Section: Pathology

Original Research Article

A STUDY OF CD56 EXPRESSION IN PAPILLARY CARCINOMA THYROID AND ITS FOLLICULAR VARIANT IN COMPARISON WITH OTHER SOLITARY FOLLICULAR PATTERNED LESIONS OF THE THYROID

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Abstract

Background: Follicular variant of papillary carcinoma thyroid has varying differential diagnoses. We analysed the role of CD56 immunostaining in cases of papillary carcinoma and solitary follicle patterned nodules and established CD56 as a tool to differentiate follicular variant of papillary carcinoma from other solitary follicle predominant mimics. **Materials and Methods:** A retrospective 2-year study of 74 thyroid lesions was carried out in a tertiary care hospital including all cases of papillary carcinoma and solitary follicular patterned lesions. Slides and blocks retrieved from archives were reviewed. Immunostaining with CD56 was performed on selected representative sections. A positive result was defined as moderate- strong membranous immunostaining in ≥ 10% of epithelial cells. **Result:** The 74 lesions studied were- hyperplastic nodule (n=14), follicular adenoma (n=11), hurthle cell adenoma (n=1), follicular carcinoma (n=8), hurthle cell carcinoma (n=4), papillary carcinoma (n=31), follicular variant of papillary carcinoma (n=4) and non-invasive encapsulated follicular variant of papillary carcinoma (n=1). There was statistically significant difference in CD56 staining between follicular variant of papillary carcinoma and its mimics (p value 0.015) and between papillary carcinoma and benign lesions (p value 0.028). **Conclusion:** CD56 proves to be a reliable negative marker of papillary carcinoma and its variants which can be used to differentiate follicular variant of papillary carcinoma from other follicle predominant lesions in ambiguous cases.

INTRODUCTION

Papillary thyroid carcinoma (PTC) comprises majority (80%) of all thyroid neoplasms.[1] Follicular variant of PTC (FVPTC) accounts for 9% to 22.5% of all PTC cases.[2] Diagnosis of FVPTC is often a challenging task as it has various mimickers; both non-neoplastic and neoplastic. Precise diagnosis is imperative to determine the prognosis, management and treatment of patients. [3,4] The mimickers of FVPTC include all solitary follicle patterned nodules such as follicular adenoma (FA), follicular carcinoma (FC), hurthle cell adenoma (HA), hurthle cell carcinoma (HC) and hyperplastic nodules (HN). [5] Immunohistochemistry (IHC) plays a cardinal role in diagnosing thyroid neoplasms. Various immunohistochemical markers have been utilized to differentiate PTC from other thyroid lesions; namely CD56, HBME-1, claudin-1, galectin-3, CK19, TPO and Ki67.[4-11] CD56 is a cell adhesion molecule expressed in thyrocytes which influences the migration of tumor cells.[6] Although most studies have established CD56 as a useful negative marker for PTC,[3-6,10,11] Etem et al found an insignificant association between PTC and follicular neoplasms.[7] In our study, we aimed 1) to determine CD56 expression in PTC, FVPTC and its mimics 2) to analyze the expression of CD56 in differentiating FVPTC from other solitary follicle patterned thyroid lesions 3) To determine the difference of CD56 staining in cases of PTC and FVPTC.

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MATERIALS AND METHODS

This study was a retrospective 2-year study conducted in a tertiary care hospital in Mangalore after obtaining institutional ethics committee clearance. All cases of total, near total and hemithyroidectomies diagnosed as HN, FA, FC, HA, HC, PTC and its variants FVPTC and non-invasive follicular thyroid neoplasm with papillary like nuclear features (NIFTP) were included in the study. All thyroid lesions, non-neoplastic and neoplastic other than those mentioned in the inclusion criteria were excluded from the study

Data was retrieved from the laboratory information system. The cases were blinded and the hematoxylin and eosin (H&E) slides were reviewed by 2 observers.

Diagnosis was based on morphological criteria as defined in literature. FA was diagnosed if the circumscribed nodule was composed of follicles with no evidence of invasion and did not have nuclear features as defined for NIFTP.[12] HA were similar nodules with 75% follicles lined by oncocytic cells with abundant granular eosinophilic cytoplasm.[6]

The criteria used for diagnosis of FC/HC was unequivocal capsular or vascular infiltration. Capsule invasion was defined as malignant cells insinuating through complete thickness of the capsule and the resultant tumor nests seen outside the capsule while being continuous with the main tumor. Vessel infiltration was defined as a malignant embolus in the capsular vessels, attached to the vessel walls, covered by endothelium, applicable for vessels in or beyond the capsule only.[13]

The diagnosis of PTC was made based on the criteria proposed by Chan et al. The major features included ovoid, crowded nuclei with loss of polarization, clear or pale chromatin and presence of psammoma bodies. The minor features included abortive papillae, irregular or elongated follicles, dark colloid, nuclear pseudoinclusions and intraluminal multinucleated histiocytes in the follicles. Fulfilment of 4 major or 3 major and 4 minor criteria qualified for a diagnosis of PTC.[6]

FVPTC was diagnosed when a lesion was predominantly composed of follicles, with papillae comprising less than 1% of the architecture and demonstrated characteristic nuclear features as described for PTC.[14]

NIFTP was diagnosed if a circumscribed nodule, composed of less than 1% true papillae and less than 30% solid growth did not have capsular, vascular or intrathyroid invasion on extensive sectioning. These lesions did not have necrosis, atypical or increased mitosis or psammoma bodies. At least 2 of 3 abnormal nuclear features were seen- size/shape (nucleomegaly, elongation, overlapping), membrane irregularity or grooving or chromatin characteristics (clearing, margination, glassy appearance).[12]

A representative section was selected for IHC staining based on well preserved morphology and good fixation. Additional sections of areas with ambiguous morphology suspicious for malignancy were also stained with CD56. Sections were cut (5 microns) from the corresponding paraffin block onto poly-l-lysine slides, incubated at 37oC overnight, deparaffinized and rehydrated. IHC was conducted according to immunoperoxidase technique. Antigen retrieval was done in a microwave with citrate buffer (ph-6). Power block was applied followed by primary (CD56; DAKO), secondary antibodies, di-amino-benzidine (DAB) staining and hematoxylin counterstain was carried out. Sections from a duodenal neuroendocrine tumor were used as positive controls. The overlying normal duodenal epithelium served as negative control.

The IHC slides were analyzed with the corresponding H&E slides using a quantitative score. A positive result was considered when moderate to strong immunostaining with CD56 (membranous with or without the cytoplasmic staining) was seen in ≥ 10% of the follicular epithelial cells, while <10% was considered negative.[8]

Statistical package for social sciences- IBM SPSS statistics for windows, version 25.0 Armonk NY: IBM corp was used for statistical analysis. For comparison across groups, Chi-square test was used. A p value less than 0.05 was considered statistically significant. The significance of CD56 in differentiating FVPTC from other solitary follicle patterned lesions was determined.

RESULTS

A total number of 74 lesions were studied. The lesions were categorized into non-neoplastic (n=14; 19%), benign (n=12; 16%) and malignant (n=48; 65%) lesions as summarized in [Table 1].

Demographic details: The mean age of presentation, male to female ratio (M: F) and mean nodule size are listed in [Table 2].

Histopathological findings

Non-neoplastic lesions: HN were seen in 14/74 cases. Histopathologically the lesions showed a mixed pattern of macro and microfollicles, nuclear anisonucleosis surrounded by a thin partial capsule without compression of the surrounding parenchyma. Of these, four were associated with Hashimoto's thyroiditis and one with lymphocytic thyroiditis. One case showed focal areas with optically clear nuclei raising a suspicion of NIFTP. However, there was no evidence of nuclear grooving or overlapping falling short of the strict diagnostic criteria of NIFTP [12]. CD56 showed strong to moderate complete membranous positivity in this lesion as seen in [Figure 1] (CD56, 200x).

Benign lesions: Benign lesions including FA(n=11) and HA (n=1) comprised 12/74 cases studies. Grossly these lesions were firm, well encapsulated, solitary nodules compressing the surrounding thyroid tissue. Few of the lesions showed secondary changes-hemorrhage, and cystic change. Histopathologically,

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the cases of FA were composed predominantly of microfollicles with occasional macrofollicles, uniform appearing nuclei, surrounded by a thick complete capsule. HA showed cells with prominent eosinophilic granular cytoplasm arranged in microfollicles and solid sheets. Four cases of follicular adenoma were associated with Hashimoto's thyroiditis.

Malignant lesions: The malignant lesions studied were PTC(n=31) and follicle patterned malignant lesions FC(n=8), HC(n=4), FVPTC(n=4) and NIFTP(n=1) and amounted to 48/74 cases.

Cases of PTC grossly showed solid lesions which were white, granular and infiltrative. Few showed cystic change and satellite nodules. Histologically the lesions showed papillary architecture with hierarchical branching and nuclear features of overlapping, crowding, clearing, inclusions and grooving. Included in this category was one case of papillary microcarcinoma. The cases of FVPTC showed grey white solid lesions grossly; two were diffusely infiltrative while one showed near total encapsulation. Microscopically these lesions were composed of follicles with classical nuclear features of papillary carcinoma as seen in [Figure 2] (H&E, 200x). The case with the capsule showed a focus suspicious of invasion. The cells comprising this lesion were negative for CD56 highlighting a breach in the capsule in contrast to the strongly positive benign thyroid follicular cells adjacent to the lesion.

The NIFTP lesions were solid grey white with a prominent complete capsule. The microscopy showed follicles with nuclear features of papillary carcinoma all limited within the capsule. The diagnosis of these lesions was made based on the criteria defined by Johnson et al. [12] [Figure 3] (H&E,400x) shows a case of NIFTP with follicular cells exhibiting nuclear clearing, grooving, nucleomegaly and powdery chromatin.

Four cases of PTC, one of NIFTP and all of FVPTC were associated with Hashimoto's thyroiditis. Two cases of PTC were associated with lymphocytic thyroiditis.

The cases of follicular carcinoma showed gross features ranging from solitary partially encapsulated to infiltrative grey white solid lesions. Microscopically lesions were composed predominantly of tumor cells arranged in microfollicles along with few macrofollicles and solid areas. Based on capsular breach and angio-invasion, 4 cases each of widely invasive and minimally invasive FC and 2 cases each of widely invasive and minimally invasive HC were studied.[13] One case each of FC and HC were associated with Hashimoto's thyroiditis.

Although all cases presented clinically as solitary thyroid nodules, 3 cases had multiple types of lesions; HN in one lobe and PTC in the other lobe, FC in one lobe with FVPTC in the other lobe, papillary microcarcinoma in one lobe and minimally invasive FC in the other lobe.

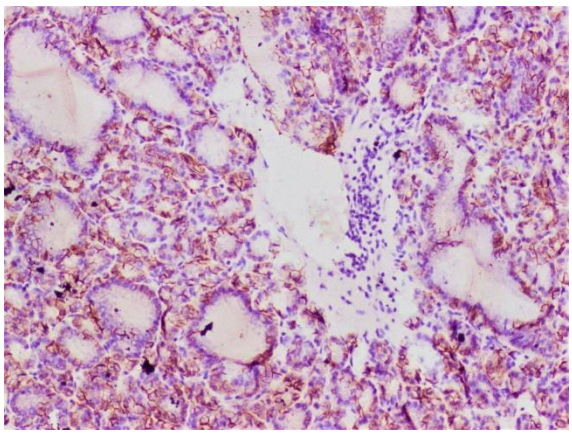

Figure 1: (200x, CD56) Strong to moderate complete membranous positivity in HN

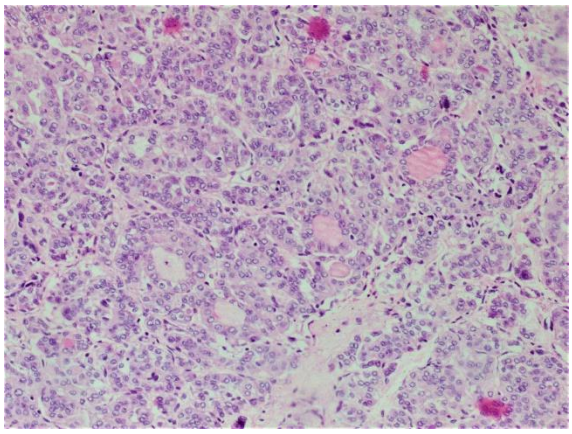

Figure 2: (200x, H&E) Follicular architecture in a case of FV-PTC

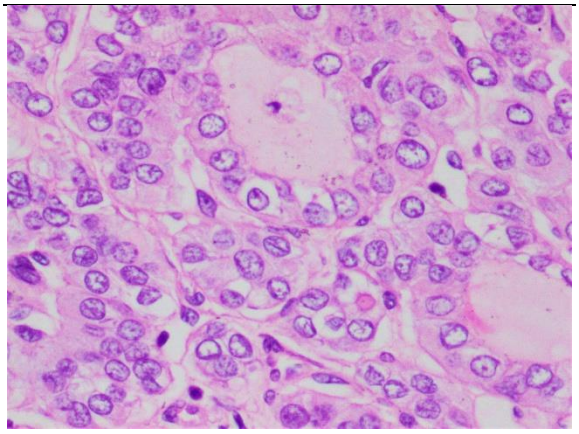

Figure 3: (H&E,400x) NIFTP with follicular cells exhibiting nuclear clearing, grooving, enlargement and powdery chromatin

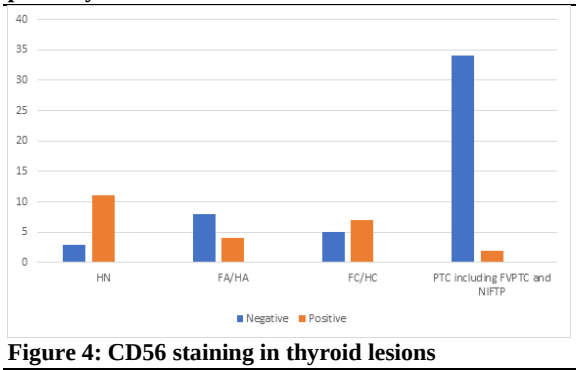

Figure 4: CD56 staining in thyroid lesions

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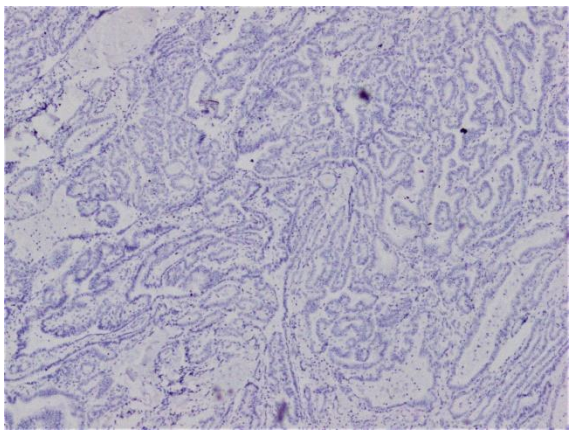

Figure 5: (CD56, 100x) PTC negative for CD56

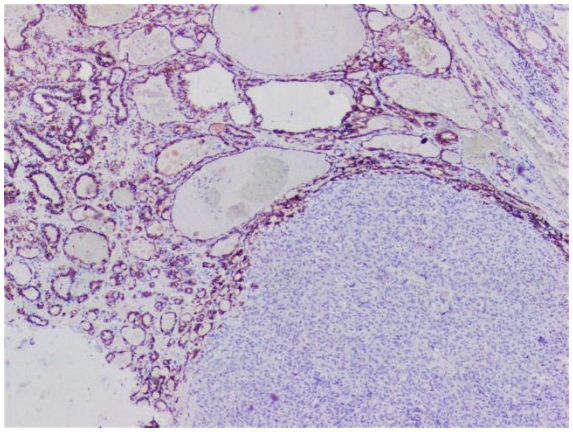

Figure 6: (CD56, 100x) Infiltrative FVPTC. CD56 negative tumor cells surrounded by CD56 positive normal thyroid follicles

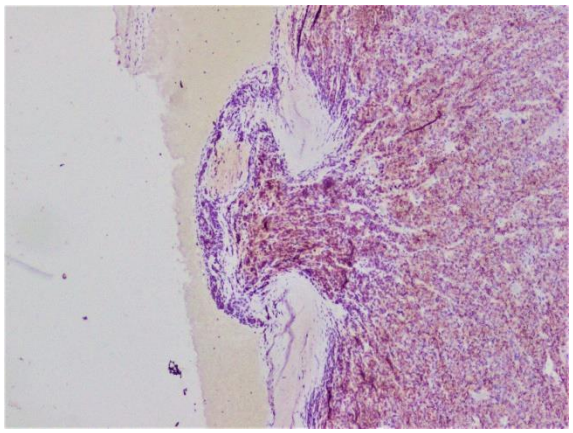

Figure 7 (CD56, 100x)]-CD 56 positive tumor cells mushrooming through the capsule in a case of FC

[Table 3] summarizes the finding of thyroiditis in the background of the various lesions studied.

Immunohistochemistry: Based on the cut off of 10%, the lesions were negative (<10% staining) for CD56 expression in 50 cases (62%) and positive in (≥10% staining) in 24 cases (38%).

One of the observations noted while evaluating the CD56 staining was the variable staining within the lesions and in the normal tissue. Normal thyroid tissue is CD56 positive.[11] However, variably stained areas in the thyroid showed strongly positive to weak to negative staining. It was observed that most of the involutional cells were negative to weakly positive and the CD56 staining was strong positive in the hyperplastic cells.

Few of the solitary lesions also showed intralesional variable staining. While troubleshooting this dilemma we reviewed the slides again, took better preserved and fixed sections, in relation to the capsule if present. The IHC was done by a different lab technician which gave more reliable results.

[Table 4 and Figure 4] summarize the findings of CD56 IHC staining seen in the thyroid lesions.

Majority of the cases of PTC (93.5%), FVPTC (100%) and NIFTP (100%) showed loss of CD56 expression in the follicular cells in comparison to non-neoplastic (HN) (21%), benign (FA, HA) (66%) and other malignant categories (FC, HC) (41%). Figure 5 shows a case of PTC negative for CD56 (CD56, 100x)

[Table 5] demonstrates the ability of CD56 in differentiating PTC from benign tumors, PTC from non-malignant lesions, PTC from FC and FVPTC from other solitary follicular patterned lesions with significant p values. Table 6 and 7 shows the results of the present study are in concordance with those with other studies in literature.

Few cases warrant further mention. One case was diagnosed as NIFTP on routine reporting. It showed strong to moderate complete membranous positivity as seen in figure 1. However, the 2 observers participating in this blinded study noted that there were no definitive nuclear features of PTC. The lesion showed a follicular pattern with varying sized follicles, anisonucleosis and a thin capsule. Artefactual nuclear changes gave a false impression of NIFTP. This lesion was a HN.

The second case was a NIFTP on morphology alone. On serial sectioning of the paraffin block during the study, the tumor cells which were negative for CD56 were seen breaching the capsule of the lesion which was surrounded by CD56 positive normal thyroid follicles as seen in figure 6 (CD56, 100x).

This focus was not apparent in the initial H&E sections. The lesion had a follicular pattern with nuclear features of PTC and was therefore reclassified as infiltrative FVPTC. This case highlights the need for extensive sampling good amount of tissue and taking deeper sections to evaluate the lesion thoroughly. CD56 staining shows a stark contrast between malignant and non-neoplastic cells aiding in identification of capsular and vascular invasion.

CD 56 was positive in majority of FC and HC cases. The mushrooming of the tumor cells through the capsule was much better appreciated in the CD56 stained slides than in the H&E sections [figure 7 (CD56, 100x)]. This feature could be used as an advantage by a Pathologist while battling between the diagnoses of adenoma versus carcinoma.

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Table 1: Categories of lesions studied

Lesion	Number of cases(n)	Percentage (%)
Non-neoplastic (HN)	14	19
Benign tumors		
FA	11	15
HA	1	1
Malignant tumors		
FC	8	11
HC	4	5.5
PTC	31	42
FVPTC	4	5.5
NIFTP	1	1
Total	74	100

Table 2: Demographic details of patients

Lesion parameters	Non-neoplastic lesions (HN)	Benign (FA, HCA)	Malignant (PTC and variants, FC , HC)
Mean Age(years)	34.21	36.33	40.10
M:F ratio	0:14	1:11	1:3.8
Mean nodule size(cm)	2.36	2.67	2.51

Table 3: Lesions associated with thyroiditis

Lesion	Number of cases associated with Hashimoto's thyroiditis	Number of cases associated with lymphocytic thyroiditis
Non-neoplastic		
HN	4	1
Benign tumors		
FA	4	0
HA	0	0
Malignant tumors		
FC	1	0
HC	1	0
PTC	4	2
FVPTC	3	0
NIFTP	1	0

Table 4: Immunohistochemical findings of CD56 staining in thyroid lesions

Lesion (Total number of cases n)	Negative (<10% staining) Number of cases (percentage)	Positive (≥10% staining) Number of cases (percentage)
Non-neoplastic HN(n=14)	3(21.4)	11(78.6)
Benign tumors FA(n=11)	8(72.7)	3(27.3)
HA(n=1)	0	1(100)
Malignant tumors FC(n=8)	3(37.5)	5(62.5)
HC(n=4)	2(50)	2(50)
PTC(n=31)	29(93.5)	2(6.5)
FV-PTC(n=4)	4(100)	0
NIFTP(n=1)	1(100)	0

Tables 5: p value of CD56 in differentiating various thyroid lesions.

Lesions	Negative	Positive	Total	p value
PTC and variants	34	2	36	0.028
FA and HA (Benign tumors)	8	4	12	
PTC and variants	34	2	36	<0.0001
FA, HA, HN (non-malignant lesions)	11	15	26	
PTC and variants	34	2	36	<0.0001
FC and HC (other follicle patterned carcinomas)	5	7	12	
FVPTC including NIFTP	5	0	5	0.015
All other solitary follicular patterned lesions	16	22	38	
PTC	29	2	31	0.559
FVPTC including NIFTP	5	0	5	

Table 6: CD56 IHC staining as compared with other studies

Study(year)	Number of positive cases/ total number of cases (Percentage)		
	PTC, FVPTC, NIFTP	Follicular neoplasms	HN
Abdel Aziz et al,[10]	4/24 (16.6)	10/14(71.43)	6/7(85.71)
Abdel Raouf et al,[11]	5/32(15.6)	19/24(79.1)	10/10(100)
Muthuswamy et al,[18]	1/31(3.2)	17/25(68)	21/29(72.4)
Present study	2/36(5.5)	8/19(42)	11/14(78.57)

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Table 7: Comparison of IHC findings (p value) of present series with other studies

Lesions compared	Present Study (p value)	Other studies (p value)	Other studies (p value)
PTC and Benign lesions	<0.0001	< 0.001 Huang et al,[9]	< 0.001 Durmus SE et al,[15]
PTC and FC	<0.0001	0.006 Dusko et al,[19]	0.046 Golu I et al,[8]
FVPTC and other follicular patterned lesions	0.015	0.008 Golu I et al,[8]	-

Table 8: Cases of FA, PTC, NIFTP and FVPTC with <10% CD56 staining.

Lesion(number)	Complete loss of CD56 expression (n; %)	CD56 positive in 1-9% of cells (n; %)
FA(n=8)	2(25)	6(75)
PTC, NIFTP, FVPTC(n=34)	29(85)	5(15)

DISCUSSION

Thyroid neoplasms being the most prevalent neoplasms of the endocrine system comprise one percent of all malignancies.[15] According to the GLOBOCAN database, the age-standardized incidence rates of thyroid cancer were 10.1/100000 women and 3.1/100000 men. The age standardized mortality rates were 0.5/100000 in women and 0.3/100000 in men.[16] Analysis of 10 years of statistics in India also show an increase in incidence rate of thyroid carcinoma from 0.9 to 1.3 in males and from 2.4 to 3.9 in females, a relative increase of 48% and 62% respectively.[17]

PTC constitutes 80% of thyroid malignancies. It can be well circumscribed/encapsulated type or infiltrative type. Further, they are divided into different morphological variants namely; conventional, follicular, hobnail, columnar, trabecular, diffuse sclerosing, tall cell and papillary micro-carcinoma. The diagnosis of PTC is relatively easy to make when a lesion exhibits classical papillary pattern with characteristic nuclear features. However, in presence of a follicular pattern, differential diagnosis is manifold and inter-observer variation is high.

FVPTC is a subtype of PTC composed predominantly of follicles, with papillae comprising less than 1% of the architecture. The diagnosis is based on characteristic nuclear features. FVPTC can be divided into infiltrative FVPTC, invasive encapsulated FVPTC and NIFTP. NIFTP is of special interest as it is often mistaken for benign follicular patterned solitary lesions of the thyroid. It follows an indolent course as compared to the infiltrative FVPTC, warranting less aggressive management. Prognosis of NIFTP is excellent as shown by a study by Nikiforov et al; Patients followed up for 10-26 years showed no recurrences whereas 12% of patients with infiltrative FVPTC had an adverse event within a follow up period of 1-18 years.[14]

With newer terminologies being introduced into the World Health Organization (WHO) classification, making an accurate diagnosis based on morphology alone is challenging. Many IHC markers have been evaluated to differentiate PTC and its variants from their mimickers including CD56, HBME-1, claudin-1, galectin-3, CK19, TPO and Ki67. [3-11,15,18] We have studied the utility of CD56 as an adjunct to morphology to help differentiate FVPTC from all other follicular patterned lesions.

CD56 is a neural cell adhesion molecule (NCAM), a surface glycoprotein encoded by a gene on chromosome 11. It multitasks as a signal receptor impacting cell adhesion, migration, proliferation, apoptosis, differentiation and survival. CD56 is expressed in the brain, natural killer (NK) cells, large granular lymphocytes, neuroendocrine cells and thyroid epithelial cells.[18]

CD56 expression has been studied in various malignancies namely medulloblastomas, astrocytomas, malignant NK/T- cell lymphomas, neuroendocrine carcinomas, leukemias, melanomas and various carcinomas. Overexpression of this marker in these lesions is correlated with poor prognosis, resistance to chemotherapy, decreased survival and metastatic potential.[19]

However, in case of thyroid carcinomas, loss of CD56 expression has been correlated with metastasis and adverse prognosis.[6,9,10,15,18] Studies have shown down-regulation or loss of expression of CD56 in PTC, FC and anaplastic carcinoma.[9] It is established that loss of this cell adhesion molecule affects lymphangiogenesis by reducing expression of vascular endothelial growth factor- D.[18] This discrepancy in the molecular workings of CD56 in the thyroid, the association of its loss with worse prognosis warrants further studies to explain opposing pathogenesis of CD56 expression in thyroid malignancies in contrast to other tumours.[19]

Majority of PTC lesions (94%) including its variants FVPTC and NIFTP were negative for CD56 whereas 58% of the non-PTC lesions and 78.5% of non-neoplastic lesions were positive for CD56. All NIFTP and FVPTC lesions showed loss of CD56 staining. This is in concordance with studies published previously. [9,15,20] CD56 staining between conventional PTC, FVPTC and NIFTP was not statistically significant. CD56 is a reliable negative marker for these lesions as supported by many other studies. [3-11]

A study conducted by Abdel Aziz et al showed significant difference of CD56 expression between PTC (including FVPTC) and non-neoplastic and follicular neoplasms (p value 0.00),

PTC versus follicular neoplasms (p value 0.001) and PTC versus HN (p value 0.001).[10]

The present study established the utility of CD56 to differentiate PTC from other malignancies, benign lesions and non-neoplastic conditions and to

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differentiate FVPTC and NIFTP from other follicular patterned lesions with significant p values. In ambiguous follicular patterned lesions with unequivocal nuclear features, CD56 can serve as a valuable adjunct to accurately diagnose the lesion.

Few observations of our study warrant specific mention. Majority of FA (72.7%) cases showed <10% CD56 expression in contrast to previous studies. Technical factors like fixation timings and fixatives, brand of IHC antigen used, antigen retrieval methods may contribute to this finding. However, heterogeneity of CD56 expression in different cells of the same thyroid lesion as described by Jung Soo Pyo et al may explain this result better.[21] There is need for additional cumulative studies addressing this phenomenon as there are numerous practical difficulties in submitting the entire thyroid tissue to compensate for this varied expression. In a study by Etem et al 35% of follicular tumours showed loss of CD56 staining and the authors concluded that CD56 is not a reliable marker in differentiation of PTC from FN. [7] However, clubbing FA and FC cases we did find a significant difference in expression from PTC in our study. [Table 8] compares the cases of FA and PTC (including NIFTP and FVPTC) which showed <10% CD56. Most cases of carcinoma showed complete loss of CD56 expression in contrast to majority of FA cases which retained at least minimal staining. A factor also to be kept in mind is the smaller number of FA cases than carcinoma cases which is a limitation of this study. Studies with comparable number of non-neoplastic, benign and malignant thyroid lesions will improve the yield of diagnostic information.

Intralesional variation in staining and prominence of staining in hyperplastic cells compared to involutional cells posed an interpretation dilemma. As explained above, technical issues such as varied fixation timings and types of fixatives, brands of IHC antigen, methods of antigen retrieval and heterogenous expression of CD56 in thyroid follicular cells are possible explanations apart from the fact that more chromogen is more visually obvious in the larger hyperplastic cells than smaller involutional cells. Our recommendation is to always look at the hyperplastic cells for evaluation of CD56 stain; either as an internal positive control adjacent to the papillary neoplasms or for assessment of expression in non-neoplastic conditions. We recommend assessing 5 high power fields with the strongest staining and calculating the number of positive cells as a percentage to evaluate the lesion.

The limitations of this study are lesser number of non-neoplastic and benign lesions compared to carcinoma, limited number of FVPTC and NIFTP cases and non-availability of various histological variants of PTC for CD56 analysis.

Additional cumulative studies with larger case numbers on CD56 heterogenous expression in thyroid lesions is warranted for more conclusive information. Molecular studies to determine the pathogenesis and relation between loss of CD56 and worse prognosis with metastatic potential is an interesting concept to research.

In limited specimens like fine needle aspirates and core biopsies from thyroid a large panel of immunocytochemical/IHC markers are not feasible. In such conditions usage of a single marker like CD56 may help the pathologist classify ambiguous lesions.

CONCLUSION

CD56 is a reliable negative marker for PTC and its variants FVPTC and NIFTP. In case of an ambiguous solitary follicular patterned lesion, CD56 can be used as a valuable adjunct to morphology for accurate diagnosis and appropriate management of the patient.

REFERENCES

1. Siegel RL, Miller KD, Jemal A. Cancer statistics, 2017. CA: a cancer journal for clinicians. 2017 1; 67(1):7-30.
2. Sebastian SO, Gonzalez JM, Paricio PP, Perez JS, Flores DP, Madrona AP et al. Papillary thyroid carcinoma: prognostic index for survival including the histological variety. Arch Surg. 2000; 135:272–277.
3. Rahman MM, Banu SG, Barua AR, Kamal M, Baqui MN, Parveen S. Follicular Variant of Papillary Thyroid Carcinoma on the Basis of Histopathological and Immunohistochemical Diagnosis. Bangladesh Medical Research Council Bulletin. 2016; 42(1):21-7.
4. Ozolins A, Narbuts Z, Strumfa I, Volanska G, Stepanovs K, Gardovskis J. Immunohistochemical expression of HBME-1, E-cadherin, and CD56 in the differential diagnosis of thyroid nodules. Medicina. 2012 Oct; 48(10):74.
5. El Atti RM, Shash LS. Potential diagnostic utility of CD56 and claudin-1 in papillary thyroid carcinoma and solitary follicular thyroid nodules. Journal of the Egyptian National Cancer Institute. 2012 Dec 1; 24(4):175-84.
6. El Demellawy D, Nasr A, Alowami S. Application of CD56, P63 and CK19 immunohistochemistry in the diagnosis of papillary carcinoma of the thyroid. Diagn Pathol 2008; 3:5.
7. Etem H, Özekinci S, Mizrak B, ŞEnTüRK S. The Role of CD56, HBME-1, and p63 in Follicular Neoplasms of the Thyroid. Turk J Pathol. 2010 Sep 1;26(3):238-42.
8. Golu I, Vlad MM, Dema A, Moleriu LC, Tudor A, Iacob M et al. The absence of CD56 expression can differentiate papillary thyroid carcinoma from other thyroid lesions. Indian Journal of Pathology and Microbiology. 2017 Apr 1;60(2):161.
9. Huang L, Wang X, Huang X, Gui H, Li Y, Chen Q Et al. Diagnostic significance of CK19, galectin-3, CD56, TPO and Ki67 expression and BRAF mutation in papillary thyroid carcinoma. Oncology letters. 2018 Apr 1;15(4):4269-77.
10. Abdel-Aziz A, Abdallah D. Role of Immunohistochemistry in Diagnosis of Papillary Thyroid Carcinoma: The Use of Ck19, CD56, P63 and CD117. Journal of Cancer and Tumor International. 2019 Mar 25:1-1.
11. Abdel Raouf SM, Atia H, Soliman SA. Diagnostic Utility of Immunohistochemical Markers Trop2 and CD56 in Differentiating Follicular Derived Thyroid Lesions. Prensa Med Argent. 2020; 106:6.
12. Johnson DN, Furtado LV, Long BC, Zhen CJ, Wurst M, Mujacic I, Kadri S, Segal JP, Antic T, Cipriani NA. Noninvasive follicular thyroid neoplasms with papillary-like nuclear features are genetically and biologically similar to adenomatous nodules and distinct from papillary thyroid carcinomas with extensive follicular growth. Archives of pathology & laboratory medicine. 2018 Jul;142(7):838-50.
13. Fletcher CD, Chan JK, Fletcher C. Tumors of the thyroid and parathyroid glands. Diagnostic Histopathology of Tumors. 5th ed. Philadelphia, PA: Elsevier. 2021.

ISSN (O): 2687-5365; ISSN (P): 2753-6556

14. Nikiforov YE, Seethala RR, Tallini G, Baloch ZW, Basolo F, Thompson LD et al. Nomenclature Revision for Encapsulated Follicular Variant of Papillary Thyroid Carcinoma: A Paradigm Shift to Reduce Overtreatment of Indolent Tumors. JAMA Oncol. 2016 Aug 1;2(8):1023-9.
15. Erdogan-Durmus S, Ozcan D, Yarikkaya E, Kurt A, Arslan A. CD56, HBME-1 and cytokeratin 19 expressions in papillary thyroid carcinoma and nodular thyroid lesions. Journal of research in medical sciences: the official journal of Isfahan University of Medical Sciences. 2016;21.
16. Pizzato M, Li M, Vignat J, Laversanne M, Singh D, La Vecchia C, Vaccarella S. The epidemiological landscape of thyroid cancer worldwide: GLOBOCAN estimates for incidence and mortality rates in 2020. The Lancet Diabetes & Endocrinology. 2022 Apr 1;10(4):264-72.
17. Sekkath Veedu J, Wang K, Lei F, Chen Q, Huang B, Mathew A. Trends in thyroid cancer incidence in India. J Clin Oncol [Internet]. 2018 May 20;36(15_suppl): e18095–e18095.
18. Muthusamy S, Shah SA, Suhaimi SN, Kassim N, Mahasin M, Saleh MF et al. CD56 expression in benign and malignant thyroid lesions. The Malaysian journal of pathology. 2018 Aug 1;40(2):111-9.
19. Gattenlöhner S, Stühmer T, Leich E, Reinhard M, Etschmann B, Völker HU, Rosenwald A, Serfling E, Bargou RC, Ertl G, Einsele H. Specific detection of CD56 (NCAM) isoforms for the identification of aggressive malignant neoplasms with progressive development. The American journal of pathology. 2009 Apr 1;174(4):1160-71.
20. Dunđerović D, Lipkovski JM, Boričic I, Soldatović I, Božic V, Cvejić D, Tatić S. Defining the value of CD56, CK19, Galectin 3 and HBME-1 in diagnosis of follicular cell derived lesions of thyroid with systematic review of literature. Diagnostic pathology. 2015 Dec;10(1):196.
21. Pyo JS, Kim DH, Yang J. Diagnostic value of CD56 immunohistochemistry in thyroid lesions. The International journal of biological markers. 2018 May;33(2):161-7.

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