Ependymoma with diffuse signet-ring features: report of a case and review of the literature

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Key words
Ependymoma • Signet-ring cell • Metastatic adenocarcinoma • Frozen sections • Small biopsies

Summary
Signet-ring cell ependymoma is a rare variant of ependymoma with only seven cases described in literature. Biological behavior and prognosis of this entity are not well-known until now. We present a case of a 49-year-old female with a history of headache and gait instability. Magnetic resonance imaging showed an upper cervical tumor with cystic component and mural nodule. The patient underwent surgery. Microscopically some cells displayed an eccentric nucleus compressed to the periphery by vacuolated cytoplasm. Perivascular pseudorosettes and ependymal rosettes were seen only focally. The cells were positive for glial fibrillary acidic protein and epithelial membrane antigen. The diagnosis was ependymoma with diffuse signet-ring features, grade II according to the World Health Organization. It may be difficult to diagnose this unusual variant of ependymoma especially on small biopsies or frozen sections. A complete examination of the specimen is recommended with immunohistochemical confirmation to rule out potential morphologic mimics, such as metastatic adenocarcinomas and gliomas in the differential diagnosis.

Introduction
Ependymomas are tumors with uncertain malignant potential arising from the cells that line the ventricles and central canal of the spinal cord. They account for 6% to 9% of primary central nervous system (CNS) neoplasms 1,2. The World Health Organization (WHO) 2007 classification of central nervous system tumors recognizes in this group rare histological variants: ependymoma with lipomatous differentiation, giant cell ependymoma, ependymoma with extensive tumor cell vacuolization, melanotic ependymoma, signet-ring cell ependymoma, ovarian ependymoma, ependymoma with neuropil-like islands 2. Other proposed variants are: ependymoma with condroid metaplasia, ependymosarcoma, epithelioid ependymoma, ependymoma with “granular cell” features and oncocytic ependymoma 3,7. Diagnosis of these rare variants is often difficult in the absence of the typical histological features of ependymoma (perivascular pseudorosettes, ependymal rosettes and canals) 2. We report a case of an intramedullary upper cervical cord ependymoma rich in signet-ring cells. Issues related to diagnostic problems are discussed with a review of signet-ring cell ependymomas previously described in literature.

Clinical history
A 49-year-old female was referred to the Department of Neurosurgery for a history of recurrent headache. Neurological examination revealed uncoordinated movements, gait instability, speech impairment and difficulty with eyes movements. Magnetic resonance imaging (MRI) showed an intramedullary tumor of the upper cervical cord with cystic component and mural nodule, T1-isointense and T2-hyperintense, suggesting the diagnosis of pilocytic astrocytoma (Fig. 1) 8,9. The patient underwent surgery for total tumor excision via sub-occipital craniotomy.
Materials and methods

Surgical specimen obtained was fixed in 10% formalin and embedded in paraffin. Five-micrometer sections were stained using hematoxylin-eosin, periodic acid-Schiff (PAS) and immunohistochemistry. Immunohistochemical staining were performed with GFAP (clone 6F2, dako), EMA (clone E29, dako), GATA-3 (clone L50-823, zeta corporation), TTF-1 (clone 8G7G3/1, dako), CK AE1/AE3 (clone AE1/AE3, dako), cytokeratin CAM 5.2 (clone Cam 5.2, ventana) and Ki67 (clone MIB-1, dako). All samples were processed using a “Bond Polymer Refine” detection system in an automated bond immunostainer (Vision Biosystem, Menarini, Florence, Italy). Ki67 immunoexpression was considered as low (< 1% of cells), intermediate (1-5%) and high (> 5%).

Results

Macroscopically the tumor was a soft, whitish nodule, 20 millimeter diameter. At microscopic examination, in four out of five sections the majority (> 50%) of tumor cells showed an eccentrically located nucleus compressed by clear vacuolated cytoplasm (Fig. 2A-B-C) in a glial fibrillary stroma, admixed with round cells (Fig. 2D). On special stain many vacuoles were PAS positive. Perivascular pseudorosettes and ependymal rosettes were focally seen (Fig. 2D). Necrosis and vascular proliferation were absent, with a Ki-67 labeling index of 2-3%. Immunohistochemical analysis of neoplastic cells showed an intracytoplasmic dot-like pattern positivity for EMA (Fig. 3A). About one third of the cells expressed GFAP, with minor intensity in signet-ring cell areas (Fig. 3B). No cytokeratins nor GATA-3 and TTF-1 expression was revealed. The final diagnosis was “ependymoma with signet-ring cell features”, grade II according to WHO classification 2007.

Discussion

Signet-ring cell ependymoma is a rare entity with seven cases reported in literature. Six cases out of seven occurred in women, with an age range between 2 and 58 years. The most commonly involved sites were the parieto-occipital region and the 4th ventricle. In almost all cases the preoperative radiologic workup was performed with MRI and the common finding was a nodule with solido-cystic pattern, with a ring contrast enhancement, T1-isointense and T2-hyperintense. These radiological features are however not specific and reported in different tumors such as pilocytic astrocytoma, gliomas, metastatic tumors, gangliogliomas, hemangioblastomas and cystic meningiomas. Available clinico-pathological features of the reported cases are summarized in Table I and Table II. All cases were classified as ependymoma grade II according to the 2007 WHO classification. As shown in Table I two cases were submitted to radical resection and exhibited an indolent behavior with no recurrence. Complete surgical resection represents the standard treatment for ependymomas, and should be carried out whenever possible. Adjuvant radiotherapy improves local control as well as overall survival, it is commonly used after complete resection of localized high-grade ependymoma and indicated for low- and high grade lesions where residual disease is suspected after surgery. Chemotherapy has been employed at the time of relapse and platinum-based regimens are considered the best available option. In two cases a subtotal resection followed by gamma-knife radiosurgery was performed. In the case reported by Hirato et al. the tumor recurred after one year, leading to a total resection, while in the case presented by Cenacchi et al. there was no recurrence. Follow-up information were available in two out of seven patients and the longest follow-up period was one year with no evidence of recurrence. Ependymomas are histologically characterized by the presence of perivascular pseudorosettes, ependymal rosettes and canals. The first consist of neoplastic ependymal cells with tappered processes radiating around a wall of a centrally placed vessel, in a spoke-wheel arrangement; the last two are tubule-like structures of neoplastic cells surrounding an empty lumen. These typical features easily allow to distinguish ependymomas from other
Fig. 2. Morphological features. (A, B, C) Signet-ring cells (D) Round cells with perivascular pseudorosettes.

Fig. 3. Immunohistochemical features. (A) EMA with intracytoplasmic dot-like pattern (B) GFAP with fibrillary pattern.
Tab. I. Summary of clinical features of reported cases of signet-ring ependymoma.

<table>
<thead>
<tr>
<th>Case No./ authors</th>
<th>Sex/age</th>
<th>Symptoms/signs</th>
<th>Site</th>
<th>Imaging</th>
<th>Treatment/recurrence</th>
</tr>
</thead>
<tbody>
<tr>
<td>1) Zuppan et al.</td>
<td>F/12</td>
<td>NA*</td>
<td>Left parieto-occipital region</td>
<td>NA*</td>
<td>NA*</td>
</tr>
<tr>
<td>2) Zuppan et al.</td>
<td>F/44</td>
<td>NA*</td>
<td>Fourth ventricle</td>
<td>NA*</td>
<td>NA*</td>
</tr>
<tr>
<td>3) Hirato et al.</td>
<td>F/2</td>
<td>Tonic-clonic seizure disorder</td>
<td>Left parieto-occipital region</td>
<td>1) Solid mass</td>
<td>2) Cyst, homogeneous enhancement</td>
</tr>
<tr>
<td></td>
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<td></td>
</tr>
<tr>
<td>4) Vajtai et al.</td>
<td>M/64</td>
<td>Headache, dizzy spells, gait problems, cerebellar disorders</td>
<td>Posterior fossa</td>
<td>Cyst with mural nodule, T2-h† ring enhancement</td>
<td>OP1: radical resection → no recurrence</td>
</tr>
<tr>
<td>5) Cenacchi et al.</td>
<td>F/5</td>
<td>Vomit, headache, gait problems, endocranial hypertension</td>
<td>Posterior fossa, fourth ventricle</td>
<td>Cyst, T2-h† ring enhancement</td>
<td>OP1,2: ventriculocisternostomies → ND 1 OP3: subtotal resection → gkrs§ → no recurrence</td>
</tr>
<tr>
<td>6) Mizuno et al.</td>
<td>F/58</td>
<td>Respiratory distress, absent gag reflex, movement/sensory disorders</td>
<td>Medulla oblongata</td>
<td>Cyst, T2-h† ring enhancement presence of hemorrhage</td>
<td>OP1: subtotal resection after IOFSA* revised diagnosis on definitive histology</td>
</tr>
<tr>
<td>7) Ertan et al.</td>
<td>F/35</td>
<td>Headache, nausea, bilateral mild papilla edema</td>
<td>Foramen magnum, fourth ventricle</td>
<td>Solid mass, T2-h† heterogeneous enhancement</td>
<td>OP: radical resection → no recurrence</td>
</tr>
</tbody>
</table>

* not available; † T2- h yperintense; ‡ operation; § gamma-knife radiosurgery; † diagnostic; ‡ intraoperative frozen section analysis.

Tab. II. Summary of pathological features of reported cases of signet-ring ependymoma.

<table>
<thead>
<tr>
<th>Case No./ authors</th>
<th>IOFSA*</th>
<th>H&amp;E</th>
<th>EMA</th>
<th>GFAP</th>
<th>Ki67</th>
<th>Necrosis/mitosis</th>
</tr>
</thead>
<tbody>
<tr>
<td>1) Zuppan et al.</td>
<td>NA†</td>
<td>NA†</td>
<td>NA†</td>
<td>NA†</td>
<td>NA†</td>
<td>NA†</td>
</tr>
<tr>
<td>2) Zuppan et al.</td>
<td>NA†</td>
<td>NA†</td>
<td>NA†</td>
<td>NA†</td>
<td>NA†</td>
<td>NA†</td>
</tr>
<tr>
<td>3) Hirato et al.</td>
<td>NO</td>
<td>SP1: signet-ring cells, clear cells, smaller cells, fibrillary stroma, perivascular pseudorosettes</td>
<td>Focally positive</td>
<td>Positive</td>
<td>SP1: 0,1% SP2: 0,66%</td>
<td>Not found</td>
</tr>
<tr>
<td>4) Vajtai et al.</td>
<td>NO</td>
<td>Signet-ring cells, clear cells, rudimentary perivascular pseudorosettes</td>
<td>Positive</td>
<td>Positive</td>
<td>&lt; 1%</td>
<td>Not found</td>
</tr>
<tr>
<td>5) Cenacchi et al.</td>
<td>NO</td>
<td>Signet-ring cells, clear cells, smaller cells, perivascular pseudorosettes</td>
<td>Focally positive</td>
<td>Positive</td>
<td>3%</td>
<td>Occasional mitosis necrosis absent</td>
</tr>
<tr>
<td>6) Mizuno et al.</td>
<td>YES</td>
<td>FSI: signet-ring cells with atypical nuclei SP1,2: signet-ring cells, smaller cells, perivascular pseudorosettes</td>
<td>Focally positive</td>
<td>Intensely positive</td>
<td>NA†</td>
<td>Low mitotic count necrosis absent</td>
</tr>
<tr>
<td>7) Ertan et al.</td>
<td>NO</td>
<td>Signet-ring cells, clear cells, pigmented cells, focal perivascular pseudorosettes, ependymal rosettes, rosenthal fibers</td>
<td>Positive</td>
<td>Positive</td>
<td>1%</td>
<td>Not found</td>
</tr>
</tbody>
</table>

* intraoperative frozen section analysis; † not available; † specimen; † frozen section
primary brain malignancies and metastatic ependymomas, but a correct diagnosis of rare histological ependymoma may be challenging in insufficient material or when the presence of typical features are lacking. In our case, the microscopic examination showed many aggregates of signet-ring cells intermixed with areas composed of round cells. Signet-ring features could be identified as a minor component in classical ependymomas. It may be predominant, leading to diagnostic pitfalls when small biopsies, representing just part of the lesion, are evaluated by the pathologist. In the case described by Mizuno et al. at intraoperative analysis, the tumor was first considered a metastatic adenocarcinoma with a signet-ring configuration leading to a subtotal removal of the mass. Afterwards, on permanent sections, the diagnosis was revised to a signet-ring cell ependymoma with a subsequent complete surgical tumor excision. It is therefore important to assess the whole tumor mass for a correct diagnosis looking for typical histological features and an associated immunohistochemical panel. The most important neoplasms to be differentiated from signet-ring cell ependymoma are metastatic adenocarcinomas and other primary brain tumors with a signet-ring cell pattern (glio-blastoma, astroblastoma, oligodendroglioma). GFAP and EMA are extremely helpful in this setting. Various investigators showed two distinct patterns of GFAP immunoreactivity in ependymomas: one was fibrillary and related to perivascular pseudorosettes, the other was a diffuse cytoplasmic pattern. GFAP is constantly expressed in glioblastomas and variably expressed in oligodendrogliomas and astroblastomas, while no expression was found in signet-ring adenocarcinomas. EMA is a sensitive and specific marker of ependymal differentiation. Immunoreactivity pattern may vary depending on the subtypes and on neoplastic cell differentiation: ring-like membrane reactivity, dot-like intracytoplasmic staining or packed cytoplasmic pattern may all be detected. EMA staining has also been described in meningioma, chordoid glioma, rhabdoid tumor, chordoma and signet-ring adenocarcinoma.

Conclusions

We reported an unusual case of ependymoma with signet-ring cell features. Although the reported cases are relatively few and long-term follow-up is not available for all, the cases treated with total surgical resection show a favorable prognosis like the counterpart of classical ependymoma. Signet-ring cell ependymoma may mimic signet-ring cell adenocarcinoma and other brain tumors, with the risk of a misdiagnosis, especially on small biopsies or frozen sections.

The preliminary abstract of this work has been accepted as presentation in form of poster at the ECP 2014 London meeting (August 30- September 3, 2014).
Signet-ring ependymoma


