

Original Research Article

ROLE OF CK19 IN THE DIAGNOSIS OF PAPILLARY THYROID CARCINOMA

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ABSTRACT

Background: Papillary thyroid carcinoma (PTC) is the most common type of thyroid cancer, and its accurate diagnosis is crucial for effective management.

Objective: This study aims to evaluate the semiquantitative expression of CK19 in different thyroid lesions, including PTC, the follicular variant of PTC (FVPTC), follicular adenoma, and multinodular goiter (MNG), and to assess its diagnostic accuracy.

Materials and Methods: This cross-sectional study was conducted at tertiary care hospital from June 2020 to June 2023. The sample size for this study comprised 50 thyroid cases. Tissue samples were collected from patients who underwent thyroid lobectomy or total thyroidectomy. The samples were then processed and fixed in formalin, followed by paraffin embedding. Serial tissue sections were prepared for IHC staining.

Results: The study revealed that CK19 expression was strongly positive in 80% of usual-type PTC cases (moderate to strong expression). In the FVPTC group, CK19 was moderately expressed in 75% of cases. The follicular adenoma and MNG groups showed minimal to no CK19 expression, with 66.7% and 77.3% of cases, respectively, showing no CK19 staining. The sensitivity of CK19 in diagnosing PTC was 84%, with a specificity of 91%. The positive predictive value (PPV) was 82%, and the negative predictive value (NPV) was 89%.

Conclusion: It is concluded that CK19 is a highly sensitive and specific marker for diagnosing papillary thyroid carcinoma, particularly in distinguishing it from benign thyroid lesions such as follicular adenoma and multinodular goiter. The semiquantitative assessment of CK19 expression demonstrated strong, diffuse positivity in usual-type PTC and moderate expression in FVPTC, making CK19 a valuable diagnostic tool.

Keywords: Carcinoma, Patients, PTC, FNAC, Metastasis, Diagnosis.

INTRODUCTION

The most widespread kind of thyroid cancer is papillary thyroid carcinoma (PTC), which makes up about 80 percent of all thyroid cancers.^[1] PTC is usually correlated with the good prognosis, because it has the tendency to grow slowly and have a low metastatic rate. Nevertheless, it is important that they are properly diagnosed and detected early in order to guarantee their appropriate management, a low level of recurrence and better chances of survival.^[2] PTC can be mostly diagnosed with the help of clinical examinations, imaging (ultrasound),

and histopathological examination with fine-needle aspiration cytology (FNAC) as the gold standard of preoperative diagnosis. However useful FNAC may be; it may at times give inconclusive results especially in follicular variants of PTC or in differentiating between benign lesions and malignancies.^[3] This points out to the necessity of additional diagnostic markers that would facilitate the identification and distinction between PTC and other thyroid diseases. Cytokeratin 19 (CK19) is an intermediate filament protein of type I whose predominant expression is seen on epithelial cells. It helps in the structural integrity of cells and tissues as a contribution to the cytoskeleton.^[4] Alterations of

the expression pattern of cytokeratins are frequently related to tumor differentiation, invasion and metastasis in malignancies. In particular, it was demonstrated that CK19 is overexpressed in PTC, thus it can serve as a potential biomarker of the diagnosis of this carcinoma.^[5]

Immunohistochemical (IHC) research has been extensively conducted to determine the utility of CK19 in helping to diagnose PTC [6]. Immunohistochemistry is a technology that enables visualization of particular proteins in tissue specimens with the aid of antibodies, and it provides a method of identification of CK19 expression in tumor tissues.^[7] It has also been shown that CK19 is much more highly expressed in PTC than in normal thyroid tissue and its expression in thyroid lesions can be used to distinguish malignant from benign disease, particularly where conventional FNAC findings are indeterminate. CK19 is thought to be especially helpful in identifying the follicular variant of PTC, in which the standard diagnostic methods may be unclear. In this regard, CK19 is an adjunct diagnostic marker that enhances the specificity and sensitivity of diagnosing thyroid cancer.^[8] Further, the expression of CK19 in PTC has been thought to be associated with tumor differentiation and aggressiveness. CK19 overexpression has also been linked to the more aggressive variants of PTC, which are lymph node metastatic and extrathyroidal invasion.^[9] Therefore, CK19 could have not only a diagnostic value but also a prognostic one; this means that it could give the clinician an idea of how the tumor will behave and, therefore, what treatment approach will be more favorable. It should be noted, though, that CK19 is not completely specific to PTC. Other forms of thyroid cancers including medullary thyroid carcinoma and poorly differentiated thyroid carcinoma and some benign diseases can also express elevated levels of CK19, which could represent a limitation to its specificity as a diagnostic agent.^[10,11]

In order to increase the diagnostic specificity of CK19, scholars have suggested that it should be applied alongside other markers, including galectin-3, thyroglobulin and HBME-1. The markers when measured as a panel provide a more detailed characterization of thyroid tumors enhancing sensitivity and specificity of the diagnosis.^[12] Combination of CK19 and galectin-3, e.g., is very useful in distinguishing between PTC and other thyroid malignancies as well as benign lesions, thus minimizing the chances of false-negative outcome.^[13] Although CK19 holds promise as a diagnostic tool it does have limitations which require to be taken into consideration. The false-positive findings it may show include one limitation, such as in the case of thyroid adenomas or other benign tumors. Also, the discrepant expression of CK19 in the various tumor subtypes of PTC can make its application as a single diagnostic entity challenging. Hence, it is necessary to consider the

role of CK19 in the diagnosis of PTC as an addition to other clinically and diagnostically applied tools, not as an independent marker.

Objective

This study aims to evaluate the semiquantitative expression of CK19 in different thyroid lesions, including PTC, the follicular variant of PTC (FVPTC), follicular adenoma, and multinodular goiter (MNG), and to assess its diagnostic accuracy.

MATERIALS AND METHODS

This cross-sectional study was conducted at tertiary care hospital from June 2020 to June 2023. The sample size for this study comprised 50 thyroid cases, categorized as follows:

- 19 cases of PTC, including 15 classic PTC cases and 4 cases of the follicular variant of PTC (FVPTC).
- 22 cases of multinodular goiter (MNG).
- 9 cases of follicular adenoma.

Inclusion Criteria:

1. Patients with histologically confirmed cases of thyroid disease (PTC, FVPTC, MNG, and follicular adenoma).
2. Age range: 18 years and above.
3. Patients who underwent thyroidectomy or had available tissue samples for histopathological examination.

Exclusion Criteria

4. Patients with previous thyroid surgery or known history of other thyroid malignancies.
5. Patients with severe systemic diseases or other malignancies that could interfere with the diagnosis of thyroid carcinoma.
6. Patients with incomplete clinical data or missing histopathological tissue samples.

Data Collection

Tissue samples were collected from patients who underwent thyroid lobectomy or total thyroidectomy. The samples were then processed and fixed in formalin, followed by paraffin embedding. Serial tissue sections were prepared for IHC staining. The primary antibody used for CK19 detection was a monoclonal antibody specific for CK19, and the staining procedure followed standard IHC protocols. The intensity of CK19 expression in the thyroid tissue samples was evaluated by two independent pathologists, who were blinded to the clinical details of the patients. Staining was assessed on a scale of 0 to 3, where 0 indicated no staining, 1 indicated weak staining, 2 indicated moderate staining, and 3 indicated strong staining.

Statistical Analysis

Data were analyzed using SPSS v26. The diagnostic performance of CK19 was assessed by calculating its sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) in diagnosing PTC. The expression levels of CK19 in PTC, FVPTC, MNG, and follicular adenoma were compared using statistical tests, such as the chi-

square test for categorical variables and the t-test for continuous variables. A p-value of less than 0.05 was considered statistically significant.

RESULTS

A total of 50 cases were added in the study with mean age of 45.6 ± 12.3 years, with a slightly higher proportion of male participants (56%) compared to female participants (44%). Among the thyroid lesion

types, 30% were diagnosed with usual-type PTC, 8% with the follicular variant of PTC, 18% with follicular adenoma, and 44% with multinodular goiter (MNG). Clinical histories revealed that 16% of participants had a history of thyroid surgery, 12% had a family history of thyroid cancer, and 20% were smokers. Regarding body mass index (BMI), the majority (54%) were in the normal weight range, while 28% were overweight, 16% were obese, and only 2% were underweight.

Table 1: Demographic and Baseline Characteristics of the Study Population

Characteristic	Value (n = 50)
Age (Mean \pm SD, years)	45.6 \pm 12.3
Gender	
Male	28 (56%)
Female	22 (44%)
Thyroid Lesion Type	
Usual Type PTC	15 (30%)
Follicular Variant PTC	4 (8%)
Follicular Adenoma	9 (18%)
Multinodular Goiter (MNG)	22 (44%)
Clinical History	
History of Thyroid Surgery	8 (16%)
Family History of Thyroid Cancer	6 (12%)
Smoking History	10 (20%)
Body Mass Index (BMI)	
Underweight (<18.5 kg/m ²)	1 (2%)
Normal (18.5–24.9 kg/m ²)	27 (54%)
Overweight (25–29.9 kg/m ²)	14 (28%)
Obese (≥ 30 kg/m ²)	8 (16%)

The sensitivity was 84%, indicating that CK19 was able to correctly identify 84% of PTC cases. The specificity was 91%, demonstrating that 91% of non-cancerous lesions were correctly identified as negative for CK19 expression. The Positive Predictive Value (PPV) was 82%, meaning that

when CK19 was positive, there was an 82% likelihood that the lesion was indeed PTC. The Negative Predictive Value (NPV) was 89%, indicating that when CK19 was negative, there was an 89% likelihood that the lesion was benign, making CK19 a reliable marker for PTC diagnosis.

Table 2: Diagnostic Accuracy of CK19 in Diagnosing PTC

Diagnostic Parameter	Value (%)
Sensitivity	84%
Specificity	91%
Positive Predictive Value (PPV)	82%
Negative Predictive Value (NPV)	89%

Papillary Thyroid Carcinoma (PTC) exhibited the highest CK19 expression, with 80% of cases showing moderate to strong staining ($p < 0.01$). The Follicular Variant of PTC (FVPTC) showed a moderate CK19 expression in 75% of cases ($p < 0.01$), suggesting that while CK19 is still a useful

marker for this variant, its expression is less pronounced compared to the usual type of PTC. In contrast, Multinodular Goiter (MNG) and Follicular Adenoma showed minimal to no CK19 expression, with 22.7% and 33.3% of cases exhibiting weak or absent staining, respectively ($p < 0.01$).

Table 3: CK19 Expression in PTC vs. Other Thyroid Lesions

Thyroid Lesion	CK19 Expression (%)	p-value
Papillary Thyroid Carcinoma (PTC)	80% (moderate/strong)	< 0.01
Follicular Variant of PTC (FVPTC)	75% (moderate)	< 0.01
Multinodular Goiter (MNG)	22.7% (weak/none)	< 0.01
Follicular Adenoma	33.3% (weak/none)	< 0.01

All cases of Papillary Thyroid Carcinoma (PTC) exhibited CK19 expression (100%), indicating that CK19 is highly sensitive for diagnosing this type of thyroid cancer. In contrast, only 33.3% of Follicular

Adenoma cases and 22.7% of Multinodular Goiter (MNG) cases showed CK19 expression, with the majority of these lesions showing no CK19 staining.

Table 4: Presence/Absence of CK19 Expression in PTC, Follicular Adenoma, and MNG

Thyroid Lesion	CK19 Expression Present (n, %)	CK19 Expression Absent (n, %)	Total (n)
Papillary Thyroid Carcinoma (PTC)	15 (100%)	0 (0%)	15
Follicular Adenoma	3 (33.3%)	6 (66.7%)	9
Multinodular Goiter (MNG)	5 (22.7%)	17 (77.3%)	22

Papillary Thyroid Carcinoma (PTC) demonstrated a strong CK19 expression in 80% of cases, with 20% exhibiting moderate expression, indicating its high sensitivity for PTC diagnosis. In contrast, Follicular Adenoma and Multinodular Goiter (MNG) showed

minimal or no CK19 expression, with 66.7% and 77.3% of cases respectively exhibiting no expression (score 0), and only a small percentage showing weak expression.

Table 5: Semiquantitative Expression of CK19 in PTC, Follicular Adenoma, and MNG

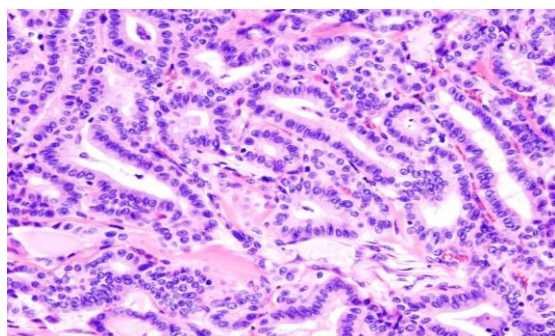
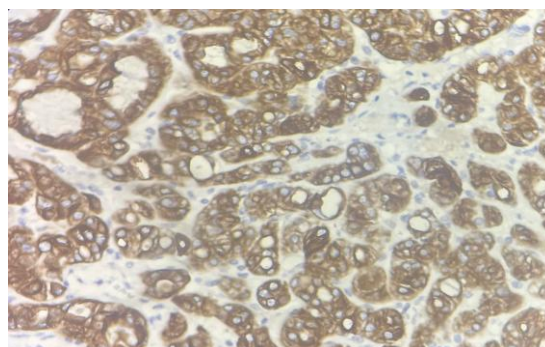
Thyroid Lesion	No Expression (Score 0) (n, %)	Weak Expression (Score 1) (n, %)	Moderate Expression (Score 2) (n, %)	Strong Expression (Score 3) (n, %)	Total (n)
Papillary Thyroid Carcinoma (PTC)	0 (0%)	0 (0%)	3 (20%)	12 (80%)	15
Follicular Adenoma	6 (66.7%)	3 (33.3%)	0 (0%)	0 (0%)	9
Multinodular Goiter (MNG)	17 (77.3%)	5 (22.7%)	0 (0%)	0 (0%)	22

In Usual Type Papillary Thyroid Carcinoma (PTC), 80% of cases exhibited strong CK19 expression, with 20% showing moderate expression. For the Follicular Variant of PTC (FVPTC), 75% of cases had moderate expression, while 25% had weak

expression. In contrast, Follicular Adenoma and Multinodular Goiter (MNG) displayed minimal to no CK19 expression, with 66.7% and 77.3% of cases, respectively, showing no expression.

Table 6: Semiquantitative Expression of CK19 in Usual Type PTC, Follicular Variant of PTC, Follicular Adenoma, and Multinodular Goiter

Thyroid Lesion	No Expression (Score 0) (n, %)	Weak Expression (Score 1) (n, %)	Moderate Expression (Score 2) (n, %)	Strong Expression (Score 3) (n, %)	Total (n)
Usual Type Papillary Thyroid Carcinoma (PTC)	0 (0%)	0 (0%)	3 (20%)	12 (80%)	15
Follicular Variant of PTC (FVPTC)	0 (0%)	1 (25%)	3 (75%)	0 (0%)	4
Follicular Adenoma	6 (66.7%)	3 (33.3%)	0 (0%)	0 (0%)	9
Multinodular Goiter (MNG)	17 (77.3%)	5 (22.7%)	0 (0%)	0 (0%)	22

**1. High power view(H&E-40x)of Papillary Thyroid Carcinoma****2. High power view (IHC- 40x) of Papillary Thyroid Carcinoma showing CK19 positivity**

DISCUSSION

The most prevalent kind of thyroid carcinoma is papillary thyroid carcinoma (PTC), which has specific histopathological features, such as papillary architecture, nuclear grooves, and orphan Annie eye nuclei. Proper diagnosis and staging of PTC are essential in the proper management of a patient.

Immunohistochemistry (IHC) has become a powerful modality to differentiate PTC and other thyroid lesions especially those cases that have cytological or histological ambiguous results. Among these markers which have attracted some interest is cytokeratin 19 (CK19), a low molecular weight protein which is often overexpressed in a variety of epithelial cancers including PTC. Here, semiquantitative CK19 expression was evaluated in various thyroid disorders such as the normal-type PTC, follicular variant PTC (FVPTC), follicular adenoma, and multinodular goiter (MNG). The results showed intense and scattered CK19 staining in most of the usual-type PTC cases and 80 percent of the tumors exhibited moderate to intense staining (score 23). This agrees with the fact that CK19 is presently considered an important indicator of PTC because it is usually present in the cytoplasm of tumor cells and this is in relation to the malignant thyroid epithelial transformation.^[14,15]

CK19 expression was somewhat less in the follicular variant of PTC, 75% of cases showing moderate staining (score 2). This indicates that although CK19 can still be applied in the identification of FVPTC, it might not be as prominent as in the common form of PTC. The lower expression in FVPTC might be explained by the various histological appearances of this variant which frequently fails to exhibit the classical nuclear changes observed in classic PTC and might exhibit an increased follicular-patterned growth. This notwithstanding, detection of CK19 in FVPTC may still be useful adjunct in establishing the diagnosis, especially in difficult cases where cytological and histological appearances overlap with benign follicular lesions. On the other hand, CK19 expression was low or negative in benign thyroid lesions including follicular adenoma and MNG. No CK19 expression was observed in 66.7 percent of cases in follicular adenoma, whereas weak staining was evident in the rest of the cases (33.3 percent). This confirms the idea that the expression of CK19 is restrained in benign thyroid neoplasms, and thus it is a consistent marker to differentiate between malignant and benign lesions.^[16] In MNG, the negative rate of CK19 expression in 77.3 percent also helps to cement its diagnostic value. This low level of CK19 in these benign lesions gives high possibilities that it is specific in the diagnosis of thyroid malignancies especially PTC. Semiquantitative analysis of CK19 expression demonstrated that CK19 is a highly sensitive marker of usual-type PTC because 100 percent of cases exhibited positive staining. This correlates with other past findings which have indicated strong expression of CK19 in PTC and thus it forms a significant diagnostic marker.^[17,18] CK19 was also very specific in distinguishing between PTC and the benign disorders, including MNG and follicular adenoma, with a close relationship between CK19 expression and the existence of malignant thyroid disease. This renders CK19 especially helpful in

cases where fine-needle aspiration cytology (FNAC) or histology only can yield inconclusive results, e.g. in the follicular variant of PTC. Though CK19 is the promising markers, it should be noted that its expression is not absolutely specific to PTC. Other thyroid cancers that have shown an increase in CK19 expression include the medullary thyroid carcinoma and the poorly differentiated thyroid carcinoma.^[19] As a result, CK19 is to be applied together with other markers and clinical observations to determine a more precise diagnosis. Galectin-3, HBME-1, thyroglobulin have also been investigated as markers to be used together with CK19 to determine PTC with increased sensitivity and specificity. Combination of these markers may increase the overall accuracy of the diagnosis, especially the distinction between PTC and benign follicular lesions, as well as other thyroid cancers.

CONCLUSION

It is concluded that CK19 is a highly sensitive and specific marker for the diagnosis of Papillary Thyroid Carcinoma (PTC), particularly for distinguishing it from benign thyroid lesions such as Follicular Adenoma and Multinodular Goiter (MNG). The semiquantitative assessment of CK19 expression demonstrated strong, diffuse positivity in the usual type of PTC and moderate expression in the follicular variant of PTC. This pattern of expression makes CK19 an effective diagnostic tool, especially in cases where conventional methods such as fine-needle aspiration cytology (FNAC) or histopathological examination may be inconclusive. The absence or minimal expression of CK19 in benign thyroid conditions further supports its utility in distinguishing malignant from non-malignant lesions. The findings also suggest that CK19, when combined with other markers such as galectin-3 and HBME-1, can improve the diagnostic accuracy of PTC, especially in difficult-to-diagnose cases. However, CK19 should not be used in isolation, as its expression is not entirely specific to PTC and can also be seen in other thyroid malignancies.

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