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How Many Papillae in Conventional Papillary Carcinoma? A Clinical Evidence-Based Pathology Study of 235 Unifocal Encapsulated Papillary Thyroid Carcinomas, with Emphasis on the Diagnosis of Noninvasive Follicular Thyroid Neoplasm with Papillary-Like Nuclear Features

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Background: The percentage of papillae is a crucial criterion in differentiating noninvasive follicular thyroid neoplasm with papillary-like nuclear features (NIFTP) from papillary thyroid carcinomas (PTCs) and in subclassifying PTC into classic and follicular variant. Since the description of NIFTP, three studies have shown that the presence of any papillae may be associated with nodal metastasis, which led to modification of the NIFTP criterion from <1% papillae to no true papillae allowed. We aim at providing clinical evidence-based data on the impact that papillary growth has on nodal spread and tumor genotype in tumors previously diagnosed as encapsulated unifocal PTC.

Methods: A meticulous histopathologic examination was performed on 235 cases previously diagnosed as unifocal encapsulated PTC (U-EPTC). One hundred of these cases were subjected to $BRAF^{V600E}$ and $NRAS^{Q61R}$ immunohistochemistry.

Results: In our cohort, 27 patients (12%) had lymph node metastasis (N1) at the time of initial resection. Overall, 89% of the tumors in the N1 group contained ≥50% papillae, compared with 13% in the N0/Nx group. Nodal metastases were only present in tumors with ≥1% papillae. In noninvasive U-EPTC ($n = 161$), N1 disease was seen only in tumors with ≥10% papillae. A higher percentage of papillae within the tumor also correlated with an increased frequency of $BRAF^{V600E}$ and decreased rate of $NRAS^{Q61R}$. None of the 26 NRAS-positive cases had nodal disease, including the invasive tumors. Among 216 patients with follow-up (median: 5.2 years), 3 patients (1.5%) had distant metastases, all detected at the initial presentation. All three tumors displayed 100% follicular growth, and capsular or vascular invasion. There was no locoregional recurrence in the entire cohort.

Conclusion: In U-EPTC, there is a strong correlation between high percentage of papillary growth, presence of nodal metastasis, and BRAF+/RAS− genotype regardless of invasive status. Nodal metastases were not seen in tumors with <1% papillae irrespective of invasive status. These findings indicate that the initial criterion of <1% papillae is still valid for the diagnosis of NIFTP. Reinstituting this criterion will spare a carcinoma diagnosis and unnecessary therapy with its side effects on patients who have negligible clinical risk.

Keywords: encapsulated variant, papillary thyroid carcinoma, papillary, follicular

Introduction

Papillary thyroid carcinoma (PTC) is the most common thyroid cancer and it has shown a dramatic increase of incidence for the past several decades (1,2). For the past 40 years, the pathologic diagnosis of PTC has been based purely on its nuclear features: (i) enlarged elongated shape; (ii) chromatin clearing; and (iii) nuclear membrane irregularity (1,3,4). On the other hand, the subtyping of PTC factors depends on several parameters, including architectural pattern (e.g., follicular variant with follicular growth pattern and conventional [classic] PTC with well-formed papillae),

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cytomorphology (e.g., oncocytic variant, columnar variant, hobnail variant, and tall cell variant), encapsulation (e.g., encapsulated variant surrounded completely by a tumor capsule), and size (papillary microcarcinoma that measures 1 cm or less) (1,4). In particular, and pertinent to this study, follicular variants of PTC (FVPTC) are those PTCs with ''exclusively or almost exclusively follicular growth pattern'' (1,3) that have been traditionally translated into a cutoff of 1% papillae to separate FVPTC (<1% papillae) and conventional PTCs with a follicular predominant growth pattern (≥1% papillae) (4).

In 2014, a comprehensive genomic analysis by The Cancer Genome Atlas (TCGA) has shown that the follicular variant and conventional PTC have distinct molecular signatures. The FVPTC is associated with *RAS* mutations, an RAS-like molecular signature, and is highly differentiated with a high thyroid differentiation score (TDS). The classical PTC (CPTC) has a high frequency of $BRAF^{V600E}$ mutations, a BRAF-like molecular signature, and a low TDS (5). Clinically, FVPTC, especially the encapsulated and noninvasive form, tends to follow a highly indolent clinical course with negligible risk of recurrence and lymph node metastasis (3,6,7). Based on these compelling clinical and molecular evidence, a subset of noninvasive encapsulated FVPTC with (near) exclusive follicular growth pattern, <30% solid growth, and ≤1% papillae was renamed as noninvasive follicular thyroid neoplasm with papillary-like nuclear features (NIFTP) in 2016, to highlight the indolent nature of these lesions by eliminating the word ''carcinoma'' from the diagnosis, with the aim of avoiding overtreatment (3,8).

However, since the publication of the NIFTP consensus paper, three studies have reported that encapsulated follicular lesions with less than 1% of true papillae and PTC nuclei have a small risk (2% to 5.5%) of lymph node metastasis (9–11). As a result, commentaries were published subsequently on behalf of the NIFTP consensus group to revise the diagnostic criteria from less than 1% of true papillae to no true papillae allowed (8,12). This revision practically means that the finding of a single papilla in an otherwise noninvasive encapsulated follicular-patterned lesion with papillary-like nuclear features will result in a diagnosis of cancer.

In this retrospective cohort of 235 patients with unifocal encapsulated PTC (U-EPTC), we aimed at studying the correlation between architectural patterns, in particular the percentage of true papillae, with the risk of nodal metastasis and underlying BRAF/NRAS alterations. Our focus was to establish a clinical evidence-based cutoff for the percentage of papillae that can be used to define NIFTP and subclassify PTC. This, in turn, would lead to a better patient stratification, potentially sparing unnecessary therapy to individuals at a negligible clinical risk.

Materials and Methods

Characteristics of the study cohort

The study was approved by the institutional review board of Memorial Sloan-Kettering Cancer Center (MSKCC). The institutional database was searched for all cases with a diagnosis of U-EPTC operated at MSKCC between 1980 and 2015. All cases with adequate material were examined microscopically by two head and neck pathologists with special interests in thyroid neoplasia (RG and BX). Cases with multifocal carcinoma or NIFTP, thyroid carcinomas with infiltrative growth pattern, and other types of thyroid carcinoma (e.g., follicular carcinoma, Hürthle cell carcinoma, and poorly differentiated thyroid carcinoma) were excluded from the study to minimize the impact of other pathologic confounding factors in predicting outcome.

A tumor was categorized as encapsulated if it was completely surrounded by a fibrous capsule, or if it had a well-demarcated border without a definite capsule. A total of 235 cases of unifocal EPTC were included in the study. The sampling of these tumors was as follows: the entire tumor sampled, including its capsule in 158 (67%) cases, entire tumor capsular interface sampled in 19 (8%), and representative sampling with a median of 9 sections sampled per tumor (range: 1–23) in 58 (25%) cases. Among tumors with <10% papillae, 113 (66%) were entirely sampled, including their capsule, 17 (10%) had tumor capsular interface entirely sampled; and 42 (24%) were representatively sampled with a median of 9 sections per tumor (range 2–21). One hundred and twelve cases fulfilled the original diagnostic criteria of NIFTP proposed in 2016 (3).

Pathologic and clinical review

The architectural patterns and their respective percentage were collected for each tumor. The papillary pattern was defined by the presence of projections lined by lesional follicular epithelium and a well-defined fibrovascular core (''true'' papillae) (Fig. 1A–D). Obliquely cut septa with fibrovascular cores and abortive papillae lacking a fibrovascular core were not included (Fig. 1E, F). It is our experience that in the day-to-day practice it may be difficult to differentiate ''true'' papillae from obliquely-cut septa with fibrovascular cores and abortive papillae. Further, it is not uncommon to find foci of papillary hyperplasia in follicular-patterned lesions lacking the PTC nuclear features. For the purpose of this study as stated earlier, only ''true'' papillae lined by lesional follicular cells and a fibrovascular core were considered (Fig. 1). For cases with 0.1% to 10% papillae, the presence and percentage of papillae were determined at consensus review sessions between BX and RG. Follicular pattern was characterized by follicles of any size (macrofollicles or microfollicles) with central colloid surrounded by follicular cells. A solid pattern consisted of areas of solid, trabecular, or insular growth.

Tumor size was measured as the maximum diameter of the resected tumor specimen. Capsular invasion was defined as complete penetration of the capsule by tumor, and the number of these foci was recorded. Capsular invasion was recorded as ''focal'' when there were <4 foci of complete penetration of the tumor capsule and was recorded as ''extensive'' when the foci were ≥4. Vascular invasion (blood vessel invasion) was defined according to the criteria outlined by the Armed Forces Institute of Pathology (AFIP) fascicle (13) and the WHO classification (1). In brief, only when the invasive focus protruded into the lumen of the vessel in a polypoid manner covered by endothelial cells, or when it was attached to the vessel wall or associated with thrombus formation within an intra-capsular or extra-capsular vasculature, it was considered true vascular invasion. Vascular invasion was recorded as ''focal'' when there were <4 foci and was recorded as ''extensive'' when there were ≥4 foci.

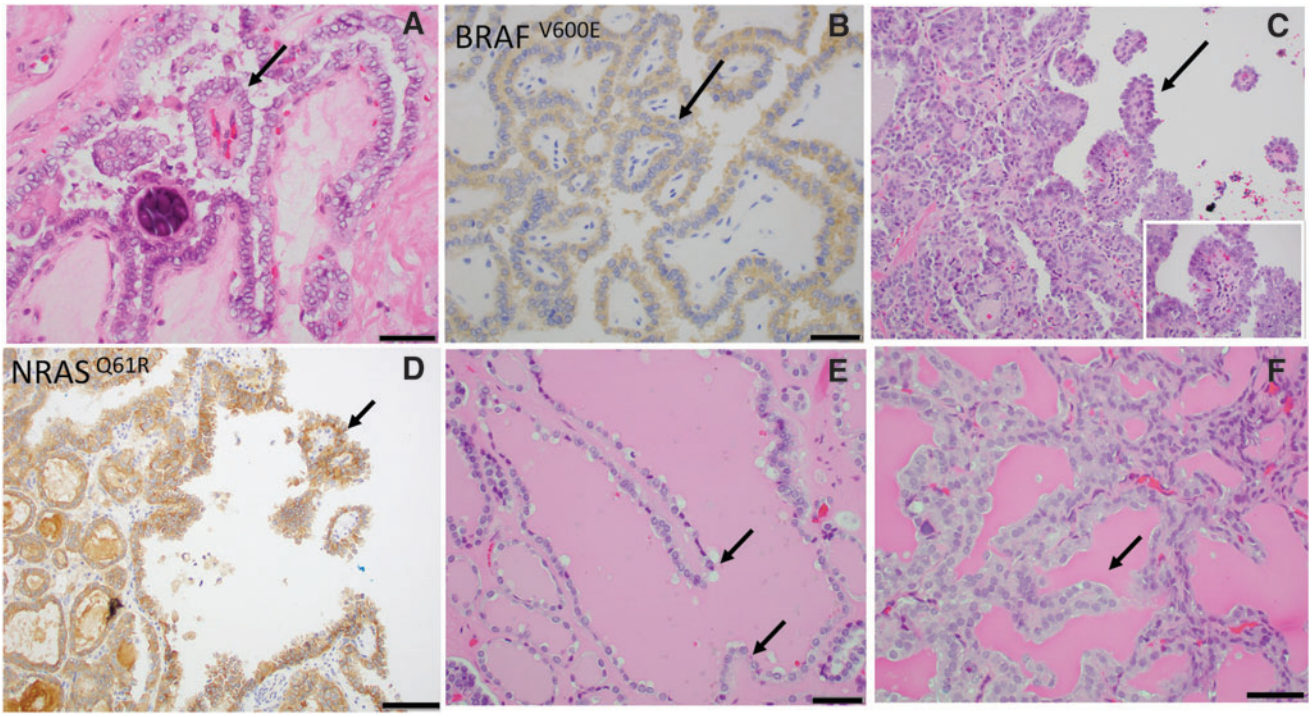


FIG. 1. Papillae in U-EPTC. (**A, B**) True papillae with fibrovascular cores (arrows) (**A**, H&E) in a classical PTC almost completely composed of papillae $BRAF^{V600E}$-positive by immunohistochemistry (**B**). (**C, D**) An encapsulated PTC with follicular predominant growth pattern containing occasional (<1%) papillary structures (arrows). Inset shows typical PTC nuclei (**C**, H&E); neoplastic cells are positive for $NRAS^{Q61R}$ by immunohistochemistry (**D**). (**E**) Pseudopapillae not fulfilling the definition of true papillae since they lack fibrovascular core (arrows) (**F**) Pseudopapilla not fulfilling the definition of true papillae since it lacks fibrovascular core and appears to represent an artefactually ruptured septa. H&E, hematoxylin and eosin; PTC, papillary thyroid carcinoma; U-EPTC, unifocal encapsulated PTC.

The patient's charts were reviewed for age at diagnosis, sex, type of surgery, radioactive iodine therapy, and outcome. The primary outcome of the study was the presence (AJCC pN1) or absence (AJCC pNx or pN0) of lymph node metastases at the time of initial resection. The secondary outcomes were disease-free survival and disease-specific survival.

$BRAF^{V600E}$ and $NRAS^{Q61R}$ immunohistochemistry and genotyping

Immunohistochemistry (IHC) staining for $BRAF^{V600E}$ and $NRAS^{Q61R}$ was performed in a subset of 100 randomly selected cases with an anti-$BRAF^{V600E}$ monoclonal antibody (clone: VE1, dilution: 1:400; Abcam, Cambridge, MA), and an anti-$NRAS^{Q61R}$ monoclonal antibody (clone: SP174, dilution: 1:25; Abcam) using the Leica Bond III system (Leica Biosystems, Inc., Buffalo Grove, IL) according to the manufacturer's recommendations. A tumor was considered as $BRAF^{V600E}$ or $NRAS^{Q61R}$ positive when diffuse granular cytoplasmic stain was identified.

BRAF and *RAS* DNA mutation status was previously examined in 19 cases by using Thyroseq version 2 in preoperative fine needle aspiration ($n=1$), Sequenom ($n=9$), or MSK-IMPACT platforms ($n=9$). The cases analyzed by Sequenom and MSK-IMPACT were included in previous publications from our group (14,15). Sequenom is a MassARRAY system based on matrix-assisted laser desorption/ionization time-to-flight mass spectrometry that was used to interrogate the presence of single nucleotide variation in 91 hot-spots of 8 oncogenes: *EGFR, KRAS, BRAF, PIK3CA, AKT1, NRAS, MEK1*, and *ERBB2* (14). MSK-IMPACT is a targeted capture massive parallel sequencing platform detecting somatic genetic alterations in 410 cancer-related genes (15). The DNA mutation status was documented and correlated with the IHC results.

Statistics

All statistical analyses were performed by using the SPSS software 24.0 (IBM Corporation, New York, NY). Clinico-pathologic characteristics, in particular the percentage of each architectural pattern and the BRAF/RAS genotype determined by using IHC, were compared between cases with and without lymph node metastases by using appropriate statistical tests, that is, Chi-square test or Fisher's exact test. The prognostic significance of papillary architecture was calculated by using log rank test for disease-free survival. *p*-Values less than 0.05 were considered statistically significant.

Results

Results are summarized in Tables 1–4, and they are illustrated in Figures 1 and 2.

Clinico-pathologic characteristics of the study cohort

The clinico-pathologic features of the 235 patients are reported in Table 1. The female to male ratio was 2:1. The

TABLE 1. CLINICO-PATHOLOGIC CHARACTERISTICS AND BRAF/NRAS MUTATION IN UNIFOCAL ENCAPSULATED PAPILLARY THYROID CARCINOMA ACCORDING TO THE NODAL STATUS AT THE TIME OF INITIAL SURGERY

	All patients (n = *235*)	*N0/Nx* (n = *208*)	*N1* (n = *27*)	p-*Values*
Architectural patterns				
Percentage of papillae				
0%	127 (54%)	127 (61%)	0 (0%)	**<0.001**
0.1–0.9%	31 (13%)	31 (15%)	0 (0%)	
1–9%	14 (6%)	13 (6%)	1 (4%)	
10–24%	6 (3%)	4 (2%)	2 (7%)	
25–49%	5 (2%)	5 (2%)	0 (0%)	
≥50%	52 (22%)	28 (13%)	24 (89%)	
Percentage of follicles				
0–24%	52 (22%)	31 (15%)	21 (78%)	**<0.001**
25–49%	6 (3%)	3 (1%)	3 (11%)	
≥50%	177 (75%)	174 (84%)	3 (11%)	
Percentage of solid growth				
0–24%	222 (94%)	195 (94%)	27 (100%)	0.409
25–49%	5 (2%)	5 (2%)	0 (0%)	
≥50%	8 (3%)	8 (4%)	0 (0%)	
BRAF and NRAS mutations (*n* = 100)				
BRAF/NRAS				
Positive $BRAF^{V600E}$	24 (24%)	10 (12%)	14 (74%)	**<0.001**
Positive $NRAS^{Q61R}$	26 (26%)	26 (32%)	0 (0%)	
Negative	50 (50%)	45 (56%)	5 (26%)	
Other characteristics				
Sex				
Female	156 (66%)	141 (68%)	15 (56%)	0.206
Male	79 (34%)	67 (32%)	12 (44%)	
Age, median (range)	46 (8–82)	47 (8–82)	36 (22–59)	**<0.001**
Tumor size, cm, median (range)	2.2 (0.1–7.5)	2.3 (0.1–7.5)	2.0 (0.3–4.7)	0.088
Type of surgery (*n* = 234)				
Lobectomy	102 (44%)	94 (45%)	8 (30%)	0.15
Total thyroidectomy	132 (56%)	113 (55%)	19 (70%)	
Capsular invasion				
Absent	166 (71%)	154 (74%)	12 (44%)	**<0.001**
Focal	57 (24%)	47 (23%)	10 (37%)	
Extensive	12 (5%)	7 (3%)	5 (19%)	
Vascular invasion				
Absent	212 (90%)	186 (89%)	26 (96%)	0.481
Focal	16 (7%)	15 (7%)	1 (4%)	
Extensive	7 (3%)	7 (3%)	0 (0%)	
Extrathyroidal extension				
Absent	231 (98%)	207 (99.5%)	24 (89%)	**<0.001**
Present	4 (2%)	1 (0.5%)	3 (11%)	
Margin status (*n* = 234)				
Negative	234 (100%)	207 (100%)	27 (100%)	NA
Radioactive iodine (*n* = 222)				
No	163 (74%)	147 (76%)	16 (62%)	0.109
Yes	56 (26%)	46 (24%)	10 (38%)	
Clinical outcome (*n* = 219)				
Follow-up period, years, median (range)	5.4 (0.1–28.1)	5.4 (0.1–28.1)	6.4 (0.1–26)	0.796
Distant metastasis				
Absent	216 (99%)	190 (98%)	26 (100%)	0.520
Present	3 (1%)	3 (2%)	0 (0%)	
Locoregional recurrence				
Absent	219 (100%)	193 (100%)	26 (100%)	NA

p-Values are obtained by using Fisher's exact test or Chi-square test for categorical variables, and log rank test for outcome (distant metastasis-free survival). Significant *p*-values are highlighted in bold.

The *BRAF* and *NRAS* mutational status was determined by immunohistochemistry using monoclonal antibodies for $BRAF^{V600E}$ and $NRAS^{Q61R}$ in a subset of 100 randomly selected cases.

NA, not applicable.

Table 2. Percentage of Papillae Within the Tumor, BRAF/NRAS Mutation, Nodal Metastasis, and Outcomes According to Invasion Status in Unifocal Encapsulated Papillary Thyroid Carcinoma

Cases with invasion (capsular and/or vascular)

	All patients (n = *79*)	*N0/Nx* (n = *64*)	*N1* (n = *15*)	p-*Values*
Percentage of papillae				
0%	27 (34%)	27 (42%)	0 (0%)	**0.001**
0.1–0.9%	11 (14%)	11 (17%)	0 (0%)	
1–9%	5 (6%)	4 (6%)	1 (7%)	
10–24%	3 (4%)	2 (3%)	1 (7%)	
25–49%	3 (4%)	3 (5%)	0 (0%)	
≥50%	30 (38%)	17 (27%)	13 (87%)	
BRAF/NRAS				
Positive $BRAF^{V600E}$	13 (46%)	4 (24%)	9 (82%)	**0.007**
Positive $NRAS^{Q61R}$	6 (21%)	6 (35%)	0 (0%)	
Negative	9 (32%)	7 (41%)	2 (18%)	
Follow-up period, years, median (range)	6.7 (0.1–28.0)	6.8 (0.1–28.0)	5.5 (0.1–22.3)	0.491
Distant metastasis ($n = 75$)				
Absent	72 (96%)	58 (95%)	14 (100%)	0.451
Present	3 (4%)	3 (5%)	0 (0%)	

Noninvasive cases (capsular and/or vascular)

	All patients (n = *156*)	*N0/Nx* (n = *144*)	*N1* (n = *12*)	p-*Values*
Percentage of papillae				
0%	100 (64%)	100 (69%)	0 (0%)	**<0.001**
0.1–0.9%	20 (13%)	20 (14%)	0 (0%)	
1–9%	9 (6%)	9 (6%)	0 (0%)	
10–24%	3 (2%)	2 (1%)	1 (8%)	
25–49%	2 (1%)	2 (1%)	0 (0%)	
50%	22 (14%)	11 (8%)	11 (92%)	
BRAF/NRAS				
Positive $BRAF^{V600E}$	11 (15%)	6 (9%)	5 (62.5%)	**<0.001**
Positive $NRAS^{Q61R}$	20 (28%)	20 (31%)	0 (0%)	
Negative	41 (57%)	38 (59%)	3 (37.5%)	
Follow-up period, years, median (range)	5.2 (0.1–28.1)	5.1 (0.1–28.1)	6.5 (0.1–26.0)	0.340
Distant metastasis ($n = 144$)				
Absent	144 (100%)	132 (100%)	12 (100%)	NA

The *BRAF* and *NRAS* mutational status was determined by immunohistochemistry using monoclonal antibodies for $BRAF^{V600E}$ and $NRAS^{Q61R}$ in a subset of 100 randomly selected cases. Significant *p*-values are highlighted in bold.

median age at diagnosis was 46 (range: 8–82). The median tumor size was 2.2 cm (range: 0.1–7.5 cm). One hundred and two patients (44%) were treated with lobectomy alone, while the remaining 132 (56%) underwent total thyroidectomy.

Seventy-nine tumors (34%) demonstrated evidence of invasion, being capsular invasion (56 out of 79, 71%), vascular invasion (10 out of 79, 13%), or both (13 out of 79, 16%). Seven tumors showed extensive vascular invasion (4 to 8 foci).

Correlation between papillary architecture and risk of lymph node metastasis at the time of initial surgery

Twenty-seven patients (12%) in our cohort had lymph node metastasis (AJCC pN1) at the time of initial resection. Compared with the patients without nodal metastases (AJCC pNx or pN0), those with nodal disease were associated with younger age ($p < 0.001$), had a higher prevalence of capsular invasion (66% in N1 group and 26% in N0/Nx group, $p < 0.001$), and of microscopic extrathyroidal extension into perithyroidal fibroadipose tissue (3 out of 77, 11% in N1 group and 1 out of 208, 0.5% in N0/Nx group, $p < 0.001$, Table 1). More importantly, the N1 and N0 groups were associated with different architectural patterns: 89% of the tumors in the N1 group had a predominance (≥50% of total tumor volume) of papillary architecture, whereas 75% of Nx/N0 tumors showed (near) exclusive follicular growth with 0% to <1% papillae ($p < 0.001$). A small percentage of tumors contained a significant amount of solid architecture: 5 cases (2%) with 25–49% and 8 (3%) with ≥50% of solid growth pattern. None of these 13 cases with significant solid growth had pN1 disease. The presence of any amount of solid pattern did not correlate with nodal status (N0/Nx vs. N1, $p = 0.409$).

All tumors were further classified into several categories according to the percentage of papillae within the tumor: 127 (54%) with no (0%) papillae, 31 (13%) with 0.1–0.9% of papillae, 14 (6%) had 1–9% papillae, 6 (3%) had 10–24% papillae, 5 (2%) had 25–49% papillae, and 52 (22%) had at least 50% papillae. The risk of lymph node metastasis

Table 3. Nodal Status and BRAF/NRAS Mutation According to the Percentage of Papillae Within the Tumor in Unifocal Encapsulated Papillary Thyroid Carcinoma

	Percentage of papillae					
	0%	*0.1–0.9%*	*1–9%*	*10–24%*	*25–49%*	*≥50%*
All cases						
pN stage						
N0/NX	127 (100%)	31 (100%)	13 (93%)	4 (67%)	5 (100%)	28 (54%)
N1	0 (0%)	0 (0%)	1 (7%)[a]	2 (33%)	0 (0%)	24 (46%)
RAS/BRAF						
$BRAF^{V600E}$	1 (2.5%)[b]	1 (4%)[c]	0 (0%)	1 (25%)	3 (75%)	18 (82%)
$NRAS^{Q61R}$	15 (37.5%)	10 (42%)	1 (17%)	0 (0%)	0 (0%)	0 (0%)
Negative	24 (60%)	13 (54%)	5 (83%)	3 (75%)	1 (25%)	4 (18%)
Invasive cases						
pN stage						
N0/NX	27 (100%)	11 (100%)	4 (80%)	2 (67%)	3 (100%)	17 (57%)
N1	0 (0%)	0 (0%)	1 (20%)[a]	1 (33%)	0 (0%)	13 (43%)
RAS/BRAF						
$BRAF^{V600E}$	0 (0%)	0 (0%)	0 (0%)	1 (100%)	2 (100%)	10 (91%)
$NRAS^{Q61R}$	1 (25%)	5 (56%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Negative	3 (75%)	4 (44%)	1 (100%)	0 (0%)	0 (0%)	1 (9%)
Noninvasive cases						
pN stage						
N0/NX	100 (100%)	20 (100%)	9 (100%)	2 (67%)	2 (100%)	11 (50%)
N1	0 (0%)	0 (0%)	0 (0%)	1 (33%)[d]	0 (0%)	11 (50%)
RAS/BRAF						
$BRAF^{V600E}$	1 (3%)[b]	1 (7%)[c]	0 (0%)	0 (0%)	1 (50%)	8 (73%)
$NRAS^{Q61R}$	14 (39%)	5 (33%)	1 (20%)	0 (0%)	0 (0%)	0 (0%)
Negative	21 (58%)	9 (60%)	4 (80%)	3 (100%)	1 (50%)	3 (27%)

The difference in $BRAF^{V600E}$ positivity and $NRAS^{Q61R}$ positivity between cases with ≥10% papillae and those with <10% papillae is highly significant (Fisher's exact test, $p < 0.001$).

[a]The percentage of papillae within this encapsulated tumor with capsular invasion was 5%; both the primary tumor and the nodal metastasis were negative for $BRAF^{V600E}$ and $NRAS^{Q61R}$.

[b]A 0.2-cm noninvasive well-circumscribed tumor composed entirely of follicles. No true papillae were identified on multiple histologic levels examined.

[c]A 1.5-cm noninvasive encapsulated lesion with 10% of total tumor volume showing tall cell cytomorphology.

[d]The percentage of papillae within this noninvasive encapsulated tumor with lymph node metastasis was 10%.

according to the percentage of papillae within the tumor is illustrated in Table 3 and Figure 2A. None of the 158 patients with less than 1% papillae developed lymph node metastases, while 24 out of 52 tumors (46%) with at least 50% papillae had pN1 disease. The frequency of nodal metastasis in tumors with 1–9%, 10–24%, and 25–50% papillae was 7% (1 out of 13), 33% (2 out of 6), and 0% (0 out of 5), respectively.

When subdividing the tumors into those with invasion (capsular and/or vascular, $n = 79$) and without invasion ($n = 156$), there was a significant association between papillary architectural pattern and nodal metastasis regardless of the invasion status ($p < 0.001$, Table 2). Encapsulated PTC with ≥50% papillae had a 50% risk (11 out of 22 cases) of nodal metastasis even without invasion, whereas noninvasive encapsulated tumors with less than 10% of papillae (129 cases) did not metastasize to lymph nodes (Table 3).

The case of U-EPTC with nodal metastasis and the lowest percentage of papillae (Fig. 3 and the case marked with superscript ''C'' in Table 3) had 5% papillae, had extensive capsular invasion, and measured 2.0 cm.

The cases of U-EPTC without invasion with the lowest percentage of papillae had 0.1% papillae: They included 6 tumors and measured from 2.5 to 5.0 cm.

Table 4. Detailed Characteristics of Patients with Unifocal Encapsulated Papillary Thyroid Carcinoma and Distant Metastasis

	Age	*Sex*	*Architecture*	*Sx*	*Size (cm)*	*CI*	*VI*	*pN*	*BRAF/NRAS*	*Molecular*	*FU (years)*	*Status at last FU*	*DM at presentation*
1	64	F	100% follicle	TT	1.5	Focal	Absent	N0	NA	NA	3.5	DOD	Yes
2	62	F	100% follicle	TT	5.0	Absent	Ext	Nx	NA	NA	14.3	AWD	Yes
3	66	M	100% follicle	TT	5.0	Ext.	Ext	Nx	NRAS	$NRAS^{Q61R}$	2.0	DOD	Yes

AWD, alive with disease; CI, capsular invasion; DOD, dead of disease; DM, distant metastasis; Ext, extensive; F, female; FU, follow up; M, male; NA, not available; Sx, surgery; TT, total thyroidectomy; VI, vascular invasion.

Within each PTC group based on the percentage of papillae, the overall invasion status (capsular and/or vascular) did not have a significant impact on the risk of nodal metastasis (p ranged from 0.286 to 1.000 for each individual category, data not shown).

Phenotype-genotype correlation: increased percentage of papillae is associated with BRAF^{V600E}

Table 3 and Figure 2B summarize the association between papillary percentage and *BRAF/NRAS* mutation status detected by using IHC (Fig. 1). Tumors with ≥10% papillae were predominantly *BRAF^{V600E}*-related (22/30, 73%) and lack *NRAS* mutations (0/30, 0%), whereas PTC with <10% papillae are enriched in *RAS* mutations (26/70, 37%, $p < 0.001$). Tumors without any papillae had a high frequency (15/40, 37.5%) of *NRAS^{Q61R}* mutation and a low rate (1/40, 2.5%) of *BRAF^{V600E}* mutation. The case positive for BRAF^{V600E} IHC was a well-circumscribed 0.2-cm tumor without invasion that was composed entirely of follicles. No true papillae or tall cell cytomorphology were identified on multiple histologic levels examined. There was also a 1.5-cm U-EPTC without invasion showing predominant follicular growth, 0.1–0.9% papillae, and 10% area with tall cell cytomorphology that was positive for BRAF^{V600E} IHC.

The frequency of BRAF^{V600E} positivity increased with the increasing proportion of papillae within the tumor, being 4% in tumors with 0.1–0.9% papillae, 0% in tumors with 1–9% papillae, 25% in tumors with 10–24% papillae, 75% in tumors with 25–49% papillae, and 82% in those with ≥50% papillae. In contrast, the rate of NRAS^{Q61R} decreased. In our cohort, no tumor with ≥10% papillae was positive for NRAS^{Q61R} IHC. The difference in BRAF^{V600E} positivity and NRAS^{Q61R} positivity between cases with ≥10% papillae and those with <10% papillae is highly significant (Fisher's exact test, $p < 0.001$). Tumors with ≥10% papillae are predominantly *BRAF^{V600E}*-related (22/30, 73%) and lack *NRAS* mutations (0/30, 0%), whereas PTC with <10% papillae are enriched in *RAS* mutations (26/70, 37%).

BRAF and *RAS* genotypes were available in 19 cases that had been analyzed by mass spectrometry or next-generation sequencing before our study. All 19 cases were negative for *BRAF^{V600E}* by IHC as well as by molecular testing with mass spectrometry or next-generation sequencing.

Nine of the 19 cases were positive for NRAS^{Q61R} by IHC. The molecular alterations detected in these 9 cases were *NRAS^{Q61R}* in four, *KRAS^{Q61R}* in two, and *HRAS^{Q61R}* in one; in two cases, no RAS mutations were detected by using the Sequenom mass spectrometry platform (14), an assay with relatively low sensitivity. Molecular testing by mass spectrometry or next-generation sequencing identified *NRAS^{Q61K}*, *HRAS^{Q61K}*, *HRAS^{G13C}*, and *KRAS^{G12D}* in four cases negative for NRAS^{Q61R} IHC.

Clinical outcome and adverse events

Follow-up data were available in 216 patients, with a median of 5.4 years (range: 0.1 to 28.1 years). There was no locoregional recurrence in the entire cohort. Three patients (1.4%) developed distant metastases and two of them died of their disease. Distant metastases were identified at the time of the initial diagnosis in all three patients (Table 4). The primary tumors were encapsulated FVPTC with exclusive follicular architecture devoid of any solid or papillary component. Two tumors had extensive vascular invasion (8 foci in each tumor), while one tumor had focal capsular invasion. One patient tested was positive for *NRAS^{Q61R}* mutation by IHC and by the Sequenom assay.

Discussion

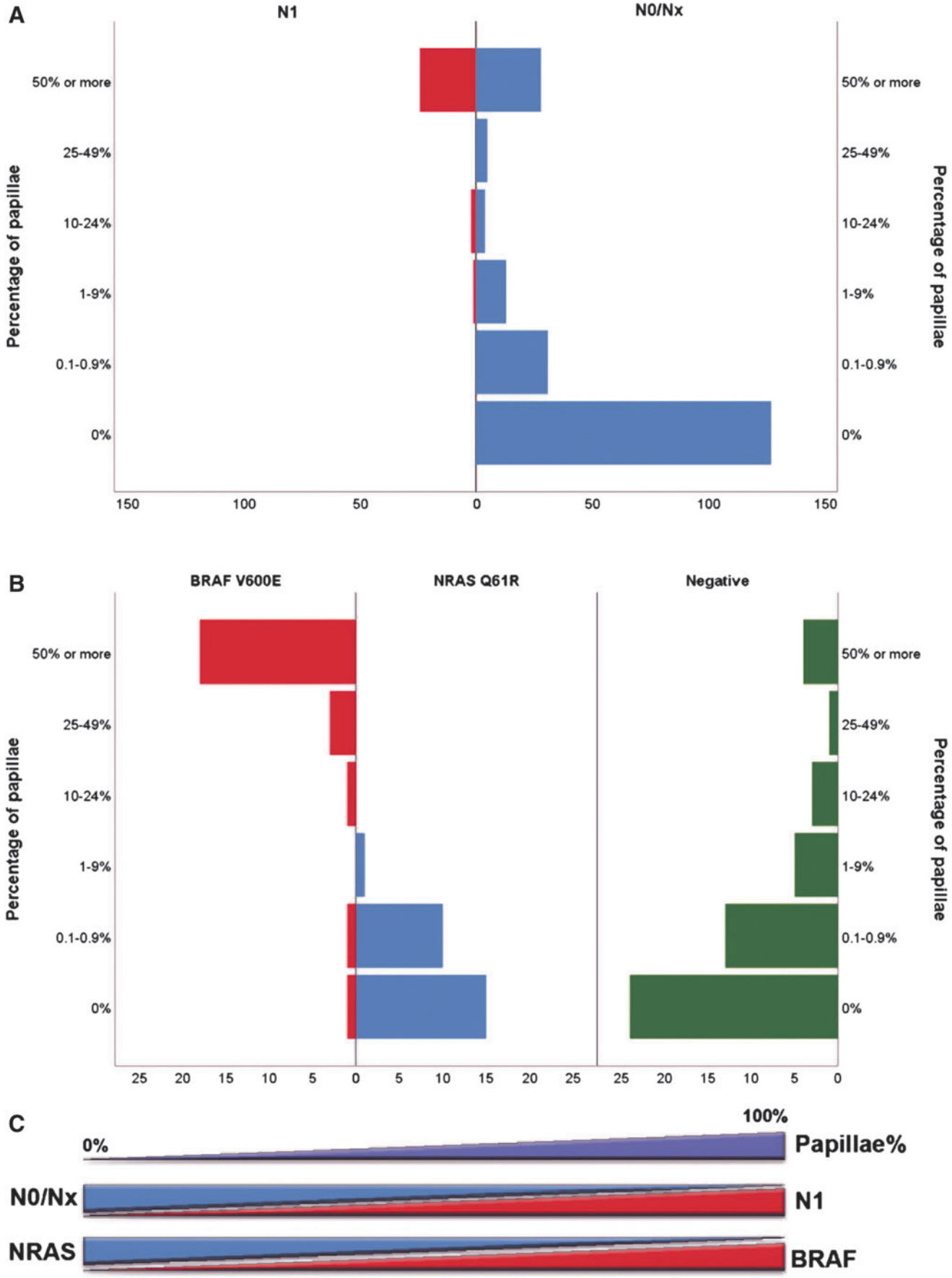
FVPTC are those PTCs with an almost exclusive follicular growth pattern (1,3) that, when encapsulated, need to be distinguished from encapsulated forms of conventional PTC, which may also have a predominantly follicular growth pattern with a few papillae. Traditionally, a cutoff of 1% has been used to separate FVPTC (<1% papillae) and conventional PTCs with a follicular predominant growth pattern (≥1% papillae). The distinction is important since FVPTC has an RAS-like molecular signature and a tendency to metastasize to distant sites rather than to regional lymph nodes, whereas conventional PTC, even when encapsulated, is BRAF-like and typically metastasizes to lymph nodes first and only later to distant sites (4). The existence of full encapsulation is relatively uncommon but well documented in classic PTC, and it has been associated with a favorable clinical outcome compared with the far more common infiltrative forms of conventional PTC (16–18).

Surprisingly, however, less than a handful of studies included a systematic evaluation of the percentage of papillae formation in their study design. Liu *et al.* defined FVPTC as a tumor with <1% papillae, and reported an overall nodal metastasis rate of 17%, including a 5% rate of nodal metastasis in the encapsulated FVPTC, and 0% nodal metastasis in the noninvasive encapsulated forms (19). Zidan *et al.* (20) allowed up to 20% papillae to define FVPTC and reported a nodal metastasis rate of 22% in FVPTC.

How many papillae are there in encapsulated PTCs that behave like classic PTC that tend to develop lymph node metastasis? The ability to reliably answer this question has gained a new relevance since the introduction of the NIFTP terminology with the clinical implication of a negligible to absent risk for the patient carrying such a tumor to develop lymph node or distant metastases.

In the attempt to address the issue, Cho *et al.* have reported a nodal metastasis risk of 3% in noninvasive encapsulated FVPTC with rare (<1%) papillae, and 2% in those without any

FIG. 2. Percentage of papillae within the tumor, nodal metastasis, and BRAF/NRAS mutation. (**A**) Papillae within the tumor and risk of nodal metastasis. (**B**) Papillae within the tumor and BRAF^{V600E}/NRAS^{Q61R} status subdivided to BRAF^{V600E} positive, NRAS^{Q61R} positive and negative for both immunostains; on the y axis, the tumor groups are subdivided based on the percentage of papillae, and the x axis shows the number of tumors. (**C**) Diagram showing how an increasing percentage of papillae within the tumor is associated with a higher rate of N1 disease, BRAF^{V600E}, and a lower frequency of NRAS^{Q61R}. The *BRAF* and *NRAS* mutational status was determined by immunohistochemistry using monoclonal antibodies for BRAF^{V600E} and NRAS^{Q61R} in a subset of 100 randomly selected cases.


A
N1
N0/Nx
50% or more
25-49%
10-24%
1-9%
0.1-0.9%
0%
Percentage of papillae
150
100
50
0
50
100
150
B
BRAF V600E
NRAS Q61R
Negative
25
20
15
10
5
0
C
100%
0%
Papillae%
N0/Nx
N1
NRAS
BRAF

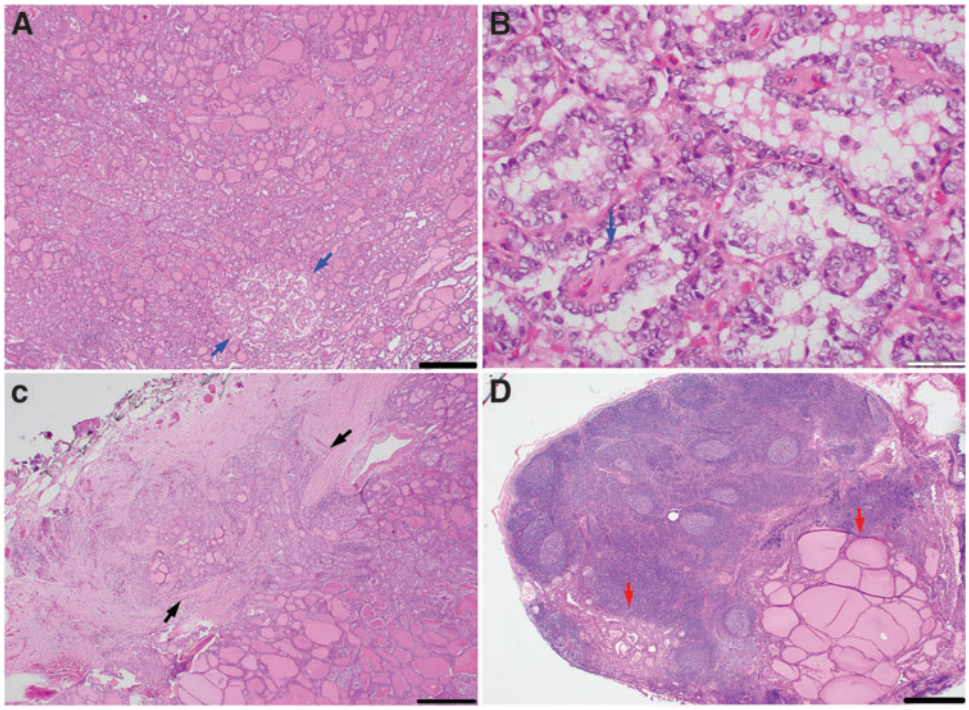


FIG. 3. Microscopic pictures of a patient with a 2-cm encapsulated PTC harboring 5% papillae with capsular invasion and nodal metastases. (**A**) Low-power view of primary tumor with area of papillae formation (blue arrows). (**B**) High-power view of papillary area in A showing papillae with fibrovascular cores (blue arrow). (**C**) Primary tumor displaying mushrooming capsular invasion (black arrows). (**D**) Lymph node with metastatic deposits (red arrows). Scale bar: 500 μm in (**A**, **C,** and **D**), 50 μm in (**B**).

papillae (10). Similarly, Kim *et al.* (11) showed a 2% risk of lymph node metastasis in noninvasive encapsulated FVPTC with 0% papillae. However, these two studies included tumors with synchronous papillary microcarcinomas. As it is well-established that papillary microcarcinoma may give rise to nodal metastasis (21–24), the reported nodal metastasis risk in noninvasive encapsulated FVPTC may be exaggerated.

Parente *et al.* (9) reported a 5% risk of nodal metastasis and 1% risk of distant metastasis in unifocal noninvasive encapsulated FVPTC without any papillae. However, molecular analyses were not done in the positive cases on the primary tumor and the metastases to confirm that the metastatic deposit emanated from the identified primary tumor. Since metastatic nodal and even distant disease has been reported in total thyroidectomy specimens negative for carcinoma and entirely submitted for microscopic examination (25,26), one cannot exclude without molecular analysis that the metastatic deposits may have originated from separate infiltrative small carcinomas embedded in the paraffin block or which had undergone regression. Thus, in our opinion, these findings remain to be validated in other studies.

In contrast to the reports just cited, multiple studies, including the NIFTP consensus cohort, showed 0% risk of nodal metastasis in NIFTP, originally defined as a noninvasive encapsulated follicular-patterned lesion with <1% papillae and papillary-like nuclear features (3,6,15,27–32). Nevertheless, essentially based on the data from Cho *et al.* (10) and Parente *et al.* (9), a proposal has been made to revise the diagnostic criteria for NIFTP from <1% papillae to no true papillae (8,12).

All this recent work highlights the relevance of our study. We provide—to our knowledge for the first time—a detailed, clinical evidence-based analysis dissecting the relationship between papillary percentage, tumor behavior, and BRAF/NRAS status in encapsulated PTC.

In this study, we demonstrate a strong correlation between the percentage of papillae, risk of nodal metastasis, and underlying *BRAF/RAS* mutation status in encapsulated PTC. Tumors with ≥50% of papillae were associated with 46% risk of nodal metastasis and frequent (82%) $BRAF^{V600E}$ mutation, whereas tumors with (near) exclusive follicular growth pattern and very few (<1%) true papillae had 0% risk of nodal metastasis and were enriched with $NRAS^{Q61R}$ (37.5%) mutations.

There may be a certain degree of subjectivity and ambiguity in defining one single true papilla and, consequently, diagnosing papillary carcinoma, and thus establishing a ''cancer'' diagnosis. We show that in any case—even when accounting for the occasional equivocal interpretation of histologic findings of papillae—tumors with rare (<1%) papillae are not uncommon (13% of the entire cohort) and are highly indolent with 0% risk of recurrence or lymph node metastasis. Therefore, the <1% papillae criterion as a cutoff for NIFTP appears sound for diagnostic purposes. The discrepant nodal metastatic rates between this study and the earlier mentioned three publications (9–11) could be due to two factors. First, we selected only unifocal carcinoma, while the study of Cho *et al.* and Kim *et al.* included tumors with synchronous papillary microcarcinomas. Further, a difference in the interpretation of the percentage of papillae could be an additional reason for these variable results.

We also demonstrate that there is an incremental risk of nodal metastasis according to the percentage of papillae, being 7% in those with 1–9% papillae and 46% in those with ≥50% papillae. In our cohort, invasion by itself did not predict the risk of nodal disease. Although capsular invasion is significantly more frequent in tumors with N1 disease, such an association is no longer significant when cases are substratified by the percentage of papillae within the tumor. Taken together, the percentage of papillae is an important histologic feature, and could be more relevant than invasive status in predicting

the behavior of encapsulated PTC. As tumors with at least 1% papillae in an encapsulated follicular-predominant lesion are at risk of developing nodal metastasis, they are best classified as CPTC, with a follicular predominant growth pattern.

The link between papillae and BRAF/RAS signature has been previously implied by multiple studies (3,5,19,33,34). For example, the TCGA PTC study showed that tumors enriched with true papillae (e.g., classic and tall cell variants) have a high frequency (89% for tall cell and 67% for classic variant) of *BRAF*^{V600E} mutation, whereas PTCs with (near) exclusive follicular growth pattern (i.e., FVPTC) are enriched with *RAS* mutations (38%) (5,35). The reported frequency of *BRAF*^{V600E} mutations in a tumor fulfilling the original NIFTP diagnostic criteria (i.e., <1% papillae) ranges from 0% (15,30,36,37) to 8% (10,11). The 8% rate was reported by two Korean studies. The 0% rates were studies from North America and/or Europe. It is plausible to assume that variation in ethnic background may have an impact on the discrepancy. This study is the first to demonstrate a strong association between a measured percentage of papillae and BRAF/RAS alterations in encapsulated PTC.

Ten percent papillae in an encapsulated PTC appears to be associated with a genotype switch from *RAS* mutations to *BRAF*^{V600E} mutations. Tumors with ≥10% papillae are predominantly *BRAF*^{V600E}-related (73%) and lack *NRAS* mutations (0%), whereas PTC with <10% papillae are enriched in *RAS* mutations (37%). Two tumors with <1% papillae were positive for BRAF^{V600E}, one of which contained 10% of cells showing tall cell cytomorphology, which would exclude this tumor from a diagnosis of NIFTP. As 89% to 95% of tall cell variant PTC harbor *BRAF*^{V600E} mutations (5,38), the BRAF^{V600E} immunopositivity in this tumor is explained by the presence of tall cell cytomorphology. In our cohort, among the 119 tumors meeting the original morphologic diagnostic criteria of NIFTP (i.e., <1% of papillae), one (0.8%) was positive for BRAF^{V600E}. This tumor was a 0.2-cm well-circumscribed tumor completely devoid of papillae and tall cell cytomorphology on multiple hematoxylin and eosin levels examined. It is possible that papillae had not yet developed, given the very small size of the tumor. Whatever the reason, we fully agree with the statement by some of the authors of the NIFTP consensus group that BRAF^{V600E} immunopositivity should lead to an exhaustive search for papillae and invasion (12). However, as stated by the same authors, it cannot be used solely to exclude NIFTP, which is a morphologic diagnosis (12).

BRAF^{V600E} and NRAS^{Q61R} IHC have been shown to be highly sensitive and specific in detecting underlying mutations in thyroid tumors (39,40). An interesting finding of our study is that NRAS^{Q61R} IHC cross-reacts with HRAS^{Q61R} and KRAS^{Q61R} mutant proteins in thyroid carcinoma. A similar phenomenon has been previously described in medullary thyroid carcinoma (41) and melanoma (42). Regardless, we demonstrate a strong correlation between BRAF/NRAS IHC status and the route and risk of metastasis. While 14 of 24 (58%) of BRAF^{V600E}-positive encapsulated PTC developed nodal metastasis, none of the 26 NRAS^{Q61R} IHC-positive tumors had nodal metastasis. Rather, one of the tumors with 100% follicular growth and distant metastasis at presentation was positive for *NRAS*^{Q61R} by IHC and genetic analysis.

The link between *BRAF*^{V600E}-mutated tumors and nodal metastasis and between *RAS*-mutated carcinoma and distant metastasis have been previously reported by Fakhruddin *et al.* (43) in PTC and by Landa *et al.* (44) in poorly differentiated thyroid carcinoma. Given these results, BRAF^{V600E} and NRAS^{Q61R} IHC may be utilized clinically as an additional tool to predict the route of metastasis and assist clinical decision regarding appropriate follow-up and management. In addition, these IHC stains may help in the diagnosis of tumors that are morphologically bordering on NIFTP. BRAF^{V600E} immunopositivity, for example, should prompt extensive histopathologic examination for true papillae.

Our cohort also contains 13 tumors (6%) with a significant amount (≥25%) of solid architecture. Although none of these 13 tumors developed nodal metastases, the number is too small to draw any definite conclusion in regard to the impact of solid growth on nodal metastasis in encapsulated PTC.

The study has some limitation. First, it is a single institution study from a large tertiary cancer center and may not reflect the population at large. Second, the number of encapsulated PTC with 1% to 49% of papillae is relatively small as most of the tumors clustered at the two ends of the spectrum (<1% or ≥50% papillae).

In conclusion, in this study, we show that an increased percentage of papillae within an encapsulated PTC is associated with a high risk of nodal metastasis and BRAF^{V600E} mutation, regardless of invasion status. U-EPTCs have an overall favorable behavior. The only patients who developed distant metastases harbored them at presentation, had 100% follicular growth, were positive for NRAS^{Q61R}, and had capsular/vascular invasion. Tumors with less than 1% papillae do not develop lymph node metastases irrespective of their invasive status. Nodal disease or recurrence are also absent in encapsulated PTC without invasion with <10% papillary growth. These findings indicate that the original criterion of <1% papillae within the tumor is still sound for a diagnosis of NIFTP. Reinstituting this criterion will spare a carcinoma diagnosis and unnecessary therapy with its side effects on patients who have a really negligible clinical risk.

Disclaimer

The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health.

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References

1. Lloyd RV, Osamura RY, Kloppel G, Rosai J 2017 WHO Classification of Tumours of Endocrine Organs. International Agency for Research on Cancer (IARC), Lyon.
2. Siegel RL, Miller KD, Jemal A 2018 Cancer statistics, 2018. CA Cancer J Clin **68:**7–30.
3. Nikiforov YE, Seethala RR, Tallini G, Baloch ZW, Basolo F, Thompson LD, Barletta JA, Wenig BM, Al Ghuzlan A, Kakudo K, Giordano TJ, Alves VA, Khanafshar E, Asa SL,

El-Naggar AK, Gooding WE, Hodak SP, Lloyd RV, Maytal G, Mete O, Nikiforova MN, Nose V, Papotti M, Poller DN, Sadow PM, Tischler AS, Tuttle RM, Wall KB, LiVolsi VA, Randolph GW, Ghossein RA 2016 Nomenclature revision for encapsulated follicular variant of papillary thyroid carcinoma: a paradigm shift to reduce overtreatment of indolent tumors. JAMA Oncol **2:**1023–1029.

4. Tallini G, Tuttle RM, Ghossein RA 2017 The history of the follicular variant of papillary thyroid carcinoma. J Clin Endocrinol Metab **102:**15–22.
5. Cancer Genome Atlas Research Network 2014 Integrated genomic characterization of papillary thyroid carcinoma. Cell **159:**676–690.
6. Thompson LD 2016 Ninety-four cases of encapsulated follicular variant of papillary thyroid carcinoma: a name change to noninvasive follicular thyroid neoplasm with papillary-like nuclear features would help prevent overtreatment. Mod Pathol **29:**698–707.
7. Rivera M, Tuttle RM, Patel S, Shaha A, Shah JP, Ghossein RA 2009 Encapsulated papillary thyroid carcinoma: a clinico-pathologic study of 106 cases with emphasis on its morphologic subtypes (histologic growth pattern). Thyroid **19:**119–127.
8. Lloyd RV, Asa SL, LiVolsi VA, Sadow PM, Tischler AS, Ghossein RA, Tuttle RM, Nikiforov YE 2018 The evolving diagnosis of noninvasive follicular thyroid neoplasm with papillary-like nuclear features (NIFTP). Hum Pathol **74:**1–4.
9. Parente DN, Kluijfhout WP, Bongers PJ, Verzijl R, Devon KM, Rotstein LE, Goldstein DP, Asa SL, Mete O, Pasternak JD 2018 Clinical safety of renaming encapsulated follicular variant of papillary thyroid carcinoma: is NIFTP truly benign? World J Surg **42:**321–326.
10. Cho U, Mete O, Kim MH, Bae JS, Jung CK 2017 Molecular correlates and rate of lymph node metastasis of noninvasive follicular thyroid neoplasm with papillary-like nuclear features and invasive follicular variant papillary thyroid carcinoma: the impact of rigid criteria to distinguish non-invasive follicular thyroid neoplasm with papillary-like nuclear features. Mod Pathol **30:**810–825.
11. Kim MJ, Won JK, Jung KC, Kim JH, Cho SW, Park DJ, Park YJ 2018 Clinical characteristics of subtypes of follicular variant papillary thyroid carcinoma. Thyroid **28:** 311–318.
12. Nikiforov YE, Baloch ZW, Hodak SP, Giordano TJ, Lloyd RV, Seethala RR, Wenig BM 2018 Change in diagnostic criteria for noninvasive follicular thyroid neoplasm with papillarylike nuclear features. JAMA Oncol **4:** 1125–1126.
13. Rosai J, DeLellis RA, Carcangiu ML, Frable WJ, Tallini G 2015 Tumor of the Thyroid and Parathyroid Gland (AFIP Atlas of Tumor Pathology Series 4). American Registry of Pathology Press, Silver Spring, MD.
14. Rivera M, Ricarte-Filho J, Knauf J, Shaha A, Tuttle M, Fagin JA, Ghossein RA 2010 Molecular genotyping of papillary thyroid carcinoma follicular variant according to its histological subtypes (encapsulated vs infiltrative) reveals distinct BRAF and RAS mutation patterns. Mod Pathol **23:**1191–1200.
15. Xu B, Reznik E, Tuttle RM, Knauf J, Fagin JA, Katabi N, Dogan S, Aleynick N, Seshan V, Middha S, Enepekides D, Casadei GP, Solaroli E, Tallini G, Ghossein R, Ganly I 2019 Outcome and molecular characteristics of noninvasive encapsulated follicular variant of papillary thyroid carcinoma with oncocytic features. Endocrine **64:**97–108.
16. Schroder S, Bocker W, Dralle H, Kortmann KB, Stern C 1984 The encapsulated papillary carcinoma of the thyroid. A morphologic subtype of the papillary thyroid carcinoma. Cancer **54:**90–93.
17. Carcangiu ML, Zampi G, Pupi A, Castagnoli A, Rosai J 1985 Papillary carcinoma of the thyroid. A clinicopathologic study of 241 cases treated at the University of Florence, Italy. Cancer **55:**805–828.
18. Evans HL 1987 Encapsulated papillary neoplasms of the thyroid. A study of 14 cases followed for a minimum of 10 years. Am J Surg Pathol **11:**592–597.
19. Liu J, Singh B, Tallini G, Carlson DL, Katabi N, Shaha A, Tuttle RM, Ghossein RA 2006 Follicular variant of papillary thyroid carcinoma: a clinicopathologic study of a problematic entity. Cancer **107:**1255–1264.
20. Zidan J, Karen D, Stein M, Rosenblatt E, Basher W, Kuten A 2003 Pure versus follicular variant of papillary thyroid carcinoma: clinical features, prognostic factors, treatment, and survival. Cancer **97:**1181–1185.
21. Gao X, Zhang X, Zhang Y, Hua W, Maimaiti Y, Gao Z 2016 Is papillary thyroid microcarcinoma an indolent tumor?: a retrospective study on 280 cases treated with radioiodine. Medicine (Baltimore) **95:**e5067.
22. Mehanna H, Al-Maqbili T, Carter B, Martin E, Campain N, Watkinson J, McCabe C, Boelaert K, Franklyn JA 2014 Differences in the recurrence and mortality outcomes rates of incidental and nonincidental papillary thyroid microcarcinoma: a systematic review and meta-analysis of 21 329 person-years of follow-up. J Clin Endocrinol Metab **99:**2834–2843.
23. Piana S, Ragazzi M, Tallini G, de Biase D, Ciarrocchi A, Frasoldati A, Rosai J 2013 Papillary thyroid microcarcinoma with fatal outcome: evidence of tumor progression in lymph node metastases: report of 3 cases, with morphological and molecular analysis. Hum Pathol **44:**556–565.
24. Tallini G, de Biase D, Durante C, Acquaviva G, Bisceglia M, Bruno R, Bacchi Reggiani ML, Casadei GP, Costante G, Cremonini N, Lamartina L, Meringolo D, Nardi F, Pession A, Rhoden KJ, Ronga G, Torlontano M, Verrienti A, Visani M, Filetti S 2015 BRAF V600E and risk stratification of thyroid microcarcinoma: a multicenter pathological and clinical study. Mod Pathol **28:**1343–1359.
25. Xu B, Scognamiglio T, Cohen PR, Prasad ML, Hasanovic A, Tuttle RM, Katabi N, Ghossein RA 2017 Metastatic thyroid carcinoma without identifiable primary tumor within the thyroid gland: a retrospective study of a rare phenomenon. Hum Pathol **65:**133–139.
26. Nishikawa M, Toyoda N, Yonemoto T, Fujiyama A, Ogawa Y, Tokoro T, Sakaguchi N, Yoshimura M, Yoshikawa N, Tabata S, Kumazawa H, Yamashita T, Sakaida N, Okamura A, Kasagi K, Inada M 1998 Occult papillary thyroid carcinoma in Hashimoto's thyroiditis presenting as a metastatic bone tumor. Endocr J **45:**111–116.
27. Xu B, Farhat N, Barletta JA, Hung YP, Biase D, Casadei GP, Onenerk AM, Tuttle RM, Roman BR, Katabi N, Nose V, Sadow P, Tallini G, Faquin WC, Ghossein R 2018 Should subcentimeter non-invasive encapsulated, follicular variant of papillary thyroid carcinoma be included in the noninvasive follicular thyroid neoplasm with papillary-like nuclear features category? Endocrine **59:**143–150.
28. Xu B, Tallini G, Scognamiglio T, Roman BR, Tuttle RM, Ghossein RA 2017 Outcome of large noninvasive follicular thyroid neoplasm with papillary-like nuclear features. Thyroid **27:**512–517.

29. Mariani RA, Kadakia R, Arva NC 2018 Noninvasive encapsulated follicular variant of papillary thyroid carcinoma: should it also be reclassified in children? Pediatr Blood Cancer **65:**e26966.
30. Johnson DN, Furtado LV, Long BC, Zhen CJ, Wurst M, Mujacic I, Kadri S, Segal JP, Antic T, Cipriani NA 2018 Noninvasive follicular thyroid neoplasms with papillary-like nuclear features are genetically and biologically similar to adenomatous nodules and distinct from papillary thyroid carcinomas with extensive follicular growth. Arch Pathol Lab Med **142:**838–850.
31. Shafique K, LiVolsi VA, Montone K, Baloch ZW 2018 Papillary thyroid microcarcinoma: reclassification to non-invasive follicular thyroid neoplasm with papillary-like nuclear features (NIFTP): a retrospective clinicopathologic study. Endocr Pathol **29:**339–345.
32. Vivero M, Kraft S, Barletta JA 2013 Risk stratification of follicular variant of papillary thyroid carcinoma. Thyroid **23:**273–279.
33. Steward DL, Carty SE, Sippel RS, Yang SP, Sosa JA, Sipos JA, Figge JJ, Mandel S, Haugen BR, Burman KD, Baloch ZW, Lloyd RV, Seethala RR, Gooding WE, Chiosea SI, Gomes-Lima C, Ferris RL, Folek JM, Khawaja RA, Kundra P, Loh KS, Marshall CB, Mayson S, McCoy KL, Nga ME, Ngiam KY, Nikiforova MN, Poehls JL, Ringel MD, Yang H, Yip L, Nikiforov YE 2019 Performance of a multigene genomic classifier in thyroid nodules with indeterminate cytology: a prospective blinded multicenter study. JAMA Oncol **5:**204–212.
34. Adeniran AJ, Zhu Z, Gandhi M, Steward DL, Fidler JP, Giordano TJ, Biddinger PW, Nikiforov YE 2006 Correlation between genetic alterations and microscopic features, clinical manifestations, and prognostic characteristics of thyroid papillary carcinomas. Am J Surg Pathol **30:**216–222.
35. Gao J, Aksoy BA, Dogrusoz U, Dresdner G, Gross B, Sumer SO, Sun Y, Jacobsen A, Sinha R, Larsson E, Cerami E, Sander C, Schultz N 2013 Integrative analysis of complex cancer genomics and clinical profiles using the cBioPortal. Sci Signal **6:**pl1.
36. Johnson DN, Sadow PM 2018 Exploration of BRAFV600E as a diagnostic adjuvant in the non-invasive follicular thyroid neoplasm with papillary-like nuclear features (NIFTP). Hum Pathol **82:**32–38.
37. Howitt BE, Chang S, Eszlinger M, Paschke R, Drage MG, Krane JF, Barletta JA 2015 Fine-needle aspiration diagnoses of noninvasive follicular variant of papillary thyroid carcinoma. Am J Clin Pathol **144:**850–857.
38. Ghossein R, Livolsi VA 2008 Papillary thyroid carcinoma tall cell variant. Thyroid **18:**1179–1181.
39. Ghossein RA, Katabi N, Fagin JA 2013 Immunohistochemical detection of mutated BRAF V600E supports the clonal origin of BRAF-induced thyroid cancers along the spectrum of disease progression. J Clin Endocrinol Metab **98:**E1414–E1421.
40. Oishi N, Kondo T, Vuong HG, Nakazawa T, Mochizuki K, Kasai K, Inoue T, Tahara I, Hirokawa M, Miyauchi A, Katoh R 2016 Immunohistochemical detection of NRAS(Q61R) protein in follicular-patterned thyroid tumors. Hum Pathol **53:**51–57.
41. Reagh J, Bullock M, Andrici J, Turchini J, Sioson L, Clarkson A, Watson N, Sheen A, Lim G, Delbridge L, Sidhu S, Sywak M, Aniss A, Shepherd P, Ng D, Oei P, Field M, Learoyd D, Robinson BG, Clifton-Bligh RJ, Gill AJ 2017 NRASQ61R mutation-specific immunohistochemistry also identifies the HRASQ61R mutation in medullary thyroid cancer and may have a role in triaging genetic testing for MEN2. Am J Surg Pathol **41:**75–81.
42. Felisiak-Golabek A, Inaguma S, Kowalik A, Wasag B, Wang ZF, Zieba S, Pieciak L, Rys J, Kopczynski J, Sarlomo-Rikala M, Gozdz S, Lasota J, Miettinen M 2018 SP174 antibody lacks specificity for NRAS Q61R and cross-reacts with HRAS and KRAS Q61R mutant proteins in malignant melanoma. Appl Immunohistochem Mol Morphol **26:**40–45.
43. Fakhruddin N, Jabbour M, Novy M, Tamim H, Bahmad H, Farhat F, Zaatari G, Aridi T, Kriegshauser G, Oberkanins C, Mahfouz R 2017 BRAF and NRAS mutations in papillary thyroid carcinoma and concordance in BRAF mutations between primary and corresponding lymph node metastases. Sci Rep **7:**4666.
44. Landa I, Ibrahimpasic T, Boucai L, Sinha R, Knauf JA, Shah RH, Dogan S, Ricarte-Filho JC, Krishnamoorthy GP, Xu B, Schultz N, Berger MF, Sander C, Taylor BS, Ghossein R, Ganly I, Fagin JA 2016 Genomic and transcriptomic hallmarks of poorly differentiated and anaplastic thyroid cancers. J Clin Invest **126:**1052–1066.

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