Recurrent Papillary Thyroid Carcinoma with Pleural Metastasis Diagnosed by Effusion Cytology: A Report of Cases with Clinicopathologic Correlation

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Abstract

Papillary thyroid carcinoma (PTC) is typically an indolent disease characterized by slow growth and a favorable prognosis. In rare instances, this disease may metastasize to the pleura and manifest as a malignant pleural effusion. We report 3 female patients of Japanese/Okinawan ancestry with a history of PTC who presented with hydrothorax. Cytologic examination in conjunction with immunohistochemical staining enabled a definitive diagnosis of metastatic PTC. Molecular analysis of the mitogen activated protein kinase (MAPK) and phosphatidylinositol 3-kinase (PI3K) pathways demonstrated the presence of the v-raf murine sarcoma viral oncogene homolog B (BRAF)^{V600E} mutation in 2 of our 3 patients, with the absence of any other clinically significant mutations in all cases. Further investigation is necessary to elucidate the molecular and environmental mechanisms involved in this aggressive manifestation of PTC.

Keywords

metastatic papillary thyroid carcinoma, pleural metastasis, effusion cytology, BRAF^{V600E} mutation

Introduction

Papillary thyroid carcinoma (PTC) accounts for 80%-90% of all thyroid cancers and is the most common endocrine malignancy in the United States.[1] Thyroid cancer is especially prevalent in Hawai'i, with a higher incidence compared to national and worldwide averages.[2] PTC tends to be a low grade malignancy with an overall 5-year survival rate of 95%-97%. However, in 5%-10% of cases, distant metastasis may occur in the bone or lung,[3] and in rare instances in the pleura with accumulation of effusive fluid. This paper reports on the clinicopathologic and molecular findings of 3 patients with PTC metastatic to the pleura who presented to the Queens Medical Center (QMC). Approval for this study was sought and obtained through the QMC Research & Institutional Review Committee (IRB# RA-2013-035).

Case Reports

Patient #1 DM

A 49-year-old woman of Japanese-Okinawan ancestry first underwent a near total thyroidectomy in 1982 for PTC followed by postoperative radioactive iodine therapy. The disease recurred at 7, 16, 24, and 25 years postoperatively, with multiple bilateral neck and pulmonary metastases. Twenty-nine years after her initial diagnosis, a chest CT scan showed a moderate sized left-sided pleural effusion; 900 cc of turbid, dark red/brown fluid was removed from the left pleura. Cytologic examination revealed cohesive tumor cell clusters with minimal nuclear pleomorphism and abundant finely vacuolated cytoplasm (Figure 1a). Papillary clusters with intranuclear inclusions (Figure 1b) and grooves (Figure 1c) were also noted. Immunohistochemical staining was positive for thyroid transcription factor-1 (TTF-1) (Figure 1d), thyroglobulin (TGB) (Figure 1e), and negative for Napsin-A (Figure 1f), thus confirming the diagnosis of metastatic PTC. A subsequent CT scan revealed enlargement of the pleural effusion, as well as additional nodules in the right lung. The patient passed away later that year at the age of 79. Molecular analysis of the mitogen activated protein kinase (MAPK) and phosphatidyl 3-kinase (PI3K) pathways within the patient's original thyroid tumor demonstrated the BRAF^{V600E} mutation as the only clinically significant mutation (Table 2).

Patient #2 SK

An 82-year-old woman of Japanese ancestry with a history of PTC diagnosed 8 years previously, first presented to our hospital with a right neck mass. Fine needle aspiration cytology showed metastatic PTC, confirmed by removal of her jugular chain lymph nodes. The patient remained asymptomatic until 9 years later, when she developed a right cervical neck mass with accompanying weight loss and early satiety. A CT scan demonstrated multiple pleural-based metastases in the right lung base with a moderate to large sized pleural effusion; 900 cc of cloudy dark red fluid was aspirated from the right pleura. Cytologic examination was positive for papillary and follicular clusters, intranuclear grooves and inclusions, and cytoplasmic vacuoles, consistent with metastatic PTC (Table 1). No immunohistochemical staining was performed in this instance. Despite treatment with radioactive iodine, the pleural fluid continued to accumulate. The patient succumbed to her disease the following year at the age of 91. Molecular analysis of the MAPK and PI3K pathways within the patient's original thyroid tumor revealed the BRAF^{V600E} mutation as the only clinically significant mutation (Table 2).

Patient #3 CF

A 50-year-old woman of Japanese-Okinawan ancestry first presented to our hospital with a right neck mass. Thyroid ultrasound-guided core needle biopsy revealed PTC with chronic thyroiditis. A subsequent total thyroidectomy showed PTC in the right thyroid lobe with lymphatic invasion, multiple parathyroidal, mediastinal, and cervical lymph node metastases, and extension into the paratracheal soft tissues. The patient received postoperative radioiodine I-131 ablation. Three years

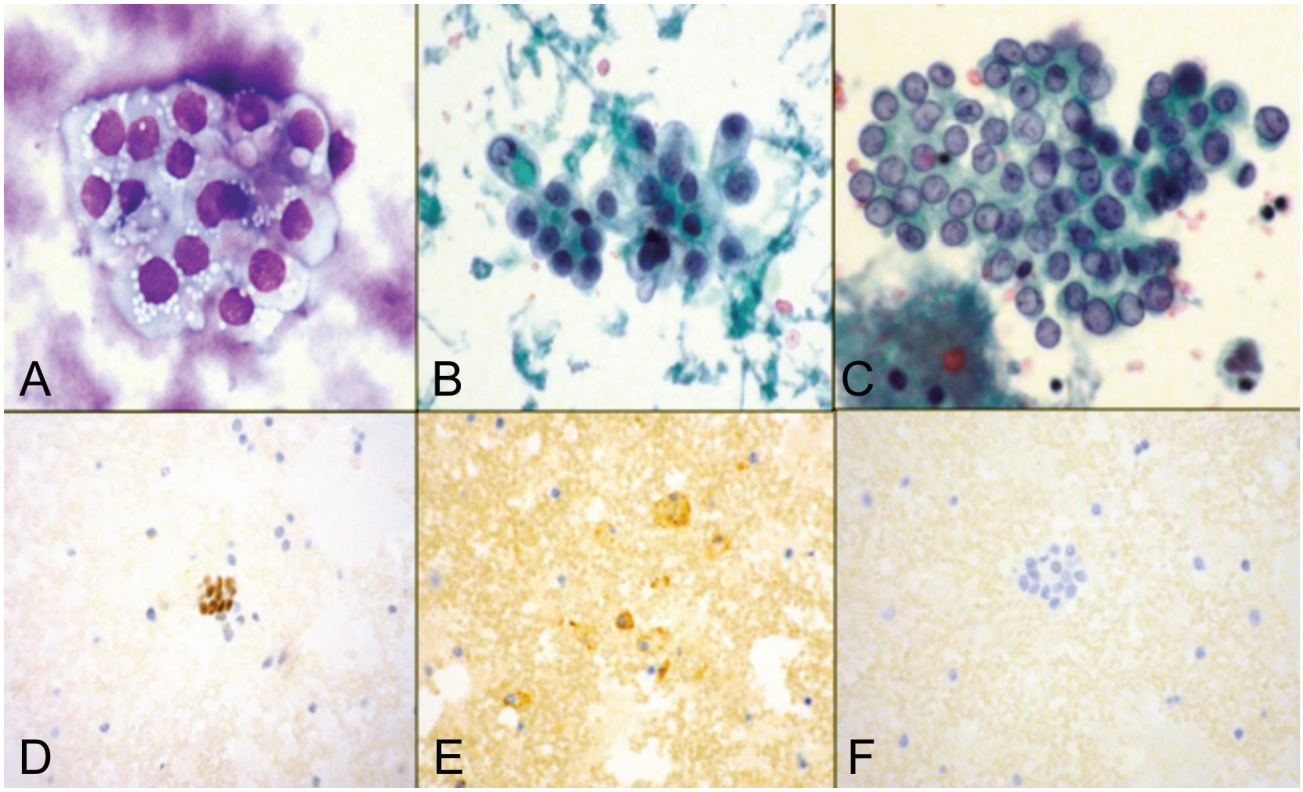

Figure 1. **Cytology of metastatic PTC to the pleura** (a) Cohesive cell clusters with minimal nuclear pleomorphism, and abundant finely vacuolated cytoplasm (Diff-Quik, 400x). (b) Papillary cluster with tumor cells displaying fine, powdery chromatin, thin nuclear membranes, and occasional intranuclear inclusion (Papanicoloau, 400x). (c) Papillary cluster with uniform bland nuclei and intranuclear grooves (Papanicolaou, 400x). (d) Immunohistochemical TTF-1 (thyroid transcriptase factor 1) staining (400x). (e) Thyroglobulin stain (400x). (f) Napsin-A stain (400x).

Table 1. Clinicopathologic Findings of Patients with Metastatic PTC to the Pleura

	PT #1 DM	PT #2 SK	PT #3 CF
Age at First Diagnosis	49	74	50
Sex	F	F	F
Histologic Type	Papillary	Papillary	Papillary
Years Until Pleural Metastasis	29	17	4
Other Sites of Distant Metastasis	Lung	Lung	Pituitary, Lungs, Liver, Ribs, Pelvis
Papillary Clusters	+	+	+
Follicular Clusters	+	+	+
Intranuclear Grooves	+	+	+
Intranuclear Inclusions	+	+	+
Psammoma Bodies	-	-	+
Cytoplasmic Vacuoles	+	+	+
TTF-1	+	N/A	N/A
TGB	+	N/A	+
Napsin-A	-	N/A	N/A

Table 2. Molecular Analysis of the MAPK and PI3K Pathways in Primary PTC Tumors									
Gene	**CHR**	**CHR STR**	**SNP**	**PT #1 DM**	**PT #2 SK**	**PT #3 CF**	**Location**	**Function**	**Cancer Association**
AKT1	14	105239894	C>T	+	-	+	Exonic	Synonymous	N/A
AKT1	14	105242966	T>C	+	-	+	Intronic, near exon		N/A
AKT1	14	105258972	G>A	-	+	-	Exonic	Synonymous	N/A
AKT2	19	40742320	T>C	+	+	+	Intronic, near exon		N/A
BRAF (V600E)	7	140453136	A>T	+	+	-	Exonic	Missense	PTC, Melanoma, Colorectal Cancer
GNAS	20	57478807	C>T	+	+	-	Exonic	Synonymous	N/A
GNAS	20	57478939	G>A	-	-	+	Intronic, near exon		N/A
KIT	4	55599436	T>C	+	+	+	Intronic, near exon		N/A
PIK3C2A	11	17111272	A>G	+	+	-	Untranslated region		N/A
PIK3C2A	11	17113483	C>T	+	+	+	Intronic, near exon		N/A
PIK3C2A	11	17126670	C>T	-	+	+	Intronic, near exon		N/A
PIK3C2A	11	17172133	T>C	-	+	+	Exonic	Synonymous	N/A
PIK3C2A (Q337R)	11	17190279	T>C	-	+	-	Exonic	Missense	N/A
PIK3C2A	11	17191019	A>G	-	+	+	Exonic	Synonymous	N/A
SNAPC5(MAP2K1)	15	66782048	C>T	-	-	+	Intronic, near exon		N/A

later, she presented with extensive metastases to the left neck, right paratracheal regions, ribs, paraspinal area, and lungs. Later that year, she developed abdominal pain, vomiting, and diarrhea. A chest X-ray revealed a right pleural effusion with diffuse pulmonary interstitial infiltrates, and 30 cc of turbid red fluid was withdrawn from the right pleura. The cytologic exam was positive for papillary and follicular clusters, intranuclear grooves and inclusions, psammoma bodies, and cytoplasmic vacuoles. Immunohistochemical staining was positive for TGB, confirming a PTC origin (Table 1). One year later, the patient was admitted to QMC with symptoms of panhypopituitarism and optic chiasm compression caused by a pituitary mass. Transphenoidal excision of this pituitary mass demonstrated PTC metastatic to the pituitary gland. A subsequent positron emission tomography (PET) scan showed extensive progression of metastatic disease, involving the right and left pleura, mediastinal and abdominal/pelvic lymph nodes, and multiple skeletal sites. The patient expired later that year at the age of 57. Molecular analysis of the MAPK and P13K pathways within the original thyroid tumor did not reveal any clinically significant mutations (Table 2).

Discussion

The exact incidence of metastatic papillary thyroid cancer to the pleura is not well known. In order to ascertain this incidence within our institution, a retrospective search of the QMC pathology database from January 2002 to December 2012 was conducted. This search revealed a total of 4,046 pleural fluids submitted to our laboratory during this time period. Of the examined pleural fluids, 82% (3311) were benign effusions and 18% (735) were malignant. Lung (39%) and breast (15%) cancers were the most common primary malignancies. Our search revealed only 3 patients with metastatic PTC to the pleura, comprising only 0.1% of all pleural fluid exams and 0.5% of all malignancies. Therefore, in our institution, metastatic PTC to the pleura is exceedingly rare.

For comparison, a literature review was performed and revealed 8 articles from 1979 to 2007, reporting a total of a mere 14 cases of metastatic thyroid carcinoma to the pleura (Table 3). In these prior reports, the patients' ages ranged from 46 to 88 years, with approximately equal numbers of men and women.[4-11] Five (36%) of the patients had an effusion as their presenting symptom. Six (43%) of the carcinomas were papillary, and one (7%) was anaplastic. The remaining 7 (50%) cases were described as being either papillary or follicular carcinomas without further distinction. Four (29%) were diagnosed by pleural effusion cytology.[4,8,10,11] The cytologic features were described in these 4 cases.[4,6,8,11] Papillary clusters and psammoma bodies were present in 2 (50%) cases, follicular clusters and intranuclear inclusions were present in one (25%), and no intranuclear grooves were noted in all cases. Fine cytoplasmic vacuoles were noted in only one (25%) of these prior studies.[11] Immunohistochemical staining was performed in one case,[8] and was positive for TGB. Mutational testing was not performed in any of these studies.

Contrary to prior studies, many of the classic cytologic features of PTC were found in 4 pleural fluid specimens from our 3 patients, including 2 malignant effusions from patient 3 (CF). Intranuclear grooves and follicular and papillary clusters, as well as fine cytoplasmic vacuoles were identified in all 4 (100%) specimens (Table 1). Intranuclear inclusions were noted in 3 (75%), and psammoma bodies in one (25%). The diagnosis was confirmed with immunohistochemical TBG staining in 2(50%) cases, and TTF-1 in one (25%). Napsin-A, performed in one case, was negative. Utilizing cytologic examination and immunohistochemical staining with TGB, TTF-1, and Napsin-A, we were able to reliably render a definitive diagnosis of metastatic PTC in all four cases.

Table 3. Literature Review (1979-2006) of Metastatic Thyroid Carcinoma to the Pleura

Authors	Year	# of Patients	Age	Sex	Histologic Type	Positive Cytology Cases	Presenting Symptoms	Other Sites of Distant Metastasis	Papillary Clusters	Follicular Clusters	Intranuclear Inclusions	Intranuclear Grooves	Psammoma Bodies	Cytoplasmic Vacuoles	TGB	TTF-1
Hyman (4)	1979	1	76	F	Papillary	1	Pleural effusion	Lung	+	-	-	-	-	-	N/A	N/A
Mizukami (5)	1990	7	-	-	Papillary and follicular	-	-	-	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Vernon (6)	1992	1	50	F	Papillary	1	No	Lung, iliac crest, cervical LN	-	-	-	-	+	-	N/A	N/A
Koppl (7)	1993	1	88	F	Anaplastic	-	Pleural effusion	-	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Kovacs (8)	1994	1	68	M	Papillary	1	Pleural effusion	Pericardium, cervical LN	+	-	+	-	+	-	+	N/A
Nomori (9)	1997	1	68	F	Papillary	-	No	Lung	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Perez (10)	2003	1	66	M	Papillary	-	Pleural effusion	-	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Siddaraju (11)	2007	1	46	M	Papillary	1	Pleural effusion	-	-	+	-	-	-	+	N/A	N/A

Certain histologic variants of PTC have been known to act more aggressively, including tall and columnar cell, as well as diffuse sclerosing variants.[12] The tall cell PTC variant presents at an older age, is associated with extrathyroid extension and nodal disease, and is comprised of tumor cells that are at least three times as tall as they are wide with abundant oxyphilic cytoplasm. The columnar cell PTC variant is more common in male patients and is associated with hematogenous spread to lungs and bone, as well as extrathyroidal extension. In this PTC variant, the tumor cells are columnar with nuclear stratification without abundant eosinophilic cytoplasm, and form parallel cords of markedly elongated follicles. The diffuse sclerosing PTC variant has a higher incidence in women, presents at a younger age, and has an increased likelihood of nodal and lung metastasis. It is characterized by a neoplastic follicular cell proliferation in a background of desmoplastic, sclerotic-type fibrosis associated with psammoma bodies, metaplastic squamous epithelium, and chronic thyroiditis. Interestingly, all of our cases presenting with metastatic pleural disease exhibited classic PTC histology. No aggressive tumor morphology was noted in any of our patients.

In addition to some of the highest rates of thyroid cancer in the world, Hawai'i also exhibits various gender and ethnic disparities within its PTC patient population. All three of our patients were women of Japanese or Japanese-Okinawan ethnicity born in the United States, ranging in age from 56 to 91 at the time of metastasis to the pleura.Women in Hawai'i have a 2-3 times higher risk of acquiring thyroid cancer than men, consistent with national data,[13] so a preponderance of women amongst our patients is not surprising. However, a study conducted between 1962 and 1966 showed that the Japanese in Hawai'i had the lowest incidence of thyroid cancer among ethnic groups at a rate of 5.5 and 2.2 per 100,000 population for women and men, respectively.[14] Interestingly, these rates were noted to be twice that of Japanese patients in Japan, where the incidence was 2.6 and 1.1 per 100,000 for women and men. [14] A more recent survey in Hawai'i performed between 2000 to 2005 appears to show an increase in the incidence of thyroid cancer among Japanese in Hawai'i, with rates of 9.1 and less than 4 per 100,000 for women and men, respectively.[13] Despite this increase, the incidence of thyroid cancer among Japanese in Hawai'i still remains the lowest among other ethnic groups. For example, by comparison, Filipino women and men displayed the highest incidence rates at 27.7 and 9.5

per 100,000. Although PTC is more common in other ethnic groups in Hawai'i, we observed this highly aggressive form of PTC solely in women of Japanese or Japanese-Okinawan ancestry. In addition to the pleural/pulmonary involvement noted in all 3 of our patients, and additional metastases to the liver, pelvic bone and ribs, and pituitary developed in one. All 3 of patients succumbed to their cancer within 1 year of development of their pleural effusions.

BRAF or rat sarcoma proteins (RAS) mutations, rearrangements to the receptor tyrosine-protein kinase (RET) proto-oncogene (RET)/PTC, and other activating mutations in the MAPK pathway are known to induce tumorigenesis in PTC.[1] Specifically, BRAF mutations are the most common tumor inducing alteration, occurring in up to 83% of all PTCs.[15] Over 90% of these BRAF mutations are of the V600E variety, which has been associated with increased regional and distant metastases as well as a higher mortality rates.[1,16] Molecular analysis confirmed the presence of $BRAF^{V600E}$ mutation in 2 of our 3 PTC tumors (Table 2). In addition to $BRAF^{V600E}$, other mutations along the MAPK and P13K pathways were studied, including AKT1 and AKT2 (v-akt murine thymoma viral oncogene homologs 1 and 2), GNAS (GNAS complex locus), KIT (v-kit Hardy-Zuckerman 4 feline sarcoma viral oncogene homolog), PIK3C2A (phosphatidylinositol 3-kinase class 2 alpha), and SNAPC5/MAP2K1 (small nuclear ribonucleic acid activating protein complex polypeptide 5/mitogen-activated protein kinase 1) (Table 2). $BRAF^{V600E}$ was the only clinically significant mutation noted in our investigation. However, despite the presence of widespread metastases to brain, liver, and lung in addition to pleura, one of our patients (Table 1, Patient #3) failed to demonstrate this $BRAF^{V600E}$ mutation. Tumors from all 3 patients exhibited AKT2, KIT, and PIK3C2A mutations, but to date, there have been no reports in the literature regarding associations between these mutations and worsened prognosis in PTC. Therefore, additional molecular or environmental factors likely exist that may provide further prognostic and even therapeutic insight into aggressively behaving PTCs.

Certain populations in Hawai'i have been shown to exhibit a higher rate of the $BRAF^{V600E}$ mutation in PTCs.[17] In a recent study by Morita, et al, patients of Filipino ethnicity demonstrated the $BRAF^{V600E}$ mutation in 83.8% of PTCs, which nearly equals the highest rates reported in the literature.[17] Additionally, the purported five-year survival rate was lower in this population, supporting the association of a more clinically aggressive malignancy with the $BRAF^{V600E}$ mutation.[17] The Japanese population in Hawai'i was not investigated in Morita's study.[17] There is no current evidence of the BRAF mutation occurring at higher rates in Japanese populations, with percentages varying from 29% to 52%.[18,19,20] Furthermore, one report by Ito, et al, a large study of 631 Japanese PTC patients, failed to demonstrate a relationship between the presence of the BRAF mutation and aggressive tumor characteristics and poor patient prognosis.[20]

To date, the etiology of thyroid cancer remains obscure. Iodine deficiency and exposure to ionizing radiation appear to be the only recognized environmental factors known to cause thyroid cancer.[13,14] However, various other environmental components have been suggested to increase risk. Several studies have revealed an association between the incidence of thyroid cancer and volcanic activity around the world.[21,22,23] In addition to Hawai'i, volcanically active areas such as Iceland, the Philippines, Sicily, New Caledonia, and French Polynesia have some of the highest rates of thyroid cancer globally,[21,23,24,25,26] ranging from 11.7 per 100,000 women in Iceland to 71.4 per 100,000 women in New Caledonia. The high incidence of thyroid cancers in these geographically distinct areas would suggest that some unknown carcinogenic agent produced by volcanic activity might be responsible for its induction. In one study, the authors speculated that a chemical carcinogen released by Mount Etna in Sicily might modulate BRAF activation in their ethnically homogeneous population, and thereby account for the higher rates of BRAF mutations found in the eastern part of the island (45.9%), compared to the west (22.7%).[27] This data is extremely interesting and relevant to Hawai'i's population due to the presence of frequently erupting volcanoes in the Hawaiian island chain. However, thus far, there have been no studies demonstrating any evidence of geographic variations of PTC across the islands. Therefore, further research is necessary to elucidate a possible relationship between volcanic eruptions and the molecular features of thyroid cancer.

Conclusions

PTC metastatic to the pleura is extremely rare, occurring in 0.1% of all pleural fluids received in our institution over a 10 year period. Contrary to the prior literature, our cases demonstrated characteristic cytologic features of PTC. Therefore, in conjunction with immunohistochemical staining, we were able to reliably make definitive diagnoses of metastatic PTC. All of our cases were in Japanese women, despite those of Japanese ethnicity historically having the lowest incidence of thyroid cancer in Hawai'i. Only 2 of our 3 patients demonstrated *BRAF* mutations. Therefore, there are likely other putative molecular markers of tumor aggressiveness that have yet to be determined. The potential association between volcanic emissions and thyroid cancer development in Hawai'i's population requires further study.

Conflict of Interest

None of the authors identify any conflict of interest.

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