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Prostate-specific Membrane Antigen Expression Is Associated with Aggressive Clinicopathological Features and Short-term Incomplete Response to Initial Radioiodine Therapy in Papillary Thyroid Carcinoma: A Retrospective Single-Center Cohort Study

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Abstract

Background Prostate-specific membrane antigen (PSMA) is expressed in tumor-associated neovasculature and has been linked to aggressive disease and incomplete response to radioiodine (RAI) therapy in thyroid cancer, but validation in larger cohorts is limited. We investigated the association between endothelial PSMA expression and aggressive clinicopathological features in papillary thyroid carcinoma (PTC), and its association with initial RAI therapy response.

Methods This retrospective single-center cohort study included 241 patients with classical PTC who underwent total thyroidectomy and initial RAI therapy. PSMA expression was assessed by immunohistochemistry in tumor endothelial cells using a composite score and categorized as none, low, medium, or high. Treatment response was classified according to the 2015 American Thyroid Association guidelines; excellent and indeterminate responses were grouped as favorable response, and biochemical and structural incomplete responses as incomplete response. Univariate and multivariate logistic regression analyses were performed.

Results PSMA was expressed predominantly in tumor-associated endothelial cells and was absent in PTC cells, normal thyroid tissue, and Hashimoto's thyroiditis; PSMA expression was detectable in 80.1% of patients. Higher PSMA expression was significantly associated with larger tumor size, multifocality, capsular invasion, extrathyroidal extension, aggressive histological variants, higher Ki-67 index, and higher recurrence risk stratification (all $P < 0.05$). In multivariate analysis, higher PSMA expression, high-risk recurrence stratification (vs intermediate-risk), and higher Ki-67 index were independently associated with short-term incomplete response to initial RAI at 6–12 months (all $P < 0.05$). The incomplete response rate increased from 12.1% in the low-PSMA group to 50.0% in the high-PSMA group. In the high-risk subgroup, nodal metastasis, higher Ki-67 index, and higher PSMA expression remained significantly associated with incomplete response.

Conclusions In this retrospective PTC cohort, endothelial PSMA expression was associated with aggressive clinicopathological features and independently with short-term incomplete response to initial RAI at 6–12 months. These findings support PSMA as a candidate stromal/vascular biomarker for further investigation. Given its predominantly vascular expression and short follow-up, prospective long-term studies are needed to define its potential clinical utility in refining risk stratification.

Introduction

Thyroid cancer (TC) has become one of the most prevalent endocrine malignancies, with a rising global incidence in recent years [1]. In China, incidence has increased by 20% annually, establishing TC as the third most frequent malignancy [2]. Histopathologically, TCs are classified into papillary (PTC, ~ 90% of cases), follicular (FTC), anaplastic, and medullary subtypes, with PTC and FTC comprising differentiated thyroid carcinoma (DTC) [3].

Although most PTC follow an indolent course with favorable responses to standard therapy (surgery, radioiodine ablation, and thyroid stimulating hormone suppression), 15–30% of patients develop radioiodine-refractory disease or metastatic recurrence [4]. Angiogenesis plays a crucial role in tumor progression, particularly in advanced and radioactive iodine-refractory DTC (RAIR-DTC) [5]. Prostate-specific membrane antigen (PSMA), a type II transmembrane glycoprotein, is extensively expressed in neovascular endothelial cells in several different cancers, including DTC [6]. Previous immunohistochemistry (IHC) studies observed that PSMA expression in the neovasculature of DTC tumors was linked to an increased probability of displaying clinically aggressive behavior and that individuals who have DTC lesions exhibiting a significant level of PSMA expression are more susceptible to developing resistance to radioiodine (RAI) therapy and increased disease-specific mortality [7, 8]. However, previous studies have largely been pilot studies with a small number of patient samples, leading to patient selection bias and heterogeneity, which have hindered the determination of the diagnostic and therapeutic value of PSMA-targeted radiopharmaceuticals for TC. To address this, we conducted a large-scale study to investigate the association between PSMA expression and aggressive clinicopathological features in PTC. Furthermore, we evaluated its potential utility as a predictive biomarker for treatment response to initial RAI therapy.

Materials and methods

Patient selection

We conducted a retrospective analysis of 241 consecutive patients with PTC who underwent total thyroidectomy followed by RAI therapy at Yantai Yuhuangding Hospital between November 2022 and November 2023. Inclusion criteria required: (1) histopathological confirmation of classical PTC (excluding follicular variant); (2) complete hematoxylin and eosin (HE) staining, CD31, and PSMA immunohistochemical data; and (3) absence of distant metastases. Patients with incomplete clinical data or concurrent malignancies were excluded.

Collected clinicopathological parameters included: Demographic characteristics (age, sex), Tumor features (maximum diameter, bilaterality, multifocality), Invasive characteristics (capsular invasion, extrathyroidal extension), Nodal status (N0/N1a/N1b), Histological subtype [aggressive (tall-cell, hobnail, columnar) vs. non-aggressive], Ki-67 index recorded in three tiers (<5%, 5%-10%, and ≥ 10%) for descriptive baseline comparisons, Initial recurrence risk stratification.

Reagents and Immunohistochemical Methods

The following primary antibodies were used for immunohistochemical staining: HE staining kit (Roche, BenchMark XT), Anti-CD31 antibody (Roche, BenchMark XT), Anti-PSMA antibody (Shanghai Yonyiwei Biotechnology Co., Ltd.).

Immunohistochemical Analysis

For PTC specimens obtained from total thyroidectomies at our institution, HE and CD31 staining were performed in-house. Representative tumor sections were subsequently processed for PSMA immunohistochemistry at Shanghai Youningwei Biotechnology Co., Ltd. All slides were independently evaluated by two board-certified pathologists from our hospital, with consensus interpretation of staining results.

Quantification of MVD and PSMA Expression

Intratumoral microvessel count values were calculated following previously described protocols [9–11]. Microvessel density (MVD) was assessed using CD31 immunohistochemistry. Three vascular “hotspots” (areas of highest microvessel density) were identified per slide, excluding tumor sclerosis areas and vessels > 8 erythrocyte diameters. MVD was calculated as the mean vessel count across these hotspots.

PSMA expression was evaluated in the same hotspots, with staining intensity graded: 0 (negative), 1 (weak), 2 (moderate), and 3 (strong). The PSMA-positive vessel percentage relative to CD31-positive vessels was categorized: 0% (0), 1–25% (1), 26–50% (2), 51–75% (3), and > 75% (4). The composite PSMA score (intensity + percentage) defined expression levels: 0 (none), 1–4 (low), 5–6 (medium), and 7 (high).

Treatment and Follow-up Protocol

Treatment and Follow-up Protocol
Prior to initial radioiodine therapy, all participants maintained a strict low-iodine diet for 2–4 weeks and discontinued medications affecting iodine metabolism. Enhanced computed tomography (CT) examinations were prohibited for over one month to prevent iodine contamination. Levothyroxine supplementation was withdrawn for 3–4 weeks to achieve therapeutic TSH levels exceeding 30 mIU/L. Radioiodine dosages were individualized according to 2015 American Thyroid Association (ATA) guidelines and patient-specific risk factors [12]. Follow-up evaluations at 6–12 months post-treatment incorporated comprehensive assessments including: serum biomarkers (TSH, thyroglobulin [Tg], and thyroglobulin antibodies [TgAb]), complete blood count, cervical ultrasonography, thoracic/neck CT, thyroid scintigraphy, and diagnostic whole-body RAI scanning (Dx-WBS).

Laboratory and Imaging Parameters

Standard reference values were established as follows: TSH 0.27–4.20 mIU/L, Tg 3.5–77 ng/mL, and TgAb 0-115 IU/mL. Imaging studies were considered negative when demonstrating no evidence of malignant lesions across all modalities: cervical/thoracic ultrasonography and CT, Dx-WBS, whole-body bone scintigraphy, and PET/CT when indicated. Two independent specialists from radiology and nuclear

medicine departments conducted blinded interpretations of all imaging results. PSMA immunohistochemical analysis followed previously established protocols.

Treatment Response Assessment

Treatment outcomes were categorized according to 2015 ATA guidelines into four distinct classes: 1) Excellent response (ER) required both negative imaging findings and suppressed Tg < 0.2 ng/mL or stimulated Tg < 1.0 ng/mL in TgAb-negative patients; 2) Indeterminate response (IDR) was designated for cases showing either nonspecific imaging findings or minimal thyroid bed uptake on Dx-WBS, accompanied by suppressed Tg 0.2-1.0 ng/mL or stimulated Tg 1.0–10.0 ng/mL with stable/declining TgAb levels; 3) Biochemical incomplete response (BIR) was assigned when imaging remained negative despite abnormal Tg levels (suppressed ≥ 1.0 ng/mL or stimulated ≥ 10.0 ng/mL) or progressively increasing TgAb; 4) Structural incomplete response (SIR) was confirmed by definitive structural/functional evidence of disease regardless of Tg/TgAb status. For analytical purposes, ER and IDR were grouped as favorable response (AR), while BIR and SIR constituted incomplete response (IR) [12].

Statistical Analysis

Statistical analyses were performed using SPSS 26.0 (IBM Corp., Armonk, NY, USA). Categorical variables were presented as frequencies and percentages. The association between PSMA expression and clinicopathological characteristics was assessed using non-parametric tests: the Wilcoxon rank-sum test for comparisons between two groups and the Kruskal-Wallis H test for multiple group comparisons. Post-hoc pairwise analyses were conducted using the Mann-Whitney U test with Bonferroni correction for multiple comparisons. The association between clinicopathological characteristics and initial radioiodine treatment response was assessed using binary logistic regression analysis. Both univariate and multivariate analyses were performed to evaluate potential predictors of treatment outcome. Results were expressed as odds ratios (OR) with corresponding 95% confidence intervals (CIs). PSMA expression was treated as an ordinal categorical variable (none, low, medium, high), with the none group as the reference. Ki-67 was treated as a categorical variable (< 5%, 5–10%, ≥ 10%), with < 5% as the reference. Recurrence risk was referenced to intermediate-risk. Lymph node metastasis was referenced to N0. Extrathyroidal extension was referenced to absent. Statistical significance was defined as a two-tailed *P* value < 0.05.

Results

Primary objective: DTC aggressiveness prediction

Clinical Characteristics

The cohort included 241 patients (60.2% female, 39.8% male), with 71.4% aged < 55 years. Most tumors were > 1.0 cm (72.6%), multifocal (72.6%), and exhibited capsular invasion (87.6%). Extrathyroidal extension was present in 44.4%, and lymph node metastasis was nearly universal (96.7%), predominantly N1b (76.8%). Histologically, 95.0% were non-aggressive PTC, while 5.0% comprised aggressive subtypes. The Ki-67 index was < 5% in 73.0%, 5% to < 10% in 18.7%, and ≥ 10% in 8.3% of patients. All patients were stratified as intermediate- (49.4%) or high-risk (50.6%) for recurrence, with no low-risk patients (Table 1).

Table 1
Comparative analysis of baseline clinicopathological characteristics and PSMA expression levels in PTC patients

Variable	Category	PSMA Expression				Test Statistic	*P* value
		None	Lowwwww	Medium	High		
Age							
< 55 years	172	41	48	71	12	-1.757	0.079
≥ 55 years	69	7	23	33	6		
Gender							
Female	145	28	45	58	14	-0.247	0.805
Male	96	20	26	46	4		
The longest diameter of the primary lesion							
≤1.0cm	66	29	23	13	1	-6.344	< 0.001
>1.0cm	175	19	48	91	17		
Lobe involvement							
Unilateral	89	22	30	32	5	-2.155	0.031
Bilateral	152	26	41	72	13		
Multifocality							
Unifocal	66	18	21	23	4	-1.996	0.046
Multifocal	175	30	50	81	14		
Capsular invasion							
Absent	30	15	10	5	0	-4.635	< 0.001
Present	211	33	61	99	18		
Extrathyroidal extension							
Absent	134	48	54	29	3	-9.546	< 0.001
Present	107	0	17	75	15		
Lymph node metastasis							
N0	8	1	2	4	1	1.674	0.433
N1a	48	7	15	21	5		

Variable	Category	PSMA Expression				Test Statistic	*P* value
		None	Lowwwww	Medium	High		
N1b	185	40	54	79	12		
Histological type							
Non-aggressive	229	47	70	96	16	-2.227	0.026
Aggressive	12	1	1	8	2		
Recurrence risk stratification							
Low-risk	0	0	0	0	0	-9.132	< 0.001
Intermediate-risk	119	44	50	22	3		
High-risk	122	4	21	82	15		
Ki-67 index							
< 5%	176	47	67	62	0	93.024	< 0.001
5% to < 10%	45	1	4	30	10		
≥ 10%	20	0	0	12	8		

Association Between PSMA Expression and Clinicopathological Characteristics in PTC

PSMA was exclusively expressed in tumor stromal endothelium, absent in PTC cells, normal tissue, and Hashimoto's thyroiditis. This study analyzed 241 PTC tissue specimens, revealing PSMA expression in 193 cases (80.1%), with expression levels distributed as follows: low (71 cases, 29.5%), moderate (104 cases, 43.2%), and high (18 cases, 7.5%); PSMA was absent in 48 cases (19.9%). PSMA expression demonstrated no significant correlation with patient age, gender, or lymph node metastasis (test statistics: -1.757, -0.247, and 1.674, respectively; all $P > 0.05$). However, significant associations were observed with several aggressive clinicopathological features, including primary tumor diameter (maximum dimension), thyroid lobe involvement, multifocality, capsular invasion, extrathyroidal extension, aggressive histological variants, Ki-67 proliferation index, and initial recurrence risk stratification (test statistic range: -9.546 to 93.024; all $P < 0.05$) (Table 1). Aggressive histological variants showed stronger PSMA expression than non-aggressive PTC.

Furthermore, patients with high Ki-67 expression exhibited significantly higher PSMA expression than those with low Ki-67 expression (Fig. 1). Spearman correlation analysis revealed a significant positive correlation between PSMA expression and the Ki-67 index in differentiated thyroid cancer lesions ($\rho = 0.56$, $P < 0.05$). And, among patients with multifocal papillary thyroid carcinoma who exhibited

intrapatient heterogeneity in the Ki-67 proliferation index, PSMA expression was significantly higher in lesions with a high Ki-67 index compared to lesions with a low Ki-67 index within the same patient.

Secondary objective: Initial response to RAI treatment prediction

Univariate logistic regression analysis of each variable and the initial response to RAI treatment

Due to the absence of follow-up data after RAI treatment in 22 patients, the final analysis comprised 219 participants. Clinicopathological features of the 219 patients included in the analysis of initial RAI treatment response are shown in Table 2. Treatment response stratified by PSMA expression levels showed that the low-expression group comprised 58 (87.9%) AR and 8 (12.1%) IR cases (n = 66), the medium-expression group comprised 66 (69.5%) AR and 29 (30.5%) IR cases (n = 95), the high-expression group comprised 9 (50.0%) AR and 9 (50.0%) IR cases (n = 18), and the “None” PSMA expression category included 40 cases (35 AR and 5 IR).

Table 2
Results of the univariate and multivariate logistic regression analysis for prediction of the initial treatment response of RAI in PTC

Variable	Initial treatment response of RAI		Univariate logistic regression		Multivariate logistic regression	
	AR					
	AR	IR	OR (95% CI)	*P* value	OR (95% CI)	*P* value
Age						
< 55 years	128	33	1.690 (0.861–3.314)	0.127	-	-
≥ 55 years	40	18				
Gender						
Female	105	30	0.857 (0.452–1.624)	0.636	-	-
Male	63	21				
The longest diameter of the primary lesion						
≤1.0cm	48	10	1.640 (0.761–3.535)	0.207	-	-
>1.0cm	120	41				
Lobe involvement						
Unilateral	67	18	1.216 (0.634–2.334)	0.556	-	-
Bilateral	101	33				
Multifocality						
Unifocal	49	15	0.988 (0497-1.967)	0.973	-	-
Multifocal	119	36				
Capsular invasion						
Absent	20	8	0.726 (0.299–1.764)	0.480	-	-
Present	148	43				
Extrathyroidal extension						
Absent	107	24	1.973	0.035	0.591	0.310

Variable	Initial treatment response of RAI AR		Univariate logistic regression		Multivariate logistic regression	
	AR	IR	OR (95% CI)	*P* value	OR (95% CI)	*P* value
Present	61	27	(1.047–3.718)		(0.214–1.633)	
Lymph node metastasis						
N0	8	0	1.724	0.133	2.119	0.05
N1a	35	9	(0.847–3.510)		(0.999–4.491)	
N1b	125	42				
Recurrence risk stratification						
Low-risk	0	0	3.099	0.001	3.042	0.043
Intermediate-risk	94	15	(1.552–5.987)		(1.037–8.924)	
High-risk	74	36				
PSMA expression						
None	35	5	2.148	< 0.001	1.873	0.01
Low	58	8	(1.424–3.241)		(1.159–3.026)	
Medium	66	29				
High	9	9				
Ki-67 index						
< 5%	141	17	5.984	< 0.001	6.051	< 0.001
5% to < 10%	23	19	(3.457–10.357)		(3.010-12.163)	
≥ 10%	4	15				

Univariate analysis revealed that PSMA expression levels (OR = 2.148, 95% CI 1.424–3.241, $P < 0.001$), Ki-67 index (OR = 5.984, 95% CI 3.457–10.357, $P < 0.001$), extrathyroidal extension (OR = 1.973, 95% CI 1.047–3.718, $P = 0.035$), and initial recurrence risk stratification (OR = 3.099, 95% CI 1.552–5.987, $P = 0.001$) were significantly associated with the initial RAI treatment response. In contrast, no significant associations were observed for patient age, gender, maximum tumor diameter, thyroid lobe involvement, capsular invasion, multifocality, or lymph node metastasis (all $P > 0.05$).

Multivariate Logistic Regression Analysis

Variables demonstrating statistical significance in univariate analysis (PSMA expression levels, Ki-67 index, extrathyroidal extension, and initial recurrence risk stratification) were included as independent variables in the multivariate model. Although lymph node metastasis showed no significant association with initial RAI treatment response in univariate analysis ($P > 0.05$), it was incorporated into the multivariate analysis due to its established prognostic value in PTC outcomes. The final multivariable model identified three independent predictors of incomplete treatment response: higher PSMA expression (OR = 1.873, 95% CI 1.159–3.026, $P = 0.01$), elevated Ki-67 index (OR = 6.051, 95% CI 3.010-12.163, $P < 0.001$), and high-risk recurrence stratification compared with intermediate-risk (OR = 3.042, 95% CI 1.037–8.924, $P = 0.043$). Extrathyroidal extension (OR = 0.591, 95% CI 0.214–1.633, $P = 0.310$) and lymph node metastasis (OR = 2.119, 95% CI 0.999–4.491, $P = 0.05$) were not independently associated with incomplete treatment response in the multivariable model (Table 2).

Logistic Regression Analysis of Variables Associated with Initial RAI Treatment Response in the High-Risk Recurrence Group

Based on the elevated recurrence and mortality risks associated with high-risk stratification in PTC, a cohort of 110 high-risk PTC patients was analyzed to assess the association between initial RAI therapy efficacy and various aggressive pathological factors. Univariate and multivariate analyses consistently identified lymph node metastasis (univariate OR = 2.432, 95% CI 1.057–5.598, $P = 0.037$; multivariate OR = 2.525, 95% CI 1.075–5.927, $P = 0.033$), elevated Ki-67 index (univariate OR = 5.599, 95% CI 2.832–11.070, $P < 0.001$; multivariate OR = 5.257, 95% CI 2.429–11.381, $P < 0.001$), and higher PSMA expression (univariate OR = 2.278, 95% CI 1.175–4.418, $P = 0.015$; multivariate OR = 2.361, 95% CI 1.192–4.678, $P = 0.014$) as significant factors associated with an unfavorable initial response to RAI therapy in high-risk PTC patients (Table 3). Other variables, including age, gender, tumor diameter, lobe involvement, multifocality, capsular invasion, and extrathyroidal extension, were not significantly associated with initial RAI response in either univariate or multivariate analysis (all $P > 0.05$).

Table 3
Regression Analysis of Variables Associated with Initial RAI Treatment Response in High-Risk Patients

Variable	Initial treatment response of RAI AR		Univariate logistic regression		Multivariate logistic regression	
	AR	IR	OR (95% CI)	*P* value	OR (95% CI)	*P* value
Age						
< 55 years	34	17	1.526 (0.651–3.577)	0.331	-	-
≥ 55 years	40	19				
Gender						
Female	50	23	0.849 (0.368–1.960)	0.702	-	-
Male	24	13				
The longest diameter of the primary lesion						
≤1.0cm	12	5	1.200 (0.388–3.710)	0.752	-	-
>1.0cm	62	31				
Lobe involvement						
Unilateral	28	11	1.383 (0.591–3.239)	0.455	-	-
Bilateral	46	25				
Multifocality						
Unifocal	20	11	0.842 (0.351–2.020)	0.700	-	-
Multifocal	54	25				
Capsular invasion						
Absent	1	3	0.151 (0.015–1.503)	0.107	-	-
Present	73	33				
Extrathyroidal extension						
Absent	13	9	0.639	0.363	-	-

Variable	Initial treatment response of RAI AR		Univariate logistic regression		Multivariate logistic regression	
	AR	IR	OR (95% CI)	*P* value	OR (95% CI)	*P* value
Present	61	27	(0.244–1.675)			
Lymph node metastasis						
N0	7	0	2.432	0.037	2.525	0.033
N1a	20	7	(1.057–5.598)		(1.075–5.927)	
N1b	47	29				
PSMA expression						
None	2	2	2.278	0.015	2.361	0.014
Low	23	3	(1.175–4.418)		(1.192–4.678)	
Medium	44	22				
High	5	9				
Ki-67 index			5.599	< 0.001	5.257	< 0.001
< 5%	54	8	(2.832–11.070)		(2.429–11.381)	
5% to 10%	17	14				
≥ 10%	3	14				

Discussion

The principal findings of this study highlight the potential dual significance of PSMA in PTC management. First, PSMA expression was associated with intrinsic tumor aggressiveness, and second, it was independently associated with short-term incomplete response to initial RAI therapy. Our results support PSMA as a candidate stromal/vascular biomarker in PTC and suggest its potential utility in risk stratification, although further validation is needed.

Our study demonstrates that PSMA expression was exclusively localized to the tumor stromal endothelium, with no detectable expression in PTC cells, normal thyroid parenchyma, or Hashimoto's thyroiditis. This observation aligns with the established paradigm that PSMA expression in non-prostatic malignancies is a specific feature of the neovasculature that sustains tumor growth [10, 13]. The high prevalence of PSMA expression (80.1%) in our cohort of 241 PTC patients suggests that active angiogenesis is a common event in PTC pathogenesis. Importantly, the intensity of endothelial PSMA

expression was significantly associated with several clinicopathological hallmarks of aggression. Tumors with larger diameter, multifocality, capsular invasion, extrathyroidal extension (ETE), and aggressive histological variants demonstrated significantly higher PSMA expression levels. This result concurs with earlier findings from preliminary studies [7, 14].

Moreover, the finding that patients with higher Ki-67 expression exhibited significantly higher PSMA expression, together with the histological association of elevated PSMA in multifocal lesions with a high Ki-67 index [15], suggests a link between PSMA-positive vasculature and biologically more aggressive, proliferative tumor clones. These findings support the hypothesis that more aggressive PTC subtypes may drive the formation of a richer, more metabolically active vascular network, characterized by high PSMA expression, to support their growth and invasive behavior, consistent with the concept of the “angiogenic switch” in solid tumors [16, 17]. These correlations suggest that PSMA expression may serve as a marker of a tumor’s invasive potential.

Beyond its role as a pathological marker of aggressiveness, our investigation yields a potentially clinically relevant insight: PSMA expression was independently associated with short-term incomplete response to initial RAI therapy. In our cohort, the rate of incomplete response increased with increasing PSMA expression, from 12.1% in the low-expression group to 50.0% in the high-expression group. While ETE was significant in univariate analysis, the multivariate logistic regression model identified PSMA expression level, Ki-67 index, and initial recurrence risk stratification as independent predictors of treatment response. In the subgroup of high-risk patients, lymph node metastasis, Ki-67 index, and PSMA expression remained significantly associated with initial RAI response in both univariate and multivariate analyses (Table 3), further supporting these associations in aggressive disease.

The high-PSMA vascular phenotype may share clinical significance with the aggressive biology associated with incomplete response. Therefore, a high PSMA score on routine immunohistochemistry might serve as an indirect proxy for identifying tumors with a higher likelihood of short-term incomplete response to RAI. However, this short-term response (assessed at 6–12 months) does not equate to true radioiodine refractoriness. Any speculation regarding more extensive initial surgery or earlier use of adjuvant therapies (e.g., external beam radiotherapy or tyrosine kinase inhibitors) should be viewed as exploratory and warrants prospective validation in larger cohorts. Placing our results in a broader context, they contribute histopathological evidence to the growing literature on PSMA PET-CT imaging in non-prostate malignancies, including thyroid cancer. Our data may help explain the avidity observed in aggressive PTC metastases on PSMA PET scans and suggest that this modality could have potential value for restaging patients with suspected incomplete response. Nevertheless, head-to-head comparisons with ^{18}F-FDG PET-CT and long-term follow-up are needed to establish its clinical utility, particularly given the retrospective nature of our study.

Several limitations of our study warrant acknowledgment. Its retrospective, single-center design necessitates validation in a prospective, multi-institutional setting. The relatively small number of patients in the high-PSMA expression group (n = 18) limits the precision of the estimates for this

subgroup. In addition, the high-risk subgroup analysis included 110 patients; these limited event numbers may increase the risk of sparse-data bias and model overfitting. The generalizability of our findings to populations with a different distribution of risk factors needs to be established. Furthermore, the precise molecular crosstalk between aggressive tumor cells and endothelial cells that leads to upregulated PSMA expression remains to be fully elucidated. Future research should focus on the pathways regulating PSMA in tumor endothelium. Given the predominantly vascular expression of PSMA, whether there is a rationale for PSMA-targeted theranostics in PTC remains uncertain; this question requires further biological investigation.

Conclusions

In this PTC cohort, endothelial PSMA expression was associated with aggressive clinicopathological features and proliferative activity, and independently with short-term incomplete response to initial RAI at 6–12 months. These findings suggest that PSMA may be a candidate stromal/vascular biomarker for further investigation. However, short-term incomplete response should not be equated with true radioiodine-refractory disease, and the predominantly vascular expression pattern limits direct therapeutic inference. The present data are therefore hypothesis-generating and not yet sufficient to guide changes in initial management based on PSMA immunohistochemistry. Prospective studies with long-term follow-up are needed to define its potential clinical utility.

Declarations

Ethical approval

This retrospective study was conducted in accordance with the Declaration of Helsinki. The need for ethics approval and informed consent was waived by the Medical Ethics Committee of The Affiliated Yantai Yuhuangding Hospital of Qingdao University, in accordance with national regulations in China regarding the use of de-identified retrospective clinical data. All patient data were anonymized prior to analysis, and no direct patient contact was involved.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

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None.

Author Contribution

M.L. conceived the concept of the study. W.L. contributed to the design of the research. L.Y. contributed to data collection and data analysis. All authors edited and approved the final version of the manuscript.

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None.

Data Availability

The data that support the findings of this study are available from the corresponding author upon reasonable request. The data are not publicly available due to ethical and patient privacy restrictions.

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Figures

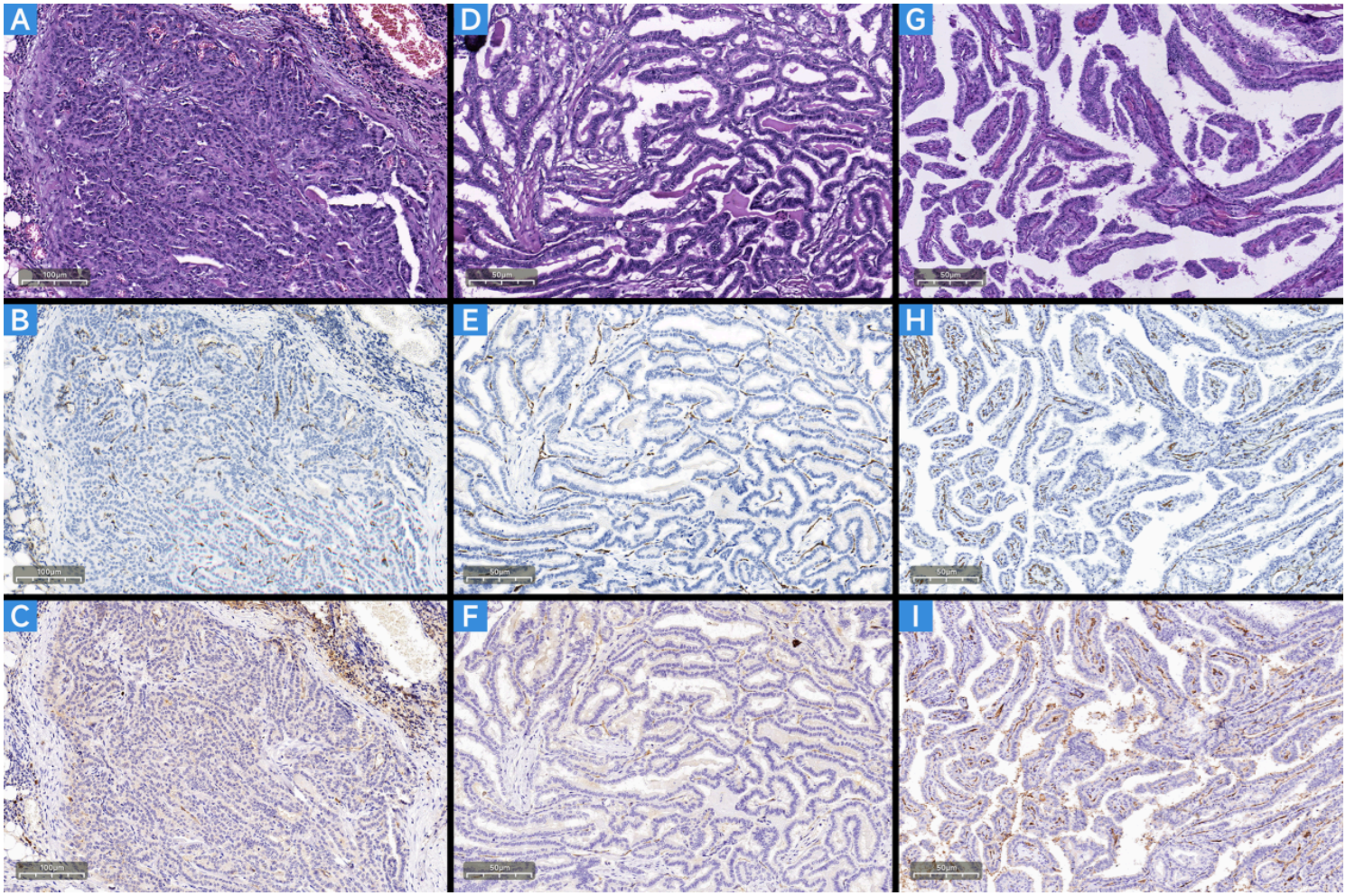


Figure 1

Representative histopathological and immunohistochemical images of PTC tissues stratified by Ki-67 proliferation index. (A–C) Low Ki-67 group: (A) H&E staining, (B) CD31 immunostaining, and (C) PSMA immunostaining (PSMA score 2). (D–F) Intermediate Ki-67 group: (D) H&E staining, (E) CD31 immunostaining, and (F) PSMA immunostaining (PSMA score 5). (G–I) High Ki-67 group: (G) H&E staining, (H) CD31 immunostaining, and (I) PSMA immunostaining (PSMA score 7).