Atypical Papillary Thyroid Carcinoma with Squamous Metaplasia and RET/NRAS/TERT/PIK3CA Mutations: A Case Report

Zhenyu Li

Chongqing University Cancer Hospital

Haiping Huang

Chongqing University Cancer Hospital

Dongfang Guo

Chongqing University Cancer Hospital

Yujie Feng

Chongqing University Cancer Hospital

Jing Zhang

Chongqing University Cancer Hospital

Qingming Jiang (✉ jiangqm80@126.com)

Chongqing University Cancer Hospital <https://orcid.org/0000-0002-5783-0633>

Research Article

Keywords: Squamous metaplasia, PTC, Immunohistochemistry, Genetic detection, Case report

Posted Date: January 3rd, 2022

DOI: <https://doi.org/10.21203/rs.3.rs-880031/v2>

License: © ⓘ This work is licensed under a Creative Commons Attribution 4.0 International License.

[Read Full License](#)

Abstract

Background: Papillary thyroid carcinoma (PTC) with squamous metaplasia is a relatively rare and special subtype adenocarcinoma of thyroid which involves multiple genetic changes. We reported a rare case of atypical PTC with squamous metaplasia and RET/NRAS/TERT/PIK3CA mutations which was confirmed after surgical resection pathologically.

Case summary: A 2.5×2.5×2 cm³, smooth, hard, clear boundaries and solid nodule on the left thyroid gland was found on relevant physical examination of the patient. And then, the unilateral radical resection of thyroid carcinoma was performed after diagnosing. The tumor cells were squamous metaplasia and arranged in structures with diffuse growth pattern microscopically. At high magnification, stretched nucleus, nuclear grooves and internuclear pseudoinclusions in tumor cells were detected, and the follicular epithelium cells were atypical. Immunohistochemical staining shown strong positiveness of CK19, TTF-1, P40 and Galectin-3, partial positiveness of TG of tumor cells and negativeness of Calcitonin, which could exclude medullary thyroid carcinoma (MTC). Furthermore, first generation sequencing of 18 PTC related genes techniques shown RET, NRAS, TERT and PIK3CA was mutated.

Conclusion: Genetic detection is vital to the diagnosis of thyroid adenocarcinoma, especially for the PTC with atypical morphology or rare metaplasia

Introduction

Papillary thyroid carcinoma (PTC) with squamous metaplasia is not only a very rare and special subtype adenocarcinoma of thyroid, but also belongs to diffuse sclerosing variant papillary thyroid carcinoma (DSVPTC) which is considered a poor prognosis (1–4). DSVPTC is recognized by prominent squamous metaplasia, innumerable psammoma bodies, in a background of lymphocytic thyroiditis and stromal fibrosis (3). To our knowledge, only dozens of relevant literature about PTC with squamous metaplasia or DSVPTC have been published in National Center for Biotechnology Information (NCBI) database so far, and mostly focus on pathologic findings since first authoritatively introduced at 1987 (5, 6). DSVPTC has different clinical, pathological and molecular profiles when compared to conventional PTC (7). DSVPTCs should be considered high-risk PTCs because of high propensity for tumor invasion, metastasis, relapse and mortality. Aggressiveness of DSVPTCs might be related to a different molecular pathway than that in conventional PTCs (8). On genetic analysis, the occurrence of BRAF and RAS mutations are uncommon events in DSVPTC and activation of RET/PTC rearrangements are common (9). Genetic detection is not only crucial to the diagnosis for PTC, and also used to assess the prognosis (10, 11). When the morphology is atypical, genetic detection can be of great help to pathologists. Here, we used a first generation sequencing of thyroid carcinoma containing 18 genes techniques as a support to diagnose the case of PTC with squamous metaplasia after histology and immunohistochemistry, and reported an unique typical papillary thyroid carcinoma with squamous metaplasia and RET/NRAS/TERT/PIK3CA mutations.

Case Description

This is a case of a 60-years-old male who had diagnosed with diabetes 7 years ago, treated with metformin orally, and presented with a left anterior neck mass more than 1 year ago inadvertently. No pain, redness, fever, hoarseness, dysphagia, dyspnea, excitability, irritability and hunger was found during the period, and the patient did not go to the hospital and receive any treatment. The patient noticed an enlargement of the mass recently, and then came to our hospital for further treatment. There was no abnormal carotid pulse, venous engorgement, and hepatojugular reflux on relevant physical examination. However, a $2.5 \times 2.5 \times 2 \text{ cm}^3$, smooth, hard, clear boundaries and solid nodule on the left thyroid gland was found. The patient had no familial genetic, psychosocial and exposure to radiation and toxins history. Furthermore, he was conducted in line with the care check-list. A timeline with relevant information, and follow-up process of the patient was shown in Fig. 1A. The pathological diagnosis process was shown in Fig. 1B.

Laboratory tests of the tumor biomarkers such as CA125 (17.20 U/ml) shown slightly increased, and cytokeratin 19 (CK19) fragment shown a critical value. The levels of thyroid function indexes and parathyroid hormone were normal. Thyroid function determination shown a free thyroxine (FT4) level of 112.70 nmol/L, thyroid-stimulating hormone (TSH) level of 1.83 $\mu\text{IU/ml}$, thyroglobulin (TG) level of 20.6 ng/ml and AbTg level of 15.50U/ml.

Then, he was admitted and underwent gray scale ultrasonography and color doppler flow imaging (CDFI). The gray scale ultrasonography shown a sized $2.1 \times 2.6 \times 2.7 \text{ cm}^3$ low echoic shadow and a few striped strong echoes (Fig. 2A). CDFI shown streaks of blood flow signal in and around the thyroid, and the pulsed wave shown the arterial spectrum with maximum speed of 21 cm/s (Fig. 2B). Hypoecho of lymph nodes in submandibular space and the lymphatic hilar blood flow signals were detected by CDFI (Fig. 2C).

The patient refused the fine needle aspiration (FNA) administration. And then, the unilateral radical resection of thyroid carcinoma was performed. After surgery, thyroid function determination shown a FT4 level of 100.20 nmol/L, TSH level of 3.19 $\mu\text{IU/ml}$, TG level of 3.76 ng/ml and AbTg level of 19.20U/ml.

The mass was about $3 \times 2 \times 2 \text{ cm}^3$ in size with a 0.2 cm capsule of the left thyroid. The histological characteristics were sclerosing with clear boundaries by the capsule (Fig. 3A). At low magnification, the extensive vitrification, calcification, and necrosis was in the background, and the tumor cells were squamous metaplasia, scattered, and arranged in clusters with diffuse growth pattern (Fig. 3B). Besides, some tumor cells and psammoma bodies have infiltrated to the capsule (Fig. 3C). At high magnification, the tumor cells were enlarged and shown stretched nucleus, the follicular epithelium cells of thyroid were atypical (Fig. 3D). Furthermore, the nuclear grooves (Fig. 3E) and internuclear pseudoinclusion (Fig. 3F) was detected.

Immunohistochemical staining shown strong positiveness of CK19, TTF-1, P40 and Galectin-3, partial positiveness of TG of tumor cells and negativeness of Calcitonin (Fig. 4A-F). Besides, CK-pan, Pax-8, P63,

P16, P53 and TPO was strong positive, CK5/6 and CD56 was partial positive, and then Syn and CgA was negative.

First generation sequencing techniques of thyroid carcinoma test containing AKT1, GNAS, KRAS, PIK3CA, PTEN, TERT, BRAF, HRAS, NRAS, PAX8/PPARG, RET, TP53, CTNNB1, NTRK, ZNF148, SPOP, EZH1 and TSHR was used in this case. The RET gene shown Exon 11c.2071G>A p.G691S mutation, and abundance of mutation was 52.53% (S1. A). The NRAS gene revealed Exon 3c.182A>G p.Q61R mutation, and abundance of mutation was 19.80% (S1. B). The TERT gene indicated c.-124C>T (C228T) mutation, and abundance of mutation was 15.08% (S1. C). The PIK3CA gene resulted Exon 21c.3140A>G p.H1047R mutation, and abundance of mutation was 1.50% (S1. D). Finally, we made the diagnosed of typical papillary thyroid carcinoma with squamous metaplasia and RET/NRAS/TERT/PIK3CA mutations pathologically.

The patient remained under careful observation by ultrasonic examination follow-up and treated with levothyroxine sodium tablets orally. and there was no recurrence or metastasis in the 6 months follow-up. The patient got appropriate perspective including the assessment and the episode of care in every 3 months. No adverse and unanticipated events happened during the period. Finally, the patient was satisfied with the treatment plan, process and prognosis, and will continue to follow up as prescribed by the doctors.

Discussion

DSVPTC is an uncommon variant of PTC which is macroscopically involved the thyroid extensively without forming a dominant mass, and microscopically revealed extensive fibrosis, squamous metaplasia and numerous psammoma bodies (12). But in this case, we macroscopically got a distinct mass with an incrassated capsule of the left thyroid.

Histopathology of DSVPTC always shows the numerous psammoma bodies, lymphoplasmacytic infiltrates with germinal centre, fibrous stroma, squamoid differentiation and nuclear characteristic of PTC (6, 13). In this case, the extensive vitrification, calcification, and necrosis was in the background, and the tumor cells were squamous metaplasia, scattered, some arranged in clusters with diffuse growth pattern. Besides, The tumor cells were atypical except the nuclear grooves and internuclear pseudoinclusion. Histology of this case seemed impossible to make an accurate diagnosis because of the morphometric atypism. Immunohistochemical staining results supported the thyroid primary adenocarcinoma, nonetheless, we still needed the genetic detection to distinguish the subtype of PTC.

BRAF mutation was significantly associated with increased cancer-related mortality among patients with PTC which support further investigation of the prognostic and therapeutic implications of BRAF status in PTC (14, 15). Several drugs have been developed, which inhibit signaling kinases or oncogenic kinases (BRAfV600E, RET/PTC), such as those associated with platelet-derived growth factor receptor and vascular endothelial growth factor receptor (16). But in this case, BRAF mutation was not found in PTC formalin-fixed, paraffin embedded tumor tissues.

Alterations of RET gene or protein have been found in diverse thyroid cancer subtypes, and observed in PTC, which result in RET fusion products (17). RET variant c.2071G>A (p.G691S) have been described in the general population as well as in patients with MTC and with Hirschsprung syndrome (18). MTCs produce calcitonin, measurement of which indicates the presence of tumor in at-risk individuals and the effectiveness of therapy in treated patients (19). In this case, the negativeness of Calcitonin could exclude the diagnosis of MTC, besides, there was no Hirschsprung syndrome in this patient. p.G691S also could act synergistically in the development or progression of follicular thyroid cancer (FTC) (20). But in this case, we were unable to diagnose the FTC because of the histopathology. Significantly, this is the first description of a p.G691S mutation in association with DSVPTC. Furthermore, NRAS and TERT mutations are related to higher PTC aggressiveness, and are potential use in diagnostics in PTC patients (21, 22). Mutational activation of PI3K signaling, through mutational activation of PIK3CA or loss of PTEN, are well described in aggressive thyroid cancer (23). In this case, we found the mutations of RET, NRAS, TERT and PIK3CA gene, it was useful for the diagnosis of PTC. Taking together, RET, NRAS, TERT and PIK3CA mutation happened on the same patient, and combined with histopathology and immunohistochemical results, it would be more reasonable to diagnose PTC although its histology was not typical. But we did not use the next generation sequencing to detect the gene mutation, that was the limitation of this case.

In summary, we presented an unusual and rare case of PTC with squamous metaplasia and RET, NRAS, TERT and PIK3CA mutation which was first reported. Although the tumor has characteristic histological features, awareness is important for its diagnosis. Therefore, genetic detection is vital to the diagnosis of thyroid adenocarcinoma, especially for the PTC with atypical morphology or rare metaplasia.

Abbreviations

NCBI

National Center for Biotechnology Information

PTC

papillary thyroid carcinoma

DSVPTC

diffuse sclerosing variant papillary thyroid carcinoma

MTC

medullary thyroid carcinoma

FTC

follicular thyroid cancer

FT4

free thyroxine

TSH

thyroid-stimulating hormone

CDFI

color doppler flow imaging

FNA
fine needle aspiration
TTF1
transcription termination factor 1
TG
thyroglobulin
TPO
thyroid peroxidase
P40
40s ribosomal protein SA
P16
biomineralization protein SpP16
CK
cytokeratin
P63
hypothetical protein 63
Pax-8
paired box 8
Syn
Synapsin
CgA
glycoprotein hormones, alpha polypeptide.

Declarations

Ethics approval and consent to participate

Informed consent was obtained in this case, and protocols were approved by the Ethics approval of Chongqing University Cancer Hospital. The patient provided informed consent for the publication of this report and any accompanying images.

Consent for publication

The patient consented all the individual person's data to publish.

Availability of data and materials

The datasets used or analysed during the current study are available from the corresponding author on reasonable request.

Competing interests

The authors declare that there is no conflict of interest.

Funding

No funding information.

Author's contributions

ZL contributed to acquisition, analysis and interpretation of patient data and the drafting of the manuscript. DG and YF contributed to the acquire the ultrasonic examination data. JZ contributed to the immunohistochemistry and molecular pathological methods. QJ and HH gave the final approval of the report. All authors read and approved the final manuscript.

Acknowledgements

Not applicable.

References

1. Kitahara S, Ito T, Hamatani S, Shibuya K, Shiba T. Thyroid papillary carcinoma recurring as squamous cell carcinoma: report of a case. *Surg Today* (2006) 36(2):171-174. doi: 10.1007/s00595-005-3107-3
2. Kunisue H, Mikami Y, Tanaka K, Sonoo H, Udagawa K, Yamamoto Y, et al. Metastatic papillary thyroid carcinoma of the submandibular lymph nodes with extensive squamous metaplasia: report of a case. *Surg Today* (2003) 33(10):751-754. doi: 10.1007/s00595-003-2581-8
3. Thompson LD, Wieneke JA, Heffess CS. Diffuse sclerosing variant of papillary thyroid carcinoma: a clinicopathologic and immunophenotypic analysis of 22 cases. *Endocr Pathol* (2005) 16(4):331-48. doi: 10.1385/ep:16:4:331
4. Imamura Y, Kasahara Y, Fukuda M. Multiple brain metastases from a diffuse sclerosing variant of papillary carcinoma of the thyroid. *Endocr Pathol* (2000) 11(1):97-108. doi: 10.1385/ep:11:1:97
5. Chan JK, Tsui MS, Tse CH. Diffuse sclerosing variant of papillary carcinoma of the thyroid: a histological and immunohistochemical study of three cases. *Histopathology* (1987) 11(2):191-201. doi: 10.1111/j.1365-2559.1987.tb02622.x
6. Carcangiu ML, Bianchi S. Diffuse sclerosing variant of papillary thyroid carcinoma. Clinicopathologic study of 15 cases. *Am J Surg Pathol* (1989) 13(12):1041-9. doi: 10.1097/00000478-198912000-00006
7. Spinelli C, Strambi S, Bakkar S, Nosiglia A, Elia G, Bertocchini A, et al. Surgical Management of Diffuse Sclerosing Variant of Papillary Thyroid Carcinoma. Experience in 25 Patients. *World J Surg* (2020) 44(1):155-162. doi: 10.1007/s00268-019-05230-5
8. Vuong HG, Kondo T, Pham TQ, Oishi N, Mochizuki K, Nakazawa T, et al. Prognostic significance of diffuse sclerosing variant papillary thyroid carcinoma: a systematic review and meta-analysis. *Eur J Endocrinol* (2017) 176(4):433-441. doi: 10.1530/EJE-16-0863

9. Pillai S, Gopalan V, Smith RA, Lam AK. Diffuse sclerosing variant of papillary thyroid carcinoma—an update of its clinicopathological features and molecular biology. *Crit Rev Oncol Hematol* (2015) 94(1):64-73. doi: 10.1016/j.critrevonc.2014.12.001
10. Póvoa AA, Teixeira E, Bella-Cueto MR, Batista R, Pestana A, Melo M, et al. Genetic Determinants for Prediction of Outcome of Patients with Papillary Thyroid Carcinoma. *Cancers (Basel)* (2021) 13(9):2048. doi: 10.3390/cancers13092048
11. Xing M, Haugen BR, Schlumberger M. Progress in molecular-based management of differentiated thyroid cancer. *Lancet* (2013) 381(9871):1058-69. doi: 10.1016/S0140-6736(13)60109-9
12. Joung JY, Kim TH, Jeong DJ, Park SM, Cho YY, Jang HW, et al. Diffuse sclerosing variant of papillary thyroid carcinoma: major genetic alterations and prognostic implications. *Histopathology* (2016) 69(1):45-53. doi: 10.1111/his.12902
13. Ni Y, Zhang Y, Xiang K, Zhao P, Pan G, Peng Y, et al. Axillary lymph node metastasis from papillary thyroid carcinoma with elevated CA 19-9 and CA 242 levels: a case report and literature review. *Gland Surg* (2021) 10(3):1147-1153. doi: 10.21037/gs-20-815
14. Xing M, Alzahrani AS, Carson KA, Viola D, Elisei R, Bendlova B, et al. Association between BRAF V600E mutation and mortality in patients with papillary thyroid cancer. *JAMA* (2013) 309(14):1493-501. doi: 10.1001/jama.2013.3190
15. Giannetta E, Isidori AM, Durante C, Di Gioia C, Longo F, Tombolini V, et al. Anaplastic thyroid carcinoma and foscarnet use in a multitarget treatment documented by 18F-FDG PET/CT: A case report. *Medicine (Baltimore)* (2017) 96(6):e5621. doi: 10.1097/MD.0000000000005621
16. Fallahi P, Ferrari SM, Galdiero MR, Varricchi G, Elia G, Ragusa F, et al. Molecular targets of tyrosine kinase inhibitors in thyroid cancer. *Semin Cancer Biol* (2020) S1044-579X(20)30249-2. doi: 10.1016/j.semcancer.2020.11.013
17. Salvatore D, Santoro M, Schlumberger M. The importance of the RET gene in thyroid cancer and therapeutic implications. *Nat Rev Endocrinol* (2021) 17(5):296-306. doi: 10.1038/s41574-021-00470-9
18. Elisei R, Cosci B, Romei C, Bottici V, Sculli M, Lari R, et al. RET exon 11 (G691S) polymorphism is significantly more frequent in sporadic medullary thyroid carcinoma than in the general population. *J Clin Endocrinol Metab* (2004) 89(7):3579-84. doi: 10.1210/jc.2003-031898
19. Traugott A, Moley JF. Medullary thyroid cancer: medical management and follow-up. *Curr Treat Options Oncol* (2005) 6(4):339-46. doi: 10.1007/s11864-005-0037-7
20. Khan MS, Pandith AA, Iqbal M, Naykoo NA, Khan SH, Rather TA, et al. Possible Impact of RET Polymorphism and Its Haplotypic Association Modulates the Susceptibility to Thyroid Cancer. *J Cell Biochem* (2015) 116(8):1712-8. doi: 10.1002/jcb.25130
21. Mio C, Verrienti A, Pecce V, Sponziello M, Damante G. Rare germline variants in DNA repair-related genes are accountable for papillary thyroid cancer susceptibility. *Endocrine* (2021) 73(3):648-657. doi: 10.1007/s12020-021-02705-1

22. Rusinek D, Pfeifer A, Cieslicka M, Kowalska M, Pawlaczek A, Krajewska J, et al. TERT Promoter Mutations and Their Impact on Gene Expression Profile in Papillary Thyroid Carcinoma. *Cancers (Basel)* (2020) 12(6):1597. doi: 10.3390/cancers12061597
23. Knippler CM, Saji M, Rajan N, Porter K, La Perle KMD, Ringel MD. MAPK- and AKT-activated thyroid cancers are sensitive to group I PAK inhibition. *Endocr Relat Cancer* (2019) 26(8):699-712. doi: 10.1530/ERC-19-0188

Figures

Figure 1

(A) The treatment and follow-up process. (B) The pathological diagnosis process.

Figure 2

(A) The image exhibited with gray scale ultrasonography. (B) The image exhibited with CDFI. (C) Lymph nodes in submandibular space by CDFI.

Figure 3

(A) The mass was shown microscopically ($\times 1$). (B) Extensive vitrification, calcification, and necrosis in the background ($\times 40$). (C) Tumor cells and psammoma bodies infiltrate the thyroid capsule ($\times 40$). (D) The atypical follicular epithelial cells ($\times 400$). (E) Nuclear grooves ($\times 800$). (F) Internuclear pseudoinclusion ($\times 800$). (The red arrow indicated the nuclear grooves and the blue arrow indicated the internuclear pseudoinclusion).

Figure 4

Immunohistochemical staining (×200) shown CK19 (A), TTF-1 (B), P40 (D) and Galectin-3 (E) strong positive, TG (C) partial positive of tumor cells and negative of Calcitonin (F).

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- [CAREchecklistEnglish2013.tif](#)
- [SupplementFigure1.tif](#)