYROID MALIGNANCY ARE ABOUT 5 TO 10% OF ALL THYROID NODULES. THE USUAL METHOD OF APPROACH TO SUCH PRESENTATIONS PREVIOUSLY WAS TO EXCISE THESE NODULES AND SEND TO HISTOPATHOLOGY EXAMINATION. HOWEVER WITH AID OF BETTER DIAGNOSTIC TECHNIQUES LIKE FNAC, NOW WE CAN DIFFERENTIATE BENIGN FROM MALIGNANT CASES AND AVOID UNNECESSARY SURGERIES. A SOLITARY THYROID NODULE IS DEFINED AS A PALPABLE SINGLE, CLINICALLY DETECTED NODULE IN THE THYROID. IT IS MORE CONCERN BECAUSE OF HIGH PROBABILITY OF MALIGNANCY, WHICH CAN RANGE FROM 5-35% OF ALL SOLITARY THYROID NODULES. THERE ARE DIFFERENT DIAGNOSTIC MODALITIES USED TO EVALUATE AND DIAGNOSE EFFICIENTLY THYROID NODULES. THESE INCLUDE CLINICAL EXAMINATION, THYROID HORMONE ASSAY, ULTRASONOGRAPHY (USG), FINE NEEDLE ASPIRATION CYTOTOLOGY (FNAC) AND HISTOPATHOLOGICAL EXAMINATION. HOWEVER, CLINICAL ASSESSMENT, THYROID HORMONE ASSAY AND USG HAVE BEEN POOR PARAMETERS FOR ASSESSING THYROID NODULES. FOR FINAL DIAGNOSIS REQUIRES MORPHOLOGICAL EXAMINATION OF LESIONS AND FOR THIS FNAC OR HISTOLOGICAL EXAMINATION BECOMES MANDATORY. FNAC IS AN ESTABLISHED TECHNIQUE FOR THE INVESTIGATION OF THYROID LESIONS. DESPITE MANY ADVANTAGES, FNAC HAS SOME LIMITATIONS THAT INCLUDE SPECIMEN ADEQUACY AND CYTOTOLOGICAL INTERPRETATION, AS THE SAMPLING IS VARIABLE, BLOOD MIXED AND NOT ALWAYS REPRESENTATIVE. THUS A SPECIFIC DIAGNOSIS CAN ONLY BE ARRIVED AT AFTER A HISTOLOGICAL EXAMINATION.

**Materials and Methods**

A prospective study was carried out in the department of Pathology, Jaber Al Ahmed Armed Forces Hospital, Kuwait during the two years period from January 2016 to December 2017. Total 160 cases with thyroid lesions referred to cytology laboratory were considered for this study. Detailed clinical history was procured from each patient by designed proforma before obtaining a sample for cytological study. The gland morphology is studied by USG. USG was performed for all the cases to distinguish cystic and solid lesions. On echogenic pattern to distinguish benign from malignant thyroid lesions. Specimens were obtained for cytological study by Fine needle aspiration (FNA) under USG guided or by palpation (without guided). follicular lesions and suspected malignant lesion underwent surgery and sent histopathological examination. Specimens were processed in automated tissue processing units and staining was performed with routine haematoxylin and eosin stain. The Pearson chi square test is used to compare the possible correlation between FNAC and Histopathology of thyroid lesions.

**Aim**

The aim of study is to evaluate the cytohistopathological findings, and the accuracy of fine needle aspiration cytology (FNAC) procedure in various thyroid lesions.

**Inclusion Criteria**

Those patients having thyroid lesions, irrespective of their age and sex, referred for cytological study from ENT and Surgery Outpatient Department (OPD) and admitted to ward were selected. The gland morphology was studied by USG. USG was performed for all the cases to distinguish cystic and solid lesions. All solitary nodule lesions were included the study. All suspected lobectomy or thyroidectomy specimens accepted for histopathological examination.

**Exclusion criteria**

Clinically and USG those thyroid lesions are diffuse or normal appearance, these lesions are excluded as the study based on solitary nodular thyroid pattern. Nodular lesion but inadequate cytology excluded from study.

**Fixation and Staining**

After Fine needle aspiration of thyroid, all smears were prepared, and fixed by using 95% ethyl alcohol. All surgical specimens were received in 10% buffered formalin. For the standard staining procedure hematoxylin and eosin (H&E), Papanicolaou (PAP) stain was used for the cytological smears. For histopathological sample routine hematoxylin and eosin (H&E) and sometimes special and immunohistochemistry stain were done.

**Ethical clearance:** The present study was conducted after obtaining the ethical approval from the Ethical Review Committee.

**Result**

This study was carried out over 2 years (January 2016 to December 2017). in the department of histopathology, Jaber al Ahmed Armed Forces Hospital, Kuwait. It was a prospective analysis of 160 cases with thyroid lesions referred to cytopathology department from ENT and Surgery department. The distribution of thyroid lesions diagnosed by Fine Needle Aspiration Cytology (FNAC) is shown in the Table-1:
Table -1: Distribution of Thyroid lesion by FNAC

| Serial | Category                              | No of cases (n = 160) | Percentage |
|--------|---------------------------------------|-----------------------|------------|
| 1      | Colloid nodular goiter                | 52                    | 32.5%      |
| 2      | Colloid nodular goiter with cystic changes | 46                 | 28.75%     |
| 3      | Hashimotos thyroiditis                | 6                     | 3.75%      |
| 4      | Lymphocytic thyroiditis               | 8                     | 5%         |
| 5      | Dequervain thyroiditis                | 2                     | 1.25%      |
| 6      | Hyperplastic thyroid nodule           | 25                    | 15.6%      |
| 7      | Follicular neoplasm                   | 10                    | 6.25%      |
| 8      | Hurthle cell neoplasm                 | 2                     | 1.25%      |
| 9      | Papillary carcinoma                   | 5                     | 3.3%       |
| 10     | Medullary carcinoma                   | 2                     | 1.25%      |
| 11     | Anaplastic carcinoma                  | 1                     | 0.625%     |
| 12     | Lymphoma                              | 1                     | 0.625%     |
| Total  |                                       | 160                   | 100%       |

Age incidence varied from 10-80 yrs. The most common age group affected was 21-40 yrs. The duration of symptoms ranged from 10 days to 15 years. Most of the cases presented with midline swelling of the neck and feeling discomfort during swallowing of food. Among all cases in this study, most of the cases (42.5%) between 21-40 yrs followed by 41-60 yrs (21.25%). Females are predominantly affected than male and M:F=1:2.2. The age distribution of Thyroid lesions diagnosed by FNAC is shown in table II: Table -II: Age distribution of Thyroid lesion diagnosis by FNAC (n=160)

| S/L | Category                              | 10-20 yrs | 21-40 yrs | 41-60 yrs | 61-80 yrs |
|-----|---------------------------------------|-----------|-----------|-----------|-----------|
| 1   | Colloid nodular goiter                | 10        | 25        | 12        | 05        |
| 2   | Colloid nodular goiter with cystic changes | 06   | 22        | 08        | 08        |
| 3   | Hashimotos thyroiditis                | 01        | 02        | 02        | 01        |
| 4   | Lymphocytic thyroiditis               | 02        | 02        | 02        | 02        |
| 5   | Dequervain thyroiditis                | -         | 01        | 01        | -         |
| 6   | Hyperplastic thyroid nodule           | 12        | 08        | 03        | 02        |
| 7   | Follicular neoplasm                   | 01        | 04        | 02        | 03        |
| 8   | Hurthle cell neoplasm                 | -         | 01        | 01        | -         |
| 9   | Papillary carcinoma                   | -         | 02        | 02        | 01        |
| 10  | Medullary carcinoma                   | -         | -         | 01        | 01        |
| 11  | Anaplastic carcinoma                  | -         | -         | -         | 01        |
| 12  | Lymphoma                              | 1         | 01        | -         | 01        |
|     | Total                                 | 33        | 68        | 34        | 25        |

Most of the aspiration content from lesions was hemorrhagic in 48.3% of cases, followed by blood mixed colloid in 41.2% of cases, and frank colloid in nature in 10.5% of cases. The provisional diagnosis on cytopathology was given out in the following Bethesda system: benign, atypia of undetermined significance, neoplasm, suspicious of malignancy, and malignant is shown in table -III:

Table -III: Diagnostic categorization of thyroid lesions (Bethesda system):

| Categorization                         | No of cases (n=160) | Percentage |
|----------------------------------------|---------------------|------------|
| Group A: Benign                        | 125                 | 78.125%    |
| Group B: Atypia with undetermined significance | 14                 | 8.75%      |
| Group C: Suspicious for neoplasm       | 08                  | 5%         |
| Group D: Suspicious for malignancy     | 05                  | 3.125%     |
| Group E: Malignant                    | 08                  | 5%         |
| Group F: Inadequate/non diagnostic     | -                   | -          |
| Total                                  | 160                 | 100%       |

USG of thyroid was done in all of the 160 cases. The maximum number of thyroid cases that were investigated on USG was diagnosed as solitary nodular goiter accounting for 40.5% of cases followed by nodular goiter with cystic changes (23.2%), thyroiditis 12.3%, hyperplastic thyroid nodule 10.2%, Follicular neoplasm 7.4% and nodular goiter with heterogeneity 6.4%. Conventional aspiration was done in 110 case and ultrasono-guided was done in 50 cases. Guided cytology was more accurate (92.85%) than conventional cytology (84.37%)-shown in table -IV. Table-IV: Comparison of diagnostic accuracy of conventional & guided cytology:

| Category of cytology followed by histopathology sample | No. of cases with accurate diagnosis. | %          | No. and % of false positive cases | No. and % of false negative cases |
|-------------------------------------------------------|---------------------------------------|------------|----------------------------------|----------------------------------|
| Conventional cytology                                 | 32                                    | 27         | 84.37%                           | 2 (6.25%)                        |
| Guided cytology                                       | 28                                    | 26         | 92.85%                           | 1 (3.57%)                        |

The results in Table 1 show that 93% of the thyroid FNACs were cytotologically benign and colloid goiter with or without cystic changes occupied 61.25%.
Under the malignant category, there were eleven cytologically diagnosed cases: five cases of papillary carcinoma, two cases of medullary carcinoma, one case is anaplastic carcinoma and a single case of lymphoma. Among 10 cases of follicular neoplasm, two cases were follicular carcinoma that was confirmed by biopsy and histopathology. Cytologically malignant neoplasm and suspicious lesion were 11 cases comprising 6.9% [Table 1]. We confirmed cytological diagnosis by histopathology. We received 60 biopsy cases which was diagnosed cytologically. Most lobectomy specimen were due to follicular neoplasm with suspected malignancy and all malignant neoplasm. Nodular goiter, which created pressure effects, and some granulomatous thyroiditis that involved the whole lobe, resected and send for histopathology. 100% malignant or suspicious lesion confirmed diagnose by biopsy. Some suspicious follicular neoplasm confirms malignancy (50%) and some are diagnosed as follicular adenoma (50%). The correlation of cytopathological and histopathological diagnosis is shown in the Table -V:

| SL | Diagnosis                                | Cytological diagnosis | Histopathological Diagnosis |
|----|-----------------------------------------|-----------------------|-----------------------------|
| 1  | Colloid goiter                          | 20                    | 18                          |
| 2  | Colloid goiter with cystic changes      | 18                    | 20                          |
| 3  | Granulomatosus thyroiditis              | 03                    | 04                          |
| 4  | Lymphocytic thyroiditis                 | 03                    | 02                          |
| 5  | Dequarvein thyroiditis                  | 01                    | 01                          |
| 6  | Follicular neoplasm                     | 10                    | 08                          |
|    | Follicular adenoma                      |                       | 02                          |
|    | Follicular carcinoma                    |                       |                             |
| 7  | Papillary carcinoma                     | 03                    | 03                          |
| 8  | Anaplastic carcinoma                    | 01                    | 01                          |
| 9  | Medullary carcinoma                     | 01                    | 01                          |
|    |                                         | 60                    | 60                          |

**Discussion**

FNAC is the best diagnostic method for early evaluation of solitary thyroid nodules and distinguishing between benign and malignant lesions. Most of the thyroid nodules are non-neoplastic which require no surgical intervention. Based on the FNAC findings, decreasing the rate of unnecessary surgery of benign lesion. In the present study, cytological features of thyroid lesions were reported according to Bethesda system and correlated with histopathology of available specimen, to determine its diagnostic accuracy. Thyroid nodules is more common in females and in this study there were 50 male and 110 female, with male and female ratio of 1:2.2 which is almost comparable to other studies conducted nationally and internationally. The majority of the
cases were presented in the age group 21–40 years with a mean age of 27.6 years. The repeat aspiration was done in 10 cases (6.25%) due to inadequate sample, excessive blood mixed sample, cystic lesions, suspected neoplastic lesions, or big diffuse lesions. The inadequacy of material was more due to blunt aspiration techniques. The number of needle passes was restricted up to 3 times. Repeat aspirations led to hemorrhagic smears as thyroid is a highly vascular organ. Jayaram and Orell et al.\textsuperscript{8} suggested that in thyroid lesion the average number of needle passes recommended for adequate sampling is two to five. As thyroid gland is a highly vascular organ, due to repeat aspiration the chances of hemorrhagic fluid rises each time, so it is advised to keep the number of aspiration as minimum as possible. Repeat aspiration was reported by Mondal et al.\textsuperscript{9} and Mandal et al.\textsuperscript{10} in 6.76% and 5.5% of their cases, respectively. So comparatively, repeat aspiration were little less in the present study. In the present study, 95% (n = 160) cytological smears were satisfactory for evaluation whereas 5% (n = 8) were inadequate for interpretation, which is comparable to most of the previous studies and closest to Sangalli et al.\textsuperscript{11} Most of the authors believe that adequate cellularity depends on whether the lesion is solid or cystic and whether aspirate was performed under palpation or ultrasound guided. All thyroid aspirated material should be well-preserved and well-prepared thyroid follicular epithelial cells for interpretation. If the aspirate sample contain only cyst fluid and RBCs are inadequate. The present study reported 5% aspirates as unsatisfactory smears which correlates with studies of Yassa et al.\textsuperscript{12} and Nayar and Ivanovic\textsuperscript{13} who reported 7% and 5% unsatisfactory smears in their studies, respectively. Usually, an ultrasound-guided FNAC is performed for small nodules and cytopathologist himself performs the procedure of FNAC, thereby ensuring the lower percentage of unsatisfactory smear.

In the present study, the specificity of thyroid cytology was 98.2% which is comparable to the other studies and closest to reported by Guhamallick et al.\textsuperscript{14} The accuracy of thyroid cytology reported by previous authors ranged from 64.6% to 99%. In the present study, the accuracy of thyroid cytology was 97.1% which is comparable to the other studies and closest to reported by Aravintan et al.\textsuperscript{15}. In this study patients are belonged to different category of life but solitary thyroid nodule was more common in low socioeconomic group simulating to the study organised by Mehmood Q and colleagues.\textsuperscript{16} Most of the patients complain mainly neck swelling (98%) which is similar to the study of Thompson and colleague\textsuperscript{17}. In our study the solitary nodule was observed mainly in right lobe of thyroid and similar finding is also found by TorreEM and colleagues\textsuperscript{18}.

In this study we found colloid goiter 32.5% and colloid cystic goiter 28.75% close to study of Gupta M et al\textsuperscript{19}. But in FNAC finding the malignant cases were 7.05 % but in the study of Gupta having malignant lesions 16%. Papillary carcinoma is the commonest malignancy of thyroid gland that found in Schlumberger study\textsuperscript{21}. In our study papillary carcinoma was diagnosed on FNAC in 3.3% cases which were confirmed by biopsy and it was the most common malignant tumour. One case of papillary carcinoma was misdiagnosed as Nodular goiter with cystic changes by FNA but histopathology confirmed the diagnosis as papillary carcinoma as presence of micro-papillae within the cystic cavity. On FNAC 10 cases (6.25%) were diagnosed as cellular follicular neoplasm and 4 of them suspicious for follicular carcinoma. But biopsy confirmed 2 malignant cases out of 4 suspicious malignant. This was the pitfalls of follicular carcinoma diagnosed by FNA as capsular or vascular invasion only confirm the diagnosis. Smears in which macrofollicular arrangement leads to misdiagnosis of probably suspicious nature similar observations made by Orell et al\textsuperscript{22}. In two cases of follicular adenoma FNAC did not show any conclusive evidence for definitive diagnosis and confirmed by histopathology examination. Two cases of suspicious diagnosis were later reported as nodular goiter by histopathology. However one should also keep in mind that the distinction between follicular adenoma and follicular carcinoma is extremely difficult even with experienced cytopathologist. Therefore extreme precaution should be taken to reach final conclusion. In our study Auto-immune thyroiditis found predominantly in female in 3rd and 4th decades similar to findings of Rajesh SP et al\textsuperscript{23}. Thyroiditis was successfully diagnosis by FNA and some of them are confirmed by biopsy. But most of them conform with thyroid hormonal study. But sometimes Hashimoto thyroiditis missed diagnose as lymphocytic thyroiditis and thyroid hormone level and antibodies guide the diagnosis of thyroiditis.

Anaplastic and medullary carcinoma was suspicious in FNA and it was confirmed by biopsy. The diagnosis of medullary carcinoma was made by presence of solid sheets of follicular cells and the presence of amyloid in the stroma. Presence of anaplastic
cells, multinucleated cells with large bizarre nuclei and very prominent nucleoli guided the diagnoses of anaplastic carcinoma and it was confirmed by biopsy. Only one NHL suspicious in FNA but could not confirm because biopsy was not available. From this study we find that FNAC is a very good tool for diagnosis of Thyroid related malignances however in certain suspicious cases mainly in follicular lesion a definite diagnosis may be only obtained by Histopathology.

**Table -VI: Comparison of sensitivity, specificity, and accuracy of thyroid fine-needle aspiration cytology reported in various studies**

| Author's name   | Year of publication | Number of cases | Sensitivity (%) | Specificity (%) | Accuracy (%) |
|-----------------|---------------------|-----------------|-----------------|-----------------|--------------|
| Sangalli et al  | 2006                | 5469            | 93.4            | 74.9            | 95.4         |
| Aravinthan et al| 2007                | 110             | 80.2            | 87.2            | 98.0         |
| Mandal et al    | 2011                | 120             | 90              | 84.6            | 97.0         |
| Bamanikar et al | 2014                | 300             | 65              | 98              | 94.2         |
| Present study   | 2018                | 160             | 90.2            | 98.2            | 97.1         |

Results of various case series reported by previous authors showed the sensitivity of thyroid cytology ranged widely from 50% to 97%. In the present study, sensitivity of thyroid cytology was 90.2%. On the other hand, specificity of thyroid cytology ranged from 74.9% to 100% in different case series. In the present study, the specificity of thyroid cytology was 98.2% which is comparable to the other studies and closest to reported by Sangalli et al. The accuracy of thyroid cytology reported by previous authors ranged from 64.6% to 99%. In the present study, the accuracy of thyroid cytology was 97.1% which is comparable to the other studies and closest to reported by Aravinthan et al.

**Conclusion.**

FNAC of thyroid lesions one of the most reliable method for screening of solitary thyroid nodule, but malignancy occasionally come as a surprise in postoperative histopathological examination. It is not the final diagnostic tool for thyroid lesion but guide the physicians for further treatment plan. It has certain limitations such as inability to distinguish follicular adenoma from follicular carcinoma, difficulties in detecting papillary micro-carcinoma. Therefore the malignant of thyroid nodules must not depend only on FNAC. Before surgical intervention, we should correlate the clinical findings with FNAC reports thereby reduced unnecessary surgical intervention and save the patient from thyroid hormonal deficiency.

**Conflict of Interest:**
All authors declare there is no conflict of interests regarding publication of this article.

**Authors' contribution:**
Data gathering and idea owner of this study: Karim M I, Nachev R, Fuklev N, Nargis N
Study design: Karim M I
Data gathering: Karim M I, Nachev R, Fuklev N, Nargis N
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