Histol Histopathol (2016) 31: 785-791

http://www.hh.um.es

Histology and Histopathology

From Cell Biology to Tissue Engineering

A clinicopathologic study of paragangliomas of the urinary bladder: can the clinical behavior of the tumor be predicted?

Shuangping Guo[1], Chaoliang Fan[1], Joseph Rohr[2], Linni Fan[1], Yingmei Wang[1], Mingyang Li[1], Xia Li[1], Ying Guo[1], Qingguo Yan[1], Lu Wang[1] and Zhe Wang[1]

[1]Department of Pathology, Xi Jing Hospital, the Fourth Military Medical University, Xi'an, Shaan Xi Province, China and [2]Department of Pathology and Microbiology, University of Nebraska Medical Center, Omaha, NE, USA

Summary. Paraganglioma of the urinary bladder is rare but even more unusual as no singular histologic feature is consistently characteristic of malignancy. Additionally, paragangliomas can manifest in hypertensive crisis for clinicians resecting the tumors in unusual locations without proper histologic diagnosis. Herein we report nine cases of paraganglioma of the urinary bladder with immunohistologic study and follow-up information, including one rare malignant case with liver metastasis. Comparison of the immunohistologic features reveal that the malignant case shows the common features suggested by both the Pheochromocytoma of the Adrenal Gland Scaled Score (PASS) and Grading of Adrenal Pheochromocytoma and Extra-adrenal Paraganglioma (GAPP) system. The predominant histopathologic features of malignant cases were large irregular nests with focal spindle tumor cells and a diffusely infiltrative growth pattern between smooth muscle of the urinary bladder wall with multiple necrotic areas and a high proliferative index. Eight cases without metastasis showed the classic zellballen of benign paragangliomas without irregular nests and well-demarcated nodules either in the submucosa or between smooth muscle bundles with no diffuse infiltration. We discuss the histopathologic and immunohistochemical features detecting malignant behavior, and comprehensively review the previously published cases of malignant paraganglioma of the urinary bladder. In summary, we assess some clinicopathologic features which might help to predict which neoplasms are more likely to behave in a clinically aggressive manner to avoid adverse outcomes in this rare tumor's resection.

Key words: Histopathology, Urinary bladder, Paraganglioma, Malignancy

Offprint requests to: Zhe Wang, M.D, Ph.D., Department of Pathology, Xi Jing Hospital, the Fourth Military Medical University, Xi'an, 710032, Shaan Xi Province, China. e-mail: Zhwang@fmmu.edu.cn

DOI: 10.14670/HH-11-751

Introduction

Paragangliomas usually arise from the adrenal medulla but may also arise from the carotid body, the urinary bladder and other locations. Paragangliomas of the bladder are histologically identical to paragangliomas at other sites and the commonly cited incidence is 0.006-0.10%. However, malignant paragangliomas of the urinary bladder are rare, and only 38 cases have been previously reported (Das and Lowe, 1980; Michel et al., 1990; Grignon et al., 1991; Pang and Tsao, 1993; Kato et al., 1999; Ansari et al., 2001; Salanitri et al., 2001; Yoshida et al., 2004; Segawa and Osafune, 2005; Huang et al., 2007; Kairi-Vassilatou et al., 2007; Yadav et al., 2007; Patnayak et al., 2015). Pathologic diagnosis, treatment, and prognosis of this unusual tumor are controversial (Piedrola et al., 1997). Paraganglioma is currently classified by the WHO as a borderline tumor; however, the histological and immunohistologic features to predict its biologic behavior remain controversial. Herein, we describe nine cases of paraganglioma of the urinary bladder including a malignant case with liver metastasis. In order to find the histologic and immunophenotypic features which may suggest the

malignancy, we compare the malignant cases to eight cases without metastasis during follow-up. In addition, the malignant cases of the urinary bladder reported in previous literature are reviewed.

Materials and methods

Patient materials

A retrospective search of the pathology database from 2006~2014 at Xi Jing Hospital using the key words paraganglioma of the urinary bladder, cases were reviewed by pathologists (S.P.G. and Z.W.) and nine cases including one malignant case with liver metastatic tumor were selected for study. The clinicopathological information including patients' age, tumor size, and operative model were collected. The follow-up data of these patients were obtained.

Immunohistochemistry

Resected tissues were fixed in 10% buffered formalin and embedded in paraffin and stained with hematoxylin-eosin (HE). Slides for immunohistochemical staining were prepared from formalin-fixed, paraffin-embedded tissue and immunohistochemically stained using EnVision™ . The slides were immersed in a citrate buffer at pH 6.0, pretreated in a microwave pressure cooker for 20 minutes at 92-95°C, and then stained using one of the primary antibodies summarized in Table 1. For Ki67 immunostaining, 1000 tumor cell nuclei per slide were evaluated manually at high magnification (×400), and nuclear staining was interpreted as positive. A Ki67 labeling index was determined as a percentage of positive-staining tumor cell nuclei.

Electron microscopy

The case with liver metastasis was observed using electron microscopy. Formalin-fixed tissue was subjected to electron microscopic examination. The tissue was minced into 1mm-thick cubes, re-fixed in glutaraldehyde and osmium tetroxide, and embedded in epon. Ultrathin sections were viewed and photographed on a CM120 transmission electron microscope (Phillips Electronics, Eindhoven, The Netherlands).

Clinical features

All nine patients (Table 2) had the characteristic presentation of a functional paraganglioma with

Table 1. Summary of the antibodies used in the study.

Antibody	Clone	Dilution	Source	Antigen Retrieval
EMA	E29	1:100	Dako Cytomation, Glostrup Denmark	Microwave-citrate
Ki67	MIB-1	1:200	Dako Cytomation, Glostrup Denmark	Pressure cooker-citrate
Chromogranin A	DAK-A3	1:50	Dako Cytomation, Glostrup Denmark	HIER
Synaptophysin	SY38	1:50	Dako Cytomation, Glostrup Denmark	HIER
S-100 protein	Rb polyclonal	1:100	Dako Cytomation, Glostrup Denmark	HIER
Vimentin	V9	1:50	Zymed laboratories, San Fransico, CA, USA	HIER
NSE	BBS/NC/VI-H14	1:100	Dako Cytomation, Glostrup Denmark	HIER
COX2	CX294	1:100	Dako Cytomation, Glostrup Denmark	HIER
SDHB	Rb polyclonal	1:100	Abcam,Shanghai company, China	None
P53	PAb 1801	1:100	Abcam, Shanghai company, China	HIER
GATA3	EPR17874	1:500	Abcam, Shanghai company, China	HIER

EMA, epithelial membrane antigen; HIER, heat induced epitope retrieval; NSE, neuron specific enolase; COX2, Cyclooxygenase-2.

Table 2. Clinicopathological features of paragangloimas of the bladder.

Case	Age/ gender	Diameter cm	Growth pattern	Vascular invasion	Large nest size	Necrosis	High cellularity	Ki67	S100	T stage	Recurrence	Follow-up
1	45/M	8	Diffusely infitrative	+	+	+	+	36%	Focally Negative	T2b	Liver metastasis	50 m
2	42/F	2.5	Focally invasive	-	-	-	+	5%	Positive	T2a	No	83m
3	50/F	0.5	Submucosa, well demarcated	-	-	-	-	2%	Positive	T1	No	62m
4	50/F	1	Submucosa,well demarcated	-	-	-	-	2%	Positive	T2a	No	62m
5	50/M	2	Focally invasive	-	-	-	-	5%	Positive	T2a	No	60m
6	55/F	0.5	Focally invasive	-	-	-	-	8%	Positive	T2a	No	35m
7	42/F	0.8	Focally invasive	-	-	-	+	2%	Positive	T2a	No	34m
8	73/F	1.2	Submucosa,well demarcated	-	-	-	-	5%	Positive	T2a	No	31m
9	25/F	1	Focally invasive	-	-	-	-	5%	Positive	T1	No	28 m

hypertension ranging from 150/100 to 165/110 mmHg for variable times before presentation at our institution. The median age was 48 years and the ratio of female to male was 3.5:1. In addition to hypertension, the most common symptoms were the lower urinary tract symptoms and macroscopic painless hematuria. For a 45-year-old male (Table 2, Patient 1), cystography and CT revealed a tumor measuring 4.8×8.0 cm which protruded into the lumen in the left lateral wall of the urinary bladder (Fig 1). Invasive transitional cell carcinoma was suspected and total cystectomy was performed. CT of this patient also revealed a small tumor in the left part of liver at the same time and hemangioma was considered, although the hepatic tumor grew rapidly during follow-up. The liver tumor and a part of the liver were resected two months later. For the other eight patients (Table 2, Patients 2-9), cystography and CT revealed submucosal tumors ranging from 0.5 to 2.5 cm; lumpectomy or partial cystectomy was performed in each case. Follow-up information was obtained in all nine patients. There were no recurrences except the patient with liver metastasis during follow-up period with a median of 50 months (range, 28-83 months).

Results

Gross findings

The clinicopathologic features of nine paragangliomas are summarized in Table 2. For the case with liver metastasis (Patient 1), total cystectomy was performed. Transient elevation of systolic blood pressure to 250 mmHg occurred upon touching or pressing the tumor during the operation. The blood pressure returned to normal level after tumor resection. Examination of the urinary bladder specimen obtained at surgery revealed a submucosal tumor in the left side of the urinary bladder. The lesion was soft, measuring 8.0 cm in greatest dimension. Examination of the liver specimen obtained at surgery two months after total cystectomy revealed a tumor measuring 2.5×2.5cm. The tumor was well-circumscribed from the liver tissue. For the other eight cases, the lesions were well-demarcated, submucosal, soft nodules measuring 0.5 to 2.5 cm in greatest dimension.

Histopathologic findings

For the case with liver metastasis, the bladder tumor was partially covered by normal transitional epithelium. Underneath the epithelium were nests of tumor cells. Histologic examination showed a neoplasm composed of anastomosing cell cords or a trabecular arrangement of tumor cells. The characteristic small nests pattern ("zellballen") was focally seen. The predominant histologic pattern was of irregular large nests (Fig. 2a). A "large nest" was defined as three times the size of "zellballen," or the size of medullary paraganglia nests. The clusters consisted of relatively uniform, round eosinophilic cells with abundant, finely granular cytoplasm. In some areas, a specific growth pattern was lost altogether and a diffuse pattern was dominant with focal spindle tumor cells. The cellularity was high with a high nuclear-cytoplasmic ratio (Fig. 2a). The nuclei were hyperchromatic with occasional pleomorphisms and intranuclear cytoplasmic inclusions, and bizarre nuclei and atypical mitoses were observed (Fig. 2b). Mitotic counts >10 per 10 high power fields (magnification ×400) were observed. The cell clusters were closely associated with a rich vascular sinusoidal network. Pyknosis of single tumor cell and areas of focal necrosis (Fig. 2c), as well as vascular invasion (defined by intravascular attached tumor thrombi beneath the normal transitional epithelium) were also observed Fig. 2d). The histopathologic features of the tumor in the liver were similar to that of urinary bladder except that mitotic figures could be more easily observed (Fig. 2e).

The eight cases without metastasis during follow-up showed the typical morphology of paragangliomas with predominantly small alveolar or trabecular architecture (Fig. 2f). The tumors were either located submucosally with a well-circumscribed border or grew between bundles of smooth muscles in the bladder wall. The common features suggested in both PASS and GAPP score system predicting malignancy, such as large nests, necrosis, vascular invasion and sheets of high nuclear/cytoplasm ratio cells were not observed.

Immunohistochemical findings

All nine cases were positive for neuroendocrine markers such as chromogranin A (Fig. 3a), synaptophysin and neuron-specific enolase (NSE). Eight cases including the one with liver metastasis expressed

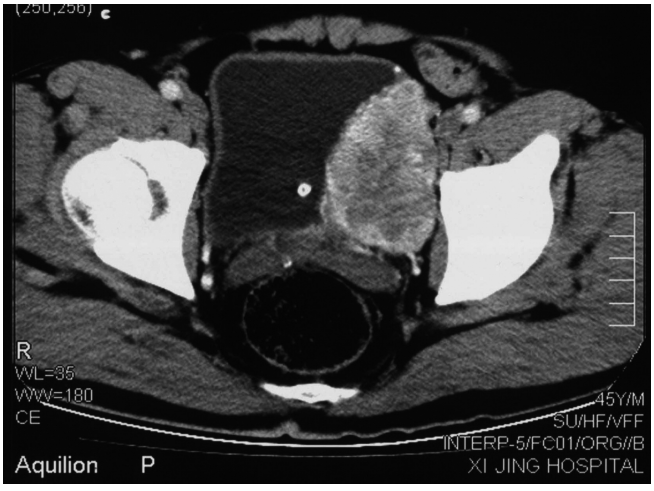

Fig. 1. CT revealed a tumor in the left lateral wall of the urinary bladder protruding into the bladder lumen.

cyclooxygenase-2 (COX2) (Fig. 3b). Eight cases expressed S-100 protein while the malignant case was only focally positive (Fig. 3c). All nine cases were negative for keratin and epithelial membrane antibody (EMA) including the metastatic tumor in the liver (Fig. 3d). Of the eight cases without metastasis, the median Ki67 labeling index was 4% (range: 2-8%), but for the malignant cases, the median was 36% (range: 34-38%). Eight cases expressed p53 with variable intensity and percentage, probably suggesting no consistent mutation, and one case was negative for p53.

Outcome analysis

Follow-up data were obtained in nine cases with a median of 50 months (range, 28-83 months). There was no recurrence or tumor-related death during follow-up except for the patient with liver metastasis.

Ultrastructural findings

The case with liver metastasis was observed under electron microscopy. Neurosecretory granules were observed in the cytoplasm. The dense-core granules tended to be round and regular and were usually 100-250 nm in diameter. Other organelles such as ribosomes and rough endoplasmic reticulum were also observed in the cytoplasm. Intercellular junctions were unremarkable (Fig. 3f).

Discussion

Paragangliomas of the urinary bladder are rare neoplasms constituting less than 0.06% of all bladder neoplasm and 10% of extra-adrenal paragangliomas (Murphy et al., 2004). It is important to recognize this rare tumor of urinary bladder clinicopathologically, both to treat it effectively and to minimize pre- and peri-operative hypertensive crisis. They are classified as borderline tumors in the current WHO classification (Davis, 2004) . However, the biological behavior varies considerably, some cases never show recurrence or metastasis while others are at high risk of both (Davis, 2004). Malignant paragangliomas (usually defined by metastasis) occur in 10-15% bladder paragangliomas Davis, 2004). Most clinicians and pathologists have little experience with the diagnosis and treatment of paragangliomas of the urinary bladder (Martucci and Pacak, 2014). Unfortunately, there are no consensus clinicopathologic features or scoring system predicting the outcomes of paraganglioma of the urinary bladder. Therefore, this study summarizes histopathologic

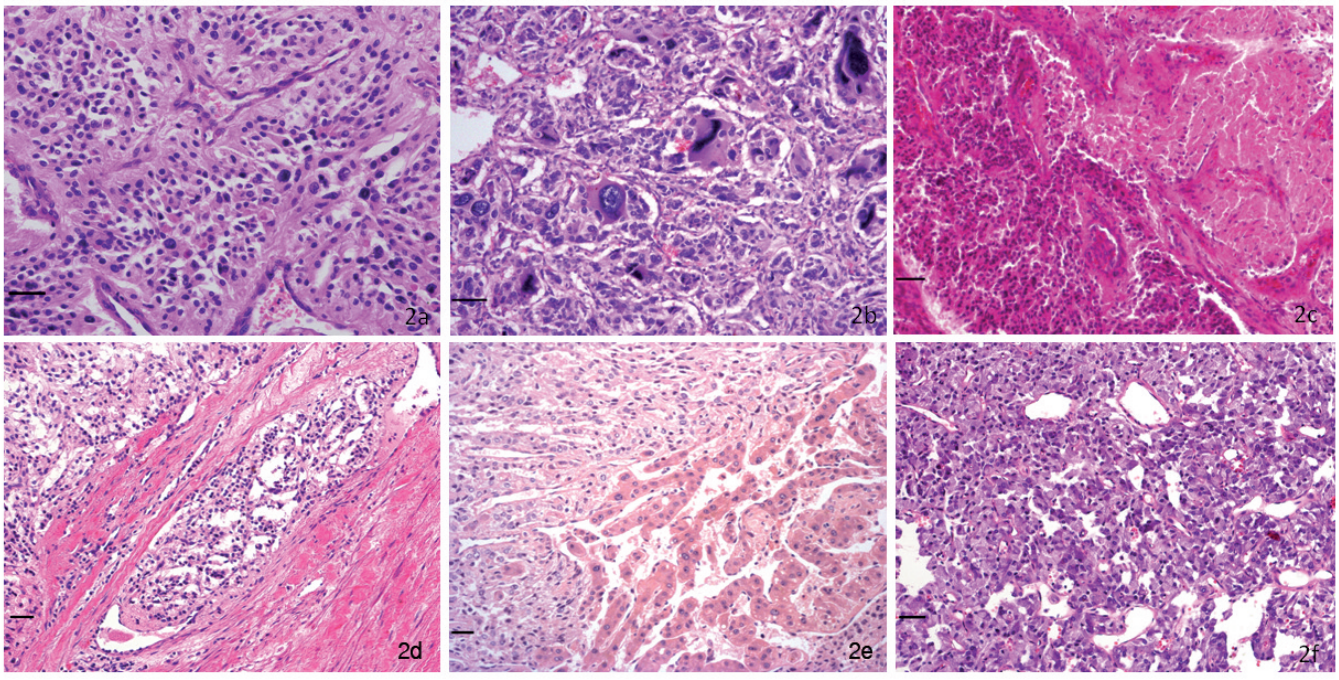

Fig. 2. Microscopic view of the malignant paraganglioma with liver metastasis **(a-e)**. The predominant histologic pattern was of large nests **(a)**. The nuclei were hyperchromatic with prominent nuclear pleomorphism and atypical mitoses **(b)**. Multiple foci of necrosis were easily observed **(c)**. Vascular invasion defined by intravascular attached tumor thrombi was evident **(d)**. The histopathologic features of the tumor in the liver was similar to that of urinary bladder **(e)**. Microscopic view of the paraganglioma without metastasis showing typical small zellballen with basophilic cytoplasm **(f)**. Scale bars: a, 12.5 μm; b, d, f, 25 μm; c, e, 50 μm.

features which might help to predict malignancy by comparing the clinicopathologic features of malignant cases to those without metastasis, and reviewing malignant paragangliomas of the urinary bladder reported in previous literature.

It is commonly accepted that the biological behavior of paraganglioma cannot be predicted on the basis of macroscopic or microscopic features alone. Indeed, there are no completely reliable histopathological criteria for differentiating malignant paragangliomas of the urinary bladder from benign cases (Cheng et al., 2000). However, we think that the malignancy should not be based solely on the development of metastasis. It is possible that there are some clinicopathologic features which may help to predict which neoplasms are more likely to behave aggressively. Thompson suggested that pheochromocytoma of the adrenal gland scaled score (PASS) weighted for these specific histologic features can be used to separate tumors with a potential for a biologically aggressive behavior (PASS≥4) from tumors that behave in a benign fashion (PASS<4) (Thompson, 2002). The pathologic features that are incorporated into the PASS correctly identified tumors with a more aggressive biological behavior (Thompson, 2002). Application of these criteria to a large cohort of cases will help to elucidate the accuracy of this grading system in clinical practice, and Salmenkivi et al. reported that all malignant (metastasized) pheochromocytomas displayed at least one histopathologically suspicious feature, i.e. confluent necrosis, over 5 mitoses/10 HPF, vascular or capsular invasion. The authors suggest that these tumors are not malignant for certain, but need closer follow-up than benign tumors (Salmenkivi et al., 2004). While PASS applies only to pheochromocytomas, recently, Kimura et al. suggested a scoring system for the grading of adrenal pheochromocytoma and extra-adrenal paraganglioma (GAPP) (Kimura et al., 2014). There are four features common to PASS and GAPP: tumor necrosis, high cellularity (a reflection of high nuclear-cytoplasmic ratio), vascular invasion, and large nest size or diffuse pattern (Tischler and de Krijger, 2015). The malignant tumor we present in this study not only displayed the common four histologic features of the PASS and GAPP scoring systems including vascular invasion, large nest size and diffuse architecture, necrosis and high cellularity, but also other features described in malignant adrenal pheochromocytomas: focal tumor cell spindling, monotony, hyperchromasia nuclei with occasional nuclear pleomorphism, mitotic counts >10 per 10 high power fields with atypical mitotic figures, and diffuse infiltrative growth between smooth muscle of the urinary bladder. Vascular invasion was also observed. The eight cases without metastasis, however, did not display the common four histologic features of the PASS and GAPP scoring systems, the typical zellballen were usually small, and there were neither tumor necrosis nor vascular invasion. Although the eight paragangliomas without metastasis might have grown between bundles of smooth muscles in the wall of bladder, the tumors were well-demarcated without a diffuse infiltrative growth pattern. No other histologic features suggested malignancy in these cases either. So, we think that the histologic features listed in the PASS and GAPP scoring system, especially the four common

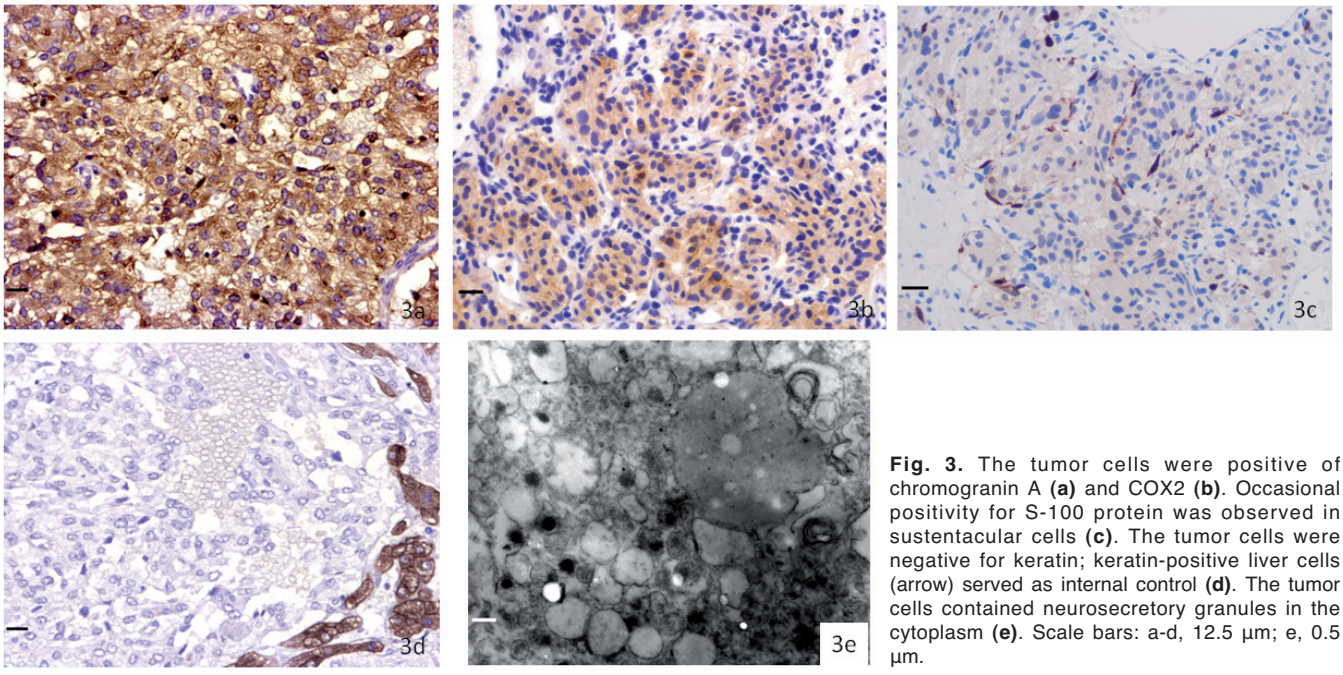

Fig. 3. The tumor cells were positive of chromogranin A **(a)** and COX2 **(b)**. Occasional positivity for S-100 protein was observed in sustentacular cells **(c)**. The tumor cells were negative for keratin; keratin-positive liver cells (arrow) served as internal control **(d)**. The tumor cells contained neurosecretory granules in the cytoplasm **(e)**. Scale bars: a-d, 12.5 µm; e, 0.5 µm.

features in both systems, may suggest the malignancy of the paraganglioma of the urinary bladder.

Immunohistochemical staining was done to find if there are any immunophenotypical features which might be useful in predicting malignancy. Pheochromocytoma cells were surrounded by sustentacular cells which are S-100 protein positive. Loss of S-100 protein positivity has been reported to correlate with unfavorable prognosis in adrenal (Unger et al., 1991) and extra-adrenal pheochromocytomas (Montresor et al., 1994). The malignant tumor we present here was only focally positive for S-100 while the eight cases without metastasis consistently expressed S-100 protein in the sustentacular cells. It is indicated that the development of large cell nests rather than the small, well-formed zellballen structures is also indicative of malignant behavior.

Cyclooxygenase (COX) is the key enzyme in the conversion of arachidonic acid to prostaglandins. Salmenkivi et al. reviewed that normal adrenal medulla does not express COX-2 immunohistochemically; most benign adrenal pheochromocytomas were negative or only weakly immunopositive for COX-2, whereas malignant tumors exhibited strongly enhanced COX-2 immunopositivity (Salmenkivi et al., 2004). The primary tumor of the malignant paraganglioma of the urinary bladder and the metastasis in the liver had strong COX-2 expression in the present series. However, 7/8 (87.5%) of paragangliomas without metastasis also expressed COX-2. According to our results, COX-2 expression in the paraganglioma of the urinary bladder fails to predict malignancy.

Some survey series have generally supported the prognostic utility of Ki67 in adrenal pheochromocytomas. In research concerning material of 105 pheochromocytomas, six out of seven malignant tumors had over 6% immunohistochemical positivity for Ki-67. Also, all benign adrenally located pheochromocytomas had at most 5% immunopositivity for Ki-67 (Salmenkivi et al., 2004). And according to their investigations, extraadrenally located tumors have a higher and more heterogeneous proliferative index than adrenally located ones (Salmenkivi et al., 2004). There was study suggested that Ki67 positivity and c-erbB-2 expression can be used as immunohistochemical markers for predicting the malignant behavior of pheochromocytoma (Tavangar et al., 2010). However, a study focused only on the urinary bladder paraganglioma found that no histologic criteria, including Ki67 labeling, could distinguish benign from malignant paraganglioma (Cheng et al., 2000). It was reported that metastases can develop in the absence of rapid multiplication of the tumor cells (Cheng et al., 2000). The malignant case we present here showed Ki67 labeling index as high as 36% and mitotic figures >10 per 10 high power fields, while the average proliferative index of Ki67 in the eight cases without metastasis was 4% (range from 2% to 8%). Our results suggest that an extraordinarily high proliferative index at least suggests rapid proliferation of the tumor. While the presence of aneuploidy has been shown to be a predictor of malignant behavior in adrenal pheochromocytomas, other studies illustrate that DNA ploidy cannot be used as a diagnostic criterion for malignancy in urinary bladder paraganglioma (Tokuno et al., 1990). According to the literature, abnormalities in vascular architecture and marked expression of VEGF in the tumor cells may be regarded as prognostic signs to predict metastasis (Kovacs et al., 2005).

A review of the literature of maliagnant paraganglioma of the urinary bladder reveals that malignant paragangliomas of urinary bladder arise in younger to middle aged adults, have a predilection for males over females and commonly present with the prototypical catecholamine crisis symptoms (Beilan et al., 2013; Feng et al., 2013; Ranaweera and Chung, 2014; Henderson et al., 2015; Quist et al., 2015), while size is not correlated with likelihood of metastasis. Furthermore, in one particular study, only patients with tumor of advanced stage ≥T3 (tumor invading perivesical tissue) were found to be at risk of recurrence or metastasis, while patients with T1 or T2 disease had favorable outcome after complete tumor resection (Cheng et al., 2000). However, in our series, the malignant tumor with liver metastasis is T2b which infiltrates the deep layer of the bladder muscle but shows a diffusely infiltrative growth pattern, so we think the diffuse infiltration of the smooth muscle of the urinary bladder probably suggests malignancy.

In conclusion, diagnosis and treatment of paraganglioma of the urinary bladder remains controversial. At present, it is not histologically feasible to differentiate benign from malignant paragangliomas. However, malignancy of the urinary bladder should not be based solely on the development of metastasis. It is possible that there are some clinicopathologic features that might help to predict which neoplasms are more likely to behave in a clinically aggressive manner. We would emphasize the predictive potential of the histologic features in this study does not provide sufficient data. However, paragangliomas of the urinary bladder which occur in younger males have features such as large nests with necrosis, vascular invasion, a very high rate of proliferation and loss reactivity for S100, all of which suggest malignancy. It is important to recognize this rare tumor of urinary bladder clinicopathologically, both to treat it effectively and to minimize pre- and peri-operative hypertensive crisis.

References

Ansari M.S., Goel A., Goel S., Durairajan L.N. and Seth A. (2001). Malignant paraganglioma of the urinary bladder. A case report. Int. Urol. Nephrol. 33, 343-345.

Beilan J.A., Lawton A., Hajdenberg J. and Rosser C.J. (2013). Pheochromocytoma of the urinary bladder: a systematic review of the contemporary literature. BMC Urol. 13, 22.

Cheng L., Leibovich B.C., Cheville J.C., Ramnani D.M., Sebo T.J., Neumann R.M., Nascimento A.G., Zincke H. and Bostwick D.G.

(2000). Paraganglioma of the urinary bladder: can biologic potential be predicted?. Cancer. 88, 844-852.

Das S. and Lowe P. (1980). Malignant pheochromocytoma of the bladder. J. Urol. 123, 282-284.

Davis C.J. (2004). Paraganglioma. In: Tumours of the urinary system and male genital organs. Eble J.N., Sauter G., Epstein J. and Sesterhem I.A. (eds). IARC press. Lyon. Pp 136-137.

Feng N., Li X., Gao H.D., Liu Z.L., Shi L.J. and Liu W.Z. (2013). Urinary bladder malignant paraganglioma with vertebral metastasis: a case report with literature review. Chin. J. Cancer 32, 624-628.

Grignon D.J., Ro J.Y., Mackay B., Ordóñez N.G., el-Naggar A., Molina T.J., Shum D.T. and Ayala A.G. (1991). Paraganglioma of the urinary bladder: Immunohistochemical, ultrastructural, and DNA flow cytometric studies. Hum. Pathol. 22, 1162-1169.

Henderson S.J., Kearns P.J., Tong C.M., Reddy M., Khurgin J., Bickell M., Miick R., Ginsberg P. and Metro M.J. (2015). Patients with urinary bladder paragangliomas: a compiled case series from a literature review for clinical management. Urology 85, e25-29.

Huang K.H., Chung S.D., Chen S.C., Chueh S.C., Pu Y.S., Lai M.K. and Lin W.C. (2007). Clinical and pathological data of 10 malignant pheochromocytomas: Long-term follow up in a single institute. International. J. Urol. 14,181-185.

Kairi-Vassilatou E., Argeitis J., Nika H., Grapsa D., Smyrniotis V. and Kondi-Pafiti A. (2007). A malignant paraganglioma of the urinary bladder in a 44-year-old female: clinicopathological and immunohistochemical study of a rare entity and literature review. Eur. J. Gynaecol. Oncol. 28, 149-151.

Kato H., Suzuki M., Mukai M. and Aizawa S. (1999). Clinicopathological study of pheochromocytoma of the urinary bladder: Immunohitochemical, flow cytometric and ultrastructural findings with review of literatures. Pathol. Int. 49, 1093-1099.

Kimura N., Takayanagi R., Takizawa N., Itagaki E., Katabami T., Kakoi N., Rakugi H., Ikeda Y., Tanabe A., Nigawara T., Ito S., Kimura I. and Naruse M, Phaeochromocytoma Study Group in Japan. collaborators (2014). Pathological grading for predicting metastasis in phaeochromocytoma and paraganglioma. Endocr. Relat. Cancer 21, 405-414.

Martucci V.L. and Pacak K. (2014). Pheochromocytoma and paraganglioma: Diagnosis, genetics, management, and treatment. Curr. Probl. Cancer 38, 7-41.

Kovacs K., Bell D., Gardiner G.W., Honey R.J., Goguen J. and Rotondo F. (2005). Malignant paraganglioma of the urinary bladder: immunohistochemical study of prognostic indicators. Endocr. Pathol. 16, 363-370.

Michel F., Gattegno B., Sicard J.F., Roland J. and Thibault P. (1990). Report of a case of malignant vesical pheochromocytoma. Diagnosis and therapy. Ann. Urol. 24, 396-399.

Montresor E., Iacono C., Nifosi F., Zanza A., Modena S., Zamboni G., Bernardello F. and Serio G. (1994). Retroperitoneal paragangliomas: role of immunohistochemistry in the diagnosis of malignancy and in assessment of prognosis. Eur. J. Surg. 160, 547-452.

Murphy W.M., Grignon D.J. and Perlman E.J. (2004). Tumors of the kidney, bladder and related urinary structures. In: AFIP atlas of the American REGISTRY of pathology. Rosai J. and Sobin L.H. (eds), Washington, DC. pp 272-273.

Pang L.C. and Tsao K.C. (1993). Flow cytometric DNA analysis for the determination of malignant potential in adrenal and extra-adrenal pheochromocytomas or paragangliomas. Arch. Pathol. Lab. Med. 117, 1142-1147.

Patnayak R., Preetam P., Jena A., Satyanarayana, Subramanian S. and Lakshmi A.Y. (2015). Space occupying lesion (paraganglioma) of the urinary bladder in a young male: A case report and brief review of the literature. J. Cancer Res. Ther. 11, 1039.

Piedrola G., Lopez E., Rueda M.D., Lopez R., Serrano J. and Sancho M. (1997). Malignant pheochromocytoma of the bladder: current controversies. Eur. Urol. 31, 122-125.

Quist E.E., Javadzadeh B.M., Johannesen E., Johansson S.L., Lele S.M. and Kozel J.A. (2015). Malignant paraganglioma of the bladder: A case report and review of the literature. Pathol. Res. Pract. 211, 183-188.

Ranaweera M. and Chung E. (2014). Bladder paraganglioma: A report of case series and critical review of current literature. World. J. Clin. Cases. 2, 591-592.

Salanitri J., Smith P. and Schlicht S. (2001). Multifocal malignant extra-adrenal paragangliomas of the Organ of Zuckerkandl and urinary bladder. Australas. Radiol. 45, 229-232.

Salmenkivi K., Heikkila P., Haglund C. and Arola J. (2004). Malignancy in pheochromocytomas. APMIS 112, 551-559.

Segawa N. and Osafune T. (2005). Malignant pheochromocytoma of the urinary bladder. Hinyokika. Kiyo. 51. 291-296.

Tavangar S.M., Shojaee A., Moradi Tabriz H., Haghpanah V., Larijani B., Heshmat R., Lashkari A. and Azimi S. (2010). Immunohistochemical expression of Ki67, c-erbB-2, and c-kit antigens in benign and malignant pheochromocytoma. Pathol. Res. Pract. 206, 305-309.

Thompson L.D.R. (2002). Pheochromocytoma of the Adrenal Gland ScaledScore (PASS) to separate benign from malignant neoplasms, a clinicopathologic and immunophenotypic study of 100 cases. Am. J. Surg. Pathol. 26, 551-566.

Tischler A.S. and de Krijger R.R. (2015). Pathology of pheochromocytoma and paraganglioma. Endocr. Relat. Cancer 22, T123-133.

Tokuno H., Kihara Y. and Arai Y. (1990). Primary paraganglioma of the urinary bladder: a report of two cases. Hinyokika. Kiyo. 36, 691-696.

Unger P., Hoffman K., Pertsemlidis D., Thung S., Wolfe S. and Kaneko M. (1991). S-100 protein-positive ustentacular cells in malignant and locally aggressive adrenal pheochromocytomas. Arch. Pathol. Lab. Med. 115, 484-487.

Yadav R., Das A.K. and Kumar R. (2007). Malignant non-functional paraganglioma of the bladder presenting with azotemia. Int. Urol. Nephrol. 39, 449-451.

Yoshida S., Nakagomi K., Goto S. and Kobayashi S. (2004). Malignant pheochromocytoma of the urinary bladder: effectiveness of radiotherapy in conjunction with chemotherapy. Int. J. Urol. 11, 175-177.

Accepted March 10, 2016