Role of CK-19, HBME-1 and Galectin-3 in Prediction of Prognosis of Papillary Thyroid Carcinoma -An Experience from a Tertiary Care Centre

Dr. Samir Ranjan Bhowmik, Dr. Keya Basu, Dr. Subhrajyoti Karmakar, Dr. Ananya Biswas, Dr. Moumita Sengupta, Dr. Sriranjan Mukherjee, Dr. Sujoy Ghosh[1]

[1] Institute of Postgraduate Medical Education and Research, Kolkata

Received: 13 December 2018 Accepted: 2 January 2019 Published: 15 January 2019

Abstract

Introduction: Papillary thyroid carcinoma (PTC) is the most common malignant neoplasm of thyroid follicular epithelium. Its incidence has increased dramatically in past few decades reaching upto 80

Index terms— papillary thyroid carcinoma (PTC), total tumor diameter (TTD), cytokeratin 19, prognostic markers.

1 Introduction

apillary thyroid carcinoma (PTC) is the most common malignant neoplasm of thyroid follicular epithelium. [1] Its incidence has increased dramatically in past few decades reaching up to 80% of malignant thyroid tumors. [2,3,4] Despite the propensity for lymphovascular invasion, majority of the patients with this tumor, if properly treated, have an excellent long-term prognosis. [5] But appropriate treatment primarily depends on the ability of the pathologist to make an accurate diagnosis. Historically, diagnosis of PTC was based on the presence of the papillary architectures but currently it also includes the presence of nuclear features i.e. nuclear overlapping, optical clearing, macronucleoli, irregular contours with pseudoinclusion and grooving. [6] Identification of these features remains difficult and controversial when they are present focally, thus distinction of PTC from other thyroid lesions i.e. benign papillary hyperplasia, some forms of thyroiditis, variants of PTC may not be possible. In this regard, implication of immunohistochemical (IHC) markers are very useful. In this study, we have used a panel of three IHC markers -Cytokertain 19 (CK-19), Hector Battifora Mesothelial-1 (HBME-1) and Galectin-3 (GAL-3) to show their role as prognostic markers of PTC.

2 II.

3 Materials and Methods

We have conducted a single institution based, retrospective study at a tertiary care centre of Eastern India. Patients with histologically confirmed papillary thyroid carcinoma treated between January 2017 and June 2018 were identified from the Department of General Surgery. All patients had undergone total thyroidectomy for the primary tumor. A total of 41 patients were reviewed for their clinical and pathological data. We have considered gender, age, total tumor diameter (TTD), capsular invasion and lymphnodal metastasis as the parameters to be studied. We have correlated the expressions of CK 19, GAL-3 and HBME-1 with those parameters to show their role in prognostication of PTC (weak and strong expression both considered as positive expression).

CK19 is a low molecular weight cytokeratin which presents widely in simple epithelia and basal cell layers of stratified epithelium.

HBME-1 is a monoclonal antibody generated against a membrane antigen of mesothelial cells. [7] It was originally found in malignant mesothelioma; several investigators showed that HBME-1 play an important role in diagnosis of papillary thyroid carcinoma.

GAL-3 is a member of oligosaccharide selective binding protein family known as lectins which plays an important role in the cell growth, apoptosis, cell-matrix interactions, neoplastic transformation and metastasis; and now it's been considered to be an effective indicator that can be available to distinguish the malignant thyroid nodules from the benign ones. [8] The aim of present study was to investigate the relationship between the expression of CK19, HBME-1 and GAL-3 and the aggressive behavior of PTCs by correlating immunohistochemical results with the clinical features.

4 a) Statistical Analysis

The Kruskal-Wallis test was performed for comparisons between multiple groups. The ? 2 test was analysed for categorical evaluation. Correlations were evaluated using Spearman's rank correlation. p-value < 0.05 was considered as significant. Statistical software (GRAPHPAD PRISM 5) was used for analysis.

5 III.

6 Results

In Table **??** we have shown the summary of the clinicopathological traits. Among 41 cases, the female: male ratio was 19.5:1 and 14.63% cases were more than 45 years old. 80.48% cases had total tumour diameter more than 1cm. capsular invasion and lymphovascular invasion were found to be present in 26.82% and 31.70% cases respectively.

In Table 2-4 we have summarized the results showing correlation of the IHC markers with five prognostic factors. All the cases with PTC were divided into positive and negative expression groups.

According to our results in Table 2, positive expression of CK19 was correlated significantly with the total tumor diameter ($p < 0.001$). This finding indicated that the larger volume of the total tumor diameter is more likely to express CK19. Positive expression of CK19 was also correlated significantly with the capsular invasion ($p<0.007$) which denotes that CK19 positivity stands for more aggressive behavior of PTC. On the other hand, expression of CK19 had no significant relationship with age, sex and lymph nodal metastasis.

In Table 3, we have shown the correlation of expression of Galectin 3 with the prognostic parameters. Similarly expression of GAL-3 was correlated significantly with total tumor diameter ($p<0.005$) and capsular invasion ($p<0.007$) in all the PTC cases, whereas its expression was found to be not related to patient's age, sex and lymph nodal metastasis.

In Table 4, correlation between the expression of HBME-1 and the prognostic parameters is shown. Expression of HBME-1 was found to be significantly correlated with capsular invasion ($p<0.01$) and lymph nodal metastasis ($p<0.002$). IV.

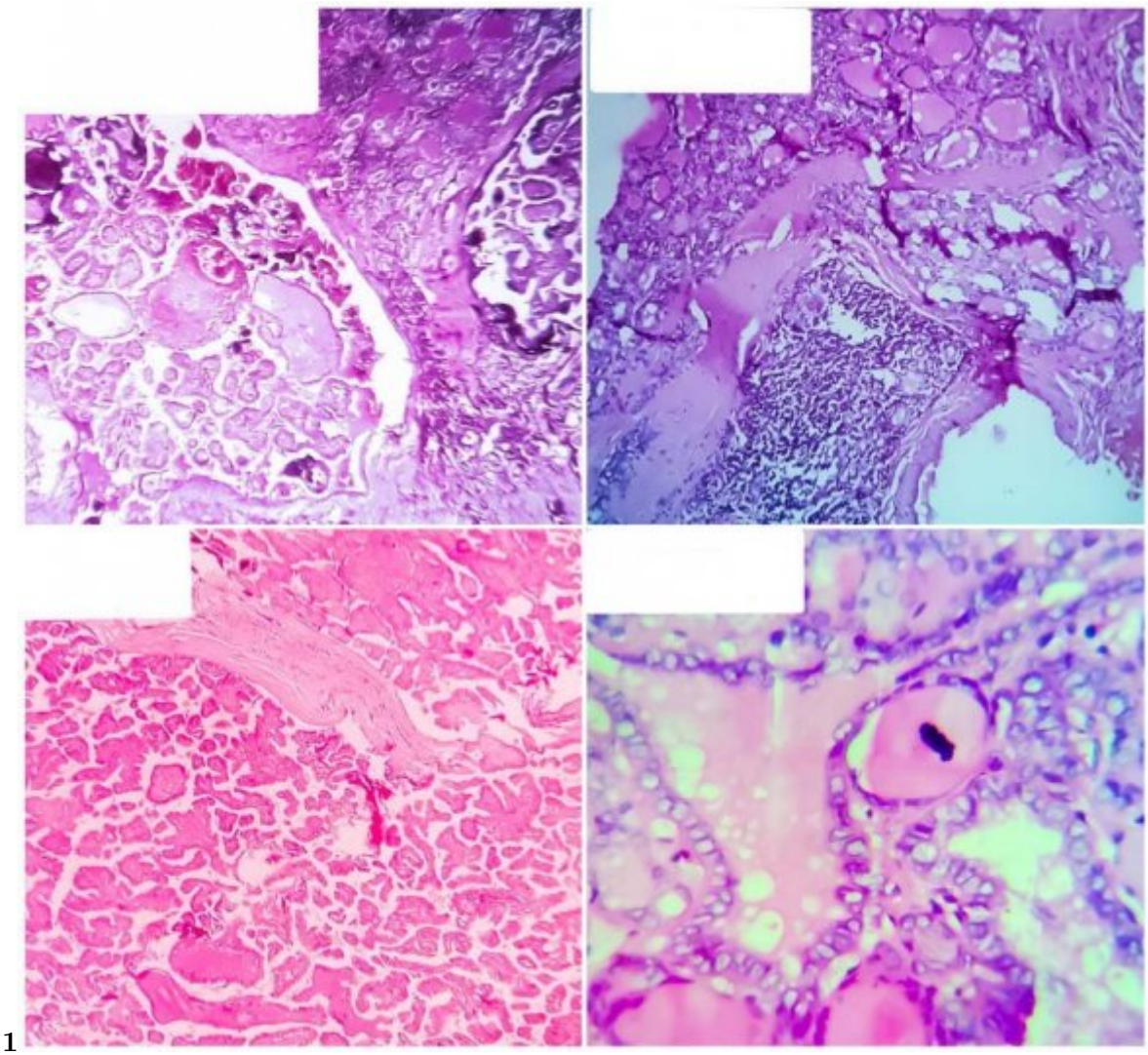
7 Table 1: Summary of clinicopathological traits

8 Discussion

Primary thyroid cancers comprise the largest group among malignancies of the endocrine system. 120,000 new cases are added each year. Thyroid carcinomas usually present in the 40-60 age group. Environmental, genetic and hormonal factors have been considered in the etiology of thyroid carcinomas. Many benign conditions like thyroidal adenomas, multinodular goiter, thyroiditis, thyroidal cysts, thyroidal malformations and focal granulomatous diseases occur clinically as solitary nodules and malignancy is found in 0.1-0.2% of these conditions. [9] Cytokeratin-19 (CK-19) expression in thyroid nodules is in general intense and diffuse in papillary carcinoma and heterogeneous labeling in follicular carcinoma and in follicular adenoma, with nil or low expression in other benign lesions. [10,11] Galectins, especially galectin-3, are suggested to play a role in the pathogenesis of well differentiated thyroid carcinoma, particularly in papillary carcinoma. [12] Therefore, it is one of the markers most commonly used to assist in distinguishing thyroid lesions. Hector Battifora mesothelial-1 (HBME-1) has been demonstrated to be important as a thyroid marker of follicular origin, with greater affinity to malignant lesions when compared to benign lesions. [13] In general, the prognosis of PTC is favorable and ten-year survival rate for PTCs is greater than 90%. [14] However, about 20% of the differentiated thyroid cancer will present with metastasis. So accurate biomarkers which can predict the aggressive behavior of thyroid carcinoma is critical for clinical management. [15] Tijana et al reported that the CK19 was a useful marker for the identification of PTCs and they suggested that the high expression of the CK19 is a predictor for the aggressive behavior of PTC and could help to identify a particular subgroup of PTCs which had a potentially worse prognosis. [16] GAL-3 could be a important tool for guiding therapeutic decisions in patients with thyroid nodules. [17] The significance of the biomarkers, such as CK19, HBME-1, GAL3, have been widely explored and debated for the differential diagnosis of thyroid neoplasms but the value of these biomarkers as prognostic factors for PTCs is not clear. [16] Thus, in our study, we attempted to investigate whether the expression of the CK19,HBME-1 or GAL3 is linked to the aggressive behavior in papillary thyroid carcinoma.

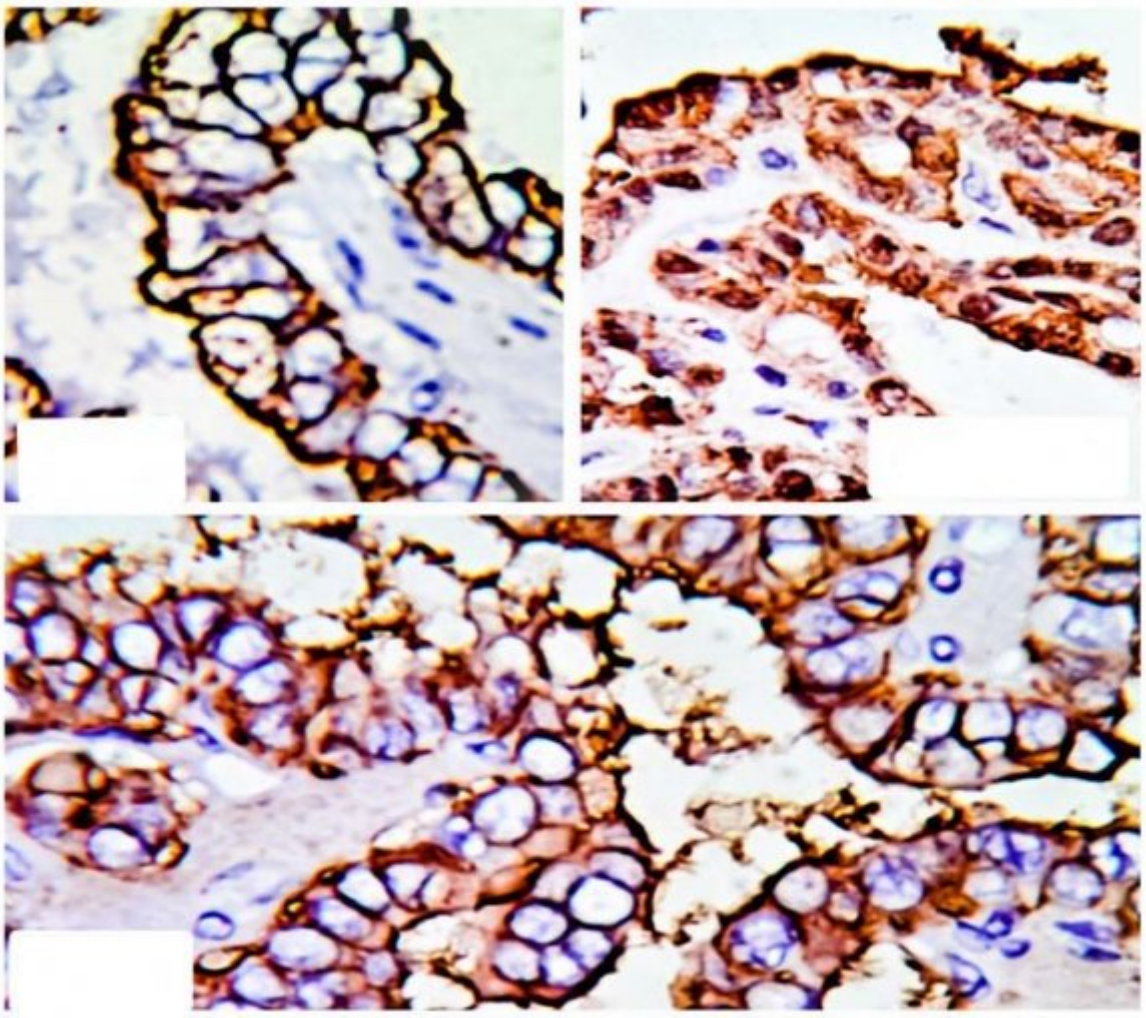
In a study performed by Prasad et al. consisting of 85 carcinoma and 21 adenoma cases, all carcinomas showed different percentage and intensity of staining and 24% of adenomas showed poor intensity of staining with HBME-1. It was concluded that HBME-1 was a very useful marker in malignancies arising from follicular

cells and its negativity in benign lesions has a specificity of 94%. [18] In a study performed by Pisani et al., specific cytoplasmic staining with galectin-3 was observed in a suspicious cell population in fine needle aspiration biopsy of a thyroid nodule. Occult papillary carcinoma was found in the operation material of this case andgalectin-3 was concluded as a marker of malignancy. [19] Thus, in our study we can conclude that this triad of IHC markers -CK19, GAL-3 and HBME-1 could be used in the prognostication of papillary thyroid carcinoma. The positive expressions of these markers have been significantly correlated with total tumor diameter, capsular invasion and lymph nodal metastasis in this study.



1

Figure 1: Figure 1



2

Figure 2: Figure 2

2

D D D D) C

(	Parameter	% (No. of cases)
Age (Yr)		
? 45		85.36 (35/41)
> 45		14.63 (6/41)
Gender		
F		95.12 (39/41)
M		4.87 (2/41)
Total Tumor Diameter		
? 1 cm		19.51 (8/41)
> 1 cm		80.48 (33/41)
Capsular Invasion		
Present		26.82 (11/41)
Absent		73.17 (30/41)
Lymph Node Metastasis		
Present		31.70 (13/41)
Absent		68.30 (28/41)

Prognostic Factors		CK 19 (N=41) Positive	Negative	P Value
Mean Age		33.88	34.78	0.76
Sex	Male Female	2 25	0 14	0.53
Diameter	? 2 cm >2 cm	2 25	12 2	<0.001 *

Figure 3: Table 2 :

3

Prognostic Factors		Galactin-3 (N=41) Positive	Negative	P Value
Mean Age		33.18	36.14	0.33
Sex	Male Female	2 25	0 14	0.53
Diameter	? 2 cm >2 cm	4 23	10 4	0.005
Capsular Invasion	Present Absent	11 16	0 14	0.0071
Lymph Node Metastasis	Present Absent	11 16	2 12	0.1559

Figure 4: Table 3 :

4

Prognostic Factors		HBME-1(N=41) Positive	Negative	P Value
Mean Age		35.54	32.29	0.26
Sex	Male Female	0 24	2 15	0.16
Diameter	? 2 cm >2 cm	6 18	8 9	0.18
Capsular Invasion	Present Absent	10 14	1 16	0.01
Lymph Node Metastasis	Present Absent	13 11	0 17	0.002

Figure 5: Table 4 :

[El Demellawy et al. ()] 'Application of CD56, P63 and CK19 immunohistochemistry in the diagnosis of papillary carcinoma of the thyroid'. D El Demellawy , A Nasr , S Alowami . *Diagn Pathol* 2008. 3 p. 5.

[Rosai et al. ()] *Armed Forces Institute of Pathology*, J Rosai , M L Carcangiu , R A Delellis . 1992. Washington, DC. (Tumors of the thyroid gland)

[Isicdencic et al. ()] 'Cytokeratin19 expression discriminates papillary thyroid carcinoma from other thyroid lesions and predicts its aggressive behavior'. T Isicdencic , D Cvejic , I Paunovic , S Tatic , M Havelka , S Savin . *Med Oncol* 2013. 30 p. 362.

[Saez ()] 'Diagnostic usefulness of tumor markers in the thyroid cytological samples extracted by fine-needle aspiration biopsy'. Gomez Saez , J . *Endocr Metab Immune Disord Drug Targets* 2010. 10 p. .

[Baloch et al. ()] 'Differential expression of cytokeratins in follicular variant of papillary carcinoma: an immunohistochemical study and its diagnostic utility'. Z W Baloch , S Abraham , S Roberts , V Livolsi . *Hum Pathol* 1999. 30 p. .

[Pisani et al. ()] 'Galectin-3 immunodetectionmay improve cytological diagnosis of occult papillary thyroid carcinoma'. T Pisani , A Vecchione , M R Giovagnoli . *Anticancer Res* 2004. 24 p. .

[Yoshii et al. ()] 'Galectin-3 maintains the transformed phenotype of thyroid papillary carcinoma cells'. T Yoshii , H Inohara , Y Takenaka , Y Honjo , S Akahani , T Nomura , A Raz , T Kubo . *Int J Oncol* 2001. 18 p. .

[Prasad et al. ()] 'Galectin-3, fibro -nectin-1, CITED-1, HBME-1 and cytokeratin-19 immunohistochemistry. is useful for the differential diagnosis of thyroid tumors'. M L Prasad , N S Pellegata , Y Huang . *Mod Pathol* 2005. 18 p. .

[Nasr et al. ()] 'Immunohistochemical markers in diagnosis of papillary thyroid carcinoma: utility of HBME1 combined with CK19 immunostaining'. M R Nasr , S Mukhopadhyay , S Zhang , A Katzenstein . *Mod Pathol* 2006. 19 (12) p. .

[Finley et al. ()] *Molecular profiling distinguishes papillary carcinoma Metab*, D J Finley , N Arora , B Zhu , L Gallagher , T J 3 Fahey , Rd . 2004. 89 p. .

[Liu et al. ()] 'MRI and ultrasonography detection of cervical lymph node metastases in differentiated thyroid carcinoma before reoperation'. Z Liu , X Xun , Y Wang , L Mei , L He , W Zeng , C Wang , H Tao . *Am J Transl Res* 2014. 6 p. .

[Balta et al. ()] 'Prognostic value of oncoprotein expressions in thyroid papillary carcinoma'. A Z Balta , A I Filiz , Y Kurt , I Sucullu , E Yucel , M L Akin . *Med Oncol* 2012. 29 (2) p. .

[Saussez et al. ()] 'Serum Galectin-1 and Galectin-3 levels in benign and malignant nodular thyroid disease'. S Saussez , D Glinoer , G Chantrain , F Pattou , B Carnaille , S Andre , H Gabius , G Laurent . *Thyroid* 2008. 18 p. .

[Saussez et al. ()] 'Serum galectin-1 and galectin-3 levels in benign and malignant nodular thyroid disease'. S Saussez , D Glinoer , G Chantrain , F Pattou , B Carnaille , S Andre , H J Gabius , G Laurent . *Thyroid* 2008. 18 p. .

[Rossi et al. ()] 'Simultaneous immunohistochemical expression of HBME-1 and Galectin-3 differentiates papillary carcinomas from hyper functioning lesions of the thyroid'. E D Rossi , M Raffaelli , A Mule , A Miraglia , C P Lombardi , F M Vecchio , G Fadda . *Histopathology* 2006. 48 p. .

[Livolsi ()] 'Surgical pathology of the thyroid'. V Livolsi . *Philadelphia: WB Saunders*, 1990. 5.

[Delellis and Williams (ed.) ()] *Tumours of the thyroid and parathyroid. In: The WHO classification of tumours of endocrine organs*, R A Delellis , E D Williams . Delellis R A, Lloyd R V, Heitz P U, Eng C (ed.) 2004. Lyon: IARC Press. p. .

[Matos et al. ()] 'Usefulness of HBME-1, cytokeratin 19 and galectin-3 immunostaining in the diagnosis of thyroid malignancy'. De Matos , P S Ferreira , A P De Oliveira Facuri , F Assumpcao , L V , Metze K Ward , L S . *Histopathology* 2005. 47 p. .

[Leboulleux et al. ()] 'Vandetanib in locally advanced or metastatic differentiated thyroid cancer: a randomised, double-blind, phase 2 trial'. S Leboulleux , L Bastholt , T Krause , C De La Fouchardiere , J Tennvall , A Awada , J M Gomez , F Bonichon , L Leenhardt , C Soufflet , M Licour , M J Schlumberger . *Lancet Oncol* 2012. 13 p. .