Small cell carcinoma of the urinary bladder: KIT and PDGFRA gene mutations

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Abstract

Primary small cell carcinoma of the urinary bladder is very rare. A 72-year-old was admitted to our hospital because of hematuria and dysuria. Cystoscopy revealed a bladder full of multiple, solid and papillary tumors. Biopsies from the deep and papillary tumors were taken. Histologically, tumor was pure small cell carcinoma. Immunohistochemically, the tumor cells were positive for cytokeratin, chromogranin, synaptophysin, neuron-specific enolase, CD56, CD117 and Ki67 (labeling 70%). The tumor cells were negative for CK7, CK20, CD3, CD20, LCA, CDX2, uroplakin, thyroid transcription factor 1, PSA and p63. The present urinary bladder tumor was small cell carcinoma histologically. Metastatic workup was performed an no primary or metastatic lung lesions were noted. Due to the clinical, radiologic and immunohistochemical findings, the patient was diagnosed as primary small cell carcinoma of bladder. A molecular genetic analysis for KIT (exons 9, 11, 13 and 17) and PDGFRA (exons 12 and 18) genes was performed, in paraffin micro dissection specimens, by the PCR-direct sequencing method. According to the sequencing analyses, two mutations were found at positions 558 (p.K558N) and 562 (p.E562D) in KIT gene exon 11 in our case. The other hand the same case presented two mutations in PDGFRA gene exon 14 at position 631 (p.F631A) and 638 (p.E638Q:639insC). The disease process was fulminating and the patient was lost due to several complications prior to any chemotherapy.

Introduction

Extrapulmonary small cell carcinoma (EPSCC) is a very rare malignant disease and comprises only 2.5% of all small cell carcinomas.1,2 Primary small cell carcinoma of the urinary bladder (SCCB), as EPSCC, is again a very rare carcinoma and accounts for less than 1% of all urinary bladder carcinomas.3,4 Since the first reported case published in 1981, only a few studies have been reported in the English literature.5-10 The disease affects mostly elderly men presenting with gross hematuria. The prognosis is usually poor due to the aggressive clinical course of SCCB with early dissemination and frequent recurrence. Therefore, knowledge of its presenting symptoms, appropriate treatment modality and predictors for survival are limited. Both immunoprofile and overall immunohistochemical studies of SCCB have not been reported. KIT protein is expressed in a significant percentage of pulmonary SCC, whereas KIT gene mutations are not present.11-15 Platelet derived growth factor receptor A (PDGFRα) protein expression of the pulmonary SCC has not been reported in immunohistochemical studies of these tumors. PDGFRA gene mutation has been only investigated in one study and was not shown to be mutated.16 It has been reported that there was no KIT and PDGFRA protein expressions. The gene mutations have been reported in two studies of the SSCBs.9,10

We have reported a case of primary small cell carcinoma of the urinary bladder with immunohistochemical studies with an emphasis on KIT and PDGFRA gene mutations.

Case Report

A 72-year-old man was admitted to our hospital because of hematuria and dysuria. Cystoscopy revealed a bladder full of multiple, solid and papillary tumors which were non-resectable in one session. Biopsies from the deep and papillary tumors were taken. Patient had an open prostatectomy and cystolithotomy 9 months before the diagnosis of bladder cancer and cystoscopy had revealed normal mucosal findings.

The pathology specimen was 4 cc and composed of irregular shaped, pale pink materials. The specimen had hemorrhagic fragments. The entire specimen was examined. Sections stained with hematoxylin and eosin showed packed cells with scant cytoplasm morphologically. Tumor was composed of pure small, round malignant carcinomas cells with hyperchromatic round to oval nuclei (Figure 1A), inconspicuous nucleoli, molded nuclei, and increased nucleo-cytoplasmic ratio. The mitotic rate was high. There were tumor necrosis, crush artifacts (Azurophary effect) and also vascular invasion. Some muscle fragments were infiltrated by tumor cells (Figure 1B). There was normal urothelium in the surface of some tumor areas. Additionally, there was a fragment with squamoid epithelium next to the tumor cells. Immunohistochemically, the tumor cells were positive for cytokeratin, chromogranin, synaptophysin, neuron-specific enolase (NSE), CD56, CD117 (Figure 1C) and CDX2, uroplakin, thyroid transcription factor 1 (TTF1), PSA and p63. The present urinary bladder tumor was small cell carcinoma histologically. Metastatic workup including chest radiograph and bone scan was negative. No primary or metastatic lung lesions were noted. Due to the clinical, radiologic and immunohistochemical findings, the patient was diagnosed as primary small cell carcinoma of bladder.

Radical cystectomy could not be done after the pathologic examination was complete due to the general health status of the patient. He experienced deep venous thrombosis and pulmonary infections during follow up. The disease process was fulminating and the patient was lost due to thromboembolic and pulmonary complications prior to any chemotherapy.

A molecular genetic analysis for KIT (exons 9, 11, 13 and 17) and PDGFRA (exons 12 and 18) genes was performed, in paraffin micro dissection specimens, by the PCR-direct sequencing method (GeXP Genetic Analysis System, Beckman Coulter, Brea, CA, USA), as previously described.17

Discussion

Although the EPSCC can present in various
organs, including the esophagus, stomach, pancreas, gallbladder, uterine cervix, urinary bladder, kidney and prostate, the most common site of EPSCC is the genitourinary tract. The SCCs of the genitourinary tract usually occur in the bladder.\textsuperscript{18} The diagnosis of SCCB is mainly accomplished via histopathological examination of specimens obtained by cystoscopy and/or TUR-BT. Because SCCB are identical to SCC of the pulmonary in histopathological examination, the diagnosis of SCCB is based on the criteria established by the WHO classification system on 2004. The SCCB must be differentiated from direct invasion of the bladder by SCC of the prostate, the metastatic SCC from another source (usually from the lung) and primary lymphomas of the bladder. Metastatic workup (chest radiograph and bone scan) was extremely helpful in establishing the diagnosis which was accomplished after the histopathological diagnosis for our case and has revealed no primary site other than the bladder or any metastatic lesions. The serum PSA level was unremarkable. There has been no comprehensive immunohistochemical study performed on the SCCB, although the immunohistochemical examinations were helpful in establishing the diagnosis. Likewise, the CK immunoprofile of the SCCB has not been clearly reported. The immunoprofile for our patient was positive for chromogranin, CD117 and Ki67 (labeling=70%), whereas it was negative for CK7, CK20, CD3, CD20, LCA, CDX2, uroplakin, PSA, TTF-1 and p63. Neuroendocrine antigens (chromogranin, synaptophysin, NSE) were all positive, indicating that the present tumor is small cell neuroendocrine carcinoma. The p63 was also negative, and this indicated that the present tumor was not an urothelial carcinoma. The CD3, CD20 and LCA were found out to be negative, which implied that the tumor cells were not hematopoietic cells. Therefore, the presented case was considered as primary small cell carcinoma of the bladder.

\textit{KIT} and \textit{PDGFRA} are mapped to 4q12, and encode the receptor for tyrosine oncproteins called \textit{KIT} (CD117) and \textit{PDGFRA}. These oncproteins are transmembranous oncproteins involved in tumorigenesis of particularly the gastrointestinal stromal tumors. \textit{KIT} is also expressed in malignant melanomas, germ cell tumors, hematopoietic malignancies and pulmonary SCC. But, the \textit{KIT} protein expression markedly varies in pulmonary SCC. Also, in the studies of the pulmonary SCCs, it is mostly reported that there has been no \textit{KIT} mutations. The present case showed KIT expression and mutations of the \textit{KIT} gene (exon 11). KIT protein expression was reported in large series of SCCs, in contrast to PDGFRA protein expression. Terada has investigated the presence of KIT and PDGFRA expression and mutations in SCCB.\textsuperscript{10-16} Sihto et al. has investigated \textit{KIT} and

\textbf{Figure 1.} A) Proliferation comprised small cells with hyperchromatic nuclei and scant cytoplasm. B) Some muscle fragments infiltrated by small monomorphic cells with hyperchromatic nuclei (Hematoxylin & Eosin 100×). C) The tumor cells are positive for CD117 (immunostaining 100×).
PDGFRA gene mutations and KIT amplifications in human solid tumors. They found no PDGFRA mutations in 31 SCLC. Therefore, the prognostic role of KIT protein positivity and KIT gene mutations or amplification is controversial. Also, much more studies are waiting to be done in the PDGFRA expression and the PDGFRA gene mutations in small cell carcinomas.

There is no standard treatment for the disease and treatment options vary greatly in the literature, due to the rarity and the poor prognosis of the SCCB. The improvement in survival may rely on the identification of new molecular markers for novel targeted therapies. But, molecular alterations in SCCB have already been the subject of few studies. A significant proportion of the SCCs have expressed the c-kit, which has suggested that it may be useful as a therapeutic target in the future. Yet, the SCCBs having the KIT and the PDGFRA mutations have been reported only 2 times so far. According to the sequencing analyses, two mutations were found at positions 558 (p.K558N) and 562 (p.E562D) in KIT gene exon 11 in our case. The other hand the same case presented two mutations in PDGFRA gene exon 14 at position 631 (p.P631A) and 638 (p.638Q_639AinsC).

Recently, overexpression of several receptors, such as the vascular endothelial growth factor receptor (VEGFR), the epidermal growth factor receptor (EGFR), the PDGFR and the fibroblast growth factor receptor (FGFR) have been evaluated on tumor cells to target new molecules in signaling pathways of pulmonary SCC. In preliminary studies, it is clearly seen that the most promising strategy is the targeting of angiogenesis. According to analog pulmonary SCC of SCCBs, the role of these targeting molecules in SCCB should be defined in the future.

Conclusions

The primary SCCB is a rare and aggressive tumor which is mostly diagnosed at advanced stages, and therefore the standard treatment regimens have not been well established yet. Diagnosis of SCC is mainly accomplished via histopathological examination. Primary SCCB must be differentiated from metastatic SCCs. The clinical and radiologic workup to investigate the presence of metastasis must be performed. Immunohistochemistry plays a major role in the diagnosis using the markers for neuroendocrine tumors. The treatment modalities vary widely in the literature because of the shortage of previous experience. Identification of new molecular markers for early diagnosis and novel targeted therapies may play an important role in the future. Furthermore, prospective studies will be needed to elucidate a more effective treatment modality for SCCB.

References