Chromophobe renal cell carcinoma-like thyroid carcinoma: A novel clinicopathologic entity possibly associated with tuberous sclerosis complex

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Abstract. We report three cases of chromophobe renal cell carcinoma-like thyroid carcinoma as a novel clinicopathologic entity possibly associated with tuberous sclerosis complex. A 15-year-old female, a 19-year-old male, and a 21-year-old male presented with primary thyroid carcinoma. Two of the patients had associated tuberous sclerosis complex. Macroscopically, the carcinomas showed invasive growth. Histologically, the carcinoma cells showed a trabecular pattern with thin vascular stroma, and were characterized by abundant eosinophilic cytoplasm with perinuclear clearing, a prominent cell border, a wrinkled nuclear membrane, and binucleation, which are all features of chromophobe renal cell carcinoma. Immunohistochemically, the carcinoma cells were positive for thyroglobulin, TTF1, and PAX8, and negative for CD10, calcitonin, and carcinoembryonic antigen. Vascular invasion was visible in all cases, but distant metastasis was not detected during follow-up. The original pathological diagnoses of the three cases were widely invasive follicular thyroid carcinoma, poorly differentiated thyroid carcinoma, and oxyphilic variant of papillary thyroid carcinoma. Thus, the cases were similar to chromophobe renal cell carcinoma associated with tuberous sclerosis complex as they were characterized by histologic findings consistent with chromophobe renal cell carcinoma, occurrence in an adolescent or young adult, and favorable prognosis regardless of the presence of vascular invasion and an infiltrating growth pattern resembling poorly differentiated carcinoma. The etiopathogenesis also seemed to suggest the presence of the tuberous sclerosis complex genetic abnormality.

Key words: Thyroid, Chromophobe renal cell carcinoma, Tuberous sclerosis complex, Oncocytic carcinoma, Poorly differentiated carcinoma
adults (AYA), and two of them had TSC. The histology was analogous to that of CRCC. Here, we report three cases of chromophobe renal cell carcinoma-like thyroid carcinoma (CRETHCA), a novel clinicopathologic entity possibly associated with TSC.

Materials and Methods

We reviewed 12,064 primary thyroid carcinoma cases resected in Kuma Hospital from 2005 to 2015, and selected two cases (cases 1 and 2) with CRCC-like appearance. CRCC-like appearance was defined as a prominent cell border, perinuclear clear cytoplasm, a wrinkled nuclear membrane, and binucleation. One case (case 3) from Fukushima Medical University School of Medicine with similar histological findings was also included in this study. Immunohistochemical study was performed using following antibodies, thyroglobulin (polyclonal, 2 dilution, Dako, Glostrup, Denmark), TTF1 (8G7G3/1, 100 dilution, Dako Cytomation, Carpinteria, CA, USA), EMA (E29, 5 dilution, Dako, Carpinteria, CA, USA), PAX8 (polyclonal, 200 dilution, Protein Tech, Chicago, IL, USA), cytokeratin 7 (OV-TL 12/30, 50 dilution, DAKO, Carpinteria, CA, USA), cytokeratin 19 (RCK108, 250 dilution, Dako, Glostrup, Denmark), the anti-mitochondria antibody (AMA) (MTC02, 1.5 dilution, NeoMarkers, Fremont, CA, USA), calcitonin (polyclonal, 2 dilution, Dako, Glostrup, Denmark), carcinoembryonic antigen (COL1, 3 dilution, HISTOFINE, Tokyo, Japan), GATA3 (L50-823, 400 dilution, Biocare Medical, Concord, CA, USA), CD10 (56C6, 100 dilution, Novocastra, Newcastle, UK), CD117 (polyclonal, 50 dilution, Dako, Carpinteria, CA, USA), p63 (4A4, 1 dilution, HISTOFINE, Tokyo, Japan), E-cadherin (NCH38, 100 dilution, Glostrup, Denmark), MUC1 (Ma695, 100 dilution, Novocastra, Newcastle, UK), p53 (DO7, 1000 dilution, Dako, Glostrup, Denmark), and Ki-67 (MIB1, 200 dilution, Glostrup, Denmark). The mutational status of the \textit{BRAF} (exon 15), \textit{NRAS}, \textit{HRAS} and \textit{KRAS} (exons 2 and 3) genes was examined as described previously (case 1 and 2, [6]; case 3, [7]).

Results

Case presentation (Table 1)

Case 1
The patient was a 15-year-old Japanese female with a history of TSC associated with epilepsy, a cardiac

<table>
<thead>
<tr>
<th>Table 1 Clinical and pathological findings of three chromophobe renal cell carcinoma-like thyroid carcinoma cases</th>
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<tr>
<td><strong>Age (years)</strong></td>
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<tr>
<td>Gender</td>
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<tr>
<td>Tuberous sclerosis complex</td>
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<tr>
<td>Location (lobe)</td>
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<td>Size (cm)</td>
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<td>Original histologic diagnosis</td>
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<td>Necrosis</td>
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<td>Extrathyroidal invasion</td>
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<td>Nodal metastasis (at surgery)</td>
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<td>Distant metastasis</td>
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<td>Outcome</td>
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<td>(follow-up period)</td>
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Lt, Left; Rt, Right; FTC, follicular thyroid carcinoma; PDTC, poorly differentiated thyroid carcinoma; PTC, Papillary thyroid carcinoma; Lo, lobectomy; TT, total thyroidectomy; MNLR, modified neck lymph node resection; CNLR, central neck lymph node resection; Bi, Bilateral; RI, Radioactive iodine therapy.
Chromophobe RCC-like thyroid carcinoma

Pathological findings

The tumors were solid and whitish tan in color. In case 1 and 3, the tumors were mostly encapsulated and had focally infiltrated into the surrounding thyroid tissue; however, they were limited to the thyroid (Fig. 1). The tumor in case 2 occupied the left lobe, was non-encapsulated, and showed extensive infiltrative growth. The cut surface was markedly lobulated (Fig. 2).

Case 1

The patient was a 52-year-old Japanese male. He had a history of TSC associated with epilepsy, bilateral renal cysts, and renal angiomyolipoma. His serum thyroglobulin, T4, and TSH levels were 11.4 ng/mL, 1.06 ng/dL, and 2.150 µIU/mL, respectively. A nodule measuring 16.9 mm in the greatest dimension was found by ultrasonographic examination of the right lobe of the thyroid. The tumor was irregular, heterogeneous, and low echoic, and diagnosed as oxyphilic variant of papillary thyroid carcinoma (PTC) based on FNAC findings. The patient underwent a right lobectomy with central neck lymph node resection. The histologic diagnosis was same as that from FNAC. Neither regional recurrences nor distant metastases were found during the 15-month follow-up.

Fig. 1 Photograph of resected tumor sections showing a solid and mostly encapsulated tumor. Satellite nodules indicating capsular invasion are visible in the upper portion of the tumor (Case 1).

Fig. 2 Photograph of resected tumor sections showing a markedly lobulated tumor with extensive infiltrative growth (Case 2).
Microscopic findings of the carcinoma cells seen in the three cases were mostly similar. The tumors were composed predominantly of large polygonal carcinoma cells showing a trabecular pattern (Fig. 3). Focally, alveolar (Fig. 4) and microfollicular patterns were seen. The tumor nests were surrounded by thin vascular stroma. The lumen of the microfollicles displayed innsipssated colloid. The cell membrane of the carcinoma cells was well defined. The cytoplasm was abundant, finely granular, and eosinophilic. The nuclei were round to irregular and had prominent nucleoli (Fig. 5). The chromatin was pale and vesicular. Binuclear carcinoma cells were frequently observed (Fig. 5). Markedly irregular nuclei with perinuclear clearing mimicked koilocytosis (Fig. 6).

In case 1, carcinoma cells with abundant eosinophilic cytoplasm were focally present (Fig. 7), and the colloid within the microfollicles was occasionally associated with calcification mimicking psammoma bodies. Vascular invasion was present in all cases, and extensive in case 1 and 2 (Fig. 8). Necrotic areas were focally observed in case 1 and 2. Case 2 showed extrathyroidal invasion. In case 1 and 3, the carcinoma was limited to the thyroid. Case 2 and 3 revealed nodal metastasis.
Chromophobe RCC-like thyroid carcinoma

Cytoplasm (eosinophilic variant) may predominate [1, 2]. Histopathology of the three cases we studied showed infiltrative growth, a trabecular pattern, rich vascular stroma, well-defined cell membranes, markedly irregular nuclei, binucleation, a koilocytotic appearance, and

Histochemical and immunohistochemical findings

Histochemical and immunohistochemical results were essentially similar for the three cases and are shown in Table 2. Hale’s colloidal iron staining was focally and weakly positive in case 2 (Fig. 9), but negative in case 1 and 3. Immunohistochemically, the tumor cells were positive for thyroglobulin (Fig. 10a), TTF1 (Fig. 10b), and EMA, but the staining was not intensive. PAX8 (Fig. 10c) was strongly immunoreactive. Cytokeratin 7 and cytokeratin 19 were detected at the periphery of the cytoplasm. Similarly, AMA was expressed at the periphery (Fig. 10d). On the other hand, the carcinoma cells tested negative for calcitonin, carcinoembryonic antigen, GATA3, CD10, CD117, and p63. E-cadherin was detected in the cell membrane. The apical cell membrane of carcinoma cells was focally positive for MUC1, while p53 staining was weakly positive. Ki-67 labeling indices for case 1, 2, and 3 were 10.5%, 8.2%, and 3.5%, respectively.

Mutational analysis

\(BRAF\), \(NRAS\), \(HRAS\), and \(KRAS\) mutations were not detected in any of the three cases.

Discussion

Microscopically, CRCC is characterized by large polygonal cells with prominent cell borders, perinuclear halos, wrinkled nuclear membranes, and binucleation, resulting in a “plant cell appearance” or koilocytosis [1-4, 8]. Carcinoma cells with granular and eosinophilic cytoplasm (eosinophilic variant) may predominate [1, 2]. Histopathology of the three cases we studied showed infiltrative growth, a trabecular pattern, rich vascular stroma, well-defined cell membranes, markedly irregular nuclei, binucleation, a koilocytotic appearance, and
Fig. 9  Hale’s colloidal iron staining is focally and weakly positive (Case 2).

Fig. 10  Carcinoma cells are weakly positive for thyroglobulin (a: Case 1) and TTF-1 (b: Case 3). PAX8 is strongly positive (c: Case 1). Anti-mitochondria antibody is expressed at the periphery of the cytoplasm (d: Case 3).
associated vascular invasion. The histologic findings of the carcinoma cells in our three cases and CRCC were similar. Metastatic renal cell carcinoma (RCC) can occur as a solitary lesion and even before a diagnosis of RCC is established [9, 10]. Therefore, we should distinguish our cases from metastatic RCC. However, there is no doubt that they primarily occurred in the thyroid, because they were immunohistochemically positive for thyroglobulin and TTF1, and negative for CD10, which is expressed in RCC.

Our cases (1, 2, and 3) had been originally diagnosed as FTC, PDTC, or oncocytic variant of PTC, respectively. However, FTC was excluded by the predominant trabecular growth pattern and marked nuclear irregularity of the tumor cells, which are features of PDTC [11-13]. Indeed, the predominant trabecular growth pattern, cellular atypism, necrosis, and vascular invasion indicated PDTC. However, compared to usual cases of PDTC, the outcome was extremely favorable and the patients were younger [11-13]. The trabecular growth pattern and abundant eosinophilic cytoplasm may point to a diagnosis of poorly differentiated oncocytic follicular carcinoma (PDOFC) [14]. However, oncocytic carcinoma cells were a minor component in our cases. In addition, perinuclear halos and wrinkled nuclear membranes, which were observed in our cases, are not features of PDOFC. In addition, the mutational analysis that we performed did not identify the genetic alterations that were usually detected in PTC and FTC. To the best of our knowledge, CRETHCA has not been described previously.

Interestingly, CRETHCA and CRCC share common clinical findings. Two of the three cases we studied were TSC patients. TSC is a genetic disorder that causes tumors in many different organs, primarily in the brain, eyes, heart, kidney, skin, and lungs [3, 4]. The incidence of RCC in TSC patients is reported to be 2% to 4%, which is higher than that in the general population [15]. CRCC comprises 5% to 10% of RCC cases, but RCCs in TSC patients are predominantly classified as CRCC [3, 16-18]. CRCC is diagnosed mainly in the sixth decade of life, but TSC-associated CRCC is diagnosed more frequently at a younger age, and our cases were also in AYA. According to a report by Guo et al. [3], there was no evidence of metastatic disease after surgery in TSC-associated RCC. Our cases included vascular invasion and a growth pattern consistent with PDC, but nevertheless, the outcome was favorable.

Patients with TSC present with multiple tumor types involving multiple organs. There are several reports of thyroid lesions in patients with TSC. Auladell et al. retrospectively reviewed chest computed tomography scans of 93 patients with TSC [19], and found thyroid abnormalities in 19 (20.4%) patients. However, a histological examination was performed only in two cases (one follicular adenoma, one papillary carcinoma). Dicorato et al. reported a case of medullary thyroid carcinoma associated with TSC [20]. Thus far, however, the relationship between thyroid tumors and TSC has not been established beyond doubt.

As two CRETHCA cases were associated with TSC, we think that the etiopathogenesis of CRETHCA could link to the genetic abnormality of TSC. The occurrence of CRETHCA in AYA and favorable prognosis suggests this possibility. The case of CRETHCA not associated with TSC in this report may be a sporadic form. This is similar to the cribriform variant of PTC, which involves familial polyposis coli-associated and sporadic cases, occurrence in AYA, and favorable prognosis regardless of the presence of solid growth and tall cell components [21, 22].

In conclusion, we report three CRETHCA cases with unique microscopic findings that have not been described previously. They involve AYA who had a favorable prognosis regardless of vascular invasion and growth patterns resembling PDTC. In two cases, the TSC genetic abnormality seemed to be related to the etiopathogenesis of CRETHCA. Further genetic studies are necessary to confirm the hypothesis.

**Disclosure**

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**References**


2. Stec R, Grała B, Maczewski M, Bodnar L, Szczyluk C (2009) Chromophobe renal cell cancer - review of the literature and potential methods of treating metastatic...


