INTRODUCTION

Neuroendocrine [NE] lesions of the GU tract represent a spectrum of neoplasms with diverse incidence, clinicopathologic presentation, and outcome. An overview of their potential origins reveals three major theories. These include: 1) Derivation from NE cells of the diffuse neuroendocrine system - such as those identified in the normal urothelial tract and prostate and which may increase in number in reactive or metaplastic settings; 2) Derivation from a multipotent stem cell – a concept crucial to understanding the nature of NE tumors arising in conjunction with epithelial or germ cell malignancies, which may express markers of both components 3) Derivation from non-APUD cell neuroendocrine structures - encompassing lesions such as bladder paraganglioma, as well as its rare counterparts in other organs.

For each entity that follows, relevant epidemiologic, clinicopathologic, immunohistochemical, and prognostic data are surveyed, including a discussion of their proposed origins. Additionally, the most significant differential diagnostic considerations have been highlighted.

NEUROENDOCRINE LESIONS OF UROTHELIUM

High grade neuroendocrine carcinoma

The majority of previous reports of high grade neuroendocrine carcinoma [NEC] involve small cell carcinoma [SmCC] of bladder, which accounts for 0.5 to 1% of primary bladder malignancies (1-3). SmCC is comparable to high grade urothelial carcinoma [UC] with regard to median age in the seventh decade, gender [M:F ratio – 2:1 to 5:1] (4-5) and clinical presentation with hematuria [≥75%] (1). Unlike SmCC in the lung (6) however, bladder SmCC is uncommonly associated with paraneoplastic syndromes (5).

Microscopically, high grade NEC represents a spectrum of findings. “Pure” SmCC cases are infrequent, with approximately 50% showing admixed carcinoma, including urothelial carcinoma, NOS, but also adenocarcinoma, squamous cell carcinoma, and/or sarcomatoid features (7-8). The co-existence of these epithelial components, as well as identification of overlying flat in situ UC, strongly suggest that the high grade NE phenotype reflects divergent differentiation of multipotent malignant urothelial cells (1, 9). Even within the NEC component, tumors exhibit a range of morphology, from “classic” SmCC features, as seen in the lung, i.e. diffuse sheets of round blue hyperchromatic cells exhibiting nuclear molding, granular chromatin, inconspicuous nucleoli, scant cytoplasm, and frequent mitoses/apoptotic debris and necrosis to lesions with better-defined organoid, palisaded, or trabecular architecture and large cells with abundant cytoplasm and macronucleoli (10). In areas, the latter may convey a “carcinoid-like” architecture at low-power, albeit with cytologic features better associated with high grade NEC.
A key differential diagnostic consideration is direct extension or metastasis from non-urothelial NEC. In addition to morphologic overlap, the difficulty in distinguishing primary from secondary lesions is compounded by immunohistochemical and ultrastructural similarities, including cytoplasmic chromogranin and synaptophysin positivity, “dot-like” cytokeratin positivity (4, 9), and sparse intracytoplasmic membrane-bound dense core granules (8-9). Although initially proposed as a specific marker for pulmonary SmCC (11-12), TTF-1 positivity has subsequently been demonstrated in 25% to 39% of bladder SmCC (13-14), limiting its utility in this differential diagnosis (15). Given these similarities, high grade pulmonary NEC secondarily involving urothelium must be excluded on clinical and radiologic grounds. As approximately half of prostatic SmCC have an associated conventional adenocarcinoma component, immunopositivity for prostate specific antigen [PSA] or prostatic acid phosphatase [PAP] in better differentiated areas may be helpful.

Bladder SmCC presents with high clinical stage and frequently displays rapid growth and metastasis at the time of or shortly after diagnosis to lymph nodes [LN], viscera, and vertebral bones (1-2, 4-5, 8, 16-17). Available outcome data suggests a poor prognosis for cases of bladder SmCC, correlating with advanced stage at presentation (2). Parallel clinicopathologic features and outcomes have been reported for exceedingly rare cases of SmCC of the ureters and renal pelvis, as well as large cell NEC [LCNEC] of lower or upper tract (3, 9-10, 18-21). Most modern series have highlighted therapeutic regimens containing cisplatin-based chemotherapy (22), in addition to surgical resection (4, 23) or radiotherapy (2, 24-25) as being beneficial. Given the propensity of SmCC for early systemic disease, some have further advocated neoadjuvant therapy with pulmonary SmCC dosages (26). However, the ideal timing and dosing of these treatments remains unknown due to the retrospective and sometimes limited nature of prior studies (27). Similarly, the low frequency of urothelial high grade NEC and its tendency to exhibit a range of morphologies have precluded clinically meaningful subclassification. Diagnostically therefore, it is reasonable to recommend reporting these cases as high grade UC with small cell/neuroendocrine differentiation, while noting explicitly the extent of the NE component, as it may affect therapeutic options.

**NEUROENDOCRINE LESIONS OF BLADDER**

**Bladder Paraganglioma**

Paragangliomas [PG] of the bladder represent 10% of extra-adrenal PG (28-29), with the classic clinical triad of sustained or paroxysmal hypertension, intermittent gross hematuria, and micturition “attacks”, characterized by headache, palpitations, blurred vision, profuse sweating, tremulousness and occasional syncope, being observed in nearly ¾ of patients (30-31). These usually well-circumscribed lesions are characterized by nests [Zellballen] of polyhedral cells with ampho- or basophilic cytoplasm and ovoid nuclei intertwined within a delicate vascular network. However, irregular growth, endocrine atypia, rare mitoses, dense fibrous septae, and focal ganglioneuromatous or neuroblast-like growth may also be seen (29). Immunohistochemical staining is identical to that seen in adrenal pheochromocytomas, with chromogranin and synaptophysin positive chromaffin cells, S-100 protein positive sustentacular cells, and negativity for keratins (32-33). While the true clinical behavior is unknown, approximately 80% of patients in small series have shown at least muscle invasive disease (33-34). This capacity to invade deeply, along with the Zellballen
architecture of PG necessitate their distinction from nested patterns of UC, especially in small biopsies or those with thermal artifact (33). While infiltrative nests of UC, often without desmoplasia, may mimic PG, the latter should not exhibit superficial urothelial disease, true cytologic atypia, or significant mitotic rates. Furthermore, other than focally, nested variants of UC lack the fine vasculature characteristic of PG. In difficult cases, immunopositivity for CK7, 20, and HMW cytokeratin will resolve the dilemma (33).

Similar to other extra-adrenal PG, bladder PG are thought to arise by malignant transformation of paraganglia (29-30, 35-37). In an elegant autopsy study of over 400 patients, Honma has demonstrated paraganglia at all levels of the bladder wall, including within detrusor muscle, in > 50% of patients (38). Occasionally, in small specimens, these collections of clear to amphophilic cells may also cause diagnostic confusion with full-fledged PG (33). In this scenario, symptomatology and cystoscopic evidence of a mass, coupled with repeat biopsy documenting extent of disease, should clarify the issue.

**Bladder Carcinoid**

Less than ten true primary bladder carcinoids have been documented (39-44) with a usual presentation as small polypoid masses at the bladder neck/trigone in patients with hematuria. Tumors demonstrate classic carcinoid architectural patterns including glandular, acinar, cribriform, or trabecular arrangements of uniform cells with basally-oriented eosinophilic cytoplasm (43).

When considering a diagnosis of primary carcinoid tumor anywhere in the GU tract, it is essential to thoroughly exclude a metastatic lesion from more common sites such as lung or GI tract. In the bladder specifically, pathologists must also bear in mind that urothelial high grade NEC may also exhibit focal “carcinoid-like” morphology (8). Unlike SmCC of the urinary bladder, which may be considered divergent differentiation from UC, an association of bladder carcinoids and UC has not been observed. It is well known however, that cells with NE features may be situated against the basement membrane of normal urothelium, as well as in reactive lesions such as von Brunn nests and cystitis cystica et glandularis (45-46). Indeed, two cases of primary bladder carcinoid have developed in a background of proliferative cystitis of the overlying urothelium (41, 43), including one case with scattered argyrophil cell staining noted in this component (41). A proposed origin of primary bladder carcinoid from innate or metaplastic mucosal urothelial NE cells is consistent with these findings.

**NEUROENDOCRINE LESIONS OF KIDNEY**

**Renal Carcinoid**

Similar to patients with renal cell carcinoma [RCC], patients with carcinoid tumor of the kidney present with abdominal, back, or flank pain, accompanied by hematuria and fever (47-48). 25-30% of renal carcinoids are incidentally detected (49) and diagnosis may be complicated with small lesions, as neither CT nor MRI reliably distinguishes these tumors from RCC (50). Evidence of carcinoid syndrome with serotonin-related flushing, generalized edema, and diarrhea, and occasional elevation of urine 5-HIAA may occur in 10-15% of patients (51). In its presence, somatostatin receptor [SR]
scintigraphy with octreotide has a high sensitivity for renal carcinoid detection, including small, clinically silent lesions (52).

Histologically, polygonal tumor cells with indistinct cell borders, round, regular nuclei, “salt and pepper” chromatin, and infrequent mitoses grow in trabeculae or nests set in a highly vascularized, yet thin fibroconnective tissue. Glandular, acinar, tubular, or rosette-like growth and densely fibrotic/sclerotic stroma may also be observed (51, 53). Ultrastructural studies demonstrate abundant neurosecretory granules and diffuse labeling for cytokeratin, chromogranin, and synaptophysin is typical (47, 51). Interestingly, these tumors have demonstrated positivity for PAP, suggesting a rectal carcinoid-like hindgut origin (47-48, 51, 54). Approximately 15% of reported cases were initially diagnosed as RCC or Wilms tumor (52, 55), likely due to under-recognition of renal carcinoid, with uniformly negative WT-1 also ruling out the latter possibility (56). Finally, as in other sites, metastasis from a remote carcinoid tumor must be excluded.

Due to the vacuous nature of the retroperitoneal space, these slow-growing neoplasms are usually diagnosed at a large size, with ~ 75% being > 4 cm (50). A recent review found approximately 45% of patients with pT3 disease [tumor invading peri-renal or sinus/hilar fat or invading renal vein]. Metastases, seen in 50-60% of cases, are usually detected at initial evaluation, though they have been reported up to 7 years post-nephrectomy. Metastases involve peri-aortic and peri-hilar LN, liver, and bone (50) and their presence may correlate with age > 40 years, tumors > 4 cm, pT3 tumors, and mitotic rates > 1 per 10 hpf. Surprisingly however, more than 90% of reported patients were without evidence of disease or alive with disease [median 34 months follow-up], while 47% of patients with LN metastases at the time of resection showed no evidence of disease [mean 43 months follow-up]. Hence, patients with renal carcinoids may experience a prolonged clinical course, even in the presence of widely metastatic disease (49-50, 57, 59).

Fascinating associations between primary renal carcinoid and horseshoe kidney [18-26%] or renal teratomas [~ 15% of patients] have been reported (58, 60-64). These findings have led some to postulate that hyperplasia of interspersed NE cells within metaplastic or teratomatous epithelium in horseshoe kidneys or nests of misplaced progenitor cells developing into teratomatous intestinal or respiratory epithelia in renal teratoma may serve as a nidus for renal carcinoids (63, 65). This view is supported by the common occurrence of renal carcinoids in the isthmus of horseshoe kidneys, a region formed by abnormal migration of posterior nephrogenic cells in utero (63). However, nearly 65% of reported lesions have not occurred in these settings. Other suggestions, including reactive NE metaplasia of the pyelocaliceal system and NE differentiation of multipotent renal stem cells have been fielded, yet neither pyelonephritis/renal calculi nor areas of renal carcinoid co-existent with UC or RCC have been observed (60, 63). While the presence of intrinsic NE cells in normal kidney may be debated, rare NE cells have been observed in urothelium of the upper tract (53, 66). It is possible therefore, that renal carcinoids bearing no relationship to congenital/acquired abnormalities arise directly from NE cells situated in the renal pelvic urothelium.

NEUROENDOCRINE LESIONS OF TESTIS

Testicular Carcinoid
Representing ~ 0.2% of all testicular neoplasms in a historical series from the AFIP (67), primary testicular carcinoids are infrequent, with reported patients ranging from 10 to 83 years (68) and, not unlike germ cell tumors, typically presenting with a mass or testicular swelling, accompanied by pain and tenderness (67). Clinical carcinoid syndrome may be seen in up to 10% of cases (68). Grossly, testicular carcinoids are solid, with an accompanying cystic component, when seen in conjunction with other teratomatous elements (67). Microscopically, they share characteristic features, immunoprofile, and ultrastructural polymorphous neurosecretory granules with carcinoids at other sites (67-70). Although most testicular carcinoids behave in an indolent manner, 10-15% metastasize, occasionally many years after orchiectomy (67-68, 71).

As with other GU tract carcinoids, metastasis from another site must be entertained, especially in the presence of bilateral tumors (72). Regarding primary lesions, early reports (67, 73-74) postulated that analogous to carcinoids arising in ovarian teratomas (75), primary testicular carcinoids may be derived from NE cells found in respiratory epithelial or enteric components of testicular teratomas (67). However, a teratomatous component has been associated with only ¼ of reported cases and intratubular germ cell neoplasia [ITGCN], a precursor lesion indicating germ cell origin, has generally been absent (76). While overgrowth of a single teratomatous component may be observed, the finding of a pure testicular carcinoid without other demonstrable germ cell tumor on initial sampling should engender extensive testicular sampling to discover ITGCN, a minute teratoma, and/or evidence of a scar, representing a “burnt out"/regressed germ cell component (77). In the event of pure testicular carcinoid in association with OCT-4-labeling ITGCN, as recently reported (78), cytogenetic studies for i12p may still be warranted, to eliminate the possibility of concurrent metastatic carcinoid. Once these efforts are exhausted, one may entertain other proposed origins, including origin from yet undiscovered NE rests (71) or designation of the lesion as a monodermal teratoma with a pathogenesis different from that of usual postpubertal teratoma [i.e. no ITGCN] (79).

NEUROENDOCRINE LESIONS OF PROSTATE

Neuroendocrine cells in the prostate

First described in the prostate by Pretl in 1944 (80), focal NE cells are now known to be widely distributed in the prostate (81-82), are members of the diffuse APUD cell system (83), and along with prostatic secretory cells, are thought to arise from endodermal-derived pluripotent prostatic stem cells (84-85). While their exact function is unknown, it is postulated that they are involved in both prostatic growth and differentiation, as well as in homeostatic regulation of the secretory process (86). NE differentiation in prostate adenocarcinoma [PCa] has three major manifestations: 1. Focal NE differentiation in PCa; 2. Prostatic carcinoid tumor; 3. High grade NE carcinoma.

Focal neuroendocrine differentiation in prostate cancer

30 to 100% of conventional PCa contain scattered NE cells (46, 81, 87), with most resembling other prostatic secretory cells on light microscopy. NE cells may be detected immunohistochemically with markers such as chromogranin and/or bioactive hormones such as serotonin and somatostatin (88). Individual cells may express both PSA and PAP as well as NE markers (89). Whether focal NE differentiation in prostatic
adenocarcinoma has prognostic import is controversial, with a few studies suggesting correlation between increasing numbers of chromogranin-positive cells and worse prognosis (90-93). Most authors however, have shown a correlation of NE differentiation with tumor grade and failed to show an independent effect on survival (85, 88, 94-96). Likewise, there is evidence that metastatic PCa contains a population of NE cells (97), which do not express the androgen receptor [AR] (98-100) and hence may not be suppressed by androgen ablation (97). It has therefore been conjectured that NE cells possess the ability to “escape” usual hormonal therapy in advanced PCa, with some reporting increased NE differentiation in androgen-insensitive PCa as well as possible prognostic significance (98, 101). However, others have argued that these relationships may depend on the agent used in androgen deprivation and have demonstrated no statistical correlation between amount of NE cells and disease specific survival (95). These varying results suggest that the often limited and focal distribution of NE cells in PCa makes its difficult to study their relevance, especially in limited specimens, such as needle biopsies (102). Nonetheless, a number of studies have shown potential roles for NE cells in PCa progression by paracrine growth stimulation of non-NE cells (97, 101, 103).

Occasionally NE cells with bland nuclei and cytoplasmic eosinophilic granules, superficially resembling Paneth cells of the GI tract (104-105) are seen in a patchy fashion in both normal and cancerous specimens. Unusually, these cells may be observed as single cells, cords, or nests of tumor cells, meeting architectural criteria for Gleason pattern 5, yet displaying bland cytology and frequent association with lower grade conventional PCa (105). A recent study found that among cases with Paneth cell-like rich areas resembling high grade PCa, none showed evidence of progression. Cases exhibiting progression reflected typical parameters such as high Gleason scores and/or extraprostatic extension/semenal vesicle invasion. Diagnostically therefore, it is suggested that one grade only the conventional carcinoma, as applying Gleason to such foci may inaccurately upgrade the tumor (105).

Prostatic Carcinoid

The literature contains a fair number of cases denoted as “prostatic carcinoid tumors” (106-111). However, as usual PCa may exhibit at least focal NE differentiation, distinguishing carcinoid-like adenocarcinomas from a true primary prostatic carcinoid is challenging (106). Especially in Gleason grade 4/5 tumors, these entities may share nested and microacinar/“rosette-like” patterns of growth with nuclear uniformity, PAP positivity, and immunohistochemical/ultrastructural evidence of NE differentiation (112-113). Most have argued therefore, that tumors with expression of both PSA and NE markers, as well as cases of histologically mixed prostatic “carcinoid” and PCa are best considered “carcinoid-like” adenocarcinomas (49, 114). This phenomenon may explain early reports of aggressive behavior for prostatic carcinoids (115), likely reflecting high Gleason score tumors with varying degrees of focal NE differentiation. Using these guidelines, only a handful of tumors exhibiting typical carcinoid architecture and features, PSA negativity and absence of admixed adenocarcinoma have been described (116-117).

High grade neuroendocrine carcinoma

First described by Wenk in 1977 (118), prostatic SmCC represents between 1 and 5% of all prostatic malignancies when mixed adenocarcinoma-SmCC are included (82). Conversely, in the constellation of extrapulmonary SmCC, the prostate is a relatively
High grade NEC of prostate histologically resembles the spectrum of SmCC/LCNEC described at other sites (120-122), with approximately ½ of prostatic SmCC being composite tumors with conventional PCa (82). Diagnostically, high grade NEC should not be assigned a Gleason grade and may be differentiated from diffuse growth of Gleason pattern 5 PCa by the large cells with lower N/C ratio, prominent nucleoli, and absent nuclear molding seen in the latter (123). Conversely, due to its constellation of features, LCNEC may be easily mistaken for Gleason score 5+5=10 PCa and hence, the likelihood of its underdiagnosis is high (120).

A number of studies have reported the immunophenotype of SmCC and conventional PCa (9, 124-125). Overall, they have found strong labeling for PSA/PAP in most PCa, and at least focal expression in ~ 25% of SmCC, as well as diffuse NE markers in a majority of SmCC with substantially less staining in PCa (124-125). Furthermore, while most have maintained that malignant NE cells do not express AR (98-99) a recent study has shown focal staining for AR in SmCC (125). The presence of mixed prostatic acinar adenocarcinoma in many high grade SmCC and LCNEC, coupled with evidence of cells that may co-express PSA/PAP/AR and NE markers (120-121, 124-125) suggests evolution of a subset of multipotent non-NE prostatic tumor cells as the derivation for prostatic high grade NEC (84-85, 120, 124, 126). As in high grade NEC of the urothelium, local extension or distant spread of NEC from other sites must be excluded clinically, as morphologic and immunohistochemical features may be identical.

Akin to SmCC at other sites, prostatic lesions present at advanced stage, are often unresectable, and display a high frequency of visceral metastases and abysmal survival (121, 127-128). Small modern series have suggested managing prostatic SmCC with a combination of ADT with cisplatin-based regimens followed by consolidative surgery or radiotherapy (127-129). As in most SmCC however, even chemotherapy-treated patients tend to progress rapidly. As such, novel small molecule therapeutic approaches, developed for pulmonary high grade NEC (130), may have activity in prostatic SmCC. To this end, Yao et al have recently demonstrated high levels of labeling for CD56, bombesin/GRP, c-KIT, Bcl-2, and EGFR in a small series of prostatic SmCC, suggesting future therapeutic targets (125).

Few examples of distinct prostatic LCNEC have been reported (120, 131-133). Among these, the majority have been an incidental finding in palliative TURP specimens in patients with androgen-independent disease (120). As reported cases have been detected late in the disease course, widespread metastases to bone and viscera, uniformly poor responses to NE specific chemotherapy and limited survival have been observed (120).
REFERENCES