Case Report

t(6; 11) renal cell carcinoma. A case report successfully diagnosed by using fluorescence in situ hybridization

Hidekazu Nishizawa,[1,2] Masaya Baba,[2] Mitsuko Furuya,[3] Ikuma Kato,[4] Ryoma Kurahashi,[1] Yumi Honda,[5] Yoshiki Mikami,[5] Yoji Nagashima,[6] Masatoshi Eto[7] and Tomomi Kamba[1]

[1]Department of Urology, Faculty of Life Sciences, and [2]Laboratory of Cancer Metabolism, International Research Center for Medical Sciences (IRCMS), Kumamoto University, Kumamoto, [3]Pathology Center, GeneticLab Co., Ltd, Sapporo, [4]Department of Molecular Pathology, Yokohama City University, Yokohama, [5]Department of Diagnostic Pathology, Kumamoto University Hospital, Kumamoto, [6]Department of Surgical Pathology, Tokyo Women's Medical University Hospital, Tokyo, and [7]Department of Urology, Graduate School of Medical Sciences, Kyushu University, Fukuoka, Japan

Abbreviations & Acronyms
ACTB = Actin Beta
AMACR = α-methyl acyl CoA racemase
CADM2 = cell adhesion molecule 2
CD10 = neprilysin
CK7 = cytokeratin 7
CLTC = clathrin heavy chain
COL21A1 = collagen type XXI alpha 1 chain
CT = computed tomography
EWSR1 = Ewing sarcoma breakpoint region1
FISH = fluorescence in situ hybridization
KHDRBS2 = KH domain containing, RNA binding, signal transduction associated 2
MALAT1 = metastasis associated in lung adenocarcinoma transcript-1
MiT = microphthalmia transcription
PPP1R10 = Protein Phosphatase 1 Regulatory Subunit 10
RCC = renal cell carcinoma
tRCC = translocation renal cell carcinoma
TFEB = transcription factor EB
TFEC = transcription factor EC

Correspondence: Masaya Baba M.D., Ph.D., International Research Center for Medical Sciences, Kumamoto University, 2-2-1 Honjo, Chuo-ku, Kumamoto City 860-0811, Japan. Email: babam@kumamoto-u.ac.jp

How to cite this article: Nishizawa H, Baba M, Furuya M *et al.* t(6; 11) renal cell carcinoma. A case report successfully diagnosed by using fluorescence in situ hybridization. *IJU Case Rep*. 2021; **4**: 375–378.

Received 18 May 2021; accepted 16 July 2021.
Online publication 12 August 2021

Introduction: Definitive diagnosis of translocation renal cell carcinoma is challenging. We herein experienced a case of translocation(6;11) renal cell carcinoma, successfully diagnosed by using fluorescence in situ hybridization.
Case presentation: During the follow-up of a 21-year-old man with Crohn's disease, computed tomography revealed a 40-mm mass in the right kidney. Since imaging could not exclude malignancy, needle biopsy was performed. The histological diagnosis from the biopsy specimen was renal cell carcinoma, but histological typing had not been done adequately. A laparoscopic partial nephrectomy was then performed. Transcription factor EB immunoreactivity was positive, transcription factor EB rearrangement was shown by break apart and fusion fluorescence in situ hybridization. As a result, a definitive diagnosis of t(6; 11) renal cell carcinoma was made. There has been no recurrence for 5 years.
Conclusion: Transcription factor EB immunohistochemistry and fluorescence in situ hybridization are useful diagnostic tools for renal tumors of young generation.

Key words: FISH, immunohistochemistry, MiT family translocation renal cell carcinoma, t(6;11), transcription factor EB (TFEB).

Keynote message

The lack of recognition of TFEB-tRCC and the technical complexity of the diagnosis hinder the accurate diagnosis of this relatively rare disease. Immunohistochemical staining for the TFEB and FISH are essential for young renal tumors and for any suspicious cases.

Introduction

RCC in children and adolescents is rare. However, MiT family translocation RCC (WHO 2016 classification) is known as a relatively frequent RCC subtype in children and adolescents (30%) than in adults (3%).[1] In t(6;11) RCC, the chimeric gene involving the TFEB gene results in a dramatic upregulation of TFEB protein levels, which is expected to be cancerous. The case reports of t(6;11) RCC are very limited, and definitive diagnosis requires the demonstration of the chromosomal rearrangement. We report a case of t(6;11) RCC, which was successfully diagnosed by using FISH.

Case presentation

A 21-year-old male was referred to our department because of a right kidney tumor on follow-up contrast-enhanced CT during treatment of Crohn's disease. Abdominal CT demonstrated a 40-mm neoplastic lesion in the lower pole of the kidney, which had slightly poor contrast in the early phase (Fig. 1). Because it is not possible to judge benign or malignant

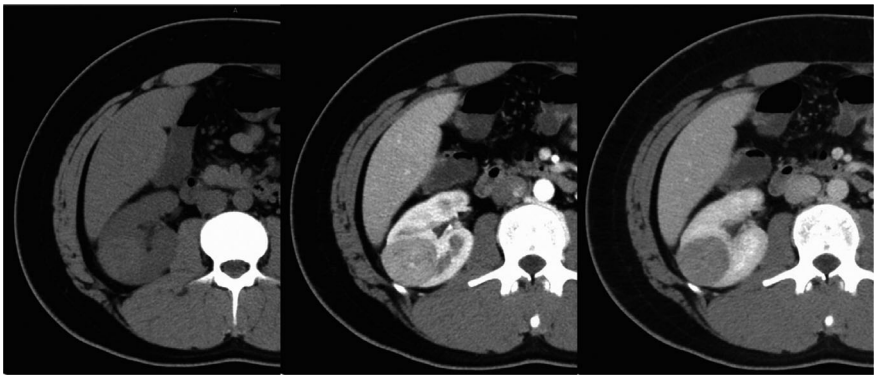

Fig. 1 Abdominal contrast-enhanced CT. A 40-mm renal tumor was detected in the right kidney, which had poor enhancement during the early phase and moderate enhancement during the late phase.

from the image, CT-guided needle biopsy was performed. Histopathological analysis demonstrated alveolar and papillary growth of tumor cells. Routine immunohistochemistry failed to suggest a definitive diagnosis. Considering its malignant potential, laparoscopic partial right nephrectomy was performed. The tumor was demarcated from the renal parenchyma with a pseudocapsule and the cut surface of the tumor was light tan, different from that of representative clear cell RCC (Fig. 2). Histologically, tumor cells with round nuclei and abundant cytoplasm with slightly bright granules proliferated in the form of alveolar lesions. Immunostaining was negative for CK7, and positive for CD10 and AMACR (clone P504S). As Melan A, a marker for MiT family translocation RCCs, was positive, we performed TFE3 and TFEB immunohistochemistry. TFE3 was negative, but TFEB was positive (Fig. 3). Based on the above results, t(6;11) RCC was highly suspected and cytogenetic examination was carried out as a definitive diagnosis. By FISH using a TFEB split probe, TFEB gene rearrangement was confirmed. Furthermore, FISH with MALAT1-TFEB fusion probe demonstrated MALAT1-TFEB gene fusion (Fig. 4). It led to the diagnosis of t(6;11) RCC.

No distant metastasis or recurrence was observed until 5 years after the operation.

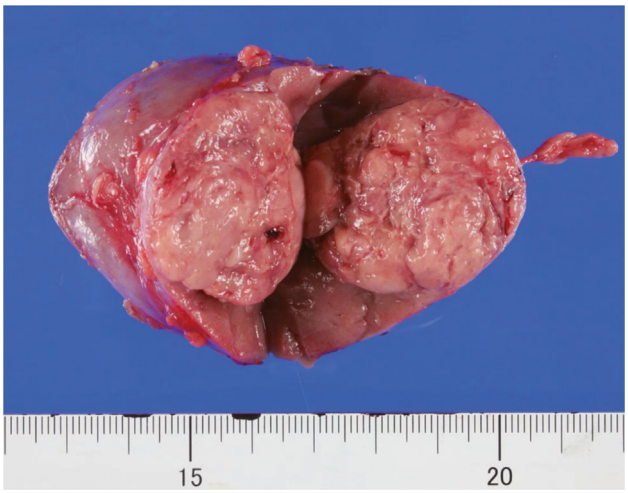

Fig. 2 Gross findings of the resected tumor. The tumor was well demarcated with a pseudocapsule. The cut surface was light tan in color.

Discussion

The t(6;11) RCC is a newly introduced histological type of MiT family translocation RCC in the 2016 WHO classification. The t(6;11) RCC is 0.02% of all RCC and the number of reported cases is under 100 worldwide.[2,3] The TFEB gene, which encodes a transcription factor TFEB belonging to the MiT family (MiTF, TFE3, TFEB, and TFEC), is located on chromosome 6. In t(6;11) RCC, it is fused with another partner gene, MALAT-1, resulting in a dramatic upregulation of TFEB protein levels. This event is considered essential in carcinogenesis. Similar to Xp11 translocation RCC, t(6;11) RCC occasionally develops in young generation. The average of patients' age at onset is 34 years. Xp11 translocation RCC tends to cause lymphatic metastases, whereas t(6;11) RCC preferentially causes hematogenous metastases.[3] Most cases are slowly progressive and have relatively favorable prognoses, but there are several reports that the 5-year survival rate is 48%.[1] In addition, approximately 17% are highly malignant.[3] Risk factors include tumor size, elderly onset, presence of mitotic activity, and tumor necrosis findings.[3,4] Besides t(6;11) RCC, TFEB-amplified RCC is another subtype and is suggested as poor prognostic.[5,6] The cause of the poor prognosis could be explained that vascular endothelial growth factor A, which exists on the short arm of chromosome 6, is amplified together with TFEB.[7]

Imaging characteristics of t(6;11) RCC are not well established yet, although the contrast effect in the early phase of contrast-enhanced CT was reported to be poor, as in this case, and a pattern that gradually increased over time was reported. As similar findings are observed in non-clear cell RCC and benign renal tumor, it is considered difficult to confirm the diagnosis by preoperative imaging modalities such as CT and magnetic resonance imaging.[8]

About histopathological features of t(6;11) RCC, no macroscopically characteristic findings have been reported so far. The tumor of the present case was well circumscribed and light tan in color. It is difficult for pathologists to diagnose rare histologic types. In the present case, characteristic histology, i.e., biphasic pattern composed of larger epithelioid cells and small lymphocyte-like cells, helped us to suspect the possibility of t(6;11) RCC. Immunoreactivity for melanosome-associated antigen (clone, HMB45), Melan A and cathepsin K also supported the diagnosis. MiT family transcription factors are involved in the differentiation of precursor cells into melanocytes and osteoclasts, and induce the expression of

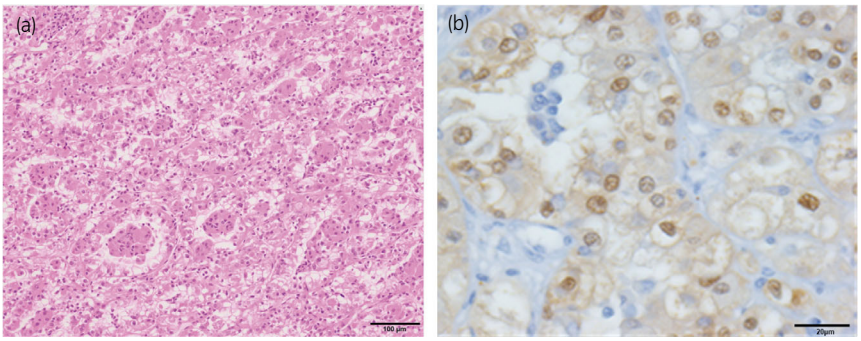

Fig. 3 Microscopic findings. (a) Hematoxylin and eosin staining demonstrated a biphasic pattern composed of small clustered cells surrounded by larger epithelioid cells with granular eosinophilic and clear cytoplasm. (b) Immunohistochemistry of the resected specimen demonstrated nuclear TFEB staining.

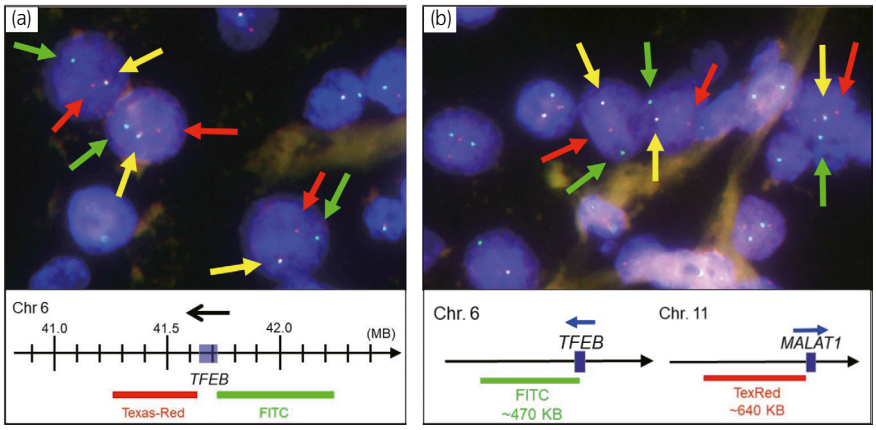

Fig. 4 FISH. (a) Split probe FISH showing TFEB gene rearrangement. Green or red: rearrangement gene. (b) Fusion probe FISH showing fusion between TFEB and MALAT1. Green: TFEB gene, red: MALAT1 gene, yellow: indicative of fusion gene.

melanocyte-related antigens and osteoclast markers.[7] Immunoreactivities for these antigens exclude clear cell RCC.[3]

As for fusion partner genes, MALAT1 accounts for 81% of TFEB translocation partners, whereas KHDRBS2, COL21A1, CADM2, CLTC, EWSR1, ACTB, and PPP1R10 have also been reported.[2] Immunostaining with anti-TFEB antibody is useful for screening, but the identification of the fusion gene by FISH or RT-PCR is needed. Unfortunately, we did not have a frozen tissue and failed to identify the exact fusion points by RT-PCR. Therefore, we performed two steps FISH analysis for diagnosis using a commercially available products from GSP Lab., Inc (Kobe, Japan). Product No SP078 and TR046. The first step involves a split probe assay to detect whether a TFEB gene rearrangement is present. The second step is a fusion probe assay using a MALAT1-TFEB probe, telomeric to TFEB and centromeric to MALAT1. A colocalized signal represents a fusion between TFEB and the partner gene. The sequential FISH analyses led us to a definitive diagnosis of t(6;11) translocation RCC.

Recent reports of Xp11.2 tRCC have reported that there is no difference in OS between partial nephrectomy and radical nephrectomy depending on the tumor size.[9] As the number of previous cases is small, the effects of drug therapy before and after surgery are unknown, and strict follow-up is required in the future.

Conclusion

In this case, immunohistochemistry for TFEB and two step FISH using TFEB split probe and MALAT1-TFEB fusion probe successfully demonstrated the t(6;11) RCC. The prognosis of this case is expected to be favorable because the risk factors such as the tumor size, elderly onset do not correspond to this case. However, careful follow-up is required for a better understanding of the rare tumor type preferentially affecting juvenile patients.

Acknowledgments

This work was supported by JSPS KAKENHI (21K06914, 18H02938) and by research grants from the Joint Usage/Research Center Program of the Advanced Medical Research Center, Yokohama City University.

Conflict of interest

The authors declare no conflict of interest.

Approval of the research protocol by an Institutional Reviewer Board

The study was approved by the Institutional Review Board of Kumamoto University (approval number, 1245).

Informed consent

Written informed consent was obtained from the all patients.

Registry and the Registration No. of the study/trial

Not applicable.

References

1 Wyvekens N, Rechsteiner M, Fritz C *et al*. Histological and molecular characterization of TFEB-rearranged renal cell carcinomas. *Virchows Arch.* 2019; **474**: 625–31.

2 Xia QY, Wang XT, Fang R *et al*. Clinicopathologic and molecular analysis of the TFEB fusion variant reveals new members of TFEB translocation renal cell carcinomas (RCCs): expanding the genomic spectrum. *Am. J. Surg. Pathol.* 2020; **44**: 477–89.

3 Calio A, Segala D, Munari E, Brunelli M, Martignoni G. MiT family translocation renal cell carcinoma: from the early descriptions to the current knowledge. *Cancers* 2019; **11**: 1110.

4 Peckova K, Vanecek T, Martinek P *et al*. Aggressive and nonaggressive translocation t(6;11) renal cell carcinoma: comparative study of 6 cases and review of the literature. *Ann. Diagn. Pathol.* 2014; **18**: 351–7.

5 Calio A, Brunelli M, Segala D *et al*. t(6;11) renal cell carcinoma: a study of seven cases including two with aggressive behavior, and utility of CD68 (PGM1) in the differential diagnosis with pure epithelioid PEComa/epithelioid angiomyolipoma. *Mod. Pathol.* 2018; **31**: 474–87.

6 Argani P, Reuter VE, Zhang L *et al*. TFEB-amplified renal cell carcinomas: an aggressive molecular subset demonstrating variable melanocytic marker expression and morphologic heterogeneity. *Am. J. Surg. Pathol.* 2016; **40**: 1484–95.

7 Calio A, Brunelli M, Segala D *et al*. VEGFA amplification/increased gene copy number and VEGFA mRNA expression in renal cell carcinoma with TFEB gene alterations. *Mod. Pathol.* 2019; **32**: 258–68.

8 Martignoni G, Bonetti F, Chilosi M *et al*. Cathepsin K expression in the spectrum of perivascular epithelioid cell (PEC) lesions of the kidney. *Mod. Pathol.* 2012; **25**: 100–11.

9 Geller JI, Cost NG, Chi YY *et al*. A prospective study of pediatric and adolescent renal cell carcinoma: a report from the Children's Oncology Group AREN0321 study. *Cancer* 2020; **126**: 5156–64.