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CASE REPORT

Synaptophysin negative central neurocytoma

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Abstract: *Background:* Central neurocytoma is a rare primary brain tumour, mostly localised in the lateral ventricles in relation to the foramen of Monro.
Objectives: To report a case of a rare central neurocytoma with a complete loss of Synaptophysin expression and provide the differential diagnosis.
Methods and results: We describe a case of a 34-year old man with a headache, unsteady gait and dim vision. MRI demonstrated a tumorous expansion localised in both lateral ventricles. The patient underwent a subtotal resection. Histology showed a picture consistent with central neurocytoma, but tumour was completely negative for Synaptophysin. We describe our approach in such a diagnostically difficult case.
Conclussions: In the rare case of Synaptophysin-negative central neurocytoma, its neuronal differentiation should be substantiated by electron-microscopic examination. Unfortunately in the routine work, biopsy samples are usually fixed in formalin fixative which does not preserve ultrastructure well. In such situations, an accurate diagnosis is disputable and based on careful assessment of the histological features, exclusion of tumours with similar morphology and detailed correlation with MRI pictures *(Fig. 4, Ref. 6)*. Full Text (Free, PDF) *www.bmj.sk.*
Key words: central neurocytoma, synaptophysin negativity, oligodendroglioma, ependymoma.

A 34-year old man presented with a few months history of headache, unsteady gait and dim vision. After admission to the hospital, MRI showed a spacious tumour in both lateral ventricles (Fig. 1). The patient underwent a subtotal resection. Four months after the operation, a continued paraparesis of the left upper limb and slight organic psychosyndrome was present. The biopsy sample was submitted for histological examination and completely processed with a standard paraffin technique and with haematoxylin-eosin (HE). For immunohistochemistry, the avidin-biotin-peroxidase complex method was used. The primary antibodies, including clone, dilution and manufacturer, were as follows: Ki67 (MIB-I, 1:75, Dako), neuron specific enolase (NSE) (BBS/NC/MH-14, prediluted, Dako), Synaptophysin (polyclonal, 1:100, Dako), PGP9,5 (polyclonal, 1:100 Dako), GFAP (polyclonal, 1:200, Dako), S100 protein (polyclonal, 1:2000, Dako), epithelial membrane antigen (EMA) (E29, 1:50, Dako), Vimentin (3B4, 1:200, Dako), Chromogranin (DAK-A3, 1:200, Dako), p53 (1801, 1:100, Biogenex), Cytokeratin (AE1/3, 1:200, Labvision), neurofilaments (NF) (NF68.04, 1:150, Labvision), Olig II (polyclonal, 1:200, Chemicon). For electron microscopy, tumour tissue was deparafinised and postfixed in 1 % osmium tetroxide, dehydrated in a graded ethanol series, and embedded in durcupan. Thin sections were stained with uranyl acetate and lead citrate and viewed in a Tesla 540 transmission electron microscope. For fluorescent in situ hybridization (FISH),

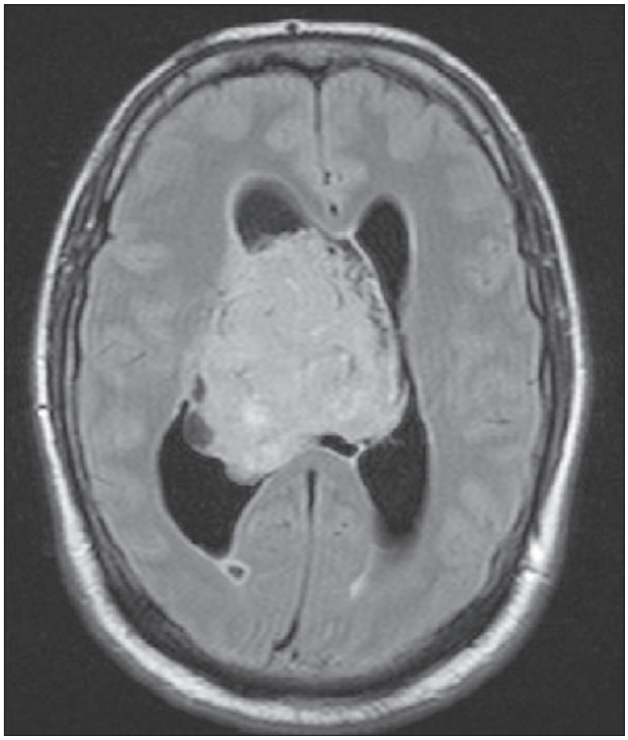

Fig. 1. MRI, FLAIR transversal section shows a spacious tumour in both lateral ventricles.

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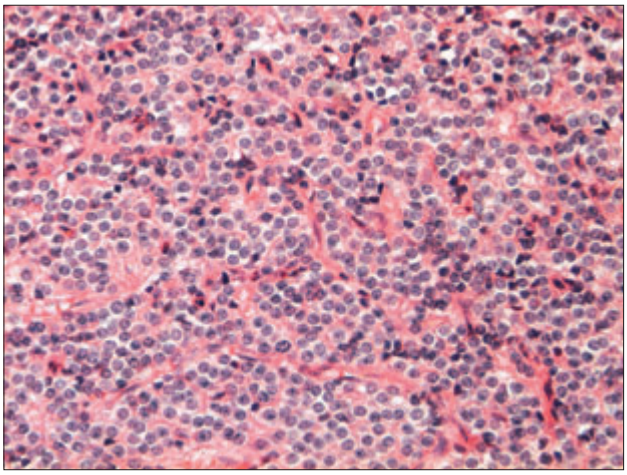

Fig. 2. Large areas of the tumour were almost indistinguishable from oligodendroglioma. Round nuclei, perinuclear haloes and delicate vasculature can be appreciated (haematoxylin eosin, original magnification x400).

eight paraffin slices were deparaffinised. 7 µl of probe LSI 1p36so/1p25sg, LSI 19p13so/19qsg and 3 µl of LSI PTENso/ /cep10sg were applied to the same area. Material was denaturated by 73 °C in thermo-block HYBriteTM (Vysis) and hybridised by 37 °C. Allelic loss was examined in a fluorescent microscope (magnification 1000x). 200 nuclei were evaluated. Two red signals and one green in the same nucleus at least in 5 % of analysed cells were considered positive.

Histology showed a tumour composed of relatively uniform cells with round to oval nuclei and scant clear cytoplasm. Perinuclear haloing was observed in some regions. Delicate thin-walled arborising vasculature pervaded the tumour. Together with a focal calcification, the resemblance to oligodendroglioma was striking (Fig. 2). However, some cells formed vague neurocytic rosettes (Fig. 3A) and the background, including occasional nuclear-free zones (Fig. 3B), was more neuropil-like than glial in appearance. A border against the brain parenchyma was quite sharp (Fig. 3D). No mitotic figutes, necrosis or vascular proliferation was observed. Tumorous tissue was completely and re-

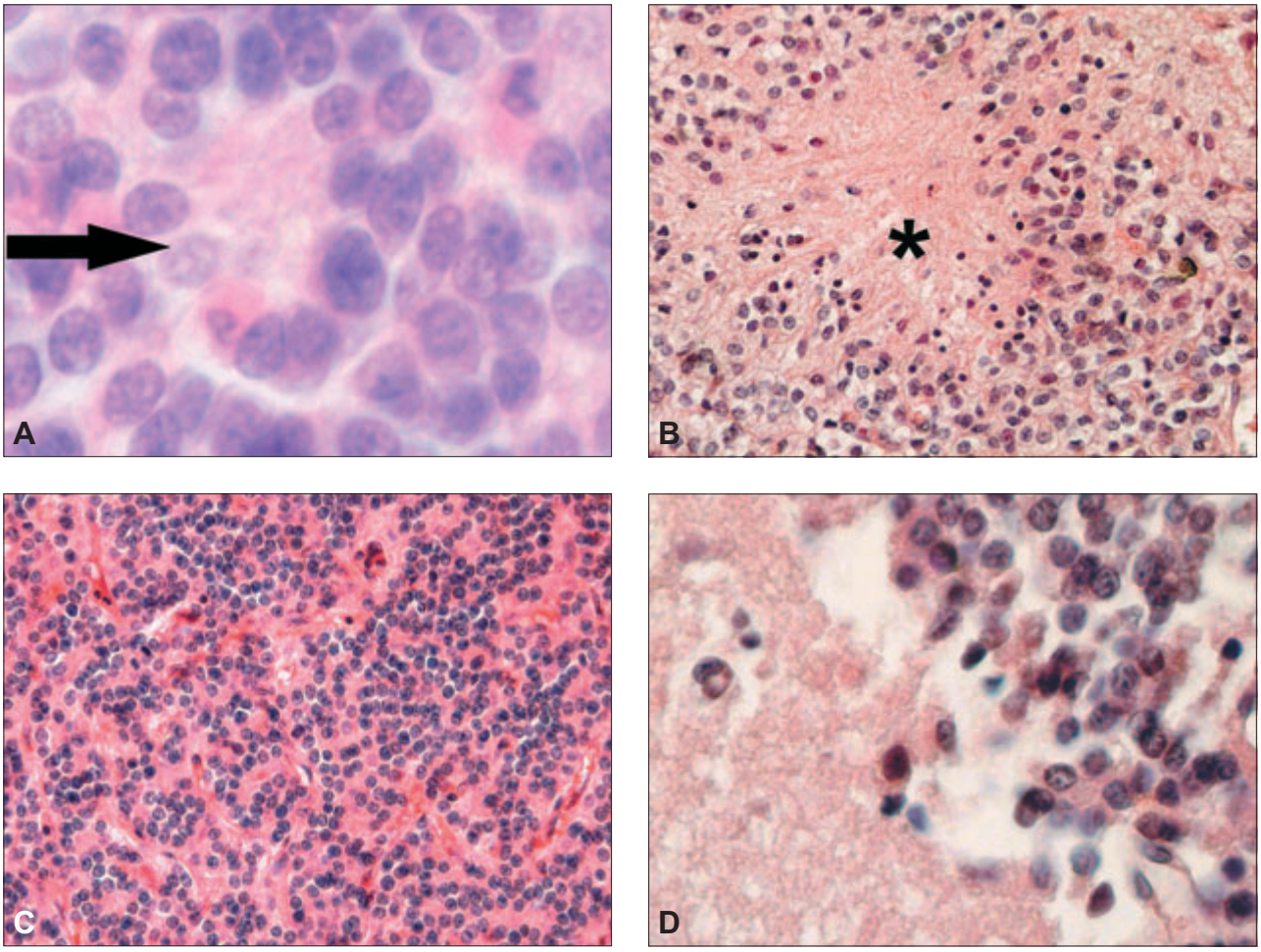

Fig. 3. Nuclei forming incomplete circles – poorly formed neurocytic rosettes (3A, arrow, HE, original magnification x600), nuclear free zones resembling neuropil (3B, asterisk, HE, original magnification x200), perivascular orientation of the cells reminiscent of ependymoma (3C, HE, original magnification x200), relative sharp border against surrounding brain tissue (3D, HE, original magnification x400).

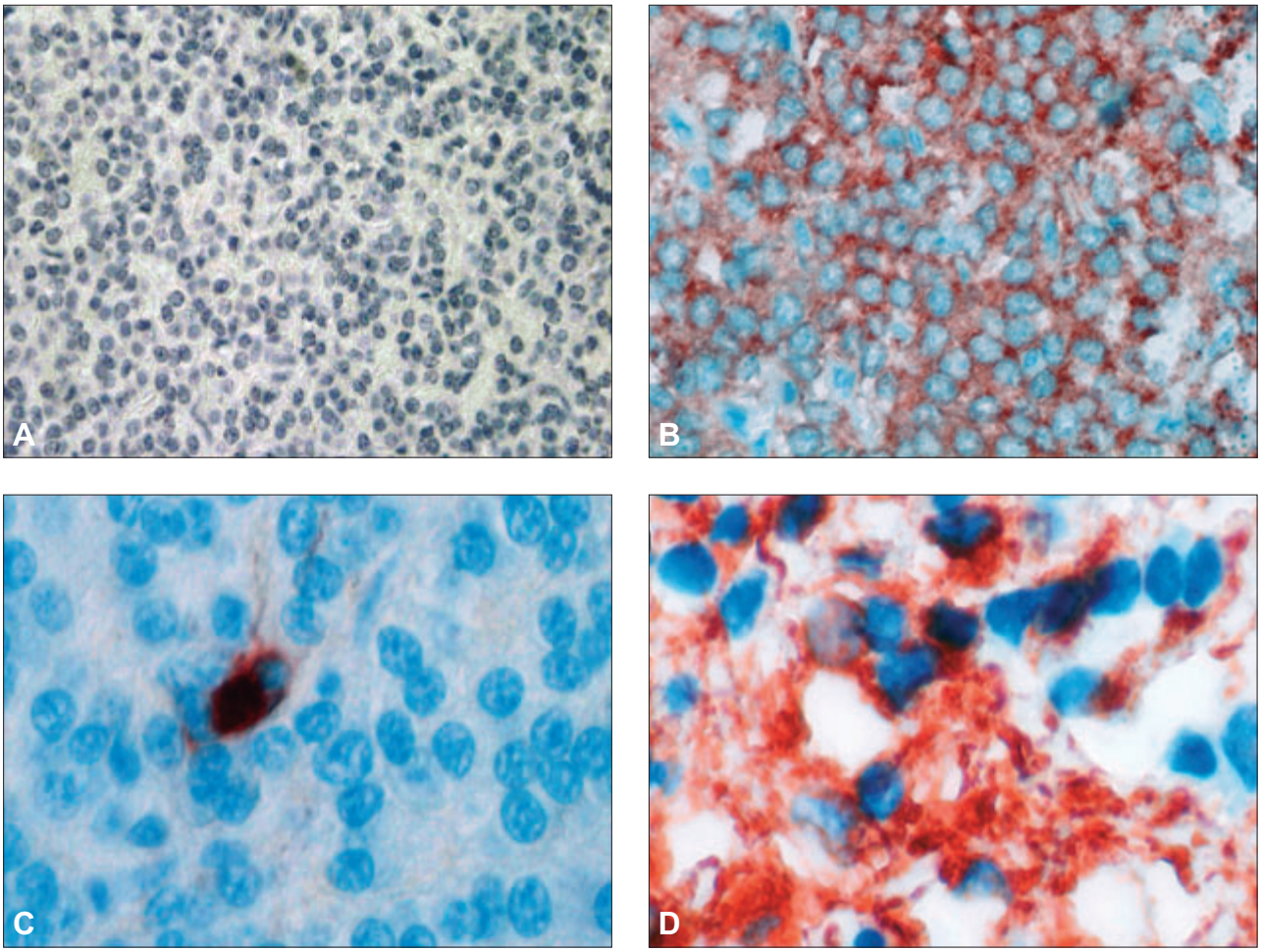

Fig. 4. Immunohistochemistry: a complete negativity for synaptophysin (4A, original magnification x200), positivity for NSE (4B, original magnification x400), rare cell positive for GFAP (4C, original magnification x600), positivity for Vimentin (4D, original magnification x600).

peatedly negative for Synaptophysin (Fig. 4A). Virtually all other markers were negative (PGP, S-100, EMA, Chromogranin, P53, CK, NF, Olig II) with the exception of diffuse positivity of NSE (4B), Vimentin (4D), and rare cells positive for GFAP (Fig. 4C). In such "immunohistochemically resistant" tumours, at least Vimentin proves antigen viability of the sample. GFAP positive cells were interpreted as tumorous and not entrapped native glial elements. Electron-microscopic examination was inconclusive, because ultrastructure was damaged by formalin fixation. Fluorescent in situ hybridisation did not show a loss of heterozygosity for short arm of chromosome 1 and long arm of chromosome 19 (LOH 1p/19q), which is thought to be the a genetic signature of the oligodendroglial tumours. The diagnosis of central neurocytoma was made despite the synaptophysin negativity. It was based on subtle histological features as fine neuropil-like background and focal ill-defined neurocytic rosettes, and the exclusion of alternative diagnoses (oligodendroglioma and clear cell ependymoma). A detailed correlation with he MRI features was essential for an accurate classification of this unusual tumour.

Discussion

Central neurocytoma (CN) is a rare tumour of the central nervous system. Presumably it originates from neuroglial precursor cells in the subependymal plate of the lateral ventricle. It was described by Haussun et al in 1982 for the first time (1). The incidence ranges from 0.25–0.5 % of all intracranial tumours. Mostly, it is diagnosed in the third decade of life. Both sexes are equally affected. Typically, it is located in the lateral ventricle in relation to foramen Monro. The majority of patients present with symptoms of an increased intracranial pressure from liquor blockade. MRI shows a heterogenous hypointensity on T1 and hyperintensity on T2-weighted images, with a well-defined margin and enhancement after a gadolinium injection (2). The mainstay of therapy is a complete surgical resection. Radiotherapy after an incomplete resection might be beneficial. A good response to chemotherapy has been described (3). The prognosis is generally favourable. Occasional CNs with an aggressive course are assigned as WHO grade II. Histology does not reliably predict biological behaviour. Histology shows a delicate neuropil back-

ground, uniform cells with round nuclei and scant eosinophilic cytoplasm (4). Perinuclear haloes are observed, and together with a delicate arborising vasculature, are responsible for the striking resemblance to oligodendroglioma. The most suitable and reliable immunohistochemical marker is Synaptophysin. However, its expression might be rarely reduced or even completely lost. CN is usually positive for NSE and NeuN. NSE can be found in virtually any type of neoplasm, due to its low sensitivity it is a poor antibody to use to screen for neuronal differentiation. Stains for Chromogranin and Neurofilament are usually negative. Electron microscopy is required when expression of specific neuronal markers is lacking. Unfortunately, in routine work, tissue fixed for electron microscopy in not always available. Ultrastructure is usually damaged by formalin fixation and its assessment might be subsequently inconclusive. In such situations, a diagnosis is challenging as in our case. There are other tumours which may demonstrate a very similar histology. In particular, differential diagnosis against oligodendroglioma and clear cell ependymoma is difficult. Nucleus-free zones as well as a background more resembling neuropil than glial fibrillarity, together with occasional ill-defined neurocytic rosettes, provides a hint of neuronal differentiation. GFAP is not helpful because its expression in oligodendrogliomas is variable and focal expression does not exclude the diagnosis of CN. The expression of neuronal markers is important for the diagnosis of CN but positivity on its own does not really distinguish CN from oligodendroglioma as expression of neuronal markers in oligodendrogliomas is a well established phenomenon (5). In fact, oligodendroglial and neurocytic lesions may form a biological and diagnostic spectrum. The oligodendroglial marker Olig II fails to differentiate astrocytoma from oligodendroglioma but its negativity was strongly against the diagnosis of oligodendroglioma (6). One biologically important distinction between oligodendroglioma and neurocytoma lies in their interaction with the surrounding brain. Oligodendroglioma is diffusely infiltrative and entrapped axons within the tumour may be demonstrated by neurofilament staining. The genetic marker LOH 1p/19q for oligodendroglioma was negative. A typical MRI picture is important for diagnosis. Intraventricular oligodendroglioma would be unusual. It is possible that intraventricular oligodendrogliomas reported in the past were in fact central neurocytomas. Because of the intraventricular location, ependymoma was also considered. In particular, clear cell ependymoma shows similar histology. Perivascular pseudorosettes made this similarity even stronger. However, on a high magnification, long tapering processes anchored to the walls of the vessels and typical of ependymoma were lacking. True ependymal rosettes or canals were not observed, but this feature in ependymomas is not very frequent. Ependymoma is a glial tumour, strongly positive for GFAP. In addition, there is a dot-like EMA positivity in ependymomas. The formation of intracytoplasmic EMA positive lumina is virtually pathognomic for ependymoma. CD 99 or the MIC2 gene product duplicates this EMA expression. No EMA or CD 99 positivity was observed in our case. Other differential diagnoses such as pineocytoma, dysembryoplastic neuroepithelial tumour, pituitary adenoma, clear cell meningioma and primitive neuroectodermal tumour should be easily differentiated by MRI, localisation, histomorphology and immunohistochemistry.

Conclusions

We describe a rare case of Synaptophysin negative central neurocytoma. Its diagnosis is usually based on histological examination and upon demonstration of neuronal differentiation by positive neuronal markers, the most suitable being synaptophysin. However, expression of neuronal markers might be sometimes reduced or rarely even completely lost. In such cases, the ultrastructure showing neuronal differentiation is required. Despite this, a specimen fixed for electron microscopy is not always available and ultrastructure might be subsequently inconclusive. In such situations, the diagnosis is challenging and should by supported by careful assessment of subtle morphological features of neuronal differentiation using light microscopy, exclusion of tumours with overlapping morphology (oligodendroglioma and ependymoma) and finally by a detailed correlation with MRI pictures.

References

1. Hassoun J, Gambarelli D, Grisoli F et al. Central neurocytoma. An electron micsroscopic study of two cases. Acta Neuropathol 1982; (56): 151—156.

2. Gourtsoyiannis NC, Ros PR (Eds). Radiologic-Pathologic Correlations from Head to Toe. Heidelberg—Berlin: Springer Verlag 2005: 56—58.

3. Rades D, Fehlauer F, Lamszus K et al. Well-differentiated neurocytoma: What is the best available treatment? Neuro-Oncology 2005; 7: 77—83.

4. Figarella-Branger D, Söylemezoglu F, Burger PC. Central neurocytoma and extraventricular neurocytoma. 106—109. In: Louis DN, Ohgaki H, Wiestler OD, Cavenee WK (Eds). World Health Organization classification of tumours. WHO Classification of Tumours of the Central Nervous System. Lyon; IARC Press 2007.

5. Wharton SB, Chan KK, Hamilton FA et al. Expression markers in oligodendrogliomas: an immunohistochemical study. Neuropathol Appl Neurobiol 1998; 24: 302—308.

6. Preusser M, Budka H, Rössler K, Hainfellner JA. OLIG2 is a useful immunohistochemical marker in differential diagnosis of clear cell primary CNS neoplasms. Histopathology 2007; 50: 365—370.

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