Clinical Case Report

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Decreased capillary density in renal cell carcinoma

Evidence from a case report with micro-computerized tomography

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Abstract

Rationale: Conventional computerized tomography (CT) examination can differentiate renal cortical tumor from urothelial carcinoma on the basis of the highly contrast-enhanced vessels in renal cortical tumors. However, the capillary distribution of renal cell carcinoma (RCC) has been under-investigated. Here, we present a micro-CT image of tumor tissue in a patient with RCC.

Patient concerns: The patient was a 72-year-old woman with a past history of diabetes mellitus and hypertension. She did not have tumor-related symptoms.

Diagnosis and interventions: The tumor was diagnosed using abdominal CT during her yearly routine health check. After radical nephrectomy, the tumor was subjected to pathological examination and micro-CT imaging. Pathological analysis confirmed a clear cell renal carcinoma. The capillary distribution of the tumor was significantly lesser than that of the normal cortex on micro-CT image.

Lessons: Microvessels of RCC can be detected by micro-CT. We also found that the distribution of microvessels was uneven and lower than that in the normal cortex in this case. For a more general diagnosis, more micro-CT images of RCC tumors are needed.

Abbreviations: CT = computerized tomography, OV = object volume, RCC = renal cell carcinoma, TV = total VOI volume of whole sample.

Keywords: capillary distribution, micro-computerized tomography, renal cell carcinoma

1. Introduction

Clear cell type renal cell carcinoma (RCC) is the most frequently seen renal malignancy.[1,2] Some of the highly vascularized tumors respond to targeted antiangiogenic therapy.[3] However, some do not, owing to the complexity of the vasculature. The prognosis may be correlated to the differentiation of vessels: undifferentiated vasculature shows a poorer prognosis than highly differentiated vasculature.[3] Qian studied the microvessel density of tumors and found that the uneven pericyte coverage in clear cell RCC indicates a poor prognosis.[4] A positron emission tomography study showed that blood perfusion is reduced in RCC when compared with normal renal tissue.[5] However, a meta-analysis of the prognostic role of microvessel density in patients with RCC by Cheng et al found that microvessel density is not reliably associated with the survival time of the patients, but

Editor: N/A.

The authors alone are responsible for the content and writing of the paper.

Informed written consent was obtained from the patient for publication of this case report and accompanying images.

This study is supported in part by China Medical University (CMU106-S-32) and Taiwan Ministry of Science and Technology (MOST 107-2314-B-039-036 and MOST 107-2320-B-039-034).

The authors have no conflicts of interest to disclose.

Supplemental Digital Content is available for this article.

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How to cite this article: Lin TC, Chen HY, Yang YR, Liu PL, Chen YH, Chen WC. Decreased capillary density in renal cell carcinoma. Medicine 2019;98:36(e16910).

Received: 17 December 2018 / Received in final form: 24 June 2019 / Accepted: 29 July 2019

http://dx.doi.org/10.1097/MD.0000000000016910

survival time depends on vessel differentiation.[6] The factor responsible for the unreliability of microvessel density as a prognostic factor may be the uneven distribution of microvessels. Although microvessel density is not a reliable prognostic factor, imaging diagnosis of RCC is still based on detecting its highly vascularized structure by enhanced contrast computerized tomography (CT).

We performed micro-CT for the study of small animals. On micro-CT scanning of the fruit fly's Malphigian tubules, small crystals can be viewed and measured.[7] The aim of this study is to apply micro-CT to investigate the distribution of microvessels in patients with clear cell type RCC.

2. Case report

A kidney sample was obtained from a patient with RCC suspected on CT before surgery (Fig. 1A and B). The patient was a 72-year-old woman with a past history of diabetes mellitus and hypertension. The tumor was found during her yearly routine health check; she had not yet presented clinical symptoms. After radical nephrectomy, a 1 cm^3 sample was excised from the tumor side along the calyx. A same-sized sample was also excised from a grossly normal cortex. Each sample was stored in a formaldehyde solution for subsequent examination. The kidney was submitted for pathological analysis.

The 2 samples were incubated for 2 hours each in 70%, 80%, and 90% ethanol and then overnight in 100% ethanol. The samples were subsequently transferred to 100% hexamethyldisilazane for a further 2 hours, before air-drying. Two samples (control and tumor, http://links.lww.com/MD/D209) were prepared for further scanning by micro-CT. A Bruker Skyscan 1272 (Kontich, Belgium) was used to scan the samples at 1.8-μm resolution. CT scanning of the control and tumor samples was performed at 40 kVp voltage, 250 μA current, and 1000 ms exposure time, without a filter. The rotation step was 0.1 per image, 3 images were averaged. Reconstruction of sections was carried out using GPU-based scanner software (NRecon).

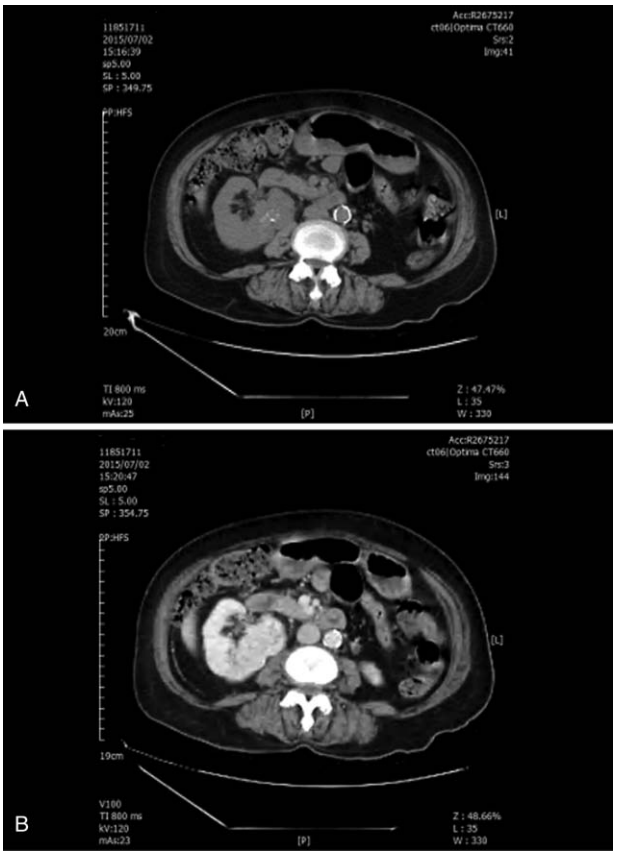

Figure 1. Images of CT diagnosis of renal cell carcinoma. (A) noncontrast CT and (B) enhanced CT. CT = computerized tomography.

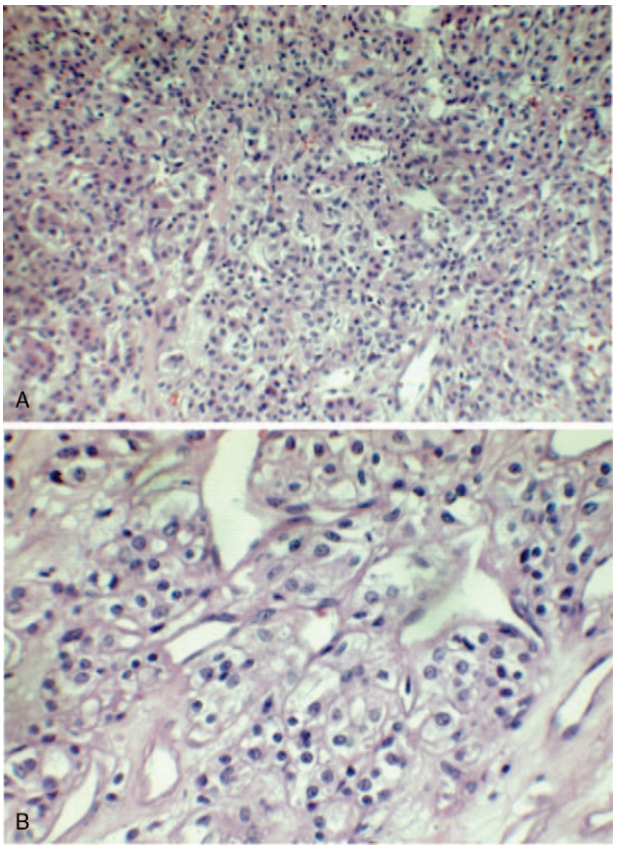

Figure 2. Histopathological features of diagnosis of clear cell type renal cell carcinoma. (A) 100× and (B) 400×.

Specific regions of interest were selected using DataViewer (Skyscan) for further analysis and illustration. CT-analyzer (Skyscan) was used to analyze the object volume (OV) and 3D morphometric indices. CTVox (Version 3.0, Skyscan) was used to provide 3D images. The grey threshold of the capillaries was set in a range of 90 to 255 on the tumor and normal cortex according to the morphology on micro-CT. We calculated the total volume of image volume of the whole sample (TV) and the OV of capillaries with diameters from 5 to 10 microns.[8] The percentage of vessels in each sample was defined as OV/TV. The distributions of capillaries in the tumor and normal cortex can, therefore, be compared.

Pathological analysis confirmed a clear cell type of RCC, Fuhrman's grade 2 (Fig. 2A and B). Micro-CT images of the normal and tumor cortices, labeled in red (120–255, scale bar = 250 μm), are shown in Figure 3.

We compared the distribution percentages of total capillaries in the tumor and normal cortices in a range of 120 to 255 (Table 1

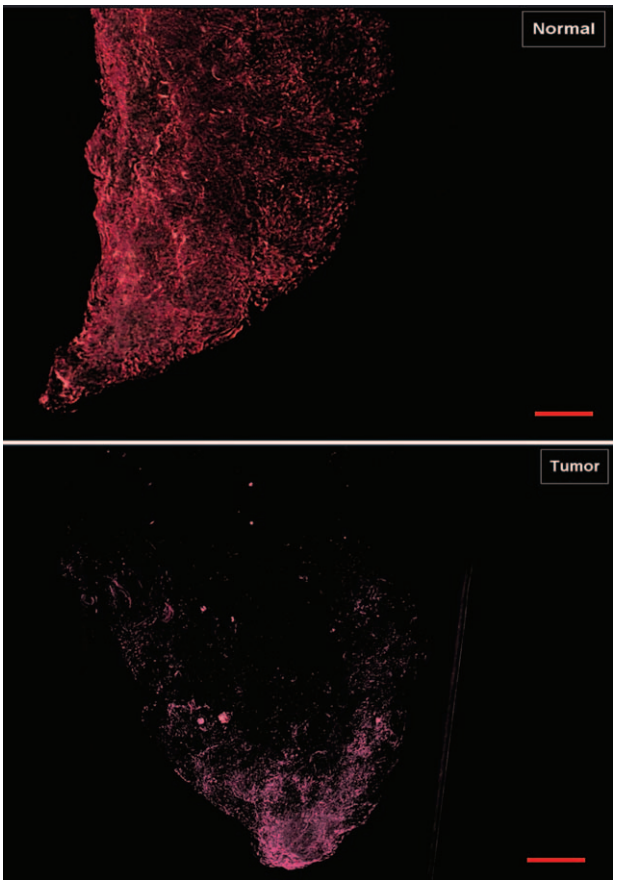

Figure 3. Two-dimensional micro-CT imaging of the normal and tumor cortex. Scale bar = 250 μm. CT = computerized tomography.

and Fig. 4). There was a large decrease in capillary distribution in the tumor cortex – (0.044%) compared with the normal cortex (2.729%). An uneven distribution of microvessles was also noted in tumor tissue. There were areas of high and low capillary density. The patient is under regular follow-up on an outpatient basis, and no local recurrence or metastasis has been found on CT examination.

Table 1

Analysis of capillary distribution of tumor and normal cortex on renal cortex from micro-CT.

Capillary	Grey threshold Range 120–255	Total VOI volume TV, mm^3	Object volume Obj.V, mm^3	Percent capillary volume Obj.V/TV, %
	Tumor	40.415	0.018	0.044
	Normal	7.8137	0.2133	2.7299

Obj.V = object volume, TV = total VOI volume of whole sample, VOI = volume of image.

3. Discussion

We found a marked decrease in capillary distribution in RCC on micro-CT image analysis. The capillary distribution in the tumor was 0.044% of the total volume, which was much lower than that in the normal renal cortex (2.73%). The lack of blood perfusion may be a tumor mass effect and therefore may be a cause of tumor necrosis. A subsequent event is high vascular endothelial growth factor expression in the tumor via the paracrine system.[9] The primary treatment for our patient was radical nephrectomy without targeted therapy. Therefore, RCC can be clinically diagnosed using CT imaging on the basis of a high vascular density. However, a lower density of capillary vessels was evident on micro-CT imaging. The capillary density was difficult to identify on abdominal CT.

The diagnosis of RCC relies on the vascularity seen in a tumor image obtained from abdominal CT, and this is a well-known diagnostic tool.[10] Like nephroureterectomy for the removal of renal urothelial cell carcinoma, which is highly prevalent in Taiwan, radical nephrectomy is indicated for RCC.[11] Therefore, a preoperative imaging study is very important for choosing the surgical treatment. The cell pathology is also important for the prediction of tumor prognosis and for planning further treatment.[12] The highly vascularized tumors of RCC can be treated by targeted therapy because of their highly expressed

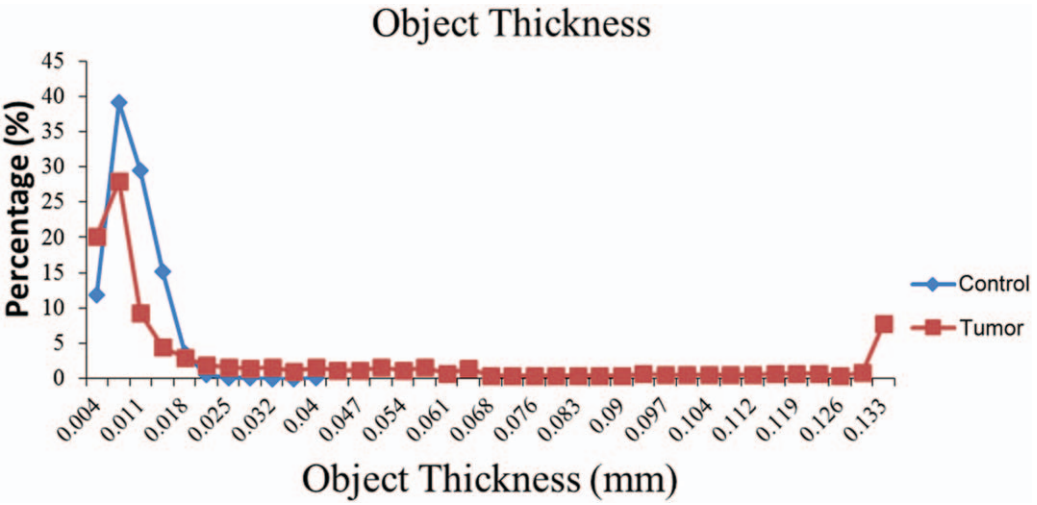

Figure 4. The distribution percentage of total capillary on normal and tumor cortex in a range of 120 to 255.

vascular endothelial growth factor receptor when the tumor has metastasized.[13] The relationship between low capillary distribution on the tumor and metastasis remains unknown. As our patient did not under targeted therapy, the clinical effect of targeted therapy on a lower capillary volume tumor is unclear. We suggest a further study on the response to targeted therapy on the basis of capillary distribution.

The uneven distribution of microvessels in the tumor indicates that predicting prognosis by density maybe not be reliable. In our patient, both high- and low-density microvessels coexisted in the tumor tissue. Therefore, predicting the prognosis using the density of microvessels might be difficult.

The resolution of micro-CT in this study was higher than that of micro-CT for progressive kidney disease. This study had a resolution of 1800 nm (1.8 μm), based on density. This resolution can be used to identify microvessels with a capillary diameter of 5 to 10 μm.[14]

There is evidence of less vascularity and uneven distribution of RCC tumor microvessles found from micro-CT. Limitations of this study are that it is a single case report with small area scanning and limited cell type. However, to our best knowledge, this is the first survey of capillary distribution of RCC using micro-CT.

In conclusion, we found that the distribution of microvessels was uneven and lower than that in the normal cortex by micro-CT. Because treatment for advanced RCC is based on vascularity in the majority of targeted therapies, the result of this imaging study may provide direction for the study of the poor response to targeted therapy in advanced RCC.

Author contributions

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Writing – original draft: Wen-Chi Chen.
Writing – review and editing: Yung-Hsiang Chen, Wen-Chi Chen.

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