Original Article

Cytological Evaluation of Thyroid Lesions by Nuclear Morphology and Nuclear Morphometry

R. Yashaswini, T. N. Suresh, A. Sagayaraj[1]

Departments of Pathology and [1]ENT, Sri Devraj Urs Medical College SDUAHER, Kolar, Karnataka, India

Abstract

Introduction: Fine needle aspiration (FNA) of the thyroid gland is an effective diagnostic method. The Bethesda system for reporting thyroid cytopathology classifies them into six categories and gives implied risk for malignancy and management protocol in each category. Though the system gives specific criteria, diagnostic dilemma still exists. Using nuclear morphometry, we can quantify the number of parameters, such as those related to nuclear size and shape. The evaluation of nuclear morphometry is not well established in thyroid cytology. **Objective:** To classify thyroid lesions on fine needle aspiration cytology (FNAC) using Bethesda system and to evaluate the significance of nuclear parameters in improving the prediction of thyroid malignancy. **Materials and Methods:** In the present study, 120 FNAC cases of thyroid lesions with histological diagnosis were included. Computerized nuclear morphometry was done on 81 cases which had confirmed cytohistological correlation, using Aperio computer software. One hundred nuclei from each case were outlined and eight nuclear parameters were analyzed. **Results:** In the present study, thyroid lesions were common in female with M: F ratio of 1:5 and most commonly in 40–60 yrs. Under Bethesda system, 73 (60.83%) were category II; 14 (11.6%) were category III, 3 (2.5%) were category IV, 8 (6.6%) were category V, and 22 (18.3%) were category VI, which were malignant on histopathological correlation. Sensitivity, specificity, and diagnostic accuracy of Bethesda reporting system are 62.5, 84.38, and 74.16%, respectively. Minimal nuclear diameter, maximal nuclear diameter, nuclear perimeter, and nuclear area were higher in malignant group compared to nonneoplastic and benign group. **Conclusion:** The Bethesda system is a useful standardized system of reporting thyroid cytopathology. It gives implied risk of malignancy. Nuclear morphometry by computerized image analysis can be utilized as an additional diagnostic tool.

Keywords: Bethesda system, computer nuclear morphometry, fine needle aspiration

Introduction

Thyroid disorders, both benign and malignant, occur in men and women of all ages and are more common in females. The prevalence of a palpable thyroid nodule is about 12.2%.[1] The cause for increase in incidence and prevalence of thyroid cancer is uncertain. Thus detection of this condition through tests such as fine needle aspiration cytology (FNAC) may decrease the morbidity and mortality. FNAC technique is minimally invasive, simple, produces a speedy result, and is cost-effective. It has been able to categorize many benign and malignant lesions, and thereby guide therapeutic protocols.[2]

To address terminology related to thyroid FNAC, the National Cancer Institute first developed Bethesda system in 2007. This reporting system for thyroid FNAC will facilitate effective communication among pathologists, surgeons, radiologists, and endocrinologists.[3,4] However, it is still difficult to establish precise diagnosis of thyroid follicular lesions by cytology. Differentiating benign thyroid adenoma from malignant follicular neoplasm on cytology was identified as a major contributor to the high false-negative results. This concern is specifically for intermediate category where the rate of malignancy is reported at 40%.[5-7] Further evaluation of these lesions is necessary to improve the diagnostic accuracy.

Computerized nuclear morphometry is a cost-effective, objective, and reproducible tool for evaluation of nuclear features. It is a scientific tool to evaluate cellular changes and it can enhance the interpretation of morphological features

Address for correspondence: Dr. T. N. Suresh,
Department of Pathology, Sri Devraj Urs Medical College SDUAHER,
Kolar - 563 101, Karnataka, India.
E-mail: sureshstn@rediffmail.com

How to cite this article: Yashaswini R, Suresh TN, Sagayaraj A. Cytological evaluation of thyroid lesions by nuclear morphology and nuclear morphometry. J Cytol 2017;34:197-202.

by the transformation of pathological changes in cells to a quantitative form. Using nuclear morphometry, a number of parameters, such as those related to nuclear size and shape, can be quantified. Evaluation of these parameters has facilitated the diagnosis and management of different neoplasm in other systems such as breast and skin.[8-10]

The evaluation of nuclear morphometry is not well established in thyroid cytology. The objective of this study is to evaluate the significance of nuclear features in cytological evaluation of thyroid lesions.

Materials and Methods

The study comprised 120 patients who presented clinically with thyroid swelling during January 2010 to May 2015. Patients who underwent both FNAC and surgery were included in the study to facilitate cytohistological correlation. Adequate FNAC sample having minimum of six groups of well-visualized follicular cluster with at least 10 cells/group is considered for the study. All the cytological cases were classified according to Bethesda system of reporting thyroid cytology.

For computerized nuclear morphometry cytology, slides stained with hematoxylin and eosin (H and E) were used and PAP smear was taken. For analysis, 81 out of 120 study cases that had 100% histocytological correlation and minimum of 100 nuclei for nuclear morphometry were included. Study group includes 54 cases of nonneoplastic group, 8 cases of benign neoplastic group, and 19 cases of malignant group.

Computerized nuclear morphometry was done by using photos captured under Olympus CX-41 research microscope, an average of 10 microscopic fields; at magnification ×400 were captured for each case. At least 100 nuclei were analyzed per case. Cells in uniform sheets with no overlapping cells, with intact whole nuclei from the actual lesion, and with nuclear characteristics are considered. These cells are outlined using the Sketch command by the computer mouse in the Aperio-Image analyzer software and four parameters were measured – area, perimeter, minimum nuclear diameter, and maximum nuclear diameter. These parameters were saved in the Excel sheet and were later used to calculate the other four parameters: mean axis ratio (MAR), mean nuclear compactness (MNC), mean shape factor (MSHF), and mean nuclear size (MNS) [$= 2 \times (MNA/\pi) \times 0.5$]. Measurements were calibrated in terms of micrometer, using a NOW micrometer slide before performing measurements in the software. Person doing nuclear morphometry is blinded for histopathology diagnosis. Institute ethical clearance was obtained for the study.

Statistics

Descriptive and inferential statistical analysis has been carried out in the present study. Analysis of variance has been used to find the significance of study parameters between three or more groups of patients. *Post hoc* Tukey test has been used to find the pairwise significance.

Results

In the present study on 120 thyroid FNAC cases, the occurrence of thyroid lesions was more in the age group of 41–50 years (41.6%). The sex distribution of thyroid aspirates showed more female predominance with male to female ratio of 1:5. Thyroid FNAC lesions were categorized according to Bethesda system.

Category II was the most common lesion constituting up to 60%, followed by category VI which was 18%. Among category II, colloid/nodular goiter is the predominant diagnosis constituting about 69%, followed by lymphocytic thyroiditis in 27% cases. Of 22 cases of papillary carcinoma, 12 cases (55%) are arranged in papillary pattern and 14 cases had cellular swirls pattern having about 50–200 tumor cells concentrically arranged, with ovoid nuclei, the long axes of which were oriented perpendicular to the radius of the swirl; 15 cases (65%) showed transpolar nuclear grooves; 13 cases (59%) showed intranuclear cytoplasmic inclusion and nuclear pleomorphism with mild anisonucleosis and dispersed chromatin. Multinucleated giant cells were seen in four cases and were larger with diverse shape, dense cytoplasm, and more nuclei compared to those seen in colloid goiter [Figure 1a–c].

On histology, among 65 cases of nonneoplastic lesion, nodular goiter was predominant in 37 cases (56.9%), followed by Hashimoto's thyroiditis in 22 cases (33.8%) and nodular hyperplasia in 6 cases (9.2%). Out of 56 neoplastic cases, 24 were of follicular adenoma (42.8%), one case each of hyalinizing trabecular adenoma and follicular carcinoma, 29 cases were of papillary carcinoma (53.5%). Among the variants of papillary carcinoma, conventional type ($n = 22$) was the most common, followed by micropapillary variant ($n = 4$), follicular variant ($n = 2$), and tall cell variant ($n = 1$).

Among 73 cases in category II, 54 cases correlated with the histopathological diagnosis with diagnostic accuracy of 73.97%. In category III, seven of 14 cases correlated with histopathological diagnosis with 50% diagnostic accuracy. In category IV, among three cases, two on histopathological examination (HPE) showed follicular adenoma. In category V, four cases on HPE were diagnosed as papillary carcinoma. In category VI, 19 cases correlated with the HPE diagnosis of papillary carcinoma thyroid with diagnostic accuracy of 86.36% [Table 1].

Of 75 cases in benign lesion category II, 56 belonged to nonneoplastic lesion on HPE. Among these, 14 cases on HPE turned out to be follicular adenoma and 10 cases had
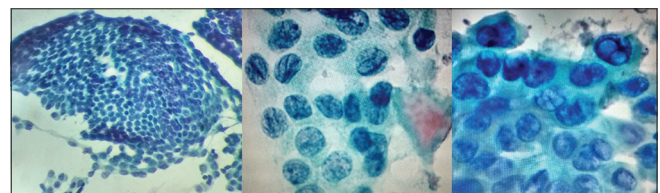
Figure 1: Cellular swirl (Pap stain ×200), nuclear groves, and intranuclear inclusion Pap stain (×400)

admixture of follicular adenoma along with colloid goiter in surrounding tissue. Cytology of these cases showed macrofollicular arrangement along with thin colloid. One case of follicular carcinoma on cytology showed cells arranged in microfollicular and macrofollicular pattern with moderate nuclear pleomorphism and moderate colloid with background of polymorphous population of lymphocytes, impinging onto follicular cells. Of four cases diagnosed as papillary carcinoma, three were micro papillary carcinoma type and none of them showed nuclear features of papillary carcinoma in cytology and other case was conventional papillary carcinoma with surrounding colloid goiter. The misdiagnosis in these 19 cases was due to aspiration from nonrepresentative areas and this can be avoided by ultrasound-guided aspiration.

Out of 14 cases in category III, two cases turned out to be follicular variant of papillary carcinoma on HPE, and these cases on cytology showed macrofollicular and microfollicular arrangement with occasional nuclear grooves.

Among three cases in category IV, one case turned out to be lymphocytic thyroiditis on HPE, which on cytology showed follicular cells in clusters and microfollicular arrangement with lymphocytic background.

Among eight cases in category V, papillary carcinoma was confirmed in four cases. In view of high cellularity, nuclear

groove and occasional nuclear inclusion suspicious of malignancy was given in another four cases.

Out of the 22 cases of papillary carcinoma on cytology, two turned out to be nodular goiter and one as lymphocytic thyroiditis. These three cases on cytology showed moderate cellularity and cells arranged in clusters and ill-defined papillae. At focal areas, these cells showed intranuclear cytoplasmic inclusions and nuclear grooves which had led to the cytological diagnosis of papillary carcinoma.

On cytohistopathological correlation, sensitivity, specificity, positive predictive value, negative predictive value, and accuracy were 62.5, 84.38, 77.78, 72, and 74.16%, respectively. The low sensitivity is mainly due to aspiration from nonrepresentative areas. Use of ultrasound guidance will improve sensitivity of thyroid FNAC.

Among groups when nuclear parameters were compared, mean minimal and maximal diameter are higher in malignant group compared to that of nonneoplastic group [Table 2]. The standard deviation (SD) of mean minimal and maximal diameter of both nonneoplastic and malignant category was higher compared to that of benign neoplastic lesion, thereby supporting the presence of monomorphism of follicular cells in benign category as compared to nonneoplastic and malignant category. The

Table 1: Correlation of cytologic diagnosis with histology and incidence of malignancy in each bethesda category

Cytology diagnosis	Number (%)	HPE diagnosis	Frequency (%)	Incidence of malignancy (%)
Category II	73 (60.83%)	Nonneoplastic	54 (73.97)	6.7
		Benign neoplasm	14 (19.1)	
		Malignant	5 (6.7)	
Category III	14 (11.6%)	Nonneoplastic	5 (35.6)	14.2
		Benign neoplasm	7 (50)	
		Malignant	2 (14.2)	
Category IV	3 (2.5%)	Nonneoplastic	1 (33.3)	NIL
		Benign neoplasm	2 (66.6)	
		Malignant	Nil	
Category V	8 (6.6%)	Nonneoplastic	2 (25)	50
		Benign neoplasm	2 (25)	
		Malignant	4 (50%)	
Category VI	22 (18.3%)	Nonneoplastic	3 (9.09%)	86.36
		Benign neoplasm	Nil	
		Malignant	19 (86.36%)	

Table 2: Nuclear morphometric parameters in thyroid lesion

Variables	Nonneoplastic group (μm) ± SD	Benign neoplastic group (μm) ± SD	Malignant Group (μm) ± SD
Mean maximal nuclear diameter	6.90±0.24	7.37±0.12	8.67±0.76
Mean minimal nuclear diameter	6.76±0.23	7.23±0.10	8.50±0.76
Mean nuclear perimeter	23.90±0.31	24.62±0.20	27.04±1.32
Mean nuclear area	81.73±5.11	84.07±2.61	114.26±18.99
Mean axis ratio	0.97±0.01	0.98±0.01	0.98±0.00
Mean nuclear compactness	6.66±0.36	7.48±0.28	8.96±0.69
Mean shape factor	1.87±0.08	1.75±0.07	1.58±0.18
Mean nuclear size	9.84±0.33	10.11±0.16	12.00±1.07

mean nuclear perimeter and area are higher in malignant group compared to that in nonneoplastic and benign neoplastic group. The SD of mean nuclear area among nonneoplastic, benign, and malignant groups were 5.11, 2.61, and 18.99, respectively. The higher mean nuclear area in malignant group suggests the enlarged nuclear size, which is a feature of malignant lesions [Figures 2 and 3].

Nuclear diameter, perimeter, and area showed statistically significant difference when compared between the groups [Table 3]. Among calculated parameters, mean axis ratio did not show any statistical significance between groups, all other remaining parameters had good statistical correlation. Nuclear diameter, perimeter, and area showed statistically significant difference when compared between the benign and malignant groups. Among calculated parameters, mean nuclear size was statistically significant between benign and malignant groups.

To evaluate the use of nuclear morphometry in improving diagnostic accuracy of thyroid cytology, the nuclear parameters were applied in cytohistological disparity cases. Nine cases were considered here out of 30 cases, because in other cases the aspirates were from nonrepresentative areas and applying morphometry on these lesions does not give any valuable information.

Among nine cases, two cases which were diagnosed as nonneoplastic lesion under Bethesda category III turned out to be papillary carcinoma in histopathology. The nuclear

parameters in these two false-negative cases are closer to the nuclear values observed in neoplastic group. Among nine cases, seven cases which were diagnosed as suspicious for malignancy and malignant lesion under Bethesda category V and VI turned out to be colloid goiter and lymphocytic thyroiditis in histopathology. The mean maximal nuclear diameter ranged from 7.29 to 7.39 μm, and mean minimal nuclear diameter ranged from 7.08 to 7.24 μm. The mean nuclear perimeter ranged from 23.97 to 24.52 μm. Mean nuclear area ranged from 82.80 to 86.12 μm^2. The nuclear parameters in seven false-positive cases were closer to the nuclear values of nonneoplastic group.

Nuclear morphometry was useful in improving the diagnostic accuracy of thyroid cytology in nine cases.

Discussion

FNAC is a safe, simple, and inexpensive technique that plays an important role in the diagnosis of thyroid lesions. The present study was conducted to know its accuracy in the diagnosis of thyroid neoplasm.

Several studies showed predominant number of cases in category II, V and VI.[11-14] These finding were correlated with the present study. Percentage of malignancy rates in different Bethesda category is also comparable to other studies.[15-17]
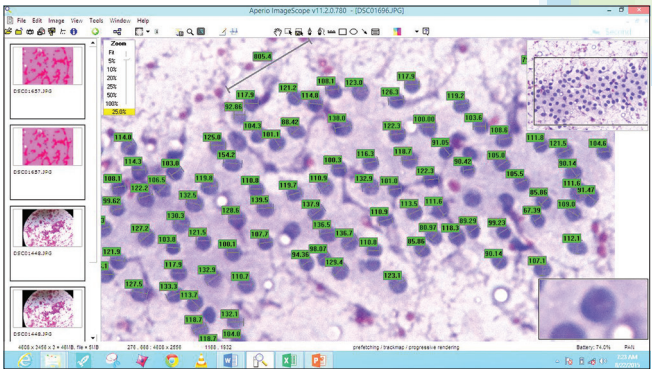

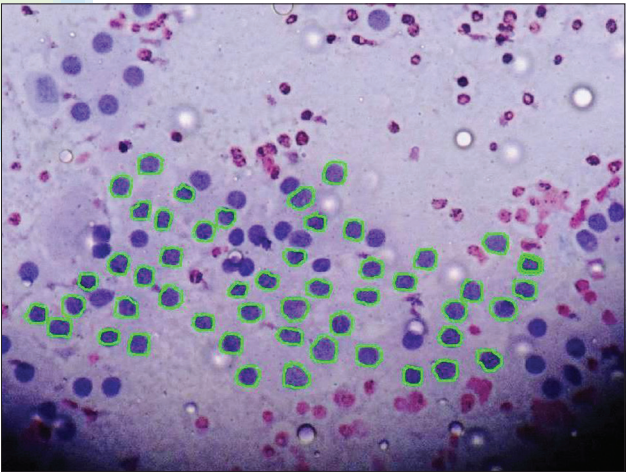**Figure 2:** Image analysis of nuclear morphometry – nuclear diameter (×400)

Figure 3: Image analysis of nuclear perimeter and area (×400)

Table 3: Pairwise comparison between nonneoplastic, benign, and malignant groups

Variables	Nonneoplastic	Malignant group	P	Benign neoplastic	Malignant group	P
Mean maximal nuclear diameter, μm	6.90±0.24	8.67±0.76	<0.001**	7.37±0.12	8.67±0.76	<0.001**
Mean minimal nuclear diameter, μm	6.76±0.23	8.50±0.76	<0.001**	7.23±0.10	8.50±0.76	<0.001**
Mean nuclear perimeter, μm	23.84±0.31	27.04±1.32	<0.001**	24.62±0.20	27.04±1.32	<0.001**
Mean nuclear area, μm^2	81.73±5.11	114.26±18.99	<0.001**	84.07±2.61	114.26±18.99	<0.001**
Mean axis ratio	0.97±0.01	0.98±0.00	0.989	0.98±0.01	0.98±0.00	0.676
Mean nuclear compactness	6.66±0.36	8.96±0.69	<0.001**	7.48±0.28	8.96±0.69	0.007
Mean shape factor	1.87±0.08	1.58±0.18	<0.001**	1.75±0.07	1.58±0.18	0.001
Mean nuclear size	9.84±0.33	12.00±1.07	<0.001**	10.11±0.16	12.00±1.07	<0.001**

**Statistically significant

The sensitivity and specificity of present study is comparable with other studies.[18,19] The low specificity rate of thyroid FNAC is due to overlap of nuclear features among the thyroid lesions. One study reported nuclear groove in papillary thyroid carcinoma (PTC) (38%), follicular neoplasm (10%), medullary carcinoma (16%), nodular goiter (22.5%), and Hashimoto's thyroiditis (14%).[20] Inclusion of new cytological features, such as cellular swirls, may improve the specificity. The sensitivity of FNAC can be improved by adapting ultrasound-guided FNAC, thereby aspirating the representative sample.[21,22]

Computerized morphometry is an objective computer image analysis to estimate the chosen parameters in every individual cell. The present study along with other study showed that minimum and maximal nuclear diameter, nuclear area, and nuclear perimeter were significantly different across benign and malignant groups, with malignant group having higher values. Karslioglu and other studies observed similar findings.[23-27]

In a study by Rajesh *et al.*, H and E stained 40 FNAC aspirates were considered and 100 cells per case was captured at $400 \times$ for morphometry and the measured values in pixels were converted to micrometer with value of 1 pixel = 0.446 micrometer.[28] They found that convex nuclear area and perimeter of follicular hyperplasia were much lower than that of follicular neoplasm and follicular variant of papillary carcinoma. However, these parameters overlap between benign neoplastic lesion – follicular adenoma and papillary carcinoma.

Dina *et al.* studied five cytology cases of tall cell variant of papillary carcinoma and 14 cases of conventional papillary carcinoma and found that nuclear area and nuclear diameter was higher in tall cell variant compared to conventional PTC which were similar to other studies.[29,30] These findings correlated with vascular invasion, which was the poor prognostic factor for tumor metastasis.

Role of nuclear morphometry in distinguishing follicular adenoma vs follicular carcinoma and diagnosing cases of follicular variant of papillary carcinoma thyroid where the nuclear features of papillary carcinoma is not overt is not well established. Multicentric study with larger number of follicular carcinoma and follicular variant of papillary carcinoma thyroid is required to establish the role of nuclear morphometry in these cytologically challenged cases.

Nuclear morphometry is helpful in cases which are of diagnostic challenge on cytology due to the overlapping cytological features. On applying morphometry in nine such cases in the present study, it was found that two false-negative cases on cytology had nuclear parameters similar to malignant group. When morphometry applied on seven false positive cases, morphometric nuclear parameter are comparable with morphometric parameters of nonneoplastic group. Whenever there is dilemma in diagnosing the lesion on cytology to various categories under Bethesda system, an adjunct of nuclear morphometry along with cytological features should be used.

Conclusion

The Bethesda system of reporting thyroid cytology is the standardized system, and implementing it in routine practice helps in clinical management of thyroid swelling as it quantifies the risk of malignancy in each category. Nuclear morphometry is helpful in cases which are of diagnostic challenge on cytology due to the overlapping cytological features.
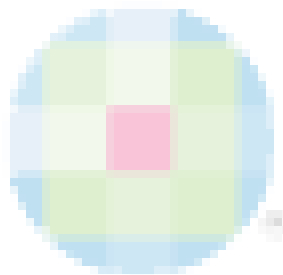
Financial support and sponsorship
Nil.

Conflicts of interest
There are no conflicts of interest.

References

1. Davies L, Welch HG. Increasing incidence of thyroid cancer. JAMA 2006;295:2164-7.
2. Handa U, Garg S, Mohan H, Nagarkar N. Role of fine needle aspiration cytology in diagnosis and management of thyroid lesions: A study on 434 patients. J Cytol 2008;25:13-7.
3. Cibas ES, Ali SZ. The Bethesda system of reporting thyroid cytopathology Am J Clin Pathol 2009;132:658-65. Doi: 10.1309/AJCPPHLWMI3JV4LA.
4. Cibas ES, Ali SZ. The Bethesda system for reporting thyroid cytopathology. Am J Clin Pathol 2009;132:658-65.
5. Mc Coy KL, Jabbour N, Ogilvie JB, Ohori NP, Carty SE, Yim JH. The incidence of cancer and rate of false negative cytology in thyroid nodules greater than or equal to 4cm in size. Surgery 2007;142:837-44.
6. Ogilvie JB, Piatigorsky EJ, Clark OH. Current status of FNA from thyroid nodule. Adv Surg 2006;40:223-38.
7. Banks ND, Kowalski J, Tsai HL, Somervell H, Tufano R, Dackiw AP, *et al*. A diagnostic predictor model for intermediate or suspicious thyroid FNA samples. Thyroid 2008;18:933-41.
8. Lira M, Schenka AA, Magna LA, Cotta AC, Cintra M, Souza EM, *et al*. Diagnostic value of combining immunostaining for CD3 and nuclear morphometry in mycosis fungoides. J Clin Pathol 2008;61:209-12.
9. Cui Y, Koop EA, Van Diest PJ, Kandel RA, Rohan TE. Nuclear morphometric features in benign breast tissue and risk of subsequent breast cancer. Breast Cancer Res Treat 2007;104:103-7.
10. Kapur V, Antic T, Venkataraman G, Durazo AR, Quat MM, Flanigan RC, *et al*. Validation of world health organisation/international society of urologic pathology 2004 classification scheme for bladder urothelial carcinomas using quantitative nuclear morphometry: Identification of predictive features using bootstrap method. Urology 2007;70:1028-33.
11. Yang J, Schnadig V, Logrono R, Wassermman PG. Fine needle aspiration of thyroid nodules: A study of 4073 patients with histologic and clinical correlations. Cancer 2007;5:306-15.
12. Yassa L, Cibas ES, Benson CB, Frates MC, Gawande AA, Larsen PR, *et al*. Long-term assessment of a multidisciplinary approach to thyroid nodule diagnostic evaluation. Cancer 2007;111:508-16.
13. Nayar R, Ivanovic M. The indeterminate thyroid fine-needle aspiration: Experience from an academic center using terminology similar to that proposed in the 2007 National Cancer Institute Thyroid Fine Needle Aspiration State of the Science Conference. Cancer 2009;117:195-202.
14. Melo-Uribe MA, Sanabria A, Perez G, Vargas EJ, Abaunza MC, Gutierrez V, *et al*. The Bethesda system for reporting thyroid cytopathology in Colombia: Correlation with histopathological diagnoses in oncology and non-oncology institutions. J Cytol 2015;32:12-6.
15. Her-Juing Wu H, Rose C, Elsheikh TM. The Bethesda system for reporting thyroid cytopathology: An experience of 1,382 cases in a community practice setting with the implication for risk of neoplasm and risk of malignancy. Diagn Cytopathol 2012;40:399-403.
16. Jo VY, Stelow EB, Dustin SM, Hanley KZ. Malignancy risk for fine-needle aspiration of thyroid lesions according to the Bethesda system for reporting thyroid cytopathology. Am J Clin Pathol 2010;134:450-6.

17. Lee K, Jung C K, Lee K Y, Bae JS, Lim DJ, Jung SL. Application of Bethesda system for reporting thyroid aspiration cytology. The Korean J Pathol 2010;44:521-7.

18. Mamoon N, Jamy R, Khan AH. Evaluation of fine needle aspiration cytology as a screening tool in thyroid lesions. J Pak Med Assoc 2013;63:1120-3.

19. Gupta M, Guptha S, Gupta VB. Correlation of fine needle aspiration cytology with histopathology in the diagnosis of solitary thyroid nodule. J Thyroid Res 2010;379051:1-8.

20. Deligeorgi-Politi H. Nuclear crease as a cytodiagnostic feature of papillary thyroid carcinoma in fine-needle aspiration biopsies. Diagn Cytopathol 1987;3:307-10.

21. Das DK. Intranuclear cytoplasmic inclusions in fine-needle aspiration smears of papillary thyroid carcinoma: A study of its morphological forms, association with nuclear grooves, and mode of formation. Diagn Cytopathol 2005;32:264-8.

22. Chan JK, Saw D. The grooved nucleolus. A useful diagnostic criterion of papillary carcinoma of thyroid. Am J Surg Pathol 1986;10:672-9.

23. Słowińska-Klencka D, Klencki M, Sporny S, Lewiński A. Karyometric analysis in the cytologic diagnosis of thyroid lesions. Anal Quant Cytol Histol 1997;19:507-13.

24. Karslioglu Y, Celasun B, Gunhan O. Contribution of morphometry in the differential diagnosis of fine-needle thyroid aspirates. Cytometry B Clin Cytom 2005;65:22-8.

25. Priya SS, Sundaram S. Morphology to morphometry in cytological evaluation of thyroid lesions. J Cytol 2011;28:98-102.

26. Sassi I, Mangili F, Sironi M, Freschi M, Cantaboni A. Morphometric evaluation of fine needle biopsy of single thyroid nodules. Pathol Res Pract 1989;185:722-5.

27. Nagashima T, Suzuki M, Oshida M, Hashimoto H, Yagata H, Shishikura T, *et al*. Morphometry in the cytological evaluation of thyroid follicular lesions. Cancer 1998;84:115-8.

28. Rajesh L, Saha M, Radhika S, Das Radotra B, Rajwanshi A. Morphometric image analysis of follicular lesions of the thyroid. Anal Quant Cytol Histol 2004;26:117-20.

29. Dina R, Capitanio A, Damiani S. A morphometric analysis of cytological features of tall cell variant and classical papillary carcinoma of the thyroid. Cytopathology 2000;11:124-8.

30. Asioli S, Righi A, Volante M, Chiusa L, Lloyd RV, Bussolati G. Cell size as a prognostic factor in oncocytic poorly differentiated carcinomas of the thyroid. Hum Pathol 2014;45:1489-95.