New and old Hereditary forms of Renal Cell Carcinoma.

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Renal cell carcinoma affects approximately 30,000 individuals per year in the United States and is responsible for close to 12,000 annual deaths. Kidney cancer, is a tumor comprising several histologic subtypes, the molecular hallmarks of which support their classification as distinct entities. While the vast majority of renal tumors occur sporadically, about 4% are found in association with syndromes of a heritable nature. Over the past 15 years considerable insight into the genetic basis of renal cancer has come from studies of renal cancer families. Identification of the genes involved in the hereditary forms of renal cell carcinoma has provided significant information about the genes involved in the common, sporadic (non-hereditary) forms of the disease.

Recognition of this group of tumors is, however, rather important both, clinically and scientifically, because it is now recognized that kidney tumors may occur in familial settings more frequently than previously contemplated. Early detection and diagnosis of these tumors, identification of specific genes that characterize each syndrome and elucidation of mechanisms responsible for the development of tumors, are important in order to establish appropriate therapies as well as for genetic counseling. At the moment, only morphology can assist in identifying and diagnosing some of the newly recognized syndromes.

Hereditary renal syndromes include:

- Von Hippel-Lindau disease,
- Hereditary papillary renal cell carcinoma,
- Birt-Hubb-Dube
- HLRCC
- SDH syndrome
- Tuberous Sclerosis
- Familial Oncocytoma
- Syndromes associated with Wilms’ tumor.

Each syndrome is associated with a specific renal pathology. Clinically, the tumors are more likely to be multifocal and bilateral and with early incipient lesions scattered throughout the adjacent renal parenchyma. The most common form of hereditary kidney cancer is VHL, which is associated with clear cell or conventional renal carcinoma (~75%). This disease has been associated with mutations in the von Hippel-Lindau gene (at 3p25) and loss of heterozygosity at chromosome 3p. Next in frequency is papillary renal cell carcinoma (~10%), associated with trisomies of chromosomes 7 and 17 and losses of chromosome Y. Chromophobe renal cell carcinoma is even more rare (~5%), is associated with numerous specific chromosomal losses (-1, -2, -6, -10, -13, -17, -21) and is seen in patients with BHD.
syndrome. Renal oncocytoma, a benign neoplasm most often without any chromosomal abnormalities, is represented at a frequency of ~3-5%.

**Von Hippel Lindau: The VHL Kidney Cancer Tumor Suppressor Gene**

VHL is an autosomal dominant cancer syndrome in which affected individuals are at risk to develop tumors in a number of organs. The most frequent clinical manifestations are:

- Bilateral, multifocal clear cell renal carcinoma
- CNS Hemangioblastomas (Cerebellum, Spine)
- Pancreatic neuro endocrine tumors
- Pheochromocytomas
- Retinal angiomas

**Pathology**

Tumors identified in VHL patients and kindred’s are frequently bilateral and multifocal. Morphologically, they are similar to sporadic RCC. Grossly the tumors have a yellow color with extensive areas of hemorrhage and necrosis. The tumor cells are large with clear cytoplasm although occasional eosinophilic cells may be identified. A variety of growth patterns, solid, tubular and papillary can be found.

Cysts are frequently present in VHL, with a multilocular growth pattern. They vary in sized and are separated by fibrous / hyalinized septa. The cysts are usually lined by a single row of low nuclear grade tumor cells. The *VHL* gene is known to be located in 3p25-p26, but changes have been reported in 5q21+ (70%) and 14q- (41%).
No deleted allele of the VHL gene shows somatic mutations or hypermethylated induced inactivation in 80% of cases; VHL is a tumor suppressor gene that encodes elongin, which inhibits the generation of a transcriptional elongation complex of vascular endothelial growth factor; loss of VHL causes an increase in VEGF, may cause the increased vascularity of these tumors. Patients with VHL associated tumors have a better prognosis and longer survivals that for sporadic RCC. Sporadic cases 38% die of disease overall and metastases develop in 27-37% of patients.

**Papillary RCC Type I**

Hereditary papillary renal cell carcinoma is an autosomal dominant disease characterized by the development of multifocal renal cell tumors. Papillary (chromophil) RCC is recognized by the formation of papillary structures lined by cuboidal small cells with scanty basophilic cytoplasm and small indolent nuclei. They can also exhibit focal tubular or solid patterns of growth. The papillae may be filled with foamy macrophages and psammoma bodies are common (type I). Marked nuclear atypia is rare. Papillary RCC as clear cell tumors derives from the proximal tubule. The tumors are multifocal in more that 80% of the cases, and incipient lesions can be found scattered throughout the adjacent renal parenchyma. Tumors stain positive for CK7, negative for CK20.

Hereditary papillary RCC (HPRC) is a rare condition characterized by type 1 papillary RCC and mutations in the tyrosine-kinase domain of the *c-MET* proto-oncogen on 7q34. Loss of chromosome 7 is the initiating event along with the loss of a sex chromosome.
It is estimated that approximately half the members of affected families will develop tumors by the age of 50 years. The 5-year survival for patients with this type of tumors is about 82-90%, much better than clear cell independent of stage. Metastases can occur to regional lymph nodes and lung.

**Birt Hogg Dubé Syndrome (BHD)**

Several years ago we described families with members affected with multiple renal oncycotomas. We termed this clinical entity “familial renal oncocytoma”. Subsequently, we found that some families with familial renal oncocytoma were also affected with the Birt Hogg Dubé Syndrome (BHD). This syndrome, first described in 1977 by Drs. Birt, Hogg, and Dubé, was initially characterized by a predisposition to develop multiple small papules on the head and neck. These papules are benign tumors of the hair follicles (fibrofolliculomas). We expanded the phenotype of Birt Hogg Dubé syndrome to include and confirm the association of lung cysts and renal carcinoma with the fibrofolliculomas described by Drs. Birt, Hogg and Dubé. Fifteen to thirty five percent of affected BHD individuals develop renal carcinoma. The histologic type of BHD associated renal carcinoma is markedly different from that of either VHL (clear cell RCC) or HPRC (Type I papillary RCC).

BHD patients are at risk to develop chromophobe RCC, oncocyic RCC and oncocyotoma. In our analysis of 134 renal tumors in 30 BHD patients from 19 families, we have noted tumors of conventional, chromophobe, papillary, and oncocyotoma histologic types, as well as tumors of a hybrid histology not previously linked to a familial syndrome the hybrid and chromophobe RCC predominated, making this the first report of a heritable syndrome conferring a predisposition to tumors with chromophobic elements or of chromophobe histology.
The size of tumors at presentation in the 30-patient subset increased with increasing malignant potential of the histologic type: The hybrid tumors averaged 2.2 cm, the chromophobe RCCs 3.0 cm, and the clear cell RCCs 4.7 cm in diameter.

Morphologically, these hybrid oncocytic tumors had zones classic for oncocytoma (large cells, poorly defined cell borders, finely granular eosinophilic cytoplasm throughout, and large nuclei with homogenous basophilic chromatin) and other zones consistent with chromophobe RCC (well-defined cell borders, “fluffy” eosinophilic cytoplasm, pyknotic nuclei, and perinuclear halos). Cells in the chromophobe zones of these tumors were arranged in the characteristic plant-like distribution of chromophobe RCC. On the other hand, the oncocytoma-like zones lacked the characteristic loose connective tissue background, central scar, and nephroid growth pattern usually seen in oncocytomas. Incipient lesions or oncocitosis, was widespread throughout the renal parenchyma.

The BHD gene has been localized to a 4cm region on the short arm of chromosome 17. The prognosis of patients with these tumors is similar to that of Papillary type I, unless clear cell cancers develop.

**Hereditary Leiomyomatosis Renal Cell Carcinoma (HLRCC)**

In 2001, an autosomal dominant hereditary syndrome involving cutaneous and uterine leiomyomas was reported to be associated with renal cancer in two families. A familial autosomal dominant predisposition to multiple cutaneous leiomyomas had been noted in the 1950’s. In 1965 Mezzadra had suggested the association of uterine leiomyomas with the cutaneous leiomyomas. Subsequent reports have confirmed a dominant familial predisposition to cutaneous and uterine leiomyomas, Multiple Cutaneous Leiomyoma (MCL).

Alam, et. al. performed a genomewide screen of 11 families with this disorder and found linkage to 1q42.3-q43. They reduced the critical region to 14cM, naming the locus as MCUL1 (multiple cutaneous and uterine leiomyomata).

In 2001 Lanonen et. Al reported on the clinical, histopathologic and, molecular features of a cancer syndrome with predisposition to uterine leiomyomas and papillary renal cancer. In the Finnish family studied, 11 members had uterine leiomyomas and 2 had uterine leiomyosarcoma. Seven individuals had a history of cutaneous nodules, two of which were confirmed to be a cutaneous leiomyomatosis. The four kidney cancer cases occurred in young (33–48) year old females and displayed a unique natural history. All these renal tumors were of papillary histology and presented as unilateral solitary lesions that had metastasized at the time of diagnosis. A second smaller family was studied also. Linkage analysis mapped the predisposition gene to chromosome 1q. Losses of the normal chromosome 1q
were observed in tumors that had occurred in the kindred displaying the phenotype. The Finnish group recently reported the finding of mutation of fumarate hydratase (fumarase) in the germline of some of the HLRCC and MCL kindred’s. It is now believed that MCL and HLRCC are the same disorder.

Hereditary Paraganglioma-Pheochromocytoma syndrome (SDH).

Hereditary paraganglioma-pheochromocytoma (PGL/PCC) syndrome is a disease characterized by the presence of paragangliomas and pheochromocytomas. Extra-adrenal parasympathetic paragangliomas are located predominantly in the head and neck. Pheochromocytomas, which arise from the adrenal medulla, typically hypersecrete catecholamines. Paraganglioma syndrome has been classified genetically into 4 entities, PGL1, PGL2, PGL3, and PGL4. To date, 3 of these 4 entities have been associated with germline mutations in the genes encoding 3 of the subunits of succinate dehydrogenase (SDH), which has a key function in the Krebs cycle and the respiratory chain. Germline mutations in SDHB, SDHC, and SDHD have been found in PGL4, PGL3, and PGL1, respectively. We have been able to study and the morphologic features of 11 cases of Renal cell carcinoma in patients with SDH. Some cases were known to be part
of family syndromes but in others, the morphology suggested SDHB disease that was confirmed later after genetic studies. Most of the tumors had a similar appearance to oncocytoma.

**Tuberous Sclerosis**

Tuberous sclerosis (TS) is an autosomal dominant disease characterized by cortical tubers, giant cell astrocytomas, retinal hamartomas, cardiac and renal tumors. The disease is associated with two predisposing genes, \textit{TSC1} at 9q34 and \textit{TSC2} at 16p13. Renal tumors are most frequently angiomyolipomas but clear cell RCC and chromophobe renal tumors have also been described.
References


Intraductal Carcinoma of the Prostate: the Whole Story

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Introduction:

Prostate lesions with cribriform or solid architectures range from benign and proliferative, such as central zone glands, clear cell cribriform hyperplasia, and basal cell hyperplasia, to invasive cribriform carcinoma. Infrequently, cribriform or solid lesions comprise cytologically malignant cells spanning or expanding glandular lumens and yet preserve at least focally a basal cell lining. These atypical cribriform/solid lesions with basal cells represent either cribriform high-grade prostatic intraepithelial neoplasia (HGPIN) or intraductal carcinoma of the prostate (IDC-P). The distinction of the two is of critical importance, especially in prostate needle biopsy, as IDC-P is almost always associated with high grade and high volume prostate cancer (PCa). A diagnosis of IDC-P in biopsy mandates immediate repeat biopsy or even definitive therapy in the absence of documented invasive PCa. In contrast, cribriform HGPIN is a putative precursor lesion to PCa. Recent data have questioned whether HGPIN on needle biopsy is associated with a significantly increased cancer risk in subsequent biopsies and whether repeat biopsy is necessary within the first year after its diagnosis, especially when HGPIN is focal \(^1,^2\).

This lecture reviews the historical perspective, histological features, diagnostic criteria of IDC-P. The incidence and clinical significance of IDC-P in prostate biopsy specimens as well as the reporting of IDC-P in prostate biopsies is also discussed.

Historical Perspective:

In earlier literature \(^3\), the term “intraductal carcinoma” was used variably to describe the extension of different carcinomas, including prostatic acinar carcinoma, prostatic ductal carcinoma and urothelial carcinoma, into prostatic ducts and acini. Currently
IDC-P refers specifically to a lumen-spanning proliferation of malignant cells within prostatic ducts and acini, resulting from spread of prostate cancer cells within pre-existing prostatic glandular structures.

The first detailed analysis of such phenomenon was credited to Kovi et al., who studied 139 cases of PCa diagnosed on transurethral resection, suprapubic prostatectomy and needle biopsy specimens and found spread of PCa cells into the pre-existing prostate ducts and acini in 48% of PCa cases. Such “intraductal spread” was positively associated with both Gleason grade and tumor extent, although only tumor extent, not Gleason grade, remained significantly associated with the intraductal spread in the final multivariate analysis.

While studying cribriform PCa, McNeal et al. found that in most cases cribriform PCa was predominantly located within prostatic ducts and acini with cancer cells following normal duct contour or showing a basal cell layer on morphological examination or basal cell immunostains. They found that the “cribriform PCa with intraductal location” was equivalent to Gleason patterns 4 and 5 PCa prognostically and was associated with high grade and high volume PCa in the majority of cases. The term “intraductal carcinoma of the prostate (IDC-P)” was introduced to emphasize the unique histological and clinical features of this lesion.

Since PIN glands also comprise cytologically atypical or malignant secretory cells within prostatic ducts and acini, the relationship between PIN and IDC-P was debated. In a subsequent study, McNeal and Yemoto presented evidence that IDC-P was different from PIN and suggested rather that it represented a distinct form of PCa with peculiar propensity for intraductal spread and growth. They noted that the presence of cancer cells within prostatic ducts and acini, or IDC-P, was almost never seen in the absence of invasive carcinoma and the concomitant invasive component was almost always high grade. Furthermore, PCa with IDC-P component had a
significantly worse prognosis than those without. The authors concluded that IDC-P was an entity with precisely defined histological criteria and unique biological and clinical significance. Morphologically, IDC-P and PIN could be reliably distinguished from each other.

Several subsequent studies also demonstrated the association of IDC-P with other adverse pathological features such as higher Gleason score, larger tumor volume, greater probability of extraprostatic extension and poorer clinical outcomes\(^9\)\(^{-12}\). The current concept is that IDC-P represents intraductal extension of large volume Gleason patterns 4 or 5 PCa. Rarely IDC-P can be an isolated finding without a concomitant PCa and may represent a form of PCa with propensity for intraductal growth\(^13\),\(^14\).

**Histological features and diagnosis of IDC-P**

The hallmark of IDC-P is the expansile proliferation of PCa cells within the native prostatic glands with an at least partially preserved basal cell layer. McNeal and Yemoto first defined IDC-P as the “complete spanning of the ductal or acinar lumen by several trabeculae of malignant epithelial cells, with foci of trabecular fusion” in radical prostatectomy specimens. More proliferative, denser lesions with cribriform or solid architecture were also included in their definition\(^8\). Later studies refined the histological features of IDC-P (Table 1)\(^10\),\(^11\),\(^14\)\(^{-17}\). Classical examples of IDC-P usually comprise many glands, often > 6 per radical prostatectomy specimen\(^17\), and the glands are larger than normal peripheral zone glands\(^13\),\(^17\) with irregular and branching contours\(^8\),\(^13\),\(^17\). The neoplastic cells in IDC-P grow in several architectural patterns, including trabecular, loose cribriform, dense cribriform and solid, that represent progressive dedifferentiation with a reciprocal increase in proliferation and correlated with cancer stage, grade, and clinical course\(^10\).
Neoplastic cells in classical IDC-P are pleomorphic, some 6X larger than adjacent non-neoplastic nuclei. Comedonecrosis is diagnostic of IDC-P, but is only present in a subset of cases.

Cohen et al. proposed a set of criteria for diagnosing IDC-P\textsuperscript{13}, which included five major and three minor criteria. The first 4 major criteria are always present in IDC-P and include 1) large-caliber glands that are more than twice the diameter of normal peripheral zone glands, 2) preserved basal cells as identified with basal cell markers, 3) cytologically malignant cells, and 4) an expansile cell mass that spans the glandular lumen. The fifth major criterion, central comedonecrosis, is diagnostic of IDC-P but not always present. Minor criteria include glands with 1) right angle branching or 2) smooth rounded outlines, and 3) 2 cell populations with an outer perimeter cell group composed of tall, pleomorphic, and mitotically active cells that stain poorly for prostate-specific antigen (PSA), and a central group that is cuboidal, monomorphic, and quiescent with abundant cytoplasm containing abundant PSA and occasional extracellular mucin.

In 2006, Guo and Epstein proposed diagnostic criteria for IDC-P in prostate biopsy (Table 2) that have subsequently been used in a larger study of IDC-P from the same institution\textsuperscript{14,16}. In these two studies, in addition to the presence of malignant epithelial cells filling large acini and prostatic ducts with preservation of basal cells, the diagnosis of IDC-P required the presence of 1) a solid or dense cribriform pattern, where punched-out luminal spaces account for $<50\%$ of the central cellular mass; or 2) marked nuclear atypia, where nuclei are at least 6x larger than adjacent benign nuclei; or 3) nonfocal comedonecrosis. Lesions that fall short of these criteria but are still felt to be more ominous than HGPIN are labeled as “atypical intraductal proliferations” where the differential
Diagnosis rests between HGPIN and IDC-P. Any atypical cribriform lesions that do not meet the diagnostic criteria for IDC-P are termed “atypical cribriform lesions” and additional work-up is warranted.

**Differential Diagnosis of IDC-P**

IDC-P should be differentiated from a broad spectrum of other prostate lesions with cribriform and/or solid architecture, ranging from normal histological structures or benign lesions to premalignant lesions and frank malignancies (Table 3).

**Normal prostatic structures and benign lesions**

Normal histological variations, such as central zone prostate glands, and benign glandular proliferations, such as cribriform clear cell hyperplasia and basal cell hyperplasia, can both present as cribriform or rarely solid structures. However, nuclear atypia, mitotic figures and comedonecrosis are absent.

**High-grade prostatic intraepithelial neoplasia**

The distinction between isolated IDC-P from cribriform HGPIN in prostate biopsy is the most important one as management for these two conditions are drastically different.

Both HGPIN and IDC-P represent the presence of cytologically atypical or malignant cells within prostatic ducts and acini, although the architectural and cytological atypia is always more pronounced in IDC-P. Cribriform HGPIN is rare. The glands are small with smooth and round contours and the cells are relatively uniform without marked nuclear pleomorphism or necrosis in cribriform HGPIN. Loose cribriform and micropapillary patterns can be seen in both HGPIN and IDC-P, but dense cribriform and solid patterns and comedonecrosis are not seen in HGPIN. To establish the diagnosis of IDC-P in the loose cribriform and
micropapillary patterns, other cytological features such as markedly enlarged and pleomorphic nuclei (> 6X of adjacent non-neoplastic glands), and non-focal comedonecrosis, are required.

These criteria can reliably distinguish IDC-P and HGPIN in most cases. However, some IDC-P glands have morphological features that overlap with HGPIN. In two recent studies, Shah et al and Han et al demonstrated that some IDC-P glands that were intermixed with and shared the same TMPRSS2-ERG gene fusion with invasive PCa were architecturally and cytologically similar to the HGPIN glands that were distant from the invasive PCa and lacked the TMPRSS2-ERG gene fusion. Both were comprised of small cribriform glands with round and smooth contours and were lined with low grade nuclei. These findings suggest that IDC-P can occasionally exhibit a “low grade” morphology that overlaps with HGPIN and does not fit the diagnostic criteria for IDC-P. Therefore, any cribriform or lumen-spanning atypical lesion may represent IDC-P and its presence in prostate needle merits aggressive work-up.

**Invasive cribriform acinar adenocarcinoma of prostate**

Infiltrating cribriform acinar adenocarcinoma (Gleason pattern 4 or 5, depending on whether comedonecrosis is present) closely mimics cribriform IDC-P. Invasive cribriform cancer, unlike IDC-P, lacks a basal cell lining. In some cases, the contour and branching pattern of normal duct architecture distinguishes IDC-P from infiltrating cribriform acinar adenocarcinoma. The distinction between invasive high grade PCa and IDC-P, though, is not critical as IDC-P is almost always associated with a high grade and high volume PCa. Most cases of IDC-P would be diagnosed as cribriform acinar adenocarcinoma if immunohistochemistry for basal cells is performed.

**Ductal adenocarcinoma of the prostate**
Ductal adenocarcinoma is an aggressive form of PCa and may occasionally arise in the peripheral zone, and ordinary acinar prostate carcinoma may focally have ductal adenocarcinoma features. It is defined by morphological features including tall pseudostratified columnar epithelium arranged in cribriform patterns with slit-like spaces and/or true papillary fronds. The papillae in ductal adenocarcinoma have true fibrovascular cores and the cells may show significant nuclear atypia with a high mitotic rate and extensive necrosis. Nuclei are large, mostly elongated or oval, and often contain a single macronucleolus. In contrast, IDC-P has cuboidal cells, cribriform pattern with rounded lumina, and micropapillary tufting without fibrovascular cores.

Similar to other high grade and high volume PCa, ductal PCa is also prone to intraductal spread. Residual basal cells are therefore often demonstrated in ductal PCa. Ductal PCa with basal cells is mechanistically IDC-P and Cohen et al proposed to classify ductal PCa as IDC-P. However, “ductal PCa” is not synonymous with IDC-P, as it has characteristic morphological features. Furthermore, not all ductal PCa cases have residual basal cells and therefore are not “intraductal carcinomas”.

Urothelial carcinoma involving the prostate

Intraductal spread of urothelial carcinoma, either from a bladder primary, or exceedingly rarely from a prostate primary, may mimic IDC-P. Urothelial carcinoma cells may fill and distend the lumen of prostatic ducts and central necrosis may occur, imparting a morphology similar to IDC-P. However, urothelial carcinoma is typically more pleomorphic than IDC-P cytologically, and it often has a dense pink, “hard” cytoplasmic quality. A panel of immunostains can often resolve the diagnostic ambiguity. IDC-P stains positive for prostate specific markers, including PSA, prostate specific acid phosphatase (PSAP), prostate specific membrane antigen (PSMA) and P501S, while stains for basal cells, such as CK5/6, 34βE12 and p63, are positive only in the basal cells at the periphery of the
cancer glands. In contrast, urothelial carcinoma is negative for prostate specific markers (PSA and PSAP) and is positive in 2/3 of cases for markers that recognize the prostate basal cells (cytokeratin 34βE12 and p63).

**Metastatic adenocarcinoma**

Metastatic adenocarcinoma from other sites, in particular colorectal adenocarcinoma, may have extensive necrosis and mimic IDC-P. On H&E stains, the presence of “dirty” necrosis, columnar appearance of cells with basally located nuclei, and mucus secretion are features that suggest the enteric origin of the lesion. Clinical history and prudent use of immunostains (CDX-2, CK20, β-catenin for colorectal adenocarcinoma) can lead to a correct diagnosis.

**Incidence and Clinical Significance of IDC-P in Prostate Biopsy**

Since the initial studies by Kovi et al and McNeal et al\(^4,8\), several other studies have investigated IDC-P in radical prostatectomy and found consistently that the presence of IDC-P correlated with other adverse pathological features, including higher Gleason score, larger tumor volume, and greater probability of extraprostatic extension, seminal vesicle invasion and pelvic lymph node metastasis. It also correlated with decreased progression free survival and postsurgical biochemical recurrence\(^10-12,15,17\).

In a recent study, we investigated the incidence of IDC-P in prospectively examined prostate biopsies\(^25\). The incidence of IDC-P in our prospective PBx cohort was 2.8% (33/1176) during a 23-month period. IDC-P was identified in 10.6% (33/312) of cancer-containing PBx. The incidence of IDC-P without associated invasive PCa on PBx was 0.26% in our study. This study demonstrates that IDC-P is uncommon in prostate biopsies, and even rarer as an isolated finding without concomitant PCa. However, IDC-P identified in prostate biopsies correlates with high grade and high volume PCa, consistent with other studies. In this study, 26 of 30
(86.7%) PBx with PCa and concomitant IDC-P had a primary Gleason pattern 4 or 5, including 4+3=7 in 12 (40%), 8 in 4 (13.3%) and 9 in 10 (33.3%) cases. The mean number of biopsy cores involved by PCa was 7.2. These findings suggest that many of these patients harbor high grade, large volume disease. This conclusion is further supported by the finding in the corresponding radical prostatectomy specimens where 8 of 9 (88.9%) patients had a primary Gleason pattern 4 or 5, including GS 4+3=7 in 3 (33.3%), GS8 in 1 (11.1%) and GS9 in 4 (44.4%). Poor prognostic factors including EPE and SVI were identified in 67% and 44% of patients, respectively. Compared with the incidence predicted by Partin Tables, the incidence of SVI in our cohort (44%) was significantly higher than that predicted by Partin Tables (12%). Our study suggests that IDC-P in prostate biopsies may provide additional prognostic values beyond that provided by conventional pathological parameters such as Gleason score and tumor volume.

Cohen et al. studied a small series of radical prostatectomy (RP) with matching pre-operative needle biopsies and found that the inclusion of IDC-P in prostate biopsies in a preoperative model could improve the prediction of pathological stage of RP specimens. Furthermore, the presence of IDC-P on biopsy correlated strongly with biochemical failure. This study also found that serum PSA levels correlated with tumor volume only when IDC-P was not present in biopsy, similar to an earlier study. These findings suggest that IDC-P identified in prostate biopsy is a very powerful parameter that counteracts the predictive values of other commonly used clinicopathological parameters, including serum PSA and biopsy Gleason score. This has potentially profound clinical significance, as virtually all nomograms for predicting prostate cancer stage and outcome include serum PSA as a variable, yet none takes into account the presence of IDC-P as potentially masking a high volume, high stage tumor with relatively low serum PSA. Similarly, a recent study by O’Brien et al also found that inclusion of several new pathological variables including IDC-P significantly improved the
predictive accuracy of a postoperative nomogram that used preoperative clinicopathological variables to predict PSA recurrence after radical prostatectomy. These studies strongly suggest that the presence of IDC-P in prostate biopsies should be reported even when it is associated with an extensive, high grade PCa, as it may provide additional prognostic information.

Guo and Epstein initially reported a small series of cases of IDC-P without invasive carcinoma on biopsy, and more recently Robinson and Epstein updated and expanded this series to include a total of 66 patients. Both studies found that the presence of IDC-P, even in the absence of documented invasive carcinoma, was associated with an aggressive clinical course. In the more contemporary of the two studies, 8 of 66 (12%) patients developed disease progression after definitive treatment, including 4 patients with distant metastasis at a mean of 22 months after diagnosis, and another 4 patients with PSA recurrence at a mean of only 8 months following definitive therapy. Furthermore, in patients who underwent radical prostatectomy, all PCa were Gleason score ≥7, and nearly half of the cases contained some Gleason pattern 5 components. Tumors were also of relatively high volume (mean: 2.85 cm3). Eight of 21 men (38%) had extraprostatic extension (pT3a), and another 3 (14%) had seminal vesicle invasion (pT3b). Nodal metastasis was seen in 1 (5%) of the men. Two patients (10%) had IDC-P only at RP without invasive PCa component. This latter finding suggests that IDC-P does not always represent intraductal spread of an invasive high grade carcinoma. In at least some cases, IDC-P represents a stage of prostate carcinogenesis that is beyond what we recognize as HGPIN morphologically but before invasive cancer develops.
Based on their studies of needle biopsy with IDC-P, and previous studies in the literature that demonstrated consistent association of IDC-P at RP with multiple adverse prognostic factors, Robinson et al recommend definitive therapy in men with IDC-P on needle biopsy even in the absence of pathologically documented invasive PCa.

**Reporting of IDC-P in prostate biopsy:**

Because of its frequent association with high grade and high volume PCa as well as adverse prognostic significance, IDC-P should be reported in prostate biopsy reports.

In the majority of cases, IDC-P is identified with a concomitant invasive PCa that almost always contains Gleason patterns 4 or 5 cancer. In these cases, reporting of IDC-P is recommended as it may provide additional prognostic information.

Rarely, IDC-P is seen on biopsy with only Gleason pattern 3 PCa. In these cases, it is imperative to document presence of IDC-P, and this can be done in two ways. Either IDC-P can be regarded as and graded like invasive PCa (Gleason patterns 4 or 5, depending on the absence or presence of solid and/or necrosis, respectively). Alternatively, one can grade only the invasive Gleason pattern 3 PCa and then mention in a comment the presence of IDC-P and its clinical significance.

When IDC-P is identified on prostate biopsy without concomitant invasive PCa, pathologists should report the presence of IDC-P with a comment stating that IDC-P is almost always associated with a high grade and high volume PCa and that definitive therapy is indicated for these patients\textsuperscript{14,16}. However, some pathologists may still recommend immediate repeat biopsy instead of definitive therapy in these rare situations.
Finally, any cribriform lesion comprising cytologically atypical cells that do not satisfy the diagnostic criteria for IDC-P, but exceed the criteria for HGPIN, should be reported on biopsy as “atypical cribriform lesion (ACLs)” with a recommendation of immediate repeat biopsy as it may represent IDC-P.

Conclusion:

IDC-P is a distinct clinicopathological entity. Morphologically it is characterized by lumen-spanning or solid proliferation of malignant cells that expand the preexisting ducts and acini. It is strongly associated with aggressive PCa with high Gleason grade and large tumor volume. Therefore, it is critical for pathologists to recognize and report this lesion in prostate specimens, especially in prostate biopsy, for patient management. Morphological criteria have been proposed to distinguish it from several other lesions with similar histological appearance such as HGPIN, invasive cribriform PCa and urothelial carcinoma involving the prostate.

Reference

Table 1. Histological Features That May Be Seen in Intraductal Carcinoma of the Prostate

<table>
<thead>
<tr>
<th>Feature</th>
<th>Description</th>
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<tbody>
<tr>
<td>No. of IDC-P glands, No.</td>
<td>Many, ; often &gt;6 / per prostate gland</td>
</tr>
<tr>
<td>Size</td>
<td>Larger than normal glands, ; may can be &gt; 1 mm</td>
</tr>
<tr>
<td>Ductal-lobular structure</td>
<td>Native ducts and acini are expanded and may show irregular and branching contours</td>
</tr>
<tr>
<td>Intraductal growth pattern</td>
<td>1. Loose cribriform with cells forming narrow strands (often two 2 cells thick), spanning lumen without stromal support and intersecting randomly to form an orderly lacework of empty spaces</td>
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<tr>
<td></td>
<td>2. Micropapillary with cells forming papillae with inconspicuous fibrovascular cores</td>
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3. Dense cribriform with cells forming small, round “punched out” lumens which comprise > 50% of the luminal space

4. Solid cell mass

**Cytology**

1. Cuboidal or low columnar

2. Significant nuclear atypia

3. Nuclei ×6 times larger than adjacent non-nonneoplastic nuclei

4. 2 cell populations with central small and uniform nuclei and peripheral pleomorphic nuclei may be seen in dense cribriform and solid patterns

**Comedonecrosis**

May be present

**Basal cell layer**

Preserved, at least focally

IDC-P: intraductal carcinoma of the prostate

**Table 2. Diagnostic Criteria for Intraductal Carcinoma of the Prostate**

Malignant epithelial cells filling large acini and prostatic ducts, with preservation of basal cells and:

- Solid or dense cribriform pattern
Or

- Loose cribriform or micropapillary pattern with either
  - Marked nuclear atypia: nuclear size ×6 normal or larger
  - Non-Nonfocal comedonecrosis
<table>
<thead>
<tr>
<th>Feature</th>
<th>IDC-P</th>
<th>HGPIN</th>
<th>or Solid PCa</th>
<th>Involving the Prostate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ductal-lobular structure</td>
<td>Expanded</td>
<td>Preserved</td>
<td>Distorted</td>
<td>Preserved (usually)</td>
</tr>
<tr>
<td>Gland size</td>
<td>Increased (&gt; 2X times size of normal glands)</td>
<td>Variable</td>
<td>Variable</td>
<td></td>
</tr>
<tr>
<td>Lumen-spanning cell mass</td>
<td>Present</td>
<td>Present in cribriform HGPIN</td>
<td>Present</td>
<td>Present</td>
</tr>
<tr>
<td>Intraductal growth pattern</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2. Cribriform (loose/, dense)</td>
<td>2. Cribriform (loose)</td>
<td>2. Solid</td>
<td>2. Cribriform</td>
<td></td>
</tr>
<tr>
<td>3. Solid</td>
<td>3. Flat</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4. Tufting</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Comedonecrosis</td>
<td>±</td>
<td>−</td>
<td>±</td>
<td>±</td>
</tr>
<tr>
<td>Basal cells</td>
<td>+</td>
<td>+</td>
<td>−</td>
<td>+</td>
</tr>
</tbody>
</table>

Abbreviations:
IDC-P: intraductal carcinoma of the prostate; HGPIN: high-grade prostatic intraepithelial neoplasia; PCa: prostate carcinoma.
Intraductal Carcinoma of the Prostate - the Whole Story!

Ming Zhou, MD, PhD
Departments of Pathology and Urology
New York University Langone Medical Center
New York, NY
Ming.Zhou@NYUMC.ORG

Case
➢ 75 y.o. male with persistently elevated PSA for 12 years
Differential Diagnosis of Atypical Cribriform Lesions of the Prostate

- Intraductal carcinoma of the prostate
- Invasive cribriform prostatic carcinoma
- Ductal adenocarcinoma of the prostate
- Cribriform high grade PIN
- Urothelial carcinoma involving the prostate
- Metastatic (colorectal) adenocarcinoma

Outline

- Historical perspective
- Histological features and diagnostic criteria
- Differential diagnosis
- Incidence and clinical significance of IDC-P in prostate biopsies
- Reporting
**Intraductal Carcinoma of the Prostate (IDC-P)
Historical Perspective**

- Used variably for extension into prostatic ducts and acini of prostatic acinar carcinoma, ductal carcinoma, urothelial and colorectal carcinoma in early literatures
- Currently only refers to lumen-spanning proliferation of prostate cancer cells within prostatic ducts and acini caused by the spread of prostate cancer cells into preexisting prostatic glandular structures

**Intraductal Carcinoma of the Prostate (IDC-P)
Current Concept**

- Intraductal spread and growth of advanced-stage PCa
  (Cohen, Br J Urol, 1998; Rubin, AJSP, 1998; Wilcox, Hum Pathol, 1998; Cohen, Prostate, 2000)
- Rarely a form of PCa with predominant/exclusive intraductal growth with little/no invasive PCa
  (Cohen, Arch Pathol Lab Med, 2007; Robinson and Epstein, J Urol, 2010)

**Intraductal Carcinoma of the Prostate (IDC-P)
Histological Features**

**Hallmark**

1. Expansile proliferation of PCa cells
   - Cribriform or solid architecture
2. Within native prostate glands
   - Basal cell layer at least partially preserved
Many atypical cribriform/solid glands

Intraductal Carcinoma of the Prostate

Marked variation in nuclear size
Pleomorphic nuclei >6X adjacent nuclei

Intraductal Carcinoma of the Prostate (IDC-P)
Diagnostic Criteria

- Morphological features not specific
- Use a constellation of morphological features
- Use stringent diagnostic criteria to ensure its unique clinical significance, i.e., association with adverse outcomes
Diagnostic Criteria for IDC-P
(Cohen RJ et al, Arch Pathol Lab Med; 2007)

Five major criteria:
• Large-caliber glands that are twice the diameter of normal glands
• Preserved basal cells as identified with basal cell markers
• Cytologically malignant cells
• Expansile cell mass that spans the glandular lumen
• Central comedonecrosis (diagnostic of IDC-P but not always present)

Three minor criteria:
• Right-angle branching
• Smooth rounded outlines
• Two cell populations with an outer pleomorphic cells and a central cuboidal monomorphic cells

Diagnostic Criteria for IDC-P
(Guo CC and Epstein JI, Mod Pathol. 2006)

Malignant epithelial cells filling large acini and prostatic ducts, with preservation of basal cells require either:
• A Solid or Classical cribriform pattern
• Marked nuclear atypia/nuclei 6x larger than adjacent benign nuclei
• Non-focal comedonecrosis

Lesions that fall short of these criteria:
“Atypical intraductal proliferations”
Atypical intraductal proliferations

Differential Diagnosis of Intraductal Carcinoma of the Prostate
(DDX for Atypical Cribriform/Solid Lesions)

- Intraductal carcinoma of the prostate
- Invasive cribriform prostatic carcinoma
- Ductal adenocarcinoma of the prostate
- Cribriform high grade PIN
- Urothelial carcinoma involving the prostate
- Metastatic (colorectal) adenocarcinoma

### Difference between IDC-P and Cribriform High Grade PIN

<table>
<thead>
<tr>
<th></th>
<th>Cribiform HGPIN</th>
<th>IDC-P</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Definition</strong></td>
<td>Putative precursor lesion to PCA</td>
<td>Intraductal spread of advanced stage PCa</td>
</tr>
<tr>
<td><strong>Clinical Significance in Prostate Biopsy</strong></td>
<td>Cancer risk associated with the diagnosis of HGPIN in prostate biopsy: ~ 25%</td>
<td>Almost always associated with high grade/volume PCa</td>
</tr>
<tr>
<td><strong>Patient Management</strong></td>
<td>HGPIN diagnosed in prostate biopsy does not mandate a repeat biopsy within 1 year when HGPIN is focal</td>
<td>Definitive treatment recommended</td>
</tr>
</tbody>
</table>

- Atypical cribriform lesion with basal cells intermixed with or within 3 mm from the border of invasive cancer
- ERG gene fusion: 75%  
- ERG fusion status concordant between IDC-P and adjacent Pca in 100% cases

- Atypical cribriform lesion with basal cells > 3 mm from the border of invasive cancer
- ERG gene fusion: 0%

IDC-P and cribriform HGPIN are genetically distinct  
ERG gene status identical between IDC-P and Pca  
IDC-P: resulting from intraductal spread of PCa

(Han et al. AJSP 2010)
Morphological Difference b/w of IDC-P and Cribriform HGPIN

(Shah, Magi-Galluzzi, Han, Zhou, ASIP 2010)

<table>
<thead>
<tr>
<th></th>
<th>IDC-P</th>
<th>Cribriform HGPIN</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td># cases</td>
<td>43</td>
<td>23</td>
<td>N.A</td>
</tr>
<tr>
<td># atypical cribriform lesion</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean</td>
<td>2.3</td>
<td>2.4</td>
<td>0.02</td>
</tr>
<tr>
<td>Range</td>
<td>0.5</td>
<td>1.6</td>
<td></td>
</tr>
<tr>
<td>Smallest size (mm)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean ± S.D.</td>
<td>0.31 ± 0.39</td>
<td>0.33 ± 0.33</td>
<td>0.48</td>
</tr>
<tr>
<td>Range</td>
<td>0.25 - 1.0</td>
<td>0.3 - 1.0</td>
<td></td>
</tr>
<tr>
<td>Largest size (mm)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean ± S.D.</td>
<td>1.5  ± 1.3</td>
<td>0.43 ± 0.15</td>
<td>0.035</td>
</tr>
<tr>
<td>Range</td>
<td>0.6 - 2.6</td>
<td>0.2 - 1.8</td>
<td></td>
</tr>
<tr>
<td>Glandular contour</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Regular</td>
<td>29 (67.4%)</td>
<td>19 (82.6%)</td>
<td>0.19</td>
</tr>
<tr>
<td>Irregular</td>
<td>14 (32.6%)</td>
<td>5 (17.4%)</td>
<td></td>
</tr>
<tr>
<td>Branching</td>
<td>36 (85.7%)</td>
<td>14 (42.4%)</td>
<td>0.019</td>
</tr>
<tr>
<td>Architecture</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cribriform</td>
<td>42 (95.5%)</td>
<td>23 (100%)</td>
<td>0.001</td>
</tr>
<tr>
<td>Dense cribriform or solid</td>
<td>15 (34.9%)</td>
<td>0 (0%)</td>
<td>0.001</td>
</tr>
<tr>
<td>Comedo necrosis</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Uniform</td>
<td>18 (41.9%)</td>
<td>14 (60.9%)</td>
<td>0.18</td>
</tr>
<tr>
<td>Variable</td>
<td>31 (71.2%)</td>
<td>9 (30.1%)</td>
<td>0.26</td>
</tr>
<tr>
<td>&gt; 6X or pleomorphic</td>
<td>12 (27.9%)</td>
<td>0 (0%)</td>
<td>0.005</td>
</tr>
</tbody>
</table>

IDC-P with “Low Grade Morphology”

Cribriform HGPIN
IDC-P with “Low Grade Morphology”

Difference between IDC-P and Cribriform High Grade PIN

- IDC-P is much more common than cribriform HGPIN
- Defining features of IDC-P
  - Large branching architecture
  - Dense cribriform/solid
  - Comedonecrosis
  - Pleomorphic nuclei (>6X adjacent nuclei)
- IDC-P may have “low grade morphology”
  - Any “atypical cribriform lesions”, even without high grade morphology, should be worked up in prostate biopsy

Atypical intraductal proliferations

Cribriform HGPIN

Cribriform PCa
Cribriform PCa

Ductal Adenocarcinoma of the Prostate

Ductal Adenocarcinoma of the Prostate

High Grade Urothelial Carcinoma Involving Prostatic Acini
Incidence and Significance of IDC-P in Prostate Biopsy

- 33/1176 (2.8%) of prospectively examined prostate biopsies
- 3 (0.26%) not associated with PCa
- 9 underwent radical prostatectomy

### Pathological Features

<table>
<thead>
<tr>
<th>Risk of adverse Pathological features</th>
<th>Patients with IDC</th>
<th>Predicted by Partin tables</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Extraprostatic extension</td>
<td>6/9 (67%)</td>
<td>41%</td>
<td>0.173</td>
</tr>
<tr>
<td>Seminal vesicle invasion</td>
<td>4/9 (44%)</td>
<td>12%</td>
<td>0.002</td>
</tr>
<tr>
<td>Lymph node metastasis</td>
<td>1/9 (11%)</td>
<td>5%</td>
<td>0.383</td>
</tr>
</tbody>
</table>

Watts KE et al, Mod Pathol 2012;25:1250A
Significance of IDC-P in Prostate Biopsy


- IDC-P in prostate biopsies should be reported even when it is associated with an extensive and high-grade PCA, as it may provide additional prognostic value

Significance of Finding Intraductal Carcinoma of the Prostate without Concomitant Invasive Carcinoma on Prostate Needle Biopsy

- 83 men with biopsies showing only IDC-P
  - Treatment (n=66)
    - RP 23, RT 15, HT 8, RT/HT 15, Repeat biopsy 3, None 2
  - 21 radical prostatectomies (RP)
    - Stage
      - T1a 9 (42.9%), T3b 3 (14.3%), T2 (42.9%)  
    - Gleason score
      - mean 7.9 (7-10)
    - 37%: primary or secondary pattern 5, 11% had tertiary pattern 5
  - Follow up
    - Available in 66 (79.5%) for 23 (1-58) months
    - 4 (6%) had bone mets (including one with RP)
    - 4 (6%) had PSA recurrence (including 3 with RP)
  - At RP, men with biopsies showing only IDC-P typically have high grade (GS>7) and advanced stage (pT3) PCA
  - Men with IDC-P as the sole finding in PBx should be treated with definitive therapy

Reporting Recommendations for Prostate Biopsy with IDC-P

<table>
<thead>
<tr>
<th>Diagnosis</th>
<th>Reporting recommendations</th>
</tr>
</thead>
<tbody>
<tr>
<td>IDC-P with high-grade PCs</td>
<td>Document IDC-P (may provide an additional prognostic value)</td>
</tr>
</tbody>
</table>
| IDC-P with Gleason pattern 3 PCs | Grade PCs and document IDC-P and its poor prognostic significance in the report  
|                                  | Grade IDC-P as pattern 4 or 5 |
| IDC-P without PCs                | Document IDC-P and its poor prognostic significance in the report  
|                                  | Advise immediate rebiopsy or recommend definitive therapy |
| Atypical cribriform lesions that do not meet the criteria for cribriform HGPIN and IDC-P | Diagnostic an "atypical cribriform lesion, cannot rule out IDC-P"  
|                                  | Recommend immediate repeat biopsy |

Intraductal Carcinoma of the Prostate: Summary

- A distinct clinicopathologic entity
- Cribriform/solid proliferation of PCa cells that expand the preexisting ducts and acini
- Strongly associated with aggressive, high grade/volume PCa
- Morphologic criteria have been established to diagnose and distinguish IDC-P from several other lesions with similar histologic appearance
- It is critical to recognize and report IDC-P in prostate specimens, especially in prostate biopsies reports, for patient management
Acknowledgement

Dr. Rajal Shah, Miraca Life Sciences, Dallas, TX

Questions?
Most of the renal cortical tumors can be easily classified on hematoxylin and eosin evaluation alone. However, there are some cases that need the support of immunohistochemistry and other ancillary tests for their proper classification. Such help is particularly important for the pathologists working in laboratories handling relatively low volumes of renal tumor specimens. The need for good, reliable immunohistochemical stains has also been progressively growing in the recent years, with pathologists increasingly being asked to render diagnosis on limited material (needle core biopsies and fine-needle aspirates).

Over the years, a large number of immunohistochemical markers have been reported to show remarkable sensitivity and specificity in discriminating between different renal cortical tumors. After the initial excitement created by the first few publications, very often the subsequent larger studies on wider range of tumors water down the initial claims. One of the common reasons for the initial “great” versus subsequent “not so great” results is that in most instances, the initial studies are performed on tumors that are morphologically typical representatives of a tumor type. As a matter of fact, in the day-to-day pathology practice, such cases hardly ever need immunohistochemical support for the diagnosis. When the same antibodies are used on tumors that are morphologically somewhat diverse and atypical, and that really need immunohistochemical support for the diagnosis, the results may be disappointing. As long as the original and subsequent investigators stick to the “factualness” of their observations, controversies will be very common. However, facts are facts; specific antibodies mark the tumors as reported in the initial publications, as well as in the subsequent papers. Only that, the authors in initial publications often did not investigate the tumors from the perspective that the subsequent authors did. And, non-acceptance of these simple facts makes a CONTROVERSY!

While none of them can be regarded as 100% sensitive and specific, in spite of the controversies, a number of antibodies are still useful in the differential diagnosis of renal cell tumors; some more so than others. A few of these are listed and discussed; the list should not be considered all inclusive.

Carbonic anhydrase-IX (CA-IX): It is a down-stream marker in the Hypoxia-Inducible Factor (HIF) pathway. HIF is generated in most body cells as a routine. However, in the presence of normal oxygen tension, it immediately gets hydroxylated. Hydroxylated HIF undergoes very quick proteosomic degradation in the presence of normal pVHL. Whenever there is tissue hypoxia, HIF does not get adequately hydroxylated; non-hydroxylated HIF cannot be degraded and is therefore over-expressed. In another scenario, at normal oxygen tension even when HIF gets hydroxylated, and normal pVHL is not present (as is usual in VHL syndrome, and very
common in sporadic clear cell RCCs), the hydroxylated HIF does not get degraded. Thus, it gets over-expressed and activates multiple down-stream proteins, including CA-IX.

CA-IX is a trans-membrane protein; therefore, the immunostain is primarily localized to the cell membranes. CA-IX is expressed in overwhelming majority (84-100%) of clear cell RCCs in a diffuse membranous pattern. Its expression has been shown to be retained in metastases from clear cell RCC. The staining is diffuse, irrespective of the nuclear grade of the tumor, although an occasional high grade tumor may show somewhat decreased staining intensity. Unlike any other antibody used for clear cell RCC, CA-IX expression is retained even in sarcomatoid areas in the overwhelming majority of sarcomatoid clear cell RCCs. The antibody is quite useful in the diagnosis of clear cell RCC even on limited material (needle core biopsies and cytological material). Although some pathologists are still concerned about its specificity (see below), the antibody (clone g250) is already being used in clinical trials by radiologists in the diagnosis of clear cell RCC at both the primary and metastatic sites. A recent multicentric phase 3 trial reported a specificity and sensitivity of greater than 86% each, in radiologic scans using a radio-labeled iodine and CA-IX compound in classifying renal tumors as clear cell RCC.

CA-IX also shows diffuse membranous immunoreactivity in clear cell papillary RCC, albeit qualitatively a little different from that seen in clear cell RCC. Positive staining has also been reported in bladder urothelial carcinoma, papillary RCC, and a number of other non-renal tumors. However, to the best of our knowledge, even when reported as positive with diffuse staining in only a small proportion of these positive tumors, none of these was reported to show expression in almost 100% of the tumor cells (as is characteristic in clear cell RCC). Such immunoreactivity likely is not related to the absence of pVHL in these tumors, but to the areas of tissue hypoxia. In most tumors other than clear cell RCC, when immunoreactivity is present, it is more prominent around the areas of tumor necrosis, and additionally in the case of papillary RCCs, close to the tips of papillae. Nevertheless, no immunostain can be used as a solitary marker in histo- or cyto-pathological diagnosis. Most of the non-clear cell RCC tumors with some CA-IX positivity will also show immunoreactivity to other markers that are appropriate for those specific tumors. In our experience, only VHL-related syndromic or sporadic tumors (e.g., clear cell papillary cystadenomas of the epididymis, cerebellar hemangioendotheliomas, etc.) show reactivity for CA-IX akin to that seen in clear cell RCC.

Another practical caveat in interpreting CA-IX staining is that in limited material (core biopsies, FNAs) that shows extensive tumor necrosis, positivity may be misleading. Most of the positive staining areas can represent peri-necrotic, hypoxic regions in the tumors. Therefore, interpretation of such biopsies requires great caution.

Overall, in our opinion, CA-IX is among the best available antibodies for recognizing clear cell RCC; primary, metastatic, high-grade, sarcomatoid, or on limited material.

PAX 8/2:
Pax8 and Pax2 are members of the paired box family of transcription factors and are very important in the fetal development of the central nervous system, eye, kidney, thyroid gland, and organs of the mesonephric (wolffian) and müllerian duct origin. They show nuclear expression in overwhelming majority of the tumors of renal cell origin, and are, therefore, very useful markers for establishing a renal cell derivation in both the primary and metastatic settings. Pax8 appears to be a more sensitive marker than Pax2, although only Pax2 may be expressed in very rare instances.

While evaluating staining for Pax markers, one always needs to remember that both Pax8 and Pax2 are not renal cell-specific, and are known to be expressed in a number of other tumors, including many tumors of the female genital tract, thyroid, a small proportion of upper urinary tract urothelial carcinoma, rare abdominal mesotheliomas, etc. At the same time, both may be absent in some tumors of renal origin, particularly some high grade tumors like collecting duct carcinoma, sarcomatoid RCCs, and tumors with rhabdoid features, as well as many chromophobe RCCs and renal oncocytomas.

Thus, interpretation of staining results on Pax 8 or 2 needs be performed in conjunction with other appropriate stains. Neither the presence of staining in isolation is definitive of renal cell origin, nor does absence of staining completely exclude such origin.

Renal Cell Carcinoma marker (RCCMa):

RCCMa, which is normally expressed in the proximal renal tubules, has been in use as a marker of renal cell origin for many years now. However, it shows sensitivity only in the range of 27-90 percent among primary renal tumors. At the same time the antibody labels a number of other tumors including lung adenocarcinoma, hepatocellular carcinoma, breast lobular carcinoma, ovarian clear cell carcinoma, parathyroid carcinoma and many others. In a more recent publication by Sharma et al, the antibody was shown to label greater than 40% of non-renal tumors with papillary architecture from various sites.

Given the availability of some better antibodies for the diagnosis of renal cell origin, RCCMa appears to have very limited clinical value now.

CK7:

CK7, a commonly utilized antibody in most surgical pathology laboratories, has multiple utilities in the differentiation of renal cell tumors. CK7 is usually diffusely expressed in type 1 papillary RCC, classical chromophobe RCC and clear cell-papillary RCC. At the 2012 Vancouver ISUP consensus conference, a majority of participants voted for performing immunohistochemical
stains (including CK7) to diagnose clear cell-papillary RCC, in addition to taking the morphologic features into consideration.

CK7 is usually negative or very focally positive in acquired cystic disease-associated RCC, even when it shows a prominent papillary architecture. CK7 is also generally negative in clear cell RCC, although it may show focal positivity in these tumors around cystic areas. Renal oncocytomas are either negative, or show only rare immunoreactive cells. While it may be useful in distinction of oncocytoma from classical chromophobe RCC, in general CK7 is not very useful in distinction of oncocytoma from eosinophilic variant of chromophobe. Such eosinophilic variants only rarely show diffuse CK7 positivity; the labeling in eosinophilic chromophobes is more often only focal or patchy. Similarly, type 2 papillary RCCs also very often show only limited staining with CK7, unlike the diffuse pattern in type 1 tumors.

CK7 remains a useful antibody in the differentiation of many renal tumors. However, too much dependence of the staining results is nor warranted in the distinction of renal oncocytoma from eosinophilic variant of chromophobe RCC, or type 2 papillary RCC from its mimics.

**CD10:**

CD10 has been used for many years in the distinction of clear cell RCC from other tumors, particularly in metastatic setting. Only membranous staining can be considered as compatible with clear cell RCC, in an appropriate clinicopathological scenario. On the other hand, cytoplasmic positivity for CD10 is seen in a large number of non-renal and renal tumors. Additionally, membranous immunoreactivity for CD10 is not limited to clear cell RCC, and may be present in papillary RCC, colorectal, pancreatic, prostatic, or some vascular tumors as well.

CD10 cannot be used in isolation to diagnose clear cell RCC, either in primary or metastatic setting. Given that better markers for renal cell (like Pax8/2) or clear cell RCC (like CA-IX) origin are now available, the dependence on CD10 may be on the wane.

**Others:**

The search for reliable immunohistochemical stains that can consistently distinguish between oncocytic neoplasms (renal oncocytoma, eosinophilic chromophobe RCC, low-grade oncocytic unclassified RCC) has been long and frustrating. A number of immunostains have been proposed in this distinction, but in the long run, none has proven to be as efficient as claimed in the initial publications. The likely reasons for that are discussed above. Multiple antibodies used in this distinction, over the past 1½ to 2 decades include, anti-mitochondrial antibodies, E-cadherin, Kidney-specific cadherin, S100A1, EpCam, etc. Kangai 1 (KAI1/CD82), product of metastasis
suppressor gene, in a few recent publications appears to have some potential. But, more evidence is needed yet.

References:


Controversies in immuno-histochemistry in renal cell tumors

Satish K. Tickoo
Controversy

Controversy: Merriam-Webster- “discussion marked especially by the expression of opposing views”

So… on controversial issues- what is true

- Most often all points of view; generally the truth, but often not “the whole truth” and “nothing but the truth”
- Example; 20°F temperature in NYC- both cold and “quite warm”

Therefore, controversy very often the creation of one’s unidirectional thought processes
Immunohistochemistry in renal tumors

- Most stains “help” in tumors where no help is really needed
- Some antibodies - put in the controversy list - may actually be useful if used appropriately
  - Carbonic Anhydrase-IX (CA-IX)
  - Pax-8/2
  - CK7
  - CD10
  - RCC Ma
Carbonic Anhydrase-IX (CA-IX)

- CA-IX, a trans-membranous enzyme/protein with functions to regulate intracellular and extracellular pH
- Expression induced by hypoxic/pseudohypoxic conditions; downstream to HIF protein: overexpression related to hypoxia or lack of pVHL
- Diffuse membranous positivity in 84-100% clear cell RCCs
CA-IX; Pros....

- Diffuse membranous + in clear cell RCCs
  - Both primary and metastatic (84 vs. 89%)
    - (Tickoo et al. USCAP 2007)
  - Even in high-grade areas with solid, pseudo-papillary, papillary areas (3+ expression; 88 vs. 95%)
  - 3/4ths of sarcomatoid clear cells in sarcomatoid areas (3+ expression; 73 vs. 86%)
    - (Tickoo et al. J Urol. 2007;177:1224)
CA-IX; Pros......

- Highly useful on limited material
  - Biopsies
  - Cell blocks (Personal experience)

- CA-IX (g250) (chimeric$^{124}$I-cG250) in phase 2 and 3 trials for radio-diagnosis of clear cell RCC
Positive in non-renal tumors
- More concentrated around necrosis; cells away from necrosis, mostly negative

Positive in urothelial carcinomas
- Almost never 100% or close to 100% cells (26% HIF-1, 3+; Tickoo et al. BJU International 2010;107:844; 14-18% CA-IX, 3+; Donato et al. 2011 Histopathology;59:1229-- +3= >50%)
- Usually CK7/CK20/HMWCK/p63 positive

Reportedly positive in papillary RCC
  - Around necrosis and papillary tips; never anywhere close to 100% cells in our experience
  - CK7/AMACR positive
CA-IX; Cons......

- Positive in translocation-associated RCC and HLRCC-associated carcinomas
  - Usually absent or patchy staining
- Clear cell papillary RCC
  - Always positive
CA-IX

- To date- the best marker in our hands/opinion for clear cell RCC, particularly when used in combination with H&E and other markers
Pax 8/2; Pros......

- Overwhelming majority, but not all, of renal tumors positive
- Clean nuclear stain
- In most cases both positive, but rarely only one and not the other (Pax 8 generally > Pax 2)
- If clinically renal tumor a consideration, positivity strongly supports a renal origin
Pax8/2; Cons

- Tumors from other organs positive (Female genital tract, thyroid, etc.)
- May not be positive in high grade tumors of renal cortical origin
- Positive in a small proportion of upper urinary tract urothelial carcinomas
Table 2: PAX 8 expression in primary neoplasms

<table>
<thead>
<tr>
<th>Organ</th>
<th>Diagnoses</th>
<th>Positive/total cases (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kidney</td>
<td>RCC, clear cell</td>
<td>90/95 (95%)</td>
</tr>
<tr>
<td></td>
<td>RCC, papillary(^a)</td>
<td>38/38 (100%)</td>
</tr>
<tr>
<td></td>
<td>RCC, chromophobe</td>
<td>22/25 (88%)</td>
</tr>
<tr>
<td></td>
<td>RCC, collecting duct</td>
<td>5/7 (71%)</td>
</tr>
<tr>
<td></td>
<td>RCC, sarcomatoid</td>
<td>4/9 (44%)</td>
</tr>
<tr>
<td></td>
<td>RCC, rhabdoid</td>
<td>0/4 (0%)</td>
</tr>
<tr>
<td></td>
<td>RCC, mucinous tubular</td>
<td>6/6 (100%)</td>
</tr>
<tr>
<td></td>
<td>RCC, ACK-related</td>
<td>8/8 (100%)</td>
</tr>
<tr>
<td></td>
<td>RCC, translocation</td>
<td>1/2 (50%)</td>
</tr>
<tr>
<td></td>
<td>RCC, tubulocystic</td>
<td>1/1 (100%)</td>
</tr>
<tr>
<td></td>
<td>Metanephric neoplasms</td>
<td>3/3 (100%)</td>
</tr>
<tr>
<td></td>
<td>Mixed epithelial and stromal tumors</td>
<td>4/4 (100%)</td>
</tr>
<tr>
<td></td>
<td>Cystic nephroma</td>
<td>4/4 (100%)</td>
</tr>
<tr>
<td></td>
<td>Urothelial carcinoma</td>
<td>0/13 (0%)</td>
</tr>
<tr>
<td></td>
<td>Oncocytoma</td>
<td>8/13 (61%)</td>
</tr>
<tr>
<td></td>
<td>Angiomyolipoma</td>
<td>0/8 (0%)</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>194/240 (89%)</td>
</tr>
<tr>
<td>Female genital tract(^b)</td>
<td>Endometrioid carcinoma</td>
<td>72/77 (93%)</td>
</tr>
<tr>
<td></td>
<td>Endometrial polyp</td>
<td>8/8 (100%)</td>
</tr>
<tr>
<td></td>
<td>Serous carcinoma</td>
<td>101/102 (99%)</td>
</tr>
<tr>
<td></td>
<td>Serous adenoma/borderline tumors</td>
<td>22/23 (95%)</td>
</tr>
<tr>
<td></td>
<td>Mucinous carcinoma</td>
<td>4/10 (40%)</td>
</tr>
<tr>
<td></td>
<td>Mucinous adenoma/borderline tumors</td>
<td>3/13 (23%)</td>
</tr>
<tr>
<td></td>
<td>Clear cell carcinoma</td>
<td>16/16 (100%)</td>
</tr>
<tr>
<td></td>
<td>Other types(^c)</td>
<td>12/18 (66%)</td>
</tr>
<tr>
<td></td>
<td>Cervical squamous cell carcinoma</td>
<td>0/9 (0%)</td>
</tr>
</tbody>
</table>

Seminal vesicles: 27/27 (100%)
Epididymis: 17/17 (100%)

Ozcan et al. Modern Pathol. 2011;24:751
<table>
<thead>
<tr>
<th></th>
<th>PAX2*</th>
<th></th>
<th>PAX8†</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n</td>
<td>Positive Cases (%)</td>
<td>n</td>
</tr>
<tr>
<td>Clear cell RCC, primary</td>
<td>330</td>
<td>288 (87)</td>
<td>248</td>
</tr>
<tr>
<td>Papillary RCC, primary</td>
<td>179</td>
<td>108 (60)</td>
<td>59</td>
</tr>
<tr>
<td>Chromophobe RCC, primary</td>
<td>117</td>
<td>19 (16)</td>
<td>43</td>
</tr>
<tr>
<td>Collecting duct RCC, primary</td>
<td>13</td>
<td>5 (38)</td>
<td>30</td>
</tr>
<tr>
<td>Tubulocystic RCC, primary</td>
<td>33</td>
<td>13 (39)</td>
<td>1</td>
</tr>
<tr>
<td>Mucinous tubular and spindle cell RCC, primary</td>
<td>7</td>
<td>6 (86)</td>
<td>6</td>
</tr>
<tr>
<td>Xp11 translocation RCC, primary</td>
<td>36</td>
<td>14 (39)</td>
<td>2</td>
</tr>
<tr>
<td>Sarcomatoid RCC, primary</td>
<td>7</td>
<td>0 (0)</td>
<td>18</td>
</tr>
<tr>
<td>Clear cell RCC, metastatic</td>
<td>323</td>
<td>202 (63)</td>
<td>329</td>
</tr>
<tr>
<td>Papillary RCC, metastatic</td>
<td>8</td>
<td>6 (75)</td>
<td>13</td>
</tr>
<tr>
<td>Sarcomatoid RCC, metastatic</td>
<td>5</td>
<td>0 (0)</td>
<td>8</td>
</tr>
<tr>
<td>Urothelial carcinoma, upper urinary tract</td>
<td>47</td>
<td>0 (0)</td>
<td>64</td>
</tr>
</tbody>
</table>

RCC indicates renal cell carcinoma.

*From references.18,20,22-33
†From references.30,32,33,43-47
Pax 8/2

- Positivity cannot be used in isolation as diagnostic of renal origin
- Absent staining does not exclude renal origin, particularly so in high grade tumors
RCCMa: Pros and Cons......

• Renal Cell Carcinoma marker (RCCMa) an antibody for RCC with reported sensitivity and specificity of approximately 75%
• Reasonable, but not great sensitivity and specificity
• Positive in a number of non-renal primary tumors
  • lung adenocarcinomas
  • hepatocellular carcinomas
  • lobular carcinoma breast
  • parathyroid carcinomas
  • ovarian clear cell carcinomas
  • testicular and extra-gonadal seminomas, yolk sac tumor
<table>
<thead>
<tr>
<th>Histologic Subtype</th>
<th>Positive</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Classic clear cell</td>
<td>46/64</td>
<td>72</td>
</tr>
<tr>
<td>Papillary renal cell</td>
<td>40/42</td>
<td>95</td>
</tr>
<tr>
<td>Unclassified</td>
<td>33/38</td>
<td>85</td>
</tr>
<tr>
<td>Sarcomatoid</td>
<td>6/30</td>
<td>20</td>
</tr>
<tr>
<td>Oncocytic</td>
<td>18/24</td>
<td>75</td>
</tr>
<tr>
<td>Chromophobe</td>
<td>20/22</td>
<td>91</td>
</tr>
<tr>
<td>Metatastastic</td>
<td>10/23</td>
<td>40</td>
</tr>
<tr>
<td>Total (all types)</td>
<td>173/241</td>
<td>72</td>
</tr>
</tbody>
</table>
RCC Ma: Cons…

- RCCMa positive in 28/66 (42%) of non-renal papillary neoplasms, including:
  - 9/9 (100%) papillary thyroid carcinomas
  - 8/10 (80%) ovarian papillary serous carcinomas
  - 4/9 (44%) uterine papillary serous carcinomas
  - 1/10 (10%) papillary urothelial carcinomas
  - 1/2 (50%) intraductal papillary mucinous carcinomas of the pancreas
  - 3/3 (100%) choroid plexus papillomas
  - 1/1 (100%) pituitary adenoma with papillary features
  - 1/2 (50%) lung adenocarcinomas with papillary features

CK7

- Is a commonly used antibody in most pathology labs
- Usually positive in papillary and chromophobe RCC
- Mostly negative in clear cell RCC
- Very focally positive in renal oncocytoma
CK7; Pros……..

- Very reliable antibody for clear cell papillary RCC- diagnostic utility (ISUP 2012)
- Quite reliable for papillary RCC, particularly type 1
- Often diffusely positive in classical chromophobe RCC
- Variably positive in collecting duct carcinoma
CK7; Cons

- Clear cell RCCs with cystic components, often positive
  - (Fine et al. 2012, USCAP)
- Eosinophilic chromophobe RCC, often very focal or negative
  - Not of much use in distinction from oncocytoma
- Type 2 papillary RCC, often negative
CK7

- Use and interpret with caution in tumors where it is needed the most (oncocytoma vs. eosinophilic chromophobe; high grade papillary)
CD10

- **Common Acute Lymphoblastic Leukemia Antigen (CALLA)**
- **Labels clear cell RCC in a membranous pattern (approximately 80%)**
  - (Liu et al. Arch Pathol Lab Med. 2007;131:1290; Tickoo et al. 2007, USCAP)
- **Labels papillary RCC**
  - Usually along the apical membrane
- **Variable expression apical/membranous/cytoplasmic in chromophobe RCC and oncocytoma (0-35%)**
CD10; Pros……

- In metastatic setting, may differentiate from clear cell adenocarcinoma of GYN tract
CD10: Cons…..

- Apical surface only: well-differentiated colonic, pancreatic and prostatic adenocarcinoma
- Canalicular pattern in HCC may mimic membranous positivity, particularly in limited tissue
- Diffuse cytoplasmic or membranous/Golgi patterns: poorly differentiated adenocarcinoma, melanoma, urothelial carcinoma, renal cell carcinoma, endometrial stromal sarcoma
  - (Am J Clin Pathol 2000;113:374)
- Epithelioid hemangioendothelioma can be strong/diffuse CD10+ and mimic metastatic renal cell carcinoma
  - (Arch Pathol Lab Med 2009;133:1965)
- Solid and pseudopapillary pancreatic tumor is CD10+
  - (Am J Surg Pathol 2000;24:1361)
CD10

- To support the diagnosis of clear cell RCC, positivity has to be membranous; cytoplasmic positivity is seen in a number of other tumors
- Cannot depend on CD10 alone
Anti-Mito, E-Cad, Ksp-Cad, S100A1, KAI1…

- Multiple markers claimed to distinguish between oncocytoma and chromophobe RCC
- Initial publications raise expectations, but ultimately none proven to be great
  - Kidney specific-Cadherin: “The findings argue against the use of Ksp-cad in differentiating chromophobe renal cell carcinoma and renal oncocytomas”
    (Kuehn et al. Am J Surg Pathol. 2007;31:1528)
- Currently, KAI1 being touted as potentially useful
Conclusions

- “When a thing ceases to be a subject of controversy, it ceases to be a subject of interest”. William Hazlitt
- “In a controversy the instant we feel anger we have already ceased striving for the truth, and have begun striving for ourselves”. Buddha