Human Pathology (2017) **61**, 205–209

ELSEVIER

Human PATHOLOGY

www.elsevier.com/locate/humpath

Case study

Metachronous anaplastic sarcoma of the kidney and thyroid follicular carcinoma as manifestations of *DICER1* abnormalities☆

Misa Yoshida MD[a,b], Satoshi Hamanoue MD, PhD[c], Masafumi Seki MD[b], Mio Tanaka MD, PhD[a], Kenichi Yoshida MD, PhD[d], Hiroaki Goto MD, PhD[c], Seishi Ogawa MD, PhD[d], Junko Takita MD, PhD[b], Yukichi Tanaka MD, PhD[a,*]

[a]*Department of Pathology, Kanagawa Children's Medical Center, Kanagawa 232-8555, Japan*
[b]*Department of Pediatrics, Graduate School of Medicine, The University of Tokyo, Tokyo 113-8655, Japan*
[c]*Department of Hematology/Oncology and Regenerative Medicine, Kanagawa Children's Medical Center, Kanagawa 232-8555, Japan*
[d]*Department of Pathology and Tumor Biology, Kyoto University, Kyoto 606-8501, Japan*

Received 18 January 2016; revised 2 May 2016; accepted 15 June 2016

Keywords:
Anaplastic sarcoma of the kidney;
Thyroid carcinoma;
Pleuropulmonary blastoma;
DICER1;
RNase III

Summary Anaplastic sarcoma of the kidney (ASK) is a tumor found in the pediatric age group and shows many histopathological similarities to pleuropulmonary blastoma (PPB). We present a 12-year-old girl diagnosed with ASK and, 3 years later, with thyroid follicular carcinoma (TFC) with *DICER1* abnormalities. Germline insertion/deletion (p. G1809_S1814delinsA) and independent somatic mutations (p. E1705K in ASK, p. E1813D in TFC) were identified. All of these abnormalities are in the catalytic domain of RNase IIIb. Single-nucleotide polymorphism genotyping microarray revealed independent copy number alterations (trisomy 8, monosomy 10, loss of 17p, and partial gain of 17q in ASK; trisomy 5 and partial loss of Xq in TFC). The copy number alteration pattern of ASK was similar to the pattern previously reported in PPB. The present case implies that ASK is a renal counterpart of PPB and that ASK with *DICER1* abnormalities should be suspected in a broader age group than PPB.

1. Introduction

Anaplastic sarcoma of the kidney (ASK) is a rare tumor first described in 2007 [1]. It mainly occurs in children and adolescents. It is histologically characterized by widespread anaplastic changes with no neoplastic epithelial structure, nephrogenic rests, or blastemal elements. Its histological similarity to pleuropulmonary blastoma (PPB) has been noted.

In 2009, germline mutations of *DICER1* in familial PPB were detected in 4 families [2]. Later, *DICER1* became known as the causative gene for 1 type of familial cancer syndrome presenting with increased risk of PPB, cystic nephroma (CN), thyroid tumors, ovarian Sertoli-Leydig cell tumor, and others [3]. Recently, Doros et al [4] reported 4 cases of "CN-associated renal sarcoma" with histology similar to that of

☆ Disclosures: The authors have no financial relationship and no funding to disclose.

* Corresponding author: Department of Pathology, Kanagawa Children's Medical Center, 2-138-4, Mutsukawa, Yokohama Minami-ku, Kanagawa 232-8555, Japan.
E-mail address: ytanaka@kcmc.jp (Y. Tanaka).

http://dx.doi.org/10.1016/j.humpath.2016.06.024
0046-8177/

PPB, detecting *DICER1* mutations in 2 cases. Wu et al [5] reported 2 cases of ASK both with *DICER1* germline and somatic mutations and suggested that ASK and CN-associated renal sarcoma are part of the same entity.

We previously reported a case of ASK with cystic thyroid nodules [6]. Later, this patient developed thyroid carcinoma. We suspected that this case may be an example of *DICER1*-related disorders and therefore performed combined sequence-based and array-based genetic analyses.

2. Materials and methods

This study was approved by the ethics boards of Kanagawa Children's Medical Center (approval no. 82-04) and the University of Tokyo (approval no. 1598-9).

2.1. Case report

A 12-year-old girl was diagnosed with ASK, stage 4 (metastases to the presacral soft tissues, lungs, and bone) [6]. Her medical history was unremarkable. Her family history was significant for her mother having undergone thyroidectomy at the age of 17 years (details unknown). Other family members reportedly had no history of malignancy. She underwent nephrectomy and chemoradiotherapy with peripheral blood stem cell transplantation. Multiple cystic-appearing thyroid nodules were detected by ultrasonography at the onset of ASK, but aspiration biopsy revealed no malignancy. At the age of 15 years, a large mass arose in the right thyroid lobe and hemithyroidectomy was performed. The diagnosis was minimally invasive thyroid follicular carcinoma (TFC). She is currently disease-free 8 years after initial diagnosis.

2.2. Pathology and immunohistochemistry

Slide preparation and histological/immunohistochemical study were performed as previously described [6]. We reviewed the formalin-fixed renal tumor macroscopically and found some cyst-like lesions. These lesions were histologically examined to evaluate for the presence of CN.

2.3. Targeted deep sequencing

Genomic DNA of the ASK and TFC tumors was extracted from stored fresh-frozen samples. Deep sequencing of *DICER1* in these tumors was performed using a genome analyzer (Hiseq 2000; Illumina Inc, San Diego, CA). Primers were designed to cover all the coding exons of *DICER1* [7]. Each amplicon was amplified using *Not* I–tagged primer and mixed together after confirmation of successful amplification by gel electrophoresis. These amplicons were digested with *Not* I followed by ligation with T4 DNA ligase. They were sonicated into fragments of an average size of 200 base pairs using a Covaris sonicator and used for the generation of sequencing libraries with a kit (NEBNext Ultra DNA library Prep Kit for Illumina; New England Biolabs, Ipswich, MA). Data processing was performed by previously described methods [8].

2.4. Sanger sequencing

To confirm the results of deep sequencing and to investigate germline materials, Sanger sequencing was performed. Germline DNA was obtained from blood samples from the patient, her parents, and siblings.

2.5. Single-nucleotide polymorphism genotyping microarray

Single-nucleotide polymorphism (SNP) genotyping microarray was performed by hybridizing DNA from tumors to a microarray chip (Affymetrix GeneChip, CytoScan HD; Affymetrix, Inc, Santa Clara, CA). Signal ratios between tumors and normal references were calculated in an allele-specific manner, and allele-specific copy numbers were inferred from the observed signal ratios based on a hidden Markov model using CNAG/AsCNAR software [9].

3. Results

3.1. Pathological features

The pathological features of the renal tumor were described previously (Fig. 1A and B) [6]. Additional examination of the cyst-like lesions showed that they were cavities within the tumor that lacked any lining; there were no true cysts. The resected right lobe of the thyroid measured 35 × 25 × 20 mm and weighed 9.8 g. On cut surface, a 20 × 20–mm grayish-white encapsulated solid mass was noted.

Histologically, the tumor was encapsulated and showed cellular growth of follicular architecture (Fig. 1C and D). The neoplastic cells had mildly enlarged hyperchromatic nuclei. There were few mitotic figures and no calcifications. Although the tumor cells invaded the fibrous capsule in some areas, no tumor extension beyond the capsule was seen. Minimal vascular invasion was found. A diagnosis of minimally invasive follicular carcinoma was made from these findings.

3.2. Sequence-based genetic analyses

Sequencing revealed *DICER1* compound heterozygous aberrations in both the ASK and TFC, which consisted of a common 17–base pair in-frame insertion/deletion (indel) (c.5426_5442del GGGATATTTTGAGTCGinsCA, p. G1809_S1814delinsA) and independent missense mutations (c. G5113A, p. E1705K in ASK and c. G5439T, p. E1813D in TFC) (Fig. 2A-C). The common indel was detected in the