




RESEARCH ARTICLE

Evaluation of serum B7-H3 expression, ultrasound and clinical characteristics to predict the risk of cervical lymph node metastases in papillary thyroid carcinoma by nomogram

Yingcheng Huang¹ | Zehao Huang¹ | Huizhu Cai¹ | Lingdun Zhuge¹  | Shixu Wang¹ | Dangui Yan¹ | Xiwei Zhang¹ | Changming An¹  | Lijuan Niu² | Zhengjiang Li¹ 

¹Department of Head and Neck Surgery, National Cancer Center/National Clinical Research Center for Cancer/Cancer Hospital, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing, China

²Department of Ultrasound, National Cancer Center/National Clinical Research Center for Cancer/Cancer Hospital, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing, China

Correspondence

Lijuan Niu, Department of Ultrasound, National Cancer Center/National Clinical Research Center for Cancer/Cancer Hospital, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing 100021, China.
Email: niulijuan8197@126.com

Zhengjiang Li, Department of Head and Neck Surgery, National Cancer Center/National Clinical Research Center for Cancer/Cancer Hospital, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing 100021, China.
Email: lizhengjiang@cicams.ac.cn

Funding information

grant provided by the Capital Clinical Features Application Research, Grant/Award Number: Z171100001017211

Abstract

Background: Improving the preoperative diagnosis of cervical lymph node metastasis (LNM) will help improve the clinical outcomes of papillary thyroid carcinoma (PTC) patients. B7-H3, as an immune checkpoint of the B7 family, is highly expressed in PTC tissues and related to LNM and prognosis. We aimed to explore the clinical values of serum B7-H3 (sB7-H3) in predicting LNM in PTC by a nomogram prediction model.

Methods: From September 2019 to May 2021, a total of 344 PTC patients with primary surgery in our hospital were enrolled in this research. Enzyme-linked Immunosorbent Assay (ELISA) was used to detect sB7-H3 from the peripheral blood of PTC patients and normal controls. We created a nomogram prediction model in combination with sB7-H3 expression, clinical and ultrasound characteristics to predict LNM in the early stage.

Results: Gender ($p = 0.001$), age ($p = 0.015$), tumor size ($p < 0.001$), number of tumors ($p = 0.021$) and sB7-H3 expression ($p = 0.003$) were independent risk factors for LNM in PTC. All the factors were included in the nomogram. The area under the curve (AUC) was 73.9% (95% CI, 68.12%–79.69%).

Conclusion: The nomogram is helpful in assessing the risk of LNM in PTC. sB7-H3 has excellent potential in predicting LNM in patients with PTC as an adjunctive ultrasound tool.

KEYWORDS

lymph nodes metastasis, nomogram, papillary thyroid carcinoma, sB7-H3

Authors Yingcheng Huang and Zehao Huang contributed equally to this work and should be co-first authors.

This is an open access article under the terms of the [Creative Commons Attribution-NonCommercial-NoDerivs](https://creativecommons.org/licenses/by-nc-nd/4.0/) License, which permits use and distribution in any medium, provided the original work is properly cited, the use is non-commercial and no modifications or adaptations are made.

© 2022 The Authors. *Journal of Clinical Laboratory Analysis* published by Wiley Periodicals LLC.

1 | INTRODUCTION

Thyroid carcinoma is the most common malignant tumor of the head and neck. With the development of sophisticated ultrasound instruments and technology,¹ the number of patients with early-stage thyroid cancer has gradually increased in recent years. As of 2020, there were 586,000 cases of thyroid carcinoma worldwide, ranking 9th in the cancer incidence.² In China, the incidence of thyroid carcinoma ranks 7th, but the mortality rate is low and stable.³ Papillary thyroid carcinoma (PTC) is the most common type, accounting for 80%–90% of all thyroid cancers,⁴ and most of PTC patients have favorable prognosis after standardized treatment. However, PTC patients with aggressive clinicopathological features, including large tumor size, cervical lymph node metastasis (LNM), and local external invasion, may still have adverse outcomes such as recurrence, metastasis, and even death.⁵ Although ultrasonography and fine needle aspiration cytology are the most commonly used methods for PTC diagnosis and the accuracy of ultrasound in diagnosing malignant thyroid nodules is about 80%, the sensitivity of ultrasound and CT in the diagnosis of LNM is only about 27%.⁶ Additionally, the incidence of occult central lymph node metastasis (CLNM) is approximately 30%–80% and the incidence of occult lateral neck lymph node metastasis (LLNM) is between 18.6% and 64%.⁷ Thus, improving the preoperative diagnosis of LNM will help improve the clinical outcomes of PTC patients.

B7-H3 (CD276), a member of the B7 family, has received extensive attention as a factor regulating tumor immune response.⁸ B7-H3 contains a transmembrane structure and intracellular part and belongs to the transmembrane glycoprotein of the immunoglobulin superfamily.⁹ The primary function of B7-H3 is to stimulate T cell proliferation, selectively improve IFN- γ production and act as a co-stimulatory molecule to exhibit anti-tumor immunity.^{9–11} The expression of B7-H3 is found in multiple types of cancer including ovarian cancer, prostate cancer, endometrial cancer, kidney cancer, etc.^{12–15} and high expression of B7-H3 is associated with poor tumor prognosis.^{16,17} B7-H3 is highly expressed, and its expression level is related to LNM and prognosis of PTC.¹⁸ Given that B7-H3 could be detected in peripheral blood¹⁹ this study aimed to explore the clinical values of serum B7-H3 (sB7-H3) in predicting LNM in PTC. In the meantime, we created a nomogram prediction model with sB7-H3 expression, clinical and ultrasound characteristics to predict LNM in the early stage.

2 | MATERIALS AND METHODS

2.1 | Clinical samples

From September 2019 to May 2021, a total of 344 PTC patients with primary surgery in the Department of Head and Neck Surgery, Cancer Hospital, Chinese Academy of Medical Sciences were enrolled in this research, with an average age of 41.1 years old (range, 7–83 years old). The inclusion criteria were as follows: (1)

Postoperative pathology confirmed papillary thyroid carcinoma. (2) Preoperative peripheral blood samples were retained. (3) All surgery were performed by the same surgical team. The follow-up time was 8–30 months (median 15 months). Fourteen patients (4.1%) were lost to follow-up. Clinical characteristics (age, gender, preoperative diagnosis, and preoperative fine needle aspiration results) and ultrasound characteristics were recorded. All patients were staged using the 8th Edition of the American Joint Committee on Cancer (AJCC) TNM staging system. The patients' stage and the status of cervical lymph nodes were determined by postoperative pathology. At the same time, we included 215 healthy volunteers as the normal control who had no underlying diseases and no history of malignant tumors during the same period of physical examination in our hospital. The healthy volunteers were 20–79 years old (average 37.7 years old). After 1:1 matched by age and sex, sB7-H3 levels of 160 PTC patients and 160 people in normal controls were compared. All peripheral blood samples were stored at -80°C immediately after centrifugation. This study was approved by the Ethics Committee of Cancer Hospital, Chinese Academy of Medical Sciences and was conducted in accordance with the principles of the Declaration of Helsinki.

2.2 | Enzyme-linked Immunosorbent Assay (ELISA)

Peripheral blood was collected from PTC patients and normal controls. Plasma was separated by centrifugation and stored at -80°C . Enzyme-linked Immunosorbent Assay (ELISA, Proteintech) was used to detect human sB7-H3 (Human CD276/B7-H3). Microtiter plates were coated with anti-B7-H3 antibodies, serum or B7-H3 standards were added and incubated for 2 h at 37°C . After washing, B7-H3 detection antibody conjugated to biotin was added, and the antibody was incubated for 40 min before washing. After washing, tetramethylbenzidine substrate (TMB) was added and incubated for 15–20 min. H_2SO_4 (2 N) was added to stop the reaction. The optical density of each well was recorded with a spectrometer at 450 nm wavelength with a microplate reader (BioTek SynergyH1). Each sample was analyzed by three rewells.

2.3 | Statistical analysis

Papillary thyroid carcinoma patients and people in normal controls were matched 1:1 according to age and gender (Table S1). The paired *t* test was used to compare the differences between paired samples by GraphPad Prism 8 software (GraphPad Software Inc.). Continuous variables are presented as mean \pm standard deviation. Univariate analysis was used to investigate the risk factors of lymph node metastasis in PTC by χ^2 test. All variables found to be statistically significant were included in multivariate logistic regression analysis. Statistical significance was defined as a two-sided *p*-value < 0.05 for all results in this study, unless otherwise stated. Nomogram of PTC with LNM was established by the multivariate logistic regression model. Its predictive accuracy and discriminatory capacity were

determined using the concordance index (C-index) and the calibration curve. Statistics were calculated using SPSS 23.0. and R statistical software (The R Foundation for Statistical Computing).

3 | RESULTS

3.1 | Patient characteristics and sB7-H3 expression

Table 1 showed the basic characteristics of 344 PTC patients and 215 normal controls. In PTC patients, 240 (69.8%) had LNM, and 340 (98.8%) were in stage I/II. As shown in Figure 1, the average expression level of sB7-H3 in peripheral blood samples of matched PTC patients (2845.83 ± 722.95 pg/ml, range 1689.07–5359.72 pg/ml, $n = 160$) was higher than that of matched normal controls

(2375.28 ± 453.85 pg/ml, range 1386.11–4481.83 pg/ml, $n = 160$; $p < 0.0001$). The optimal cutoff for sB7-H3 expression (2545 pg/ml) was identified by Cutoff Finder. sB7-H3 > 2545 pg/ml was defined as

TABLE 1 PTC patients and Healthy donors' characteristics

	PTC Patients ($n = 344$)	Normal Controls ($n = 215$)
Gender		
Male	98 (28.5%)	75 (34.9%)
Female	246 (71.5%)	140 (65.1%)
Age (years)		
<55	297 (86.3%)	192 (89.3%)
≥55	47 (13.7%)	23 (10.7%)
Tumor size (cm)		
≤1	180 (52.3%)	NA
>1	164 (47.7%)	NA
Number of tumors		
Multifocal	184 (53.5%)	NA
Unifocal	160 (46.5%)	NA
Capsule invasion		
Y	257 (74.7%)	NA
N	87 (25.3%)	NA
Strap muscle invasion		
Y	20 (5.8%)	NA
N	324 (94.2%)	NA
Pathological stage		
I	314 (91.3%)	NA
II	26 (7.6%)	NA
III	4 (1.2%)	NA
IV	0 (0.0%)	NA
Pathological T stage		
T1	293 (85.2%)	NA
T2	14 (4.1%)	NA
T3	16 (4.7%)	NA
T4	21 (6.1%)	NA
Pathological N stage		
N0	104 (30.2%)	NA
N1	240 (69.8%)	NA

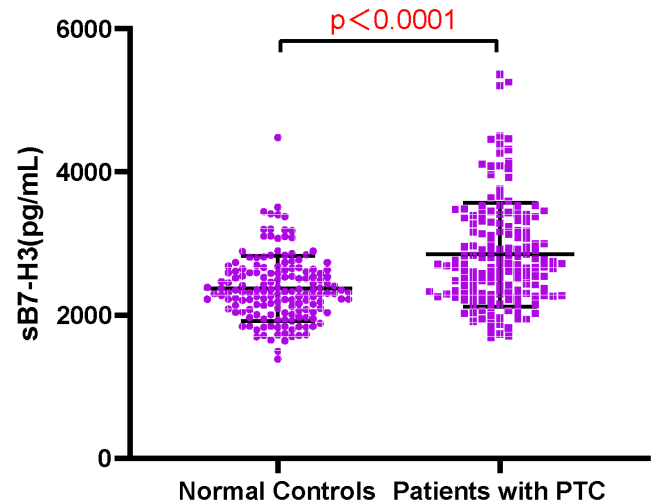


FIGURE 1 Detection of circulating B7-H3. The scatter plot displays the distribution of serum B7-H3. Significantly elevated sB7-H3 levels were found in serum from patients with PTC compared with those in normal controls. PTC, papillary thyroid carcinoma

TABLE 2 Univariate analysis for PTC patients with cervical lymph node metastases

	N0 ($n = 104$)	N1 ($n = 240$)	<i>p</i> value
Gender			
Male	15 (14.4%)	83 (34.6%)	<0.001
Female	89 (85.6%)	157 (65.4%)	
Age (years)			
<55	84 (80.8%)	213 (88.8%)	0.048
≥55	20 (19.2%)	27 (11.3%)	
Tumor size (cm)			
≤1	72 (69.2%)	108 (45.0%)	<0.001
>1	32 (30.8%)	132 (55.0%)	
Number of tumors			
Multifocal	46 (44.2%)	138 (57.5%)	0.023
Unifocal	58 (55.8%)	102 (42.5%)	
Capsule invasion			
Y	73 (70.2%)	184 (76.7%)	0.205
N	31 (29.8%)	56 (23.3%)	
Strap muscle invasion			
Y	3 (2.9%)	17 (7.1%)	0.126
N	101 (97.1%)	223 (92.9%)	
sB7-H3 expression			
High expression	40 (38.5%)	137 (57.1%)	0.002
Low expression	64 (61.5%)	103 (42.9%)	

The bold values indicated the statistical significance.

sB7-H3 high expression (51.5%, $n = 177$), and sB7-H3 ≤ 2545 pg/ml was defined as sB7-H3 low expression (48.5%, $n = 167$).

3.2 | Nomogram for PTC with cervical lymph node metastases

Univariate analysis (Table 2) revealed that gender ($p < 0.001$), age ($p = 0.048$), tumor size ($p < 0.001$), number of tumors ($p = 0.023$), and sB7-H3 expression ($p = 0.002$) were significantly associated with LNM. In multivariate analysis, gender (HR 0.342, 95% CI 0.182–0.645, $p = 0.001$), age (HR 0.425, 95% CI 0.213–0.847, $p = 0.015$), tumor size (HR 2.962, 95% CI 1.765–4.972, $p < 0.001$), number of tumors (HR 1.805, 95% CI 1.093–2.979, $p = 0.021$) and sB7-H3 expression (HR 2.128, 95% CI 1.285–3.524, $p = 0.003$) were independent

risk factors for LNM in PTC (Table 3). These factors in the multivariate logistic regression analysis were used to create a nomogram to predict LNM in PTC (Figure 2). The area under the curve (AUC) was 73.9% (95% CI, 68.12%–79.69%; Figure 3). The C-index of the nomogram was 0.739 (95% CI, 0.733–0.745). The calibration curve for the probability of LNM showed the satisfied calibration (Figure 4).

4 | DISCUSSION

Surgery was the primary treatment for thyroid carcinoma. In daily clinical work, surgeons paid great attention to the high-risk factors affecting LNM and adjusted the surgical plan accordingly. Age and gender were essential factors for the evaluation of thyroid carcinoma. Patients (age < 55 years) were more likely to have LNM^{20,21} and

	HR	95%CI	<i>p</i> value
Gender			
Male	Reference		
Female	0.342	0.182–0.645	0.001
Age (years)			
< 55	Reference		
≥ 55	0.425	0.213–0.847	0.015
Tumor size (cm)			
≤ 1	Reference		
> 1	2.962	1.765–4.972	< 0.001
Number of tumors			
Unifocal	Reference		
Multifocal	1.805	1.093–2.979	0.021
sB7-H3 expression			
Low expression	Reference		
High expression	2.128	1.285–3.524	0.003

TABLE 3 Multivariate analysis for PTC patients with cervical lymph node metastases

Abbreviations: HR, Hazard ratio. CI, Confidence interval.

The bold values indicated the statistical significance.

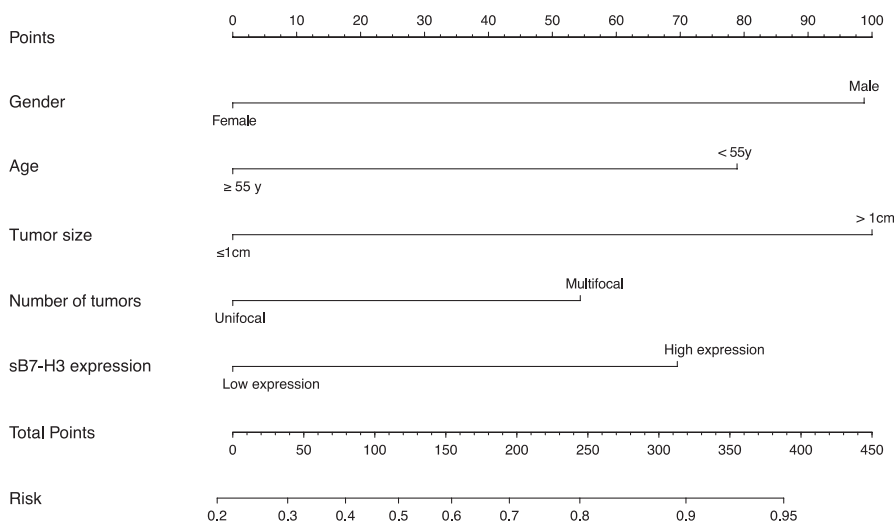


FIGURE 2 Nomogram for predicting the risk of papillary thyroid carcinoma patients with cervical lymph node metastases. According to the patient value, the corresponding values of each variable were marked and summed at each axis, and the sum was marked on the total points axis. Draw a line perpendicular to the probability axis to get the corresponding probability value

male PTC patients had a higher risk of LNM than female patients.²² This was consistent with our findings. This might be because male patients were generally at the advanced T stage, or they might be affected by sex hormones.²³ We believed that tumor size was also an essential factor in evaluating LNM. Nie et al. found that tumor size >1.5 cm was an independent risk factor for LNM.²⁴ Other studies have found that tumor size >2 cm increased the risk of LNM in PTC patients.⁷ For PTC patients with tumor diameter ≤1 cm, Siddiqui et al. considered that the tumor size of thyroid microcarcinoma had no significant statistical difference with the LNM.²⁵ According to

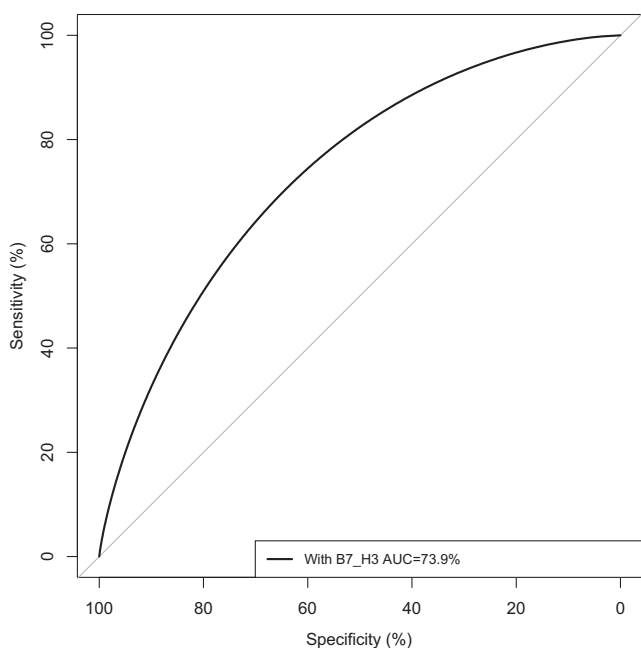


FIGURE 3 ROC curve for the prediction model. AUC was 73.9% (95% confidence interval, 68.12%–79.69%)

this study, tumor size (>1 cm) was an independent risk factor for predicting LNM. Different studies have different definitions of tumor size cutoff, but generally, it was consistent that LNM was more likely to occur in PTC patients with larger tumors. This suggested that the risk of LNM should be carefully evaluated before surgery in patients with large tumors. Thus, we added this variable to the prediction model. Multifocal tumor was also a risk factor for LNM.^{26–28} Patients with multifocal PTC tended to have a higher clinical stage and were more likely to have tumor recurrence and death.²⁹ Lombardi et al.³⁰ found that patients with multifocal PTC were 17.9 times more likely to develop LNM than other PTC patients. Our study also showed that patients with multifocal PTC have a higher proportion of LNM. We considered that some multifocal cases were independent primary tumors, while others were due to the spread of single cancer into the thyroid gland. However, the latter was often accompanied by multiple LNM.³¹

In the diagnosing and treating PTC, practice specific molecular markers still needed to be used to identify potential high-risk patients. Thus, it was crucial to find new and valuable diagnostic markers. Immune checkpoint B7-H3 has shown promising prospects in a variety of carcinoma. Our previous studies have also shown that B7-H3 was highly expressed in PTC tissues and was associated with LNM at the tissue level. To further explore the expression of B7-H3 in blood, we used ELISA to quantitatively measure the soluble concentration of B7-H3 in peripheral blood samples in this study. The results showed that sB7-H3 expression was significantly up-regulated in the peripheral blood of PTC patients compared with healthy donors. Moreover, the multivariate analysis also suggested that sB7-H3 expression was an independent risk factor affecting LNM in PTC patients. Therefore, sB7-H3 in peripheral blood of PTC patients might be used as a new diagnostic marker for future screening of high-risk patients.

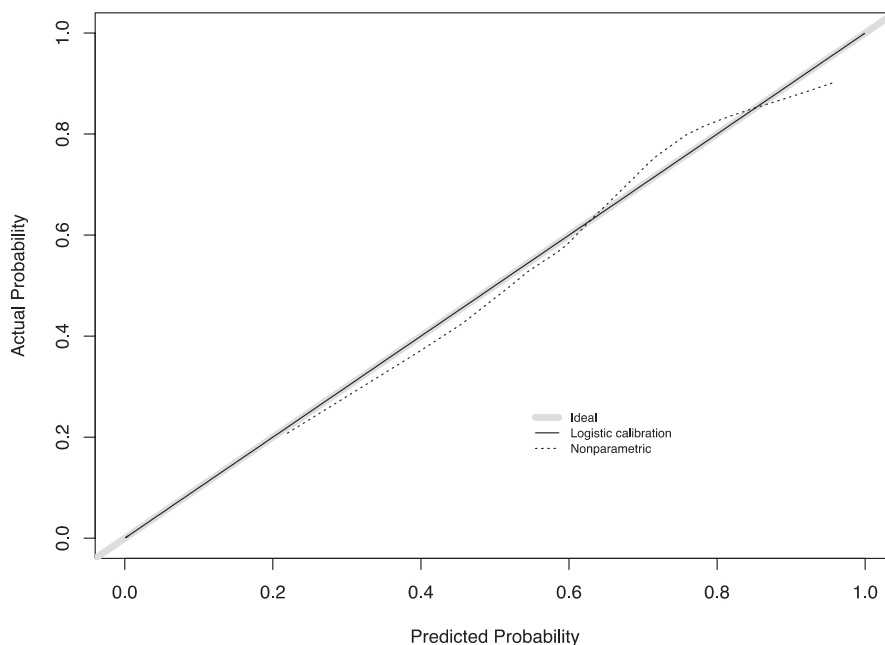


FIGURE 4 Calibration curve for the nomogram of LNM. The X-axis showed the predicted probability of LNM. The Y-axis showed the actual probability of LNM. LNM, lymph node metastasis

Nomogram had the advantage of accurately predicting the risk of LNM in PTC patients which has been used in diagnosing of thyroid carcinoma.³² In this study, gender, age, tumor size, number of tumors and sB7-H3 expression were considered as potential risk factors for PTC with LNM. Nomogram in this study was composed of clinical characteristics, preoperative ultrasound characteristics, and sB7-H3 expression which could be used to assess the risk of LNM before surgery. When nomogram showed a high risk of LNM, surgeons can perform cervical lymph node dissection in combination with intraoperative exploration and intraoperative frozen pathological results. For example, a 25-year-old (79 scores) woman (0 scores), of which the preoperative ultrasound showed multiple malignant nodules (55 scores) with the maximum diameter of more than 1 cm (100 scores). Her sB7-H3 expression level was high (70 scores). This patient's total score was 304. The probability of LNM was about 85% to 90%. Unlike previous studies, we used clinical characteristics, ultrasound characteristics and sB7-H3 expression to increase the accuracy of LNM prediction. It was more convenient and accurate to make a personalized diagnosis and treatment plan for patients. This study had some limitations. Firstly, this study lacked postoperative patients' peripheral blood samples to evaluate the dynamic changes of sB7-H3, which was to monitor recurrence and predict prognosis. Secondly, this study lacked patients' peripheral blood samples with benign thyroid nodules as a control to evaluate the feasibility of sB7-H3 in diagnosing of benign and malignant thyroid nodules. In addition, the sample size of this study was insufficient and the follow-up time was short, which might cause some bias. Therefore, the multicenter prospective study was the direction of our future research.

In conclusion, the nomogram is helpful in assessing the risk of LNM in PTC. sB7-H3 has excellent potential in predicting LNM in patients with PTC as an adjunctive ultrasound tool.

AUTHOR CONTRIBUTION

YH and ZH designed the study, analyzed the patient's data, and drafted the manuscript. YH and HC performed the ELIZA. ZH, GZ, SW, DY, XZ and CA collected the data. ZL and LN supervised the study. All authors contributed to the article and approved the submitted version.

ACKNOWLEDGMENTS

We thank all the donors who participated in our study.

FUNDING INFORMATION

This work was supported by a grant provided by the Capital Clinical Features Application Research (Z171100001017211).

CONFLICT OF INTEREST

All authors declare that there is no conflict of interest.

DATA AVAILABILITY STATEMENT

The data used and/or analyzed in this study are available from the corresponding author via reasonable request.

ORCID

Lingdun Zhuge  <https://orcid.org/0000-0003-4422-3591>

Changming An  <https://orcid.org/0000-0002-8353-4547>

Zhengjiang Li  <https://orcid.org/0000-0002-6792-5257>

REFERENCES

- Burgess JR, Tucker P. Incidence trends for papillary thyroid carcinoma and their correlation with thyroid surgery and thyroid fine-needle aspirate cytology. *Thyroid*. 2006;16(1):47-53.
- Sung H, Ferlay J, Siegel RL, et al. Global cancer statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *CA Cancer J Clin*. 2021;71(3):209-249.
- Zheng R, Zhang S, Zeng H, et al. Cancer incidence and mortality in China, 2016. *J Natl Cancer Center*. 2022;2(1):1-9.
- Sipos JA, Mazzaferri EL. Thyroid cancer epidemiology and prognostic variables. *Clin Oncol (R Coll Radiol)*. 2010;22(6):395-404.
- Haugen BR. 2015 American Thyroid Association management guidelines for adult patients with thyroid nodules and differentiated thyroid cancer: what is new and what has changed? *Cancer*. 2017;123(3):372-381.
- Mulla MG, Knoefel WT, Gilbert J, McGregor A, Schulte KM. Lateral cervical lymph node metastases in papillary thyroid cancer: a systematic review of imaging-guided and prophylactic removal of the lateral compartment. *Clin Endocrinol (Oxf)*. 2012;77(1):126-131.
- Wang Y, Deng C, Shu X, et al. Risk factors and a prediction model of lateral lymph node metastasis in CNO papillary thyroid carcinoma patients with 1-2 central lymph node metastases. *Front Endocrinol (Lausanne)*. 2021;12:716728.
- Greenwald RJ, Freeman GJ, Sharpe AH. The B7 family revisited. *Annu Rev Immunol*. 2005;23:515-548.
- Chapoval AI, Ni J, Lau JS, et al. B7-H3: a costimulatory molecule for T cell activation and IFN-gamma production. *Nat Immunol*. 2001;2(3):269-274.
- Luo L, Chapoval AI, Flies DB, et al. B7-H3 enhances tumor immunity in vivo by costimulating rapid clonal expansion of antigen-specific CD8+ cytolytic T cells. *J Immunol*. 2004;173(9):5445-5450.
- Lupu CM, Eisenbach C, Kuefner MA, et al. An orthotopic colon cancer model for studying the B7-H3 antitumor effect in vivo. *J Gastrointest Surg*. 2006;10(5):635-645.
- Brunner A, Hinterholzer S, Riss P, Heinze G, Brustmann H. Immunoexpression of B7-H3 in endometrial cancer: relation to tumor T-cell infiltration and prognosis. *Gynecol Oncol*. 2012;124(1):105-111.
- Fauci JM, Straughn JM Jr, Ferrone S, et al. A review of B7-H3 and B7-H4 immune molecules and their role in ovarian cancer. *Gynecol Oncol*. 2012;127(2):420-425.
- Liu Y, Vlatkovic L, Sæter T, et al. Is the clinical malignant phenotype of prostate cancer a result of a highly proliferative immune-evasive B7-H3-expressing cell population? *Int J Urol*. 2012;19(8):749-756.
- Qin X, Zhang H, Ye D, Dai B, Zhu Y, Shi G. B7-H3 is a new cancer-specific endothelial marker in clear cell renal cell carcinoma. *Oncotargets Ther*. 2013;6:1667-1673.
- Roth TJ, Sheinin Y, Lohse CM, et al. B7-H3 ligand expression by prostate cancer: a novel marker of prognosis and potential target for therapy. *Cancer Res*. 2007;67(16):7893-7900.
- Zang X, Thompson RH, Al-Ahmadie HA, et al. B7-H3 and B7x are highly expressed in human prostate cancer and associated with disease spread and poor outcome. *Proc Natl Acad Sci USA*. 2007;104(49):19458-19463.
- Zhao B, Huang Z, Zhu X, et al. Clinical significance of the expression of Co-stimulatory molecule B7-H3 in papillary thyroid carcinoma. *Front Cell Dev Biol*. 2022;10:819236.

19. Zhang G, Hou J, Shi J, Yu G, Lu B, Zhang X. Soluble CD276 (B7-H3) is released from monocytes, dendritic cells and activated T cells and is detectable in normal human serum. *Immunology*. 2008;123(4):538-546.
20. Rui ZY, Liu Y, Zheng W, et al. A retrospective study of the risk factors and the prognosis in patients with papillary thyroid carcinoma depending on the number of lymph node metastasis. *Clin Exp Med*. 2021;21(2):277-286.
21. Luo Y, Zhao Y, Chen K, et al. Clinical analysis of cervical lymph node metastasis risk factors in patients with papillary thyroid microcarcinoma. *J Endocrinol Invest*. 2019;42(2):227-236.
22. Lee DJ, Lee KH, Kim JH, Kwon KH, Yoon DY, Rho YS. Posterosuperior lesion has a high risk of lateral and central nodal metastasis in solitary papillary thyroid cancer. *World J Surg*. 2015;39(2):387-392.
23. Suteau V, Munier M, Briet C, Rodien P. Sex bias in differentiated thyroid cancer. *Int J Mol Sci*. 2021;22(23):12992.
24. Nie X, Tan Z, Ge M, Jiang L, Wang JF, Zheng CM. Risk factors analyses for lateral lymph node metastases in papillary thyroid carcinomas: a retrospective study of 356 patients. *Arch Endocrinol Metab*. 2016;60(5):492-499.
25. Siddiqui S, White MG, Antic T, et al. Clinical and pathologic predictors of lymph node metastasis and recurrence in papillary thyroid microcarcinoma. *Thyroid*. 2016;26(6):807-815.
26. Hu D, Zhou J, He W, et al. Risk factors of lateral lymph node metastasis in cNO papillary thyroid carcinoma. *World J Surg Oncol*. 2018;16(1):30.
27. Zhao L, Sun X, Luo Y, Wang F, Lyu Z. Clinical and pathologic predictors of lymph node metastasis in papillary thyroid microcarcinomas. *Ann Diagn Pathol*. 2020;49:151647.
28. Cheng F, Chen Y, Zhu L, et al. Risk factors for cervical lymph node metastasis of papillary thyroid microcarcinoma: a single-center retrospective study. *Int J Endocrinol*. 2019;2019:8579828.
29. Ng SC, Kuo SF, Chen ST, et al. Therapeutic outcomes of patients with multifocal papillary thyroid microcarcinomas and larger tumors. *Int J Endocrinol*. 2017;2017:4208178.
30. Lombardi CP, Bellantone R, De Crea C, et al. Papillary thyroid microcarcinoma: extrathyroidal extension, lymph node metastases, and risk factors for recurrence in a high prevalence of goiter area. *World J Surg*. 2010;34(6):1214-1221.
31. Kuhn E, Teller L, Piana S, Rosai J, Merino MJ. Different clonal origin of bilateral papillary thyroid carcinoma, with a review of the literature. *Endocr Pathol*. 2012;23(2):101-107.
32. Feng JW, Hong LZ, Wang F, et al. A nomogram based on clinical and ultrasound characteristics to predict central lymph node metastasis of papillary thyroid carcinoma. *Front Endocrinol (Lausanne)*. 2021;28(12):666315. doi:10.3389/fendo.2021.666315

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Huang Y, Huang Z, Cai H, et al. Evaluation of serum B7-H3 expression, ultrasound and clinical characteristics to predict the risk of cervical lymph node metastases in papillary thyroid carcinoma by nomogram. *J Clin Lab Anal*. 2023;37:e24811. doi:10.1002/jcla.24811