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Diagnostic value of beta-catenin immunohistochemical staining in papillary thyroid carcinoma

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Abstract

Background: The gold standard of diagnosing Papillary thyroid carcinoma (PTC) is achieved through pathologic evaluation using routine hematoxylin and eosin (H and E) staining. The determination of papillary carcinoma by the histological study is mainly based on architectural changes along with nuclear clearing, overlapping, grooving, and pseudoinclusions. In the absence of these changes, papillary carcinoma is difficult to distinguish from the benign thyroid lesion (BTL). **Objective:** This study aimed to assess the expression of β-catenin in BTL and PTC to evaluate its diagnostic values. **Materials and Methods:** One hundred H&E staining slides prepared from paraffin blocks (including 48 with PTC pathology, diagnosis and 52 with BTL pathology diagnosis) that met all inclusion criteria were selected. Paraffin blocks were sectioned and stained with a β-catenin marker using the immunohistochemistry method. Data were analyzed using the SPSS (V.18). Then, the sensitivity, specificity, positive predictive value, and negative predictive value were calculated to describe the diagnostic value of β-catenin. **Results:** Cytoplasmic and membranous expression of the β-catenin protein was observed in 98.1% and 84.6% of BTL, respectively. Related to PTC, this record was observed in 97.9% and 93.7%, respectively. The sensitivity and specificity of cytoplasmic expression were 97.9% and 1.92%, respectively. The sensitivity and specificity of membranous expression were 93.7% and 15.3%, respectively. **Conclusions:** β-catenin can be used as a morpho-immunohistochemical marker to detect specific structural features such as papillae and pseudoinclusion, which are not well seen in H and E staining.

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Full Text

Introduction

Thyroid cancer is the most common malignant disease in the endocrine system and is rapidly increasing in incidence.[1] The increasing incidence partially reflects the earlier detection of small asymptomatic cancers because of the prevalence of screening.[2] Most primary thyroid cancers are epithelial tumors that originate from thyroid follicular cells. These cancers include three main pathological types of carcinomas: papillary thyroid carcinoma (PTC), follicular thyroid carcinoma (FTC), and anaplastic thyroid carcinoma. Medullary thyroid carcinoma arises from thyroid parafollicular (C) cells.[3] PTC is the most common type of thyroid malignancy. Females are more affected than males. It can present in any age group. The mean age at the time of initial diagnosis is approximately 40 years.[4] Thyroid tumor expresses as thyroid nodules, but 90% of thyroid nodules are benign. Although histology is considered the standard diagnostic method for the diagnosis of thyroid cancer, it has some limitations where the morphological features are ambiguous. The diagnosis of papillary cancer by the histological study is mainly based on architectural changes along with the presence of nuclear clearing, overlapping, grooving, and pseudoinclusions. In the absence of papillary architecture, follicular variant of papillary thyroid carcinoma (FVPTC) is difficult to distinguish from nodular thyroid adenoma and also differentiation of PTC from hyperplastic papillary thyroid nodule is challenging. Moreover, this is further complicated by inter-observer variability among pathologists,[5] which can lead to unwanted treatment. Nodular goiter and thyroid cancer are one of the most critical problems of endocrine surgery due to the differences in the completeness of surgical treatment of benign and malignant thyroid tumors. Nodular goiter surgery does not require complete resection of the thyroid gland, which is performed when thyroid cancer is suspected.[6] Several studies have suggested immunohistochemical expression of cancer biomarkers as a diagnostic tool for thyroid carcinoma, but controversy regarding the usefulness of several markers exists.[5]

β-catenin was initially identified as a protein involved in the regulation of cell-cell adhesion coupled with E-cadherin.[7],[8] The effects of β-catenin on thyrocyte transformation into a cancer cell are associated with the Wnt signaling pathway. Under normal conditions, the level of β-catenin in the cytoplasm is kept low. Its increase causes migration of β-catenin to the cell nucleus which stimulates the Wnt pathway to protein production.[6] Most of the previous immunohistochemical studies showed membrane localization of β-catenin in papillary carcinoma.[8] Our current research is focused on the evaluation of diagnostic values of expression of the β-catenin and its immunohistochemical staining in PTC compared to benign thyroid lesions (BTLs).

Materials and Methods

Materials

β-catenin, liquid 3,3- diaminobenzidine tetrahydrochloride (DAB) + substrate CHROMOGEN System, EnVision, and Dual Link System, Target Retrieval Solution were purchased from Danish company DAKO (Glostrup, Denmark). Hydrogen peroxide, methyl alcohol, Entellan glue, and ethyl alcohol 99.6% were supplied from Merck (Gernsheim, Germany). Xylene and ethyl alcohol 70%–96% were purchased from Shimi-lab (Tehran, Iran). All used experimental apparatus were standard and calibrated.

Patient samples

In this retrospective study, 100 paraffin-embedded tissues were obtained from patients who underwent thyroidectomy, in the years of 2016 and 2017, at Imam Reza Hospital. These consist of 52 benign tumors (nodular or adenomatous goiter) and 48 PTC (41 classical variants, 6 follicular variants, and one encapsulated variant).

The mean age of the patients was 43.17 ± 1.28 years (range: 18–69). The patients consist of 80 women and 20 men.

A pathologist examined hematoxylin and eosin (H and E) staining slides prepared from paraffin blocks, and the diagnosis was agreed upon using well-established histopathological criteria.

The admission committee was received from Kermanshah University of Medical Sciences (KUMS) by a code number of KUMS.REC.1394.47 in 2016.

Immunohistochemistry

Immunohistological staining was performed on formalin-fixed paraffin-embedded tissue sections using antibodies against β-catenin. Tissue sections (4 μm) were deparaffinized at 60°C–65°C for 24 h and xylene for 24 h. Slides were rehydrated in a graded series of ethanol solutions and phosphate-buffered saline (PBS) for about 5 min. To retrieve antigens, slides were immersed in the jar containing Tris buffer (pH = 9) and heated in a water bath at 95°C for 20 min, followed by washing in PBS solution. To quench the intracellular activity of peroxidases, slides were immersed in a solution of 3% hydrogen peroxide in methanol for 15 min, and slides were incubated by primary (45–120 min) and secondary antibodies for 45°C and 30°C (30–45 min), respectively, in a humid and dark place at room temperature. The slides were washed in PBS and stained with the substrate–chromogen solution known as DAB for 5 min. The counterstaining was performed with hematoxylin for 30 s, lithium carbonate 5 min, and washed in water. The stained slides were immersed in a graded series of ethanol and then xylene to transparency and dehydration of tissues. Then, slides were mounted for examination under a microscope. Negative controls were exposed to antibody diluent replacing primary antibody. Membranous staining was scored semi-quantitatively based on the staining intensity of positively stained tumoral cells.

Samples were scored as: −, negative; +, weakly positive; ++, moderately positive; and +++, strongly positive. Nuclear staining was considered positive when more than 10% of nuclei stained strongly for β-catenin.[7],[8]

Statistical analysis

Data were analyzed using the SPSS software (PASW Statistics for Windows, Version 18.0. Chicago: SPSS Inc). Then, the sensitivity, specificity, positive predictive value, and negative predictive value were calculated to describe the diagnostic value of β-catenin.

Results

The results of different parameters of diagnostic value of β-catenin immunohistochemistry staining in PTC including its sensitivity, specificity, positive predictive value, and negative predictive value have come in following:

Cytoplasmic expression of β-catenin was observed in 98.1% (51/52) of BTL and 97.9% (47/48) of PTC [Table 1] and [Figure 1].{Figure 1}{Table 1}

Membranous expression of β-catenin was observed in 84.6% (44/52) of BTL (including 42.3% +, 17.3% ++, and 25% +++) and 93.7% (45/48) of PTC (including 33.3% +, 18.75% ++, and 41.7% +++) [Table 1] and [Figure 1].

The sensitivity and specificity of cytoplasmic expression of β-catenin in differentiating PTC from BTL were 97.9% and 1.9%, respectively. Accordingly, the positive predictive value is 50.5% and the negative predictive value is 72.7% [Table 2].{Table 2}

The sensitivity and specificity of membranous expression of β-catenin in differentiating PTC from BTL were 93.7% and 15.3%, respectively. Hence, the positive predictive value is 47.9% and the negative predictive value is 50.0% [Table 2].

No nuclear staining was found in any sample analyzed. Nuclear pseudoinclusions strongly were stained with β-catenin in 20.8% (10/48) of PTC [Table 3] and [Figure 1].{Table 3}

Discussion

The gold standard method for diagnosing thyroid lesions is to examine the pathology specimen using H and E staining. However, the morphological overlap between benign and malignant lesions such as follicular lesions and its neoplasms is particularly prevalent with the FVPTC and makes diagnosis impossible in some patients.[9]

Proper diagnosis of pathology leads to appropriate treatment and prevention of unwanted and unavoidable complications. Applying appropriate diagnostic methods of pathology can lead to the satisfaction of the patient and the treating physician.

Although histopathology and fine-needle aspiration are often used to diagnose thyroid cancer, distinguishing PTC from hyperplastic papillary thyroid nodule and FTC from FUPTC is challenging. To overcome the limitations of histopathology, immunohistochemistry has been proposed as an alternative diagnostic method for thyroid cancer.[5] Previous studies have shown that intracellular accumulation of beta-catenin and mutation in exon 3 beta-catenin are carcinogenic factors in thyroid and various cancers.[7]

It has been shown that beta-catenin is involved in cell adhesion and the Wnt message pathway. It was suggested that activation of the Wnt signaling pathway might be implicated in the tumorigenesis of papillary and follicular carcinomas.[7]

Membrane expression of beta-catenin is reduced in adenocarcinoma and FTC, and further reduction of membrane expression is associated with indifference.[10]

In this study, the nuclear expression of beta-catenin was seen neither in the malignant nor BTLs. This finding is similar to a study by Miyake et al., which confirms that in the differentiated papillary carcinoma, there is an accumulation of beta-catenin in the membrane and cytoplasm, but in undifferentiated and poorly differentiated carcinomas, due to a mutation in the beta-catenin coding gene, this accumulates in the nucleus.[8] Membrane expression of beta-catenin was observed in both benign and malignant lesions (from + to +++), although sometimes this staining did not induce different much from the nontumor tissue around the lesion. These findings were consistent with the results of a study by Miyake et al. and Sethi et al.[5],[8]

Membrane expression of beta-catenin was not observed in 93.7% of malignant lesions. In benign lesions, beta-catenin membrane expression was not observed in 84.6%. In a study by Rezk et al., the membrane expression of beta-catenin in malignant and benign lesions was 87% and 79%, respectively, which was similar to our study.[11]

In the present study, 97.9% of malignant lesions had the cytoplasmic expression of beta-catenin, and in the case of follicular variants and encapsulated papillary carcinoma, the cytoplasmic expression had been raised 100%. These results are consistent with the findings of previous studies by Garcia-Rostan et al. and Meirmanov et al. In their study, the cytoplasmic expression of beta-catenin in PTC was 100% increased.[12],[13]

Sethi et al. found the immunohistochemical expression of β-catenin in 96% of PTC and 100% of benign lesions. The sensitivity and specificity of β-catenin expression in differentiating PTC from benign tumors were 44% and 95%, respectively. Accordingly, the positive predictive value was 95.6% and the negative predictive value was 40%.[5] In another study, He et al. have shown that 71.7% PTC expressed β-catenin.[14]

In our study, the sensitivity of cytoplasmic and membranous expression of beta-catenin is 97.9% and 93.7%, respectively, and its specificity is 1.9% and 15.3%. On the other hand, the positive predictive value for cytoplasmic and membranous expression is 47.9% and 50.5%, respectively.

Despite the high sensitivity, due to the expression of beta-catenin in both benign and malignant lesions, low specificity, and positive predictive value, this marker alone is not able to differentiate benign and malignant lesions.

However, despite the low specificity of beta-catenin in differentiating malignant from benign lesions, this can be useful in cases where it is not possible to differentiate papillary carcinoma from other lesions by conventional staining. In these cases, it can be used as a morpho-immunohistochemical marker to detect specific structural features such as papillae and pseudoinclusion, which are not well seen in H and E staining. Conclusions: β catenin can be used as a morpho immunohistochemical marker to detect specific structural features such as papillae and pseudo inclusion, which are not well seen in H and E staining.

Conclusions

β catenin can be used as a morpho immunohistochemical marker to detect specific structural features such as papillae and pseudo inclusion, which are not well seen in H and E staining.

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Conflicts of interest

There are no conflicts of interest.

References

1 Jemal A, Bray F, Center MM, Ferlay J, Ward E, Forman D. Global cancer statistics. CA Cancer J Clin 2011;61:69-90.

2 Enewold L, Zhu K, Ron E, Marrogi AJ, Stojadinovic A, Peoples GE, *et al*. Rising thyroid cancer incidence in the United States by demographic and tumor characteristics, 1980-2005. Cancer Epidemiol Biomarkers Prev 2009;18:784-91.

3 Katoh H, Yamashita K, Enomoto T, Watanabe M. Classification and general considerations of thyroid cancer. Ann Clin Pathol 2015;3:1045.

4 Feldman AL, Eunhee SY. Rosai and Ackerman's surgical pathology. JAMA 2012;307:201.

5 Sethi K, Sarkar S, Das S, Rajput S, Mazumder A, Roy B, *et al*. Expressions of CK-19, NF-kappaB, E-cadherin, beta-catenin and EGFR as diagnostic and prognostic markers by immunohistochemical analysis in thyroid carcinoma. J Exp Ther Oncol 2011;9:187-99.

6 Bełdowski M. Assessment of plasma B-catenin concentration as biomarker of thyroid cancer. Pol Przegl Chir 2015;87:340-5.

7 Jargin S. Book review: Dabbs DJ. Diagnostic immunohistochemistry. Elsevier [Russian]. Ukrainian Med J 2010;78:96-8.

8 Miyake N, Maeta H, Horie S, Kitamura Y, Nanba E, Kobayashi K, *et al*. Absence of mutations in the β-catenin and adenomatous polyposis coli genes in papillary and follicular thyroid carcinomas. Pathol Int 2001;51:680-5.

9 Kurihara T, Ikeda S, Ishizaki Y, Fujimori M, Tokumoto N, Hirata Y, *et al.* Immunohistochemical and sequencing analyses of the Wnt signaling components in Japanese anaplastic thyroid cancers. Thyroid 2004;14:1020-9.

10 Dabbs DJ. Diagnostic Immunohistochemistry: Theranostic and Genomic Applications. Philadelphia, PA : Elsevier Health Sciences; 2018.

11 Rezk S, Brynes R, Nelson V, Thein M, Patwardhan N, Fischer A, *et al*. β-Catenin expression in thyroid follicular lesions: Potential role in nuclear envelope changes in papillary carcinomas. Endocr Pathol 2004;15:329-37.

12 Garcia-Rostan G, Camp RL, Herrero A, Carcangiu ML, Rimm DL, Tallini G. β-catenin dysregulation in thyroid neoplasms: Down-regulation, aberrant nuclear expression, and CTNNB1 exon 3 mutations are markers for aggressive tumor phenotypes and poor prognosis. Am J Pathol 2001;158:987-96.

13 Meirmanov S, Nakashima M, Kondo H, Matsufuji R, Takamura N, Ishigaki K, *et al*. Correlation of cytoplasmic β-catenin and cyclin D1 overexpression during thyroid carcinogenesis around Semipalatinsk nuclear test site. Thyroid 2003;13:537-45.

14 He F, Li H, Li WS, Dong XH. Expression of mucin-l and beta-catenin in papillary thyroid carcinoma and the clinical significance thereof. Zhonghua Yi Xue Za Zhi 2009;89:393-6.