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Low-grade Oncocytic Tumor of Kidney (CD117 Negative, Cytokeratin 7 Positive): A Distinct Entity?

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Abstract

Aim: To describe a group of distinct low-grade oncocytic renal tumors that demonstrate CD117 negative/Cytokeratin (CK) 7 positive immunoprofile.

Methods and results: We identified 28 such tumors from 4 large renal tumor archives. We performed immunohistochemistry for: CK7, CD117, PAX8, CD10, AMACR, e-cadherin, CK20, CA9, AE1/AE3, vimentin, BerEP4, MOC31, CK5/6, p63, HMB45, melan A, CD15 and FH. In 14 cases we performed array CGH; in 9 cases with successful result. Median patient age was 66 years (range 49-78 years) with a male-to-female ratio of 1:1.8. Median tumor size was 3 cm (range 1.1-13.5 cm). All were single tumors, solid and tan-brown, without a syndromic association. On microscopy, all cases showed solid and compact nested growth. There were frequent areas of edematous stroma with loosely arranged cells. The tumor cells had oncocytic cytoplasm with uniformly round to oval nuclei, but without

significant irregularities, and showed only focal perinuclear halos. Negative CD117 and positive CK7 reactivity were present in all cases (in 2 cases there was focal and very weak CD117 reactivity). Uniform reactivity was found for: PAX8, AE1/AE3, e-cadherin, BerEP4 and MOC31. Negative stains included: CA9, CK20, vimentin, CK5/6, p63, HMB45, Melan A and CD15. CD10 and AMACR were either negative or focally positive; FH was retained. On array CGH, there were frequent deletions at 19p13.3 (7/9), 1p36.33 (5/9) and 19q13.11 (4/9); disomic status was found in 2/9 cases. On follow-up (mean 31.8 months, range 1-118), all patients were alive with no disease progression.

Conclusion: Low-grade oncocytic tumors that are CD117 negative/CK7positive demonstrate consistent and readily recognizable morphology, immunoprofile, and indolent behavior.

Key words: Oncocytoma; Chromophobe renal cell carcinoma; Hybrid tumor; Hybrid oncocytic tumor; Low-grade oncocytic tumor; Unclassified oncocytic tumor; Unclassified renal carcinoma

Introduction

Diagnosing renal tumors composed of oncocytic cells can often be challenging, because oncocytic morphology is found in a spectrum of entities. It is also currently recognized that there are oncocytic tumors that do not fit into any of the existing "oncocytic" tumor categories.[1-4] There is a need to better define the morphologic and immunohistochemical profiles of such tumors, as well as their genetics and clinical behavior, in order to determine which tumors truly warrant the designation of carcinoma.[1] A frequent dilemma includes the distinction between the eosinophilic variant of chromophobe RCC (ChrRCC),[5] a malignant tumor, and renal oncocytoma,[6] a benign entity, that can be problematic solely on morphology.[7] Although eosinophilic ChrRCC is a relatively indolent

tumor, its distinction from oncocytoma is important, because of the impact of the cancer diagnosis, and the health care costs associated with patient management and surveillance.[1] It is also well recognized that there are tumors with "borderline or intermediate" features that are often reported descriptively, using usually "oncocytic" and/or "unclassified" as main diagnostic descriptors, often favoring one or the other.[1] Urologic pathologists frequently use additional modifiers such as "low-grade", "borderline features", or "uncertain/low malignant potential" to better characterize these types of tumors.[1] When facing this differential diagnosis, one has to also consider tumours labelled as "hybrid" or "hybrid oncocytoma-chromophobe tumours" (HOCT), that also show overlapping features of renal oncocytoma and ChrRCC. These tumors with "hybrid" morphology, when multiple and bilateral, typically occur in a hereditary Birt–Hogg–Dubé (BHD) syndrome and in renal oncocytosis.[8, 9] The "hybrid oncocytic tumors" can also very rarely present as sporadic, single tumours, in a non-syndromic setting.[10, 11] These sporadic "hybrid" tumors[10], which are not associated with BHD syndrome or oncocytosis, are currently considered variants of ChrRCC, based on the ISUP Vancouver 2012 and WHO 2016 classifications.[12, 13] However, a recent survey of urologic pathologists regarding the diagnostic criteria for oncocytic tumors showed quite variable use of the terms "hybrid" or "hybrid oncocytoma-chromophobe tumor" in practice.[1]

The workup of cases in the spectrum of oncocytoma and ChrRCC often includes immunohistochemistry evaluation for CD117 (KIT) and CK (cytokeratin) 7.[1] CD117 is typically positive in both eosinophilic ChrRCC and oncocytoma, as well as in some renal oncocytic tumors with "hybrid" features.[1] CD117 is generally helpful in differentiating these tumors from other renal neoplasms, which are typically CD117 negative. Diffuse reactivity for CK7 is generally considered to favor ChrRCC, whereas CK7 labelling in oncocytoma is very limited and restricted to scattered individual cells, typically representing ≤5% of cells.[1]

We have encountered in our practice a group of distinct oncocytic tumours with uniform, low-grade features, that are unexpectedly CD117 negative, or rarely show very focal and weakly positive CD117 (in less than 5% of the neoplastic cells), yet are diffusely CK7 positive (CD117-/CK7+). Although these tumors show overlapping morphologic features with oncocytoma and eosinophilic ChrRCC, they do not fit completely into either of these entities, based on the morphology and the immunoprofile. To our knowledge, this type of oncocytic tumor with low-grade features is not well characterized in the literature.[1, 14, 15]

Therefore, our aim was to better characterize a renal tumor that we designated "low-grade oncocytic tumor" (LOT), which is also characterized by CD117-/CK7+ immunoprofile. We evaluated the clinical and morphologic features of such tumors, their extended immunoprofile, and clinical behaviour. We also performed an array comparative genomic hybridization in a subset of cases, to examine for possible recurring genomic alterations, and to evaluate for possible karyotype similarities with oncocytoma, ChrRCC and other recognized renal tumors.

Material and Methods

Selection of cases and immunohistochemistry

An institutional ethics review was obtained for this study. Prompted by 2 index cases with oncocytic morphology that showed negative CD117 and diffusely positive CK7 staining, we initially searched 4 large institutional renal tumor archives and consult files of surgical pathologists with subspecialty interest in urologic pathology and identified 19 cases. In particular, we reviewed cases reported as "eosinophilic ChrRCC" (or "consistent with/ favor eosinophilic ChrRCC"), "oncocytic tumor, favor oncocytoma, (or with hybrid features)" and "low-grade oncocytic tumor (unclassified)", all of which had been reported with negative CD117 and diffusely positive CK7. Subsequent to the presentation on this entity at the

USCAP Annual Meeting in 2018[2, 16], we identified 9 additional tumors; some of these were consult cases in which the original pathologist either made or suspected this diagnosis. All cases included in the study were reviewed by 3 urologic pathologists (KT, SRW, OH). We collected the available clinicopathologic and follow-up data by review of the institutional records and by contacting the consulting pathologists.

In addition to the negative CD117 and positive CK7 immunohistochemistry, used as selection criteria for the study, we performed the following additional stains: PAX8, CD10, AMACR, e-cadherin, CK20, CA9, AE1/AE3, vimentin, BerEP4, MOC31 and Ki67. In a subset of cases (up to 50%), we also performed additional stains for CK5/6 (13), p63 (8), CD15 (14), HMB-45 (13), Melan A (13) and Fumarate Hydratase (3). 'Negative' IHC result was considered if less than 1% of cells stained; 'focal' positive was if >1% to 50% cells were reactive, and 'positive' result was if >50% of cells were reactive. Muller-Mowry colloidal iron stain was also performed.

DNA extraction

In 9 cases we performed array comparative genomic hybridization analysis (aCGH) evaluation. Tumor areas of the formalin-fixed paraffin-embedded (FFPE) samples were determined using hematoxylin-eosin stained slides and macro dissected. Deoxyribonucleic acid (DNA) from FFPE tumor tissue was extracted using QIAsymphony DNA Mini Kit (Qiagen, Hilden, Germany) on an automated extraction system (QIAsymphony SP, Qiagen) according to manufacturer's supplementary protocol for FFPE samples. Concentration and purity of the isolated DNA were measured using NanoDrop ND-1000 and DNA integrity was examined by amplification of control genes in a multiplex polymerase chain reaction (PCR). Only samples that were able to produce at least 400 bp long amplicons were used for aCGH.

Array comparative genomic hybridization (aCGH) analysis

SurePrint G3 Human CGH Microarray 8x60K was used for aCGH of sample reference pairs of opposite sex. Commercially produced MegaPool Reference DNA Female (Kreatech Diagnostics, Amsterdam, Netherlands) was used as a reference material. The labeled specimen were mixed, dried and hybridized overnight at 47 °C using Arrayit hybridization cassette (Arrayit Corporation, California, U.S.A.). Post-hybridization washing was done using SSC buffers with increasing stringency. The dried microarray was scanned with InnoScan 900 (Innopsys, France) at a resolution of 5 μm.

Image and Data analysis: Scanned images were analyzed and quantified by the Feature Extraction for CytoGenomics software (Agilent). Intensity values for each Cy5 and Cy3 labeled spot on the array were generated according to an appropriate grid template in a xml file. Minimum average absolute log ratio for deletion and amplification was set to more than 0.25. All genomic coordinates used for annotation are based on the March 2009 assembly of the reference genome GRCh37.

Results

Clinical and gross features

A total of 28 tumors were identified. The patients had a median age of 66 years (mean 65.5; range 49-78 years), and were more commonly females (M:F=1:1.8). None of the patients had a BHD syndrome, oncocytosis, Tuberous Sclerosis Complex or was known to have other hereditary condition. A single tumor was identified in each affected kidney and no tumor multifocality was found. There was a slight predilection for the left kidney (left 16; right 12). The majority of patients had either partial (11) or radical (14) nephrectomy; 3 tumors were diagnosed on biopsy. Median tumour size was 3 cm (mean 3.9; range 1.1-13.5 cm); 68% (17/25) measured less than 4 cm. On gross examination, tumor cut surface was

typically tan/yellow-brown and solid (Figure 1A-B). In 88% (22/25) of tumors the stage was either pT1a (68%; 17/25) or pT1b (20%; 5/25). Only 3 tumors had a higher stage than pT1 (1 each: pT2a, pT2b and pT3a). Follow-up was available in 27 of 28 patients (mean: 31.8; median 21; range 1-118 months) and all patients were alive with no evidence of disease progression. The clinical features and the follow-up data for all patients are shown in Supplemental Table 1.

Microscopic findings

On microscopy, all tumours lacked a peripheral capsule and showed solid, compact nested, or focal tubular, tubuloreticular, and trabecular growth (Figure 2A-F). Rare entrapped renal tubules can be seen at the periphery. The cells had homogeneous oncocytic or eosinophilic cytoplasm with uniformly round to oval nuclei, without significant irregularities or crumpling (i.e. "raisinoid shapes"). The nucleoli were either delicate or slightly more conspicuous (nucleolar grade equivalent either WHO/ISUP 2 or 3). The nuclei focally exhibited delicate perinuclear halos or clearings. A fairly characteristic finding was that of edematous stromal areas that were sharply delineated from the more solid areas. These areas were hypocellular, with loosely arranged tumour cells, exhibiting cords, loose reticular growth and individual cell arrangement; some of the cells showed an elongated (myoid cell-like) morphology (Figure 3A-C). These hypocellular areas often resembled tissue culture and focal fresh hemorrhage was also frequently noted in these areas. Lymphocytic clusters were also often seen in the solid areas, forming either compact, round/oval aggregates or, rarely, delicate and more irregular, intercellular clusters (Figure 3D-E). Muller-Mowry colloidal iron stain was either negative or only apical/luminal (or bar/blob-like) positive (Figure 3F). No worrisome or adverse pathologic features were seen in any of the cases, including coagulative necrosis, nuclear pleomorphism and significant cell atypia, multinucleation, and any mitotic

activity. In 3 cases the diagnosis was established in core biopsy material, which was based on the compatible morphology (without the hypocellular areas) and the CD117-/CK7+ profile. (Figure 4A-D).

Immunohistochemistry findings

Negative CD117 and positive CK7 reactivity were present in all cases, by study design (Figure 5A-B). Of note, in 2 cases (#23 and #24) there was focal and very weak CD117 reactivity (in less than 5% of the neoplastic cells); however, the morphology in these cases was virtually identical to the remaining cases, and the CK7 was also diffusely positive. Uniform tumor cell reactivity was found for: CK7, AE1/AE3, PAX8, e-cadherin, BerEP4 and MOC31. Negative stains included: CD117, CA9, CK20, and vimentin. CD10 and AMACR were predominantly negative or only focally positive. In all cases, Ki67 showed reactivity in less than 5% of the cells. Immunohistochemistry results for all cases are shown in Supplemental Table 2. Fumarate hydratase showed retained expression (3/3). Uniform negativity was found for CK5/6 (13/13), p63 (8/8), CD15 (14/14), HMB45 (13/13) and Melan A (13/13) (data not included in Supplemental Table 2). Of note, the lymphocytic aggregates demonstrated a variable and mixed composition of T and B lymphocytes.

Array comparative genomic hybridization (aCGH) findings

aCGH was successfully carried out in 9 out of 14 cases (result shown Table 1; cases # 2, 5, 25, 27, 28 were not successful). There were frequent deletions at 19p13.3 (7/9), 1p36.33 (5/9) and 19q13.11 (4/9). In 2/9 cases, a disomic chromosomal status was found. No other consistent chromosomal gains or losses were found in the evaluated cases.

Discussion

In this study we describe a group of renal tumors that we refer to as LOT, which demonstrate uniform morphologic and immunohistochemical features, including a CD117-/CK7+ profile, lack multiple chromosomal losses and gains, and show indolent clinical behavior. We have summarized the findings on these tumors in Table 2.

In our opinion, renal LOT shows remarkable morphologic similarities to the 4 cases included in the study by Davis et al, which were labelled "eosinophilic ChrRCC" that showed absence of any copy number alterations; however, an immunohistochemical profile was not provided for these cases.[14] In their study, one of the most comprehensive ever undertaken on ChrRCC, the remaining 62 of 66 ChrRCC cases (which also included 19 eosinophilic ChrRCC), demonstrated a classic karyotype profile of ChrRCC, with frequent multiple losses of chromosomes 1, 2, 6, 10, 13, 17 (seen in 86% of cases), and less common losses of chromosomes 3, 5, 8, 9, 11, 18, and 21 (seen in 12-58% of cases). In some studies, however, a combination of chromosomal losses and gains have been found in usual ChrRCC.[17-19] To our knowledge, the only other study that included similar tumours to the LOT described herein, was done by Kuroda et al.[15] They described 5 renal tumors, which were designated as "ChrRCC, oncocytic variant", that "entirely resembled renal oncocytomas on morphology", but were CD117 negative and diffusely CK7 positive.[15] On FISH, these tumors however exhibited multiple (at least 4) chromosomal losses of evaluated chromosomes 7, 10, 13, 17, and 21.[15]

Renal LOT indeed shows some similarities with renal oncocytoma, including: lack of well-formed capsule, diffuse growth with solid sheets and compact nests (particularly at the tumoral periphery), cells with oncocytic cytoplasm and relatively round to oval nuclei, without significant irregularities. However, in contrast to oncocytoma, LOT shows areas of rarefied connective tissue stroma, with loosely arranged tumour cells. These areas are

different from the hypocellular areas seen in oncocytoma, which contain compact cell islands and nests, typically found in the central areas (often described as "archipelaginous" growth). The cells in oncocytoma, in contrast to LOT, lack perinuclear halos and are usually diffusely reactive for CD117, whereas CK7 stains only scattered oncocytoma cells (≤5% of cells). LOT also shows similarities with the so-called eosinophilic variant of ChrRCC, which include: lack of well-formed capsule, diffuse growth with solid sheets and compact nests at the tumoral periphery, cells with focal perinuclear halos, and diffuse CK7 reactivity. However, in contrast to LOT, eosinophilic ChrRCC typically lacks the sharply delineated hypocellular areas. The cells typically show more prominent membranes and the nuclei are often more irregular. CD117 is also diffusely positive in the great majority of ChrRCC, including the eosinophilic ChrRCC.

The broader differential of the renal tumors with oncocytic or eosinophilic features also include several additional entities, such as clear cell RCC with eosinophilic features, papillary oncocytic RCC, epithelioid angiomyolipoma, eosinophilic solid and cystic RCC, and SDH-deficient RCC, but these are usually easily distinguished from the core group of renal "oncocytic" tumors, that include oncocytoma, eosinophilic ChrRCC and the broader group of "borderline/hybrid" oncocytic tumors. A summary of the helpful features in distinguishing LOT from other renal tumors with oncocytic/eosinophilic cytoplasm is included in Table 3.

In contrast to ChrRCC, renal oncocytoma frequently exhibits a diploid karyotype, or losses of chromosome 1 (or part of it, often the short arm), chromosome 14, or chromosome Y, rearrangement of 11q13, and translocation t(5;11)(q35;q13).[6] The so-called HOCT in the sporadic setting and in renal oncocytosis have been reported with chromosomal gains and losses.[9, 10] Based on our aCGH analysis, the renal LOT showed some karyotypic similarities with renal oncocytoma, including the loss of 1p36 and the diploid pattern, which were

observed in some of the evaluated cases. Importantly, we have not found multiple chromosomal losses (or gains), which are considered typical for ChrRCC. However, the number of analyzed cases was limited and additional studies are needed to further clarify the karyotype of these tumors.

The important practical questions in this setting pertain to the diagnostic "gold standard" attributes that separate oncocytoma, ChrRCC and other, possibly related oncocytic renal entities, including the tumor that we tentatively designated as "LOT" in this study. Should a diagnostic distinction be done strictly on morphology; what is the role of immunohistochemistry; should cytogenetic or molecular karyotyping be mandatory in non-straightforward and ambiguous cases, and is there still a role for ancillary studies in the contemporary practice, such as Muller Mowry colloidal iron and electron microscopy? A comprehensive diagnostic approach would also be difficult to implement routinely in many centers. A combination of different diagnostic modalities are certainly required to establish a more precise diagnosis in cases that are not easy to classify based on morphology and immunohistochemistry, and such cases should be referred for further expert evaluation and additional studies.

We have also recently described another group of distinct tumors, which we referred to as "high-grade oncocytic tumors" (HOT) that demonstrate different morphology and immunoprofile from LOT described in the current study.[3] HOT is also a morphologically distinct renal tumor, which is also not currently included in the WHO classification of renal tumors. Renal HOT is also readily recognizable on morphology and, based on the available follow-up, behaves indolently. On low power, they are well-circumscribed, but lack a well-formed capsule, and show nested to solid growth and focal tubulocystic architecture. The neoplastic cells have abundant voluminous oncocytic (or eosinophilic) cytoplasm, with frequent and prominent intracytoplasmic vacuoles, lack irregular nuclei and perinuclear

halos, and demonstrate prominent and enlarged nucleoli.[3] Nine of the 14 cases in our study were diffusely CD117 positive, while CK7 was either negative or only focally positive in some cases, in contrast to the uniform CD117-/CK7+ profile of LOT. Interestingly, Chen et al have recently reported a series of 7 morphologically identical tumors, labelled as sporadic "renal cell carcinomas with eosinophilic and vacuolated cytoplasm", which showed somatic mutations of *TSC2* or *MTOR*.[20] All cases included in both series behaved indolently during the available follow-up.

It is likely that examples of HOT and LOT were previously considered as "hybrids", "borderline" or "unclassified oncocytic" renal tumors.[1] In regard to the proposed terminology for both entities, it should be considered a working terminology, relying on easily recognizable acronyms. Should additional data become available in the future, appropriate terminology adjustments should be made. We do not currently recommend routine grading of these tumors using the WHO/ISUP grading. They demonstrated uniformly indolent behaviour, although the clinical follow-up was relatively limited. It is imperative that more studies are conducted, with larger number of cases and with longer follow-up, to fully characterize these tumors.

In summary, LOT is an oncocytic renal neoplasm which shows consistent morphology and immunoprofile (CD117-/CK7+), absence of multiple chromosomal losses and gains, and indolent clinical behavior. In our view, the diagnosis of LOT can be readily established on H&E morphology. This tumor does not fit completely into either oncocytoma or eosinophilic ChrRCC, despite showing some similarities with both entities. The incidence of LOT is difficult to estimate, because probably these cases were labelled variably, either as "eosinophilic ChrRCC", "oncocytic renal tumor, NOS", "unclassified/low-grade oncocytic tumor", "hybrid or hybrid oncocytoma-chromophobe tumor" or "borderline/uncertain/low malignant potential" tumors. We believe that LOT potentially represents a distinct type of

tumour that warrants further study and should be considered an emerging/provisional entity in the renal tumor classification. We hope that our study will promote awareness of the clinical, morphologic, and immunophenotypic features of this type of renal neoplasm, resulting in its increased recognition, as well as re-evaluation of the oncocytic renal tumors that were previously labelled using a spectrum of diagnostic terminologies.

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Tables 1-3

Table 1. Array comparative genomic hybridization results for low-grade oncocytic tumors.

Patient #	Array comparative genomic hybridization (aCGH)
3	-1p36.33 - p36.22, -19p13.3 - p12, -19q13.11 - q13.42
4	-1p36.33 - p36.22, -7p22.3 - p22.1, -17q25.1 - q25.3, -19p13.3 - p12, -19q13.11 - q13.42, -22q12.3 - q13.33
7	-1p36.33 - p36.22, -7p22.3 - p22.1, -16p13.3, -19p13.3 - p12, -22q12.3 - q13.33
8	-1p36.33 - p35.1, -4p16.3 - p16.1, -5p15.33 - p15.31, -7p22.3, - p22.1, -7q21.3- q22.1, -7q11.21- q21.11, -8q24.3, -9q33.3 - q34.3, -14q32.2 - q32.33, -16q24.1 - q24.3, -17p13.3 - p13.2, -19p13.3 - p12, -19q13.11- q13.43, -21q22.3, -22q11.21 - q11.22, -22q12.3 - q13.33
16	-4p16.3 - p16.1, +7p22.1 - p11.2, +7q21.11 - q36.2, -16q24.1 - q24.3, -19p13.3 - p12, -21q22.3, -22q12.3 - q13.33, -9q33.3 - q34.3, -14q32.2 - q32.33, -17q25.1 - q25.3, -19q13.11 - q13.42, -22q11.21 - q12.1,
17	-19p13.3 - p12,
18	-1p36.33 - p36.22, -19p13.3 - p13.11,
20	Disomic status
26	Disomic status

Table 2: Summary of the key features of low-grade oncocytic tumors.

Clinical	Older patients, non-syndromic, M:F=1:1.8, relatively small size, good prognosis
Gross	Tan-brown and solid, single tumors
Light microscopy	Architecture: Non-encapsulated, solid, compact nested or focal tubular and tubuloreticular growth. Frequent edematous stromal areas with irregular and loose reticular, cord-like and individual cell growth. Focal lymphocytic aggregates can be seen. Cytology: Homogeneous oncocytic cytoplasm, round to oval nuclei, without significant irregularities. Delicate perinuclear halos focally present.
Immunohistochemistry	Positive: CK7, PAX-8, E-cadherin, AE1/AE3, BerEP4, MOC 31 Negative: CD117 (rare cases focal weak +), CA9, CK20, Vimentin, CD10 (-/focal+), AMACAR (-/focal +), CK5/6, p63, HMB45, Melan A, CD15
Special stains	Muller-Mowry colloidal iron: negative or apical, bar or blob-like positive
aCGH	Frequent deletions at 19p13.3 (7/9), 1p36.33 (5/9) and 19q13.11 (4/9); some disomic (2/9). No other consistent chromosomal gains or losses

Table 3: Features helpful in distinguishing low-grade oncocytic tumors from other renal tumors with eosinophilic cytoplasm

Diagnosis	Key distinguishing features	Immunohistochemistry
Low-grade oncocytic tumor	Solid sheets and compact nests, with gradual transition to trabecular areas; sharply delineated edematous stromal areas with loose cell growth	CD117-, CK7+
Chromophobe RCC, eosinophilic	Solid growth, more prominent cell membranes, irregular (raisinoid) nuclei, perinuclear halos, loose stromal areas lacking	CD117+, CK7+
Oncocytoma	Can show more tubulocystic growth, lacks perinuclear halos, central stromal 'archipelaginous' areas are present, however lacks areas of loose and irregular cell growth	CD117+, CK7 -/+
Clear cell RCC, eosinophilic	At least focal clear cell areas, delicate vasculature in the background	CA9+, CD117-
Papillary RCC, oncocytic	Papillary growth	AMACR+, CD10+, Vimentin +
Epithelioid angiomyolipoma	Epithelioid cells, may be pleomorphic, lacks perinuclear halos	PAX8-, HMB45+, AE1/AE3-, CK7-
Eosinophilic, solid and cystic RCC	Great majority females, solid and cystic growth, cytoplasmic stippling, lacks perinuclear halos	CK20+, CK7-, CD117-
SDH-deficient RCC	Flocculent cytoplasm and vacuoles; lacks perinuclear halos	CD117-, SDH-, AE1/AE3- (often)

Figure Legends

Figure 1. Gross appearance of low-grade oncocytic tumor. Tumor cut surface was typically tan/yellow-brown and solid. Area of loose myxoid stroma could be appreciated even grossly in some examples (left).

Figure 2. Microscopic features. **A-B,** On low power, low-grade oncocytic tumor lacked a peripheral capsule and showed solid and focal tubuloreticular or tubular growth (B). **C-D**, On higher power, the cells demonstrated solid to vague nested and compact acinar growth. The cells had homogeneous oncocytic cytoplasm, uniformly round to oval nuclei, without significant irregularities; the nucleolar grade was either WHO/ISUP 2 or 3 (equivalent). **E,** The nuclei focally exhibited delicate perinuclear halos or clearings. **F,** In some areas there was more prominent trabecular to reticular growth, shown here on higher power (also shown in B).

Figure 3. Microscopic features of low-grade oncocytic tumor. **A,** Well-delineated hypocelluar areas were frequently found; focally, they demonstrated fresh hemorrhage. **B-C,** The cells in these areas were loosely arranged and had irregular, cell culture-like growth, including cords, individual cells and elongated cells. **D-E,** Lymphocytic clusters were frequently seen in the solid tumor areas. They formed compact aggregates (D) or, rarely, delicate and more irregular, intercellular clusters (E). **F,** Muller-Mowry colloidal iron stain was either negative or only apical/luminal or bar/blob-like positive.

Figure 4. Low-grade oncocytic tumor on core biopsy (case #6). **A-B,** The diagnosis in this case was established based on the compatible morphology, with solid and tubuloreticular growth (A) of cells with oncocytic cytoplasm and round to oval nuclei (B) **C-D**, CD117 was negative (C), while CK7 was diffusely positive (D).

Figure 5. Typical immunohistochemistry profile of low-grade oncocytic tumor. **A-B,** These tumors are CD117 negative with only scattered positive mast cells (A), while CK7 is diffusely positive (B).