

Case Report

Coexistent loss of INI1 and BRG1 expression in a rhabdoid renal cell carcinoma (RCC): implications for a possible role of SWI/SNF complex in the pathogenesis of RCC

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Received January 19, 2014; Accepted February 23, 2014; Epub March 15, 2014; Published April 1, 2014

Abstract: In this study, we analyzed the immunohistochemical and molecular profiles of an unusual RCC showed coexistent absence of INI1 and BRG1 expression, rhabdoid morphology, and poor prognosis. Histologically, the tumor had rhabdoid features, which were demonstrated by large round to polygonal cells with eccentric nuclei, prominent nucleoli, and eosinophilic cytoplasm varying from abundant to scanty. Immunohistochemically, the tumor were positive for BRM, PBRM1, ARID1A, CD10, CKpan, Vimentin, carbonic anhydrase IX (CA-IX), and P504S (AMACR) but negative for INI1, BRG1, HMB45, melan A, CK7, CD117, Ksp-cadherin, TFEB, TFE3, and Cathepsin K. We detected all three exons status of the *VHL* gene of the tumor and observed 1 somatic mutations in 1st exon. Chromosome 3p deletion, coupled with polysomy of chromosome 3 was also found. Based on these findings, it is further indicated that in some cases, rhabdoid RCC may arise from clear cell RCC. SWI/SNF chromatin remodeling complex may be an attractive candidate for being the "second hit" in RCCs and may play an important role during tumor progression. The role of SWI/SNF complex in rhabdoid RCC should be further studied on a larger number of cases.

Keywords: INI1, SMARCB1, BRG1, SMARCA4, rhabdoid, renal cell carcinoma, SWI/SNF complex, immunohistochemistry

Introduction

Rhabdoid renal cell carcinomas (RCC) are rare and have been recently identified as a morphologic variant of RCC associated with aggressive behavior [1-11]. In a previous study, Gokden and colleagues found that 5% of RCC exhibited rhabdoid features and most of them were found to have a nonrhabdoid carcinoma component, in most cases described as clear cell RCC [1]. Although there are approximately 60 reported cases of RCC with rhabdoid morphology in the English literature, the potential molecular connection associated with the rhabdoid cytologic phenotype and the aggressive biologic behavior of these tumors have not yet been elucidated [1-11].

The yeast switch in mating type (SWI)/sucrose nonfermentation (SNF) complex is one of sev-

eral chromatin-remodeling complexes, including the INI1 (also known as SMARCB1, SNF5 and BAF47), ARID1A (also known as BAF250A and SMARCF1), PBRM1 (also known as BAF180) and BRM (also known as SMARCA2)/BRG1 (also known as SMARCA4) subunits [12, 13]. It has been shown that the SWI/SNF complex plays critical roles for growth control and cancer development and complete loss of a SWI/SNF subunit can promote cancer formation [12, 13]. In previous studies, loss of INI1 or BRG1 expression has been described in malignant tumors with rhabdoid morphology including pediatric renal and extrarenal malignant rhabdoid tumors, atypical teratoid/rhabdoid tumors of the central nervous system, epithelioid sarcoma, renal medullary carcinoma and a subset (15%) of renal collecting duct carcinoma, suggesting that these protein act an important role in human cancer [12-16].

In this study, we reported a case of rhabdoid RCC showing coexistent loss of INI1 and BRG1 expression that implicate a possible role of SWI/SNF complex in the biological mechanisms driving these tumors.

Case report

Clinical history

A 65-year-old man with no significant past medical history presented with 3 months history of intermittent lumbar pain. Abdominal computed tomography (CT) scan demonstrated a 5.5×4 cm sized mass in the upper pole of the left kidney. A malignant tumor was suspected and a total nephrectomy was performed without chemotherapy or radiation therapy after surgery. The patient died of the disease 1 year after diagnosis.

Histopathological and immunohistochemical findings

Morphologically, the tumor displayed all areas with a rhabdoid histologic appearance, which were demonstrated by large round to polygonal cells with eccentric nuclei, prominent nucleoli, and eosinophilic cytoplasm varying from abundant to scanty. Densely eosinophilic nuclear pseudoinclusions were noted. These cells were arranged partly in an alveolar arrangement with delicate fibrovascular septa, compared with classic clear cell RCC with a delicate sinusoidal vascular network (**Figure 1A** and **1B**).

Immunoreaction was performed using the labelled streptavidin–biotin method and overnight incubation as previously described [17, 18]. Immunohistochemically, the tumor cells demonstrated moderately (2+) or strongly (3+) positive staining for BRM, PBRM1, ARID1A, CD10, CKpan, Vimentin, carbonic anhydrase IX (CA-IX), and P504S (AMACR) but negative for INI1, BRG1, HMB45, melan A, CK7, CD117, Ksp-cadherin, TFE3, and Cathepsin K. The presence of Ki-67 protein demonstrated a high proliferation rate (**Figure 1C-E**).

Molecular analysis

VHL sequence analysis of *VHL* gene and fluorescence in situ hybridization (FISH) detection for chromosome 3p deletion were performed as recently described [19, 20]. We detected all three exons status of the *VHL* gene and

observed 1 somatic mutation in 1st exon (**Figure 2**). The tumor demonstrated chromosome 3p deletion, coupled with polysomy of chromosome 3 (**Figure 1F**).

Discussion

We analyzed the immunohistochemical and molecular profiles of an unusual RCC showed coexistent absence of INI1 and BRG1 expression, rhabdoid morphology, and poor prognosis.

Histologically, the tumor had rhabdoid features, which were demonstrated by large round to polygonal cells with eccentric nuclei, prominent nucleoli, and eosinophilic cytoplasm varying from abundant to scanty. Densely eosinophilic nuclear pseudoinclusions were noted. These cells were arranged partly in a large alveolar arrangement with delicate fibrovascular septa, compared with classic clear cell RCC with a delicate sinusoidal vascular network. When reviewing published data with histopathologic description, RCC with rhabdoid features has been recently reported as a morphologic variant of RCC in few series and universally recognized as a highly aggressive neoplasm [1-11]. Although there are approximately 60 reported cases of RCC with rhabdoid morphology in the English literature, it is uncertain whether this subset of RCC has distinct immunophenotype, molecular genetic features and origin [1-11]. The possibility of a molecular connection between the rhabdoid cytologic phenotype and the aggressive biologic behavior has not yet been elucidated.

Immunohistochemically, the tumor were positive for BRM, PBRM1, ARID1A, CD10, CKpan, Vimentin, carbonic anhydrase IX (CA-IX), and P504S (AMACR) but negative for INI1, BRG1, HMB45, melan A, CK7, CD117, Ksp-cadherin, TFE3, and Cathepsin K. The immunophenotype of this subset is generally similar to that of clear cell RCC. At the molecular level, it was estimated that approximately 33% to 75% of all sporadic clear cell RCC harbor *VHL* defects [21-23]. Chromosome 3p deletion and the inactivation of the von Hippel-Lindau (*VHL*) tumor suppressor gene are the most common genetic alterations observed in this subtype [20, 24, 25]. We detected all three exons status of the *VHL* gene of the tumor and observed 1 somatic mutations in 1st exon. Chromosome 3p dele-

Loss of INI1 and BRG1 in rhabdoid RCC

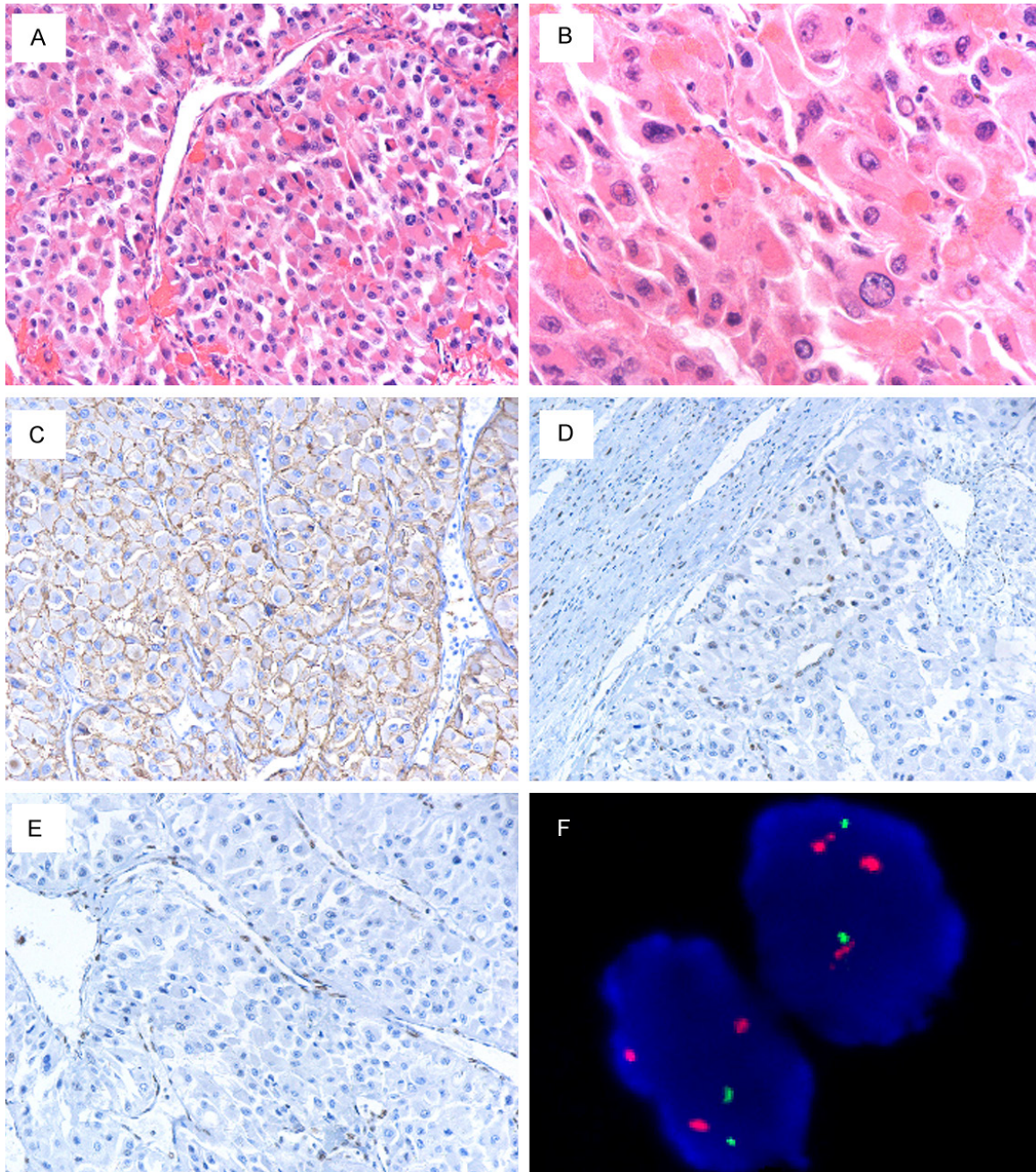


Figure 1. (A) The tumor was arranged partly in a large alveolar arrangement with delicate fibrovascular septa, compared with classic clear cell RCC with a delicate sinusoidal vascular network. (B) Neoplastic cells were large round to polygonal with eccentric nuclei, prominent nucleoli, and abundant eosinophilic cytoplasm. Densely eosinophilic nuclear pseudoinclusions were observed. (C) The tumor showed strong labeling for CA-IX. Immunostaining for INI1 (D) and BRG1 (E) was entirely negative. (F) The tumor cells showed 3p loss with polysomy of chromosome 3. FISH showed nuclei with multiple hybridization signals of centromeric probe for chromosome 3 (Spectrum Orange) and two signals of subtelomeric probe for 3p25 (Spectrum Green).

tion, coupled with polysomy of chromosome 3 was also found. These molecular findings further indicated that in some cases, rhabdoid RCC may arise from clear cell RCC. Existing evidence indicates that *VHL* inactivation is consid-

ered as a necessary but not sufficient step for clear cell RCC development and progression [26]. Indeed, exome sequencing has recently unveiled additional genes mutated in clear cell RCC. Several of those encode histone and chro-

Loss of INI1 and BRG1 in rhabdoid RCC

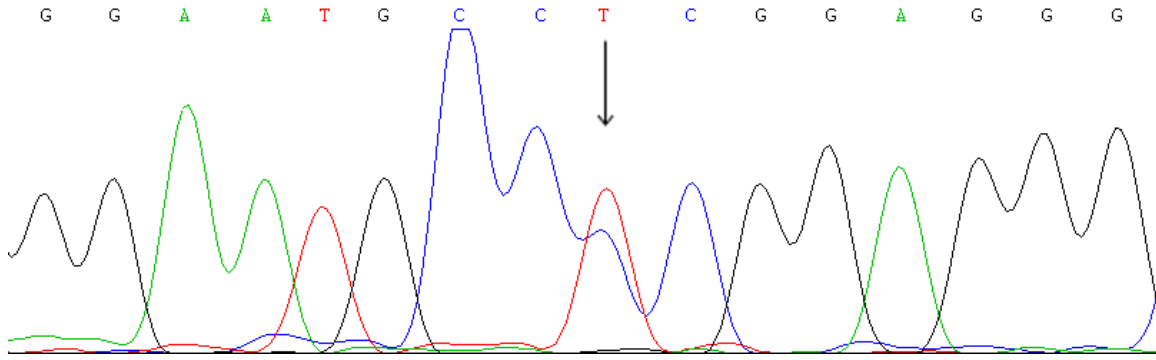


Figure 2. Analysis of the *VHL* gene mutation of the case. One synonymous mutation was identified: c.219C>T (Pro-2Pro).

matin regulators and include *SETD2*, *KDM6A*, *KDM5C*, *BAP1*, and *PBRM1* [26].

INI1 and BRG1 protein is key member of the SWI/SNF complex, which also includes other important subunits such as BRM, ARID1A, and PBRM1 and mediates gene expression by shifting the position of histones, thereby making the DNA more accessible to transcription factors and key cellular proteins [12, 13]. Several distinct tumors are associated with the loss expression of this protein family. For example, BRM has been found to be inactivated in 10-20% of many solid tumor types including lung, breast, colon, esophageal, ovarian, bladder, prostate, gastric and head/neck tumors [27]. More recently, the *ARID1A* subunit of SWI/SNF complexes was also recently found to be specifically mutated in 50% of ovarian clear cell carcinomas and 30% of endometrioid carcinomas [28, 29]. *BRG1* mutations and loss expression have been identified in primary lung cancers [13]. Mutations in *PBRM1* were identified in 41% of renal cell carcinomas, making *PBRM1* the second major clear cell RCC cancer gene [26, 30]. Moreover, loss of INI1 or BRG1 expression has been described in malignant tumors with rhabdoid morphology including pediatric renal and extrarenal malignant rhabdoid tumors, atypical teratoid/rhabdoid tumors of the central nervous system, epithelioid sarcoma, and renal medullary carcinoma [12-14, 16, 31]. In the present study, there was a complete absence of INI1 and BRG1 in the case of rhabdoid RCCs. These findings suggest a potential role of INI1 and BRG1 in the acquisition of this distinct histopathological appearance and the extremely aggressive behavior. Based on these findings, SWI/SNF chromatin remodeling com-

plex may be an attractive candidate for being the “second hit” in RCCs and may play an important role during tumor progression. The question whether genetic alterations of other members of the SWI/SNF chromatin remodeling complex might play a role in those rare cases of rhabdoid RCC remains to be determined.

In conclusion, we have reported a case of RCC with rhabdoid features showed absence of INI1 and BRG1 expression, and poor prognosis. The role of SWI/SNF complex in rhabdoid RCC should be further studied on a larger number of cases.

Acknowledgements

This work was supported by National Natural Science Foundation of China (81101933; Q Rao); and (81372743; Xiao-Jun Zhou); Natural Science Foundation of Jiangsu Province, China (BK2010463; Q Rao); and Maixin fund (m1203; Shan-Shan Shi).

Disclosure of conflict of interest

None.

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