Medulloepithelioma with peculiar clinical presentation, stem cell phenotype and aberrant DNA-methylation profile

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Practice points

- Medulloepithelioma is a rare tumor primarily arising in young children less than 5 years of age in the CNS and ciliary body of the eye.
- Can rarely present outside the CNS.
- Histomorphology resembles primitive developing neural tube.
- Previously documented to express some immunophenotypic markers of neural stem cells and other CNS tumor markers.
- More recently primitive CNS tumors ependyoblastoma, embryonal tumor with abundant neuropil and true rosettes and medulloepithelioma share similar molecular features including diffuse expression of LIN28A and similar molecular genetic features with amplification by FISH at 19q13.42 and DNA-methylation profiles demonstrating copy number variation at chromosomes 19 and 2 most commonly. This has led to the proposal that this group of tumors may be biologically the same entity and have been termed 'embryonal tumors with multilayered rosettes'.
- Poor prognosis may be related to some tumor cells expressing pluripotent stem cell phenotypes.

SUMMARY We present a 21-year-old male with a neck mass diagnosed as medulloepithelioma. Despite aggressive chemo- and radio-therapy, the tumor metastasized and proved fatal after seventeen months. The tumor demonstrated robust immunohistochemical expression of multiple markers of embryonic/neural stem cells and embryogenesis from the paraffin embedded tissue. The tumor, expressing LIN28A but negative for the 19q13.42 amplicon, also lacked the characteristic methylation profile for medulloepithelioma and other tumors with similar morphology. The expression of embryonic markers may explain its unresponsiveness to therapy and poor prognosis. Therapies targeted at embryonic cell phenotypes may hold the key for successfully treating cancers with embryonal phenotypes or tumors harboring cells with embryonal phenotypes.

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KEYWORDS

- DNA methylation
- embryonic stem cells
- ETMR
- LIN28A
- medulloepithelioma

Medulloepithelioma is a rare malignant tumor primarily of the CNS that histologically resembles the primitive neural tube. Medulloepitheliomas typically present in young children less than 5 years of age and can occur in the CNS (supra- and infra-tentorial brain, spinal cord) and peripheral CNS along nerve trunks, and commonly the ciliary body of the eye [1]. Only very rare reports of medulloepithelioma presenting in adults are found and most tumors involve the eye [2]. The prognosis for CNS medulloepithelioma is poor, although better for ciliary body tumors due to its greater ability for earlier detection and effective local treatment. Pathologically, medulloepitheliomas can show immunohistochemical expression of nestin, an intermediate filament that characterizes neural stem cell lineage and neural and glial markers including synaptophysin, neurofilament and glial fibrillary acidic protein. The differential diagnosis includes embryonal/primitive tumors of the CNS including medulloblastoma, atypical teratoid rhabdoid tumor (AT/RT), primitive neuroectodermal tumor (PNET), immature teratoma, embryonal tumor with abundant neuropil and true rosettes (ETANTR) and ependymoblastoma [1,3].

We report a case of medulloepithelioma presenting in a 21-year-old male with a neck mass. Despite aggressive multidrug chemotherapy and radiotherapy, the patient developed metastatic disease to the cervical and thoracolumbar spine and died within 17 months of presentation. Because of its histopathological appearance, we characterized it further for immunohistochemical expression of LIN28A, a marker of tumors in the medulloepithelioma spectrum termed embryonal tumors with multilayered rosetting (ETMR) as well as multiple embryonic/neural stem cell/embryogenesis markers. In addition, we performed a DNA-methylation profile and compared it with other tumors in the ETMR spectrum and other CNS tumors. This case is interesting since peripheral medulloepithelioma is a very rare tumor, presentation in adults is rare, to our knowledge, this may be the first immunophenotypic characterization from paraffin embedded tissue of a medulloepithelioma documenting diverse expression of multiple markers of embryonic stem cells (ESCs), neural stem cells (NSCs) and embryogenesis in a single tumor and the molecular profile is dissimilar to other tumors with similar morphology. In particular, embryonic/neural stem cell gene/

protein expression in tumors may have implications for pathogenesis, treatment and prognosis of this 'primitive' tumor and other tumors with embryonal phenotypes.

Case report**• Clinical**

A 21-year-old male presented to an outside hospital with a 3-month history of an expanding mass over the left posterior neck with associated left lateral neck pain, hoarseness, dysphagia and 30-lb weight loss despite antibiotics and corticosteroids for presumed lymphadenitis. A fine needle aspiration biopsy was done at an outside hospital and reviewed in consultation at our institution and given a diagnosis of 'primitive neuroepithelial cell tumor'. The patient was immediately transferred to our hospital for further evaluation and treatment. MRI demonstrated a left occipital mass, 7 cm × 4.8 cm × 4.6 cm, surrounding the left paraspinal muscle and vertebral artery, mild compression of the dural sac at C1–C3 and scalloping of the left occipital bone with T2 images showing avid enhancement. A second mass, 7 cm × 4.8 cm × 5.4 cm, was deep to and infiltrating the sternocleidomastoid muscle, displacing the paraspinal muscles and extending to the left vertebral artery (**Figure 1A**). No intracranial lesions were seen. Since no radiological evidence of direct CNS involvement was present, cerebrospinal fluid evaluation and spinal axis imaging were not pursued at this time. CT scan of the neck/chest/abdomen/pelvis demonstrated a thrombosis of the jugular vein and multiple pulmonary nodules up to 1.2 cm in greatest dimension (not shown). Pertinent laboratory workup included normal serum α -fetoprotein and human chorionic gonadotrophin and negative bone marrow biopsies.

The initial clinical presentation was concerning for airway patency and evaluation of the upper airway showed paralysis of the right vocal cord and dysfunction of the left cord. Emergent chemotherapy was started using vincristine, doxorubicin and cyclophosphamide with mesna while awaiting pathology results from an open neck biopsy (pathology findings below). The patient was initially managed on total parenteral nutrition and required a gastrostomy–jejunostomy tube. After a diagnosis of 'primitive tumor with neuroepithelial and pluripotent phenotypes' was rendered, the patient was treated per Children's Oncology Group protocol AWES0031 regimen B consisting of

Figure 1. Magnetic resonance and computed tomography of neck tumor at presentation and after treatment. (A) Coronal MRI showing two large obvious left-sided neck masses (arrows). Superior mass is a $7 \times 4.8 \times 4.6$ cm suboccipital tumor surrounding the vertebral artery on the left and pushing on the dural sac at C1–C3. The inferior mass ($7 \times 4.8 \times 5.4$ cm) infiltrates the sternocleidomastoid muscle with displacement of paraspinal muscles. A radiological differential diagnosis was not rendered. **(B)** Follow-up CT scan after chemotherapy demonstrating ill-defined 3.5×2.2 cm left suboccipital mass (between arrows). **(C & D)** MRI of the spine 14 months after initial presentation with metastases present in the intradural region of T4 (arrow in [C]) and multiple metastases in the lumbosacral cord (arrows in [D]).

etoposide and ifosfamide with mesna, alternating with vincristine, doxorubicin and cyclophosphamide with mesna, totaling 14 cycles designed to treat PNET/Ewing sarcoma. Neck CT after 1 month of treatment showed decreased size of left occipital and posterior cervical masses and lung CT after 3 months showed no evidence of lung metastases. Chemotherapy was completed 11 months after presentation and chest/abdomen/pelvis CT showed no tumor. Neck CT showed residual left suboccipital mass ~ 3.5 cm

in greatest dimension (Figure 1B). In addition, the patient was treated with localized radiation therapy to the neck/skull base with a dose of 45 Gy in 25 fractions with boost to gross residual disease with 10.8 Gy in 6 fractions, using intensity-modulated radiation therapy. The patient also received bilateral whole lung therapy with 15 Gy in 10 fractions. The patient's course was complicated by sinus venous thrombosis treated with enoxaparin and acute lung injury secondary to radiation requiring oxygen transiently.

Fourteen months after diagnosis, the patient presented with history of lower back pain 1 month earlier which then resolved, followed by bilateral leg pain which radiated down the posterior aspects of each leg and was associated with decreased sensation to the right lower extremity as well as leg weakness for two weeks. MRI showed multiple spinal metastases at T4 and lumbosacral spine extending to the cauda equina (Figure 1C & D). Neck CT showed no evidence of new nodules or lymphadenopathy. The patient was started on dexamethasone, palliative radiation and chemotherapy with temozolomide and irinotecan monthly per Children's Oncology Group protocol ACNS0821 designed to study treatment of medulloblastomas and CNS PNET.

Seventeen months after initial presentation, the patient presented to the emergency room with right arm pain/numbness and right leg weakness. Spinal cord MRI showed new extramedullary metastases at C3–C4, C7–T1, T2–T3 and T6–T9 with hemorrhage but resolution of the previously documented densities at T4, conus medullaris and cauda equina. MRI of the brain showed a possible nodular density anterior to the right pons. The patient developed progressive signs of cervical cord compression while on IV dexamethasone and supportive medications before passing away. A postmortem examination limited to the spinal cord was performed.

Pathology/molecular profile

The diagnosis from the excisional biopsy was 'primitive tumor with neuroectodermal and pluripotent cell phenotypes'. Subsequent review of the tumor present at autopsy in conjunction with reevaluation of the biopsy confirmed a diagnosis of medulloepithelioma, malignant, without teratoid features. No frozen tissue was available and insufficient paraffin-embedded tissue remained to perform gene-expression analyses. Based on the morphology, a panel of additional immunohistochemical stains were performed that included multiple markers of embryonic and NSCs, and markers noted to be expressed during embryonic development. Primary antibodies used for immunohistochemistry (IHC) are listed in Table 1. IHC was performed using standard procedures for antigen retrieval and detection. On hematoxylin and eosin sections, the tumor had tubular, papillary and trabecular architecture and 'medulloepithelial' rosettes lined by pseudostratified cells resembling neuroepithelium (Figure 2A). Germ

cell or teratomatous elements were not appreciated. Cytoplasmic apical blebs, a classic feature seen in ependymoblastomas, were not prominent. Some solid areas had cells with somewhat larger nuclei, powdery chromatin, with single small nucleoli. These cells appeared to have cytoplasmic clearing/vacuoles and numerous mitotic figures were readily identified (Figure 2B & C) particularly in regions resembling ESCs. The tumor demonstrated characteristic diffuse and apical cytoplasmic localization of nestin expression (Figure 2D). The tumor focally expressed several neural lineage markers including CD56 (neural cell adhesion molecule), PGP9.5 (protein gene product 9.5) (Figure 2E & F, respectively) and synaptophysin (not shown).

The tumor showed focal high intensity expression of several embryonic and NSC markers. The tumor cells expressed OCT4, NANOG, SOX2 and SALL4 (Figure 3A–D, respectively) with appropriate nuclear staining. OCT4 expression, in particular, was localized to regions where cells had nuclei resembling ESCs (Figure 2C). Expression of other embryonic markers, CKIT and Glypican-3 were clearly present but more focally expressed in general (Figure 3E & F, respectively) than stem cell markers. Variable expression of PAX8 was also seen (Figure 3G). Diffuse, strong intensity cytoplasmic staining for LIN28A was appreciated in tumor cells (Figure 3H). The tumor demonstrated focal cytoplasmic expression of cytokeratin and CD99, retention of INI1 staining and widespread cytoplasmic expression of vimentin, but was negative for α -fetoprotein, CD30, glial fibrillary acidic protein, and NeuN (not shown). FISH for C19MC demonstrated a balanced, nonamplified profile (Figure 3I). FISH studies for isochromosome 12p and the *EWSR1* translocation (break-apart probe for 22q12) were negative.

A DNA-methylation profile was performed as previously described [3,4]. Copy number variation analysis from the methylation profile for this tumor demonstrated none of the characteristic gains/losses described for other embryonal tumors with similar morphology including medulloepithelioma (Figure 4A & B). In addition, the methylation profile did not cluster with any of the tested CNS tumors to date including AT/RT, medulloblastoma, ependymoma, glioblastoma and other astrocytic tumors.

A limited postmortem examination confined to the spinal cord demonstrated a 5 cm intradural tumor of the spinal cord, multiple drop

Figure 2. Representative hematoxylin and eosin-stained sections and immunohistochemical stains showing neural differentiation in the neck tumor. (A) Low-magnification view of tumor showing papillary configurations and ribbons of multilayered tumor cells. An area of necrosis is present in the lower right (hematoxylin and eosin [H&E], 40×). (B) Higher magnification view showing rosettes and more solid regions (H&E, 200×). (C) High-magnification view showing clusters of cells with larger nuclei displaying smooth feathery vesicular chromatin and small nucleoli (arrows) adjacent to the ribbons of pseudostratified tumor cells with smaller nuclei having more condensed (hyperchromatic) chromatin (H&E, 400×). (D) Widespread cytoplasmic staining of cells with nestin between trabeculae and more prominently around basally oriented cells of trabeculae and papillary fronds (Nestin, 200×). (E) Focal staining for CD56 (CD56, 200×). (F) Scattered cells show nuclear and cytoplasmic staining for neuroblast and neuroendocrine marker PGP9.5 (PGP9.5, 400×).

metastases and extensive necrosis and hemorrhage with limited well-preserved tumor. An OCT4 immunohistochemical stain performed on one of the sections with more extensive tumor was negative. This result, however, should be interpreted cautiously since loss of antigenicity may have occurred secondary to tumor preservation, treatment, and postmortem interval (approximately 12 h). Conventional cytogenetics (karyotype) could not be performed on the autopsy tumor due to therapy and preservation effects.

Discussion

We present a young adult patient with a rare tumor, diagnosed as malignant medulloepithelioma without teratoid features, presenting outside the CNS likely arising from the cervical spinal roots. The pathological, immunophenotypic and molecular features of this unusual tumor may correlate with its poor response to multidrug chemotherapy and intensive radiotherapy.

Medulloepithelioma and other embryonal tumors can show variable neural and glial

differentiation as well as heterologous differentiation (cartilage, striated muscle and immature rhabdomyoblasts) that will invariably raise the differential diagnosis of immature teratoma [2,5]. In the case presented, no heterologous or differentiating elements were seen, thus teratoma or mixed malignant germ cell tumor were not diagnostic considerations. The location (lateral cervical neck) and histomorphology also excluded primary germ cell tumors (e.g., seminoma, embryonal carcinoma).

The tumor showed expression of proteins associated with neural lineage/differentiation reinforcing its histomorphology of primitive neuroepithelium. Nestin and CD56 are expressed in neuroepithelium and early differentiating human ESCs in culture conditions favorable for neural differentiation [6]. PGP9.5 and synaptophysin expression also supported neuroblastic/neuronal differentiation. PAX8 expression in this tumor was interesting since the *PAX* family of homeobox transcription genes has extensive control of neural development particularly eye/retinal development. PAX8 has been shown to

Table 1. Primary antibodies used in the evaluation of medulloepithelioma.

Antibody	Expression profile	Supplier	Type	Dilution
CD56	NCAM neural stem cell, natural killer cells	Cell Marque Corp, CA, USA	RM	1:2500
CD99 (12E7)	Ewing sarcoma/PNET marker, T-cell adhesion	Dako North America, CA, USA	MM	1:100
c-kit	Cell-surface receptor for the cytokine KIT/stem cell factor. Plays role in hematopoiesis, stem cell maintenance, gametogenesis, mast cell development and melanogenesis	Dako North America	RP	1:250
Glypican-3	Heparin sulfate glycoprotein that regulates events in embryogenesis, cancer	BioMosaics, VT, USA	MM	1:20
LIN28A (A177)	RNA binding protein suppressing let-7	Cell Signaling, Inc., MA, USA	RP	1:100
Nestin	Intermediate filament in neural stem cells	EMD Millipore, MA, USA	RP	1:200
OCT4	Marker of pluripotency, expressed in some germ cell tumors	Cell Marque Corp	MM	1:100
PAX8	Homeobox transcription factor important in eye/retinal development	Proteintech Group, IL, USA	RP	1:100
Pancytokeratin	Cytoskeletal proteins found in most epithelial cells	Ventana Medical Systems, AZ, USA	MM	1:100
PGP9.5	Marker of neuroblasts/neuroblastic differentiation, oocytes	Vector Laboratories, CA, USA	MM	1:25
SALL4	Transcription factor of SALL family; regulator of OCT-4 expression; expressed in germ cell tumors and yolk sac tumors	Biocare Medical, CA, USA	MM	1:10
SOX2	Maintenance of pluripotency; expressed in germ cell tumors, some lung carcinomas, glioblastoma	Cell Signaling Technology	MM	1:15
Synaptophysin	Neurosecretory vesicles; expressed in synapses, neuroendocrine tumors	Ventana Medical Systems	RM	Predilute
S100	Calcium binding protein; expressed in variety of tissues including neural, histiocytes	Vector Laboratories	RP	1:1000
Vimentin	Class III intermediate filament expressed in cells of mesenchymal origin	Ventana Medical Systems	MM	Predilute

MM: Mouse monoclonal; PNET: Primitive neuroectodermal tumor; RM: Rabbit monoclonal; RP: Rabbit polyclonal.

be overexpressed in gliomas while PAX5 has been reported to be overexpressed in medulloblastoma [7]. Within the CNS, several tumors in the differential diagnosis of medulloepithelioma (e.g., medulloblastomas, CNS PNET, AT/RT, ETANTR, ependymoblastoma) have been shown to have phenotypic features consistent with ESCs and NSCs [1,3,8]. These tumors can show a partial or predominant embryonal histotype composed of 'small round blue cells' with nuclei displaying powdery chromatin and small but visible nucleoli; features described for ESC/NSCs. Evidence is accruing that many tumors harbor cells that demonstrate gene and protein expression characteristics of ESCs and other primitive cell types (e.g., cancer stem cells). The tumor in this case is unique in that it demonstrated unequivocal, striking protein expression of recognized markers of pluripotency (OCT4, SOX-2, NANOG, SALL4) similar to that seen only in some germ cell tumors as well as expression of markers of embryogenesis/development

(c-KIT, Glypican-3).

OCT4 (octamer binding protein) is a master regulator gene in development and is critical for maintenance of pluripotency and self-renewal; functional elements that define ESCs. The protein product of the *OCT4* gene is a transcription factor can be readily detected immunohistochemically by its characteristic widespread nuclear expression in several germ cell tumors notably testicular seminomas, embryonal carcinomas, ovarian dysgerminoma and primary CNS germinomas [9,10]. *SOX2*, another master regulator gene, is involved in maintenance of pluripotency of ESCs and germ line. In surgical pathology, *SOX2* is also widely expressed in the nucleus of germ cell tumors such as mediastinal embryonal carcinomas and variably in lung carcinomas, glioblastoma, ovarian tumors and others [10]. *SALL4* is a member of the *SALL* (spalt-like/sall) gene family that regulates *OCT4* expression and is important in embryonic development. *SALL4* along with *OCT4*, *NANOG*

and SOX2 are established immunohistochemical markers (showing nuclear expression) of germ cell tumors, and SALL4, but not OCT4 and NANOG, is highly expressed in yolk sac tumors [9,10]. C-KIT (CD117) encodes a receptor tyrosine kinase and is crucial for normal spermatogenesis and development. C-KIT expression is seen in most testicular seminomas, ovarian dysgerminomas, intracranial germinomas and primordial germ cells [8]. Glypican-3 is a heparin sulfate proteoglycan that regulates signaling events in embryogenesis, normal physiology and cancer pathogenesis. Immunohistochemical detection of Glypican-3 serves as a surrogate marker for hepatocellular carcinoma and hepatoblastoma, another embryonal tumor of

childhood [11]. OCT4, NANOG, SOX2 and LIN28 transcription factors have been used collectively to transform somatic fibroblasts into induced pluripotent cells (iPCs) that have the capability of self-renewal, perpetuation and pluripotency [12]. Taken together, the immunohistochemical expression of these markers in this tumor is similar in quality and quantity to other germ cell tumors and signifies a tumor containing populations of cells with ESC and/or primordial germ cell phenotypes.

Because cancer stem cells share properties with ESCs (self-renewal, differentiation, drug resistance, but not pluripotency), much investigation has been devoted to the identification and characterization of putative cancer stem

Figure 3. Representative images of tumor showing expression of several stem cell markers.

(A) Regional strong intensity staining for embryonic stem cell marker, OCT4 (OCT4, 200×). (B) Focal nuclear expression for pluripotent stem cell marker, NANOG (NANOG, 400×). (C) Compared with OCT4, the tumor exhibited more diffuse, strong intensity expression of SOX2, a transcription factor important in maintenance of pluripotency. (D) Similar to OCT4, the tumor demonstrated regional expression for SALL4, a transcription factor important in the maintenance of pluripotency and modulation of *OCT4* gene expression (SALL4, 200×). (E) Regional expression of CD117 (c-kit) with appropriated membranous staining (c-kit, 200×). (F) Similar to CD117, regional and membranous staining for the glycoprotein, Glypican-3 is present (Glypican-3, 200×). (G) Focal widespread and variable nuclear staining for PAX8 (PAX8, 200×). (H) Strong diffuse cytoplasmic staining for LIN28A (LIN28A, 200×). (I) FISH for C19MC shows a balanced, nonamplified profile with same number of C19MC loci (green) and reference loci (red).

Figure 4. Plot of copy number variations generated from 450-k methylation data. (A) The tumor presented in this case showed no significant copy number variations at 19q13.42 (C19MC). **(B)** Conversely, a prototypical intracranial medulloepithelioma demonstrated amplification at the 19q13.42 locus; an amplification seen in 95% of tumors labeled as embryonal tumors with multilayered rosettes. This prototypical medulloepithelioma also demonstrated gains at chromosomes 2 and 11q, aberrations frequently observed in embryonal tumors with multilayered rosettes.

cells as therapeutic targets. Increased expression of stem cell genes (i.e., *NANOG*, *OCT4*, *SOX2*) have been identified in cells from many tumors and correlates with poorly differentiated tumors, increased risk of metastases and poor prognosis [13,14]. Within the CNS, the *SOX2* and *OCT4* expression is seen in NSCs residing in the subventricular zone and putative brain tumor stem cells in glioma. In gliomas, silencing of *SOX2* and *OCT4* gene expression decreases proliferation and tumor size [15] and *OCT4* and nestin expression correlates with worse

prognosis in patients with glioblastoma [16]. Medulloblastomas, highly malignant embryonal brain tumors, show gene expression of stem cell markers *CD133*, *LIN28*, and *OCT4*. *OCT4* is expressed in slightly less than half of medulloblastomas, but patients whose tumors have *OCT4* gene expression have a worse prognosis essentially upstaging average risk patients to high risk [17]. *OCT4* and *NANOG* expression in cancer stem cells has also been associated with resistance to cisplatin and doxorubicin treatment and increased multidrug resistance gene

expression in tumor cell lines with increased *NANOG* expression [18,19]. In addition, resistance to radiation therapy has been well documented in some cancer stem cell lines [20]. In contrast, seminoma, a germ cell tumor, that shows widespread and high expression of OCT4 along with human ESCs and NSCs in culture are highly radiosensitive [21]. This patient's tumor had unequivocal expression of multiple ESC/NSC markers and at least by imaging was apparently never completely eradicated despite aggressive chemotherapy (that included doxorubicin) and intense radiotherapy. The probable resistance to radiation therapy of this tumor may depend on other properties akin to those present in some cancer stem cell lines that confer radioresistance. It should be noted that most studies demonstrating expression of ESC markers in CNS tumors have been studies examining gene expression from fresh tumor samples and/or immunocytochemistry on cell culture preparations from tumor samples and not IHC on formalin-fixed paraffin embedded samples.

An area of exciting investigation is the role of LIN28 proteins in stem cell renewal/growth/metabolism, oncogenesis and development. Within ESCs, LIN28, an RNA binding protein suppresses let-7 thereby retarding cell differentiation. Overexpression of LIN28 along with OCT4, SOX2 and *NANOG* transforms fibroblasts into self-renewing pluripotent stem cells (iPCs). LIN28 expression persists at high levels within the neural tube and neural crest and again LIN28 overexpression can lead to increased primitive neural tissue in teratomas formed by ESCs [22]. Recently ETANTR, ependymoblastoma and medulloepithelioma have been shown to have similar immunophenotypic and molecular genetic profiles with consistent LIN28A protein expression and 19q13.42 amplification suggesting that these three entities may be interrelated and the unifying diagnostic term 'embryonal tumor with multilayered rosettes' or ETMRs has been proposed for these entities [3,23,24].

In addition, ETMRs also show remarkably similar DNA-methylation profiles unique to themselves and distinct from other embryonal and CNS tumors. Copy number variation analysis from the methylation profiles of a large number of ETMRs demonstrate consistent amplification of the 19q13.42 locus and frequent gains in chromosome 2, low level gains in 1q, 7q, 11q and loss of 6q [3]. Interestingly, in the case presented here, while LIN28A expression by IHC

was similar to that seen in other ETMRs, in stark contrast to ETMRs analyzed thus far, the methylation profile did not have significant copy number variations at any chromosomal locus. It would be interesting to compare the methylation profile of this tumor to those of ESCs, iPCs and other tumors in the category of PNETs.

Conclusion

In summary, we present a patient with a tumor that morphologically is a medulloepithelioma presenting in an unusual location at an unusual age showing incontrovertible immunohistochemical expression of multiple ESC/NSC/embryogenesis markers but lacking the characteristic DNA-methylation profile of medulloepithelioma or other similar embryonal tumors. With the available immunophenotypic and molecular genetic information, this highly unusual and enigmatic tumor might be best classified as a 'PNET with embryonic stem/germ cell and medulloepithelioma-like features'. The high level of expression of embryonic markers in this tumor may in some measure account for the eventual unresponsiveness to therapy and poor prognosis.

Future perspective

Newer therapies targeted at embryonic cell phenotypes may hold the key to successfully treating many cancers with embryonal phenotypes or tumors harboring cells with embryonal phenotypes. Evaluation for the expression of embryonal markers may be important in gauging therapy and prognosis in these tumors. As the molecular genetic signatures of CNS tumors (and other tumors) are elucidated and tumor cell populations better defined, molecular therapies tailored at removing tumor progenitor populations previously resistant to traditional therapeutic modalities will become mainstream.

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The authors state that they have obtained verbal and written informed consent from the patient/patients for the inclusion of their medical and treatment history within this case report.

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• of interest; •• of considerable interest

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