Folia Neuropathologica

Granular cell astrocytoma. A case report with immunohistochemical and ultrastructural characterization

Teresa Wierzba-Bobrowicz[1], Eliza Lewandowska[1], Ewa Matyja[4], Jacek Dziduszko[2], Waldemar Koszewski[2], Tomasz Stępień[1], Beata Błażejewska-Hyżorek[3]

[1]Department of Neuropathology, [2]Department of Neurosurgery, [3]Second Department of Neurology, Institute of Psychiatry and Neurology, [4]Department of Experimental and Clinical Neuropathology, Medical Research Centre, Polish Academy of Sciences, Warsaw, Poland

Folia Neuropathol 2008; 46 (4): 286-293

Abstract

Granular cell astrocytoma (GCA) is an uncommon type of granular cell tumours (GCTs) in the central nervous system. Granular cells in these tumours are of enigmatic origin.

We report a case of cerebral GCA in a 59-year-old man who suffered from diabetes and Addison-Biermer disease. The tumour was localized in the left parietal lobe. Microscopically, the tumour was almost entirely composed of large, polygonal cells with round to oval, granular eosinophilic, PAS-positive cytoplasm. The nuclei were located centrally or eccentrically and sometimes exhibited nucleolar vacuoles. The tumour cells were arranged in nests surrounded by blood vessels and connective tissue. Immunohistochemically, the granular tumour cells were reactive for GFAP and vimentin. They were intensively stained for ubiquitin and some of them were reactive for CD68. Moreover, a lot of stromal cells expressed CD68 reactivity.

Ultrastructurally, most tumour cells were round or oval with only a few or without filaments. Their cytoplasm was filled with electron-dense granular material limited by a single membrane and autophagic vacuoles. Another type of tumour cells, present in a significantly lower number, revealed abundant cytoplasm with numerous intermediate filaments, swollen rough endoplasmic reticulum, mitochondria and a few clusters of granular material. Cells with numerous condensed electron-dense, bizarrely-shaped mitochondria and few filaments were occasionally observed. Among granular cells, macrophages with vacuoles and/or lamellar structures were visible.

In our case, both immunohistochemical and ultrastructural analysis supported astroglial origin of the granular cell tumour.

Key words: *granular cell astrocytoma, immunohistochemistry, ultrastructure.*

Introduction

Granular cell tumour was first mentioned by Weber [33] and described by Abrikossoff , who referred to it as "granular cell myoblastoma" of the tongue [1]. The term "granular cell tumour" (GCT) was first introduced by Lack et al. in 1980 [15] and up to now about 200 cases of GCT have been described.

GCTs are uncommon lesions, which may occur in humans and domestic animals at any age in al-

Communicating author:

Teresa Wierzba-Bobrowicz, MD, PhD, Department of Neuropathology, Institute of Psychiatry and Neurology, Sobieskiego 9, 02-957 Warsaw, Poland, Email: bobrow@ipin.edu.pl

most any organs [9,10,16,20,26]. They are most frequently observed in the skin and tongue, but very rarely in the digestive and respiratory tracts or intracerebral regions.

In the central nervous system (CNS), GCTs have also been reported in the cerebral hemispheres, cerebellum, meninges, third ventricle, hypophysis and cranial nerves [2,7,11,14,26].

Morphologically, GCTs are non-encapsulated and the cells are typically grouped in a nest-like pattern, surrounded by thin strands of fibrous tissue, independently of the organs from which they originate. The tumour cells are plump and polygonal with eosinophilic and periodic acid-Schiff (PAS) positive granular cytoplasm [26,34]. Ultrastructurally, abundant mitochondria and secondary lysosomes are usually observed in the cytoplasm of tumour cells [10,28,29].

Granular cells in GCTs are of enigmatic origin. They were initially believed to be of myogenic origin and then of histiocytic and subsequently of neurogenic origin [7,22,28].

Granular cell astrocytoma (GCA) is a rare histopathological variant of astrocytoma exhibiting a prominent population of typical granular cells. These tumour cells contain numerous eosinophilic granules and show cytoplasmic immunoreactivity to glial fibrillary acidic protein (GFAP) [3,6,9,12,13,18,23,25,31].

We report a case of cerebral GCT in a man who suffered from diabetes and Addison-Biermer disease. The astroglial origin of this rare entity was confirmed by immunohistochemical and ultrastructural examinations.

Case report

A 59-year-old man was admitted to our hospital with a one-week history of progressive headache and visual disturbances. His medical history was significant for long-standing (20 years) and well-controlled insulin-treated diabetes mellitus type 2 and Addison-Biermer disease recognized 6 years before admission. Physical examinations revealed pale skin and neurological testing revealed left-sided homonymous hemianopsia. The motor strength was intact, and no sensory deficits were detected. The Babiński sign was absent. Laboratory examination showed a slight decrease of white cell count: 3.6 K/µl (4.1 to 10.9). Mild anaemia was noted with red blood cell count of 3.89 M/µl (4.2 to 5.70), haemoglobin was 11.7 g/dl (12.6 to 17.2) and haematocrit was 33.6% (38.0 to 49.0). Sedimentation rate, coagulation profile, liver tests and the results of the renal function test were insignificant. The results of the chest X-ray and abdominal computed tomography (CT) were normal. Gastric endoscopic examination revealed diffuse atrophic gastritis. Biopsies taken at the time of endoscopic examination evidenced H. pylori infection.

The magnetic resonance imaging (MRI) scan of the head revealed a tumour (15 × 25 × 15 mm) localized in the left parietal lobe (Figs. 1, 2). The tumour was shown as a well-defined, bordered, homogeneous mass, surrounded by oedematous brain tissue and compressing the posterior horn of the right lateral ventricle. Right parietal, parasagittal craniotomy was performed. A well bordered, brown-yellowish, hard pathological tissue was found 15 mm under the surface of the oedematous cerebral cortex. The tumour was completely removed within its borders.

Methods

The tumour tissue for morphological examination was fixed in 10% buffered formalin and paraffin embedded. The specimens were stained with H&E, PAS, PAS-dimedon, Gomori's method and immunohistochemically with the following antibodies: glial fibrillary acid protein (GFAP, DAKO, 1:500), vimentin (DAKO, 1:50), synaptophysin (SY, Novocastra, 1:30), CD68 (DAKO, clone PG-M1, 1:100), CD34 (Novocastra,

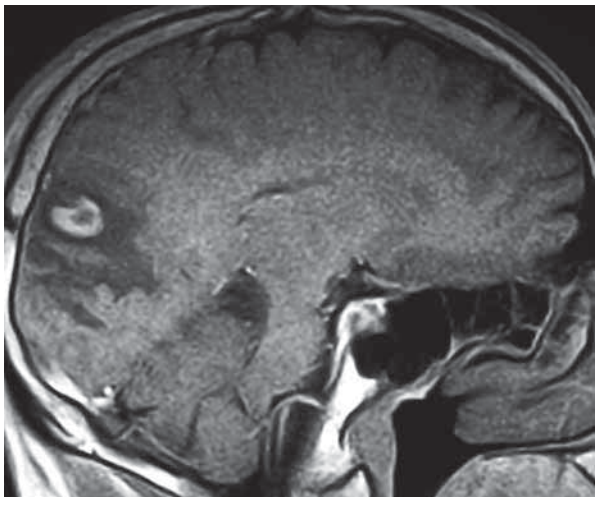

Fig. 1. Sagittal magnetic resonance imaging (MRI) revealed a tumour localized in the left parietal lobe

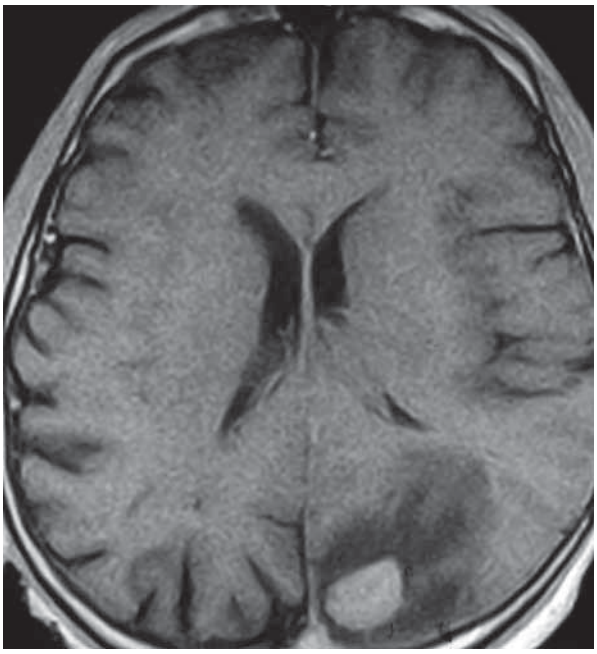

Fig. 2. MRI revealed an oval-shaped well-circumscribed tumour (15×25 mm in diameter) surrounded by brain oedema

1:300), epithelial membrane antigen (EMA, clone-GP 1,4, Novocastra, 1:300), Ki-67 (clone MIB-1, DAKO, 1:100), cytokeratin (CK, clone AE1/AE3, DAKO, 1:50) S-100 protein (DAKO, 1:400), ubiquitin (DAKO, 1: 200), and anti-neuronal nuclei (NeuN, Chemicon, clone-A60, 1:100).

For electron microscopic evaluation, a small fragment of the tumour was taken from paraffin blocks. After deparaffinizing and washing in water, the material was postfixed in 2.5% glutaraldehyde, 2% osmium tetroxide and routinely processed to Epon 812. Ultrathin sections were stained with uranyl acetate and lead citrate, then examined in a transmission electron microscope (Opton DPS 109).

Results

Microscopically, the tumour was almost entirely composed of round, oval or large polygonal cells with granular eosinophilic cytoplasm. The cells were arranged in a nest-like form (Figs. 3, 4). The blood vessels and reticular fibres compartmentalized the tumour cells (Fig. 5). The plump cytoplasm of tumour cells was eosinophilic with numerous PAS-positive granules often surrounded by a light "halo" (Fig. 6).

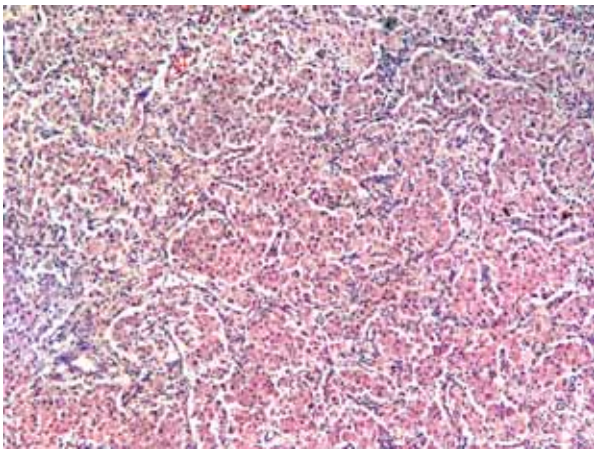

Fig. 3. The tumour was composed of large polygonal cells arranged in nest-like forms. HE ×100

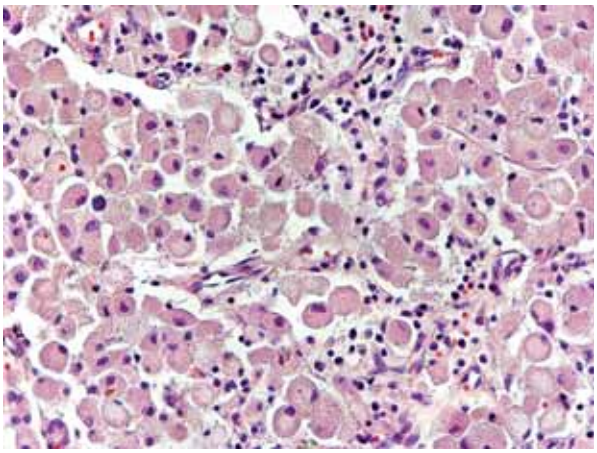

Fig. 4. Tumour cells with abundant cytoplasm and eccentrically placed nuclei. HE ×200

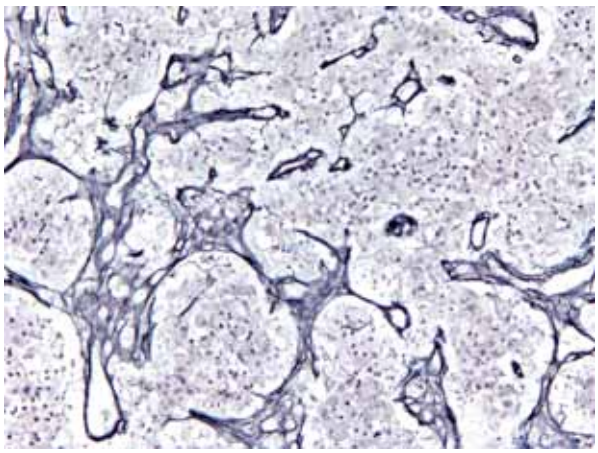

Fig. 5. Reticular fibres around nests of tumour cells. Gomori's method, ×200

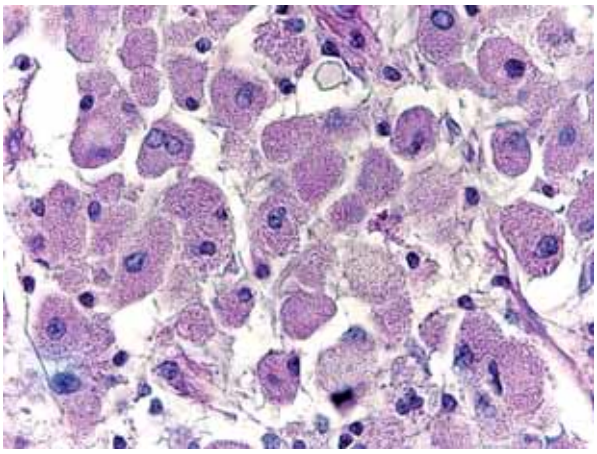

Fig. 6. Tumour cells exhibit PAS-positive granules. PAS, ×400

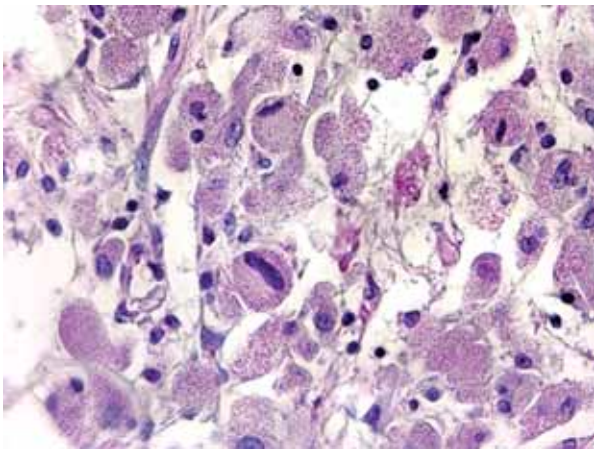

Fig. 7. Nuclei of different shapes located centrally or eccentrically in granular tumour cells. PAS, ×400

The nuclei of tumour cells were round to oval as well as bacilliform or lobular in shape (Fig. 7). They were located centrally or eccentrically and sometimes exhibited nuclear pseudovacuoles.

Immunohistochemically, the granular tumour cells were reactive for GFAP, vimentin and EMA (Figs. 8-10), whereas S-100 protein expressed weak positivity (Fig. 11). The majority of tumour cells were intensively stained for ubiquitin (Fig. 12). Cytokeratin, NeuN and SY immunostains were negative. Fibroconnective stromal cells and some of the granular cells were positively stained for CD68 (Figs. 13, 14). Labelling index of Ki-67 was low in tumour cells, while it was increased in stromal cells (Fig. 15).

At the ultrastructural level, a majority of tumour cells were round or oval and filled with electron-dense granular material limited by a single membrane (Fig. 16). Some of the vacuole-containing areas of the cytoplasm with mitochondria resembled autophagic vacuoles. Processes with bundles of intermediate filaments or collagen fibres could be seen between granular cells. In the cytoplasm of tumour cells usually sparse bundles of filaments were visible (Fig. 17). Sometimes, the filaments encircled the nucleus (Fig. 18). The nuclei of tumour cells exhibited more or less condensed chromatin and prominent nucleoli (Fig. 19). The cytoplasm of cells was often swollen with decreased electron density. Less numerous tumour cells revealed abundant cytoplasm with numerous intermediate filaments, swollen rough endoplasmic reticulum, mitochondria and a few clusters of granular material (Fig. 20). Sometimes, a few cells contained

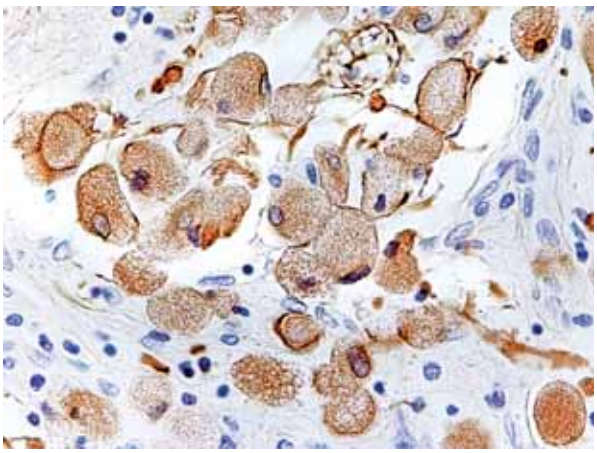

Fig. 8. The cytoplasm of tumour cells expressed GFAP, ×400

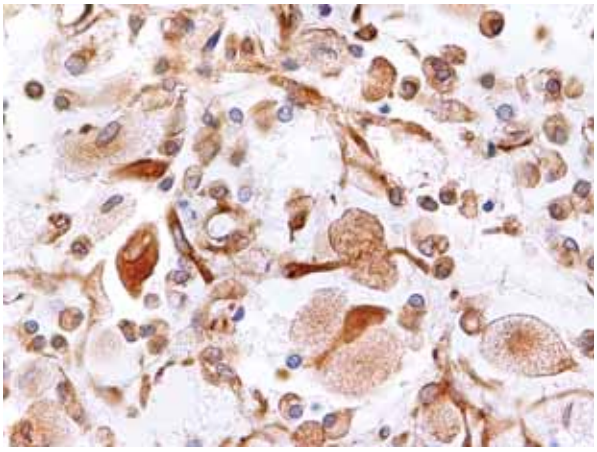

Fig. 9. Immunoreactivity of vimentin in tumour cells, ×400

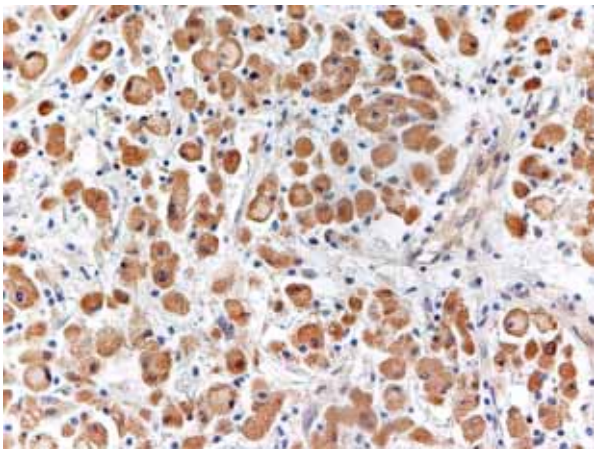

Fig. 10. Diffuse immunoreactivity for EMA, ×200

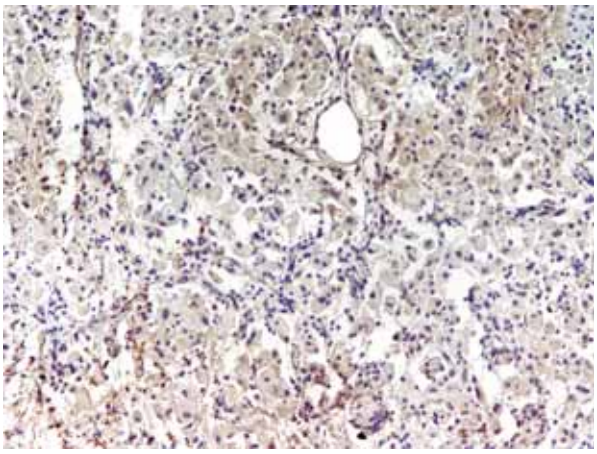

Fig. 11. Weakly expressed S-100 protein in the tumour, ×100

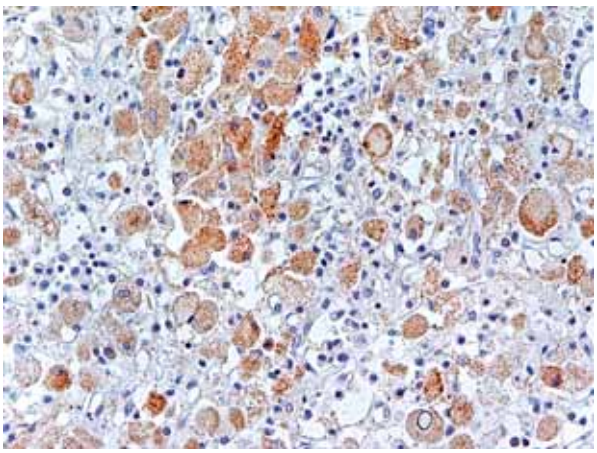

Fig. 12. Tumour cells intensively positive for ubiquitin, ×20

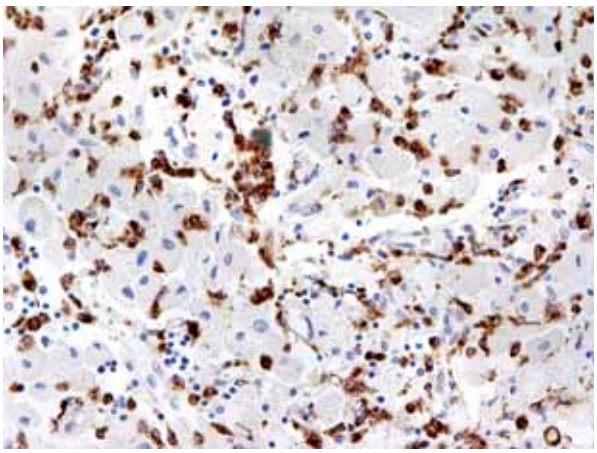

Fig. 13. Strong immunoexpression of CD 68 in stromal cells, ×200

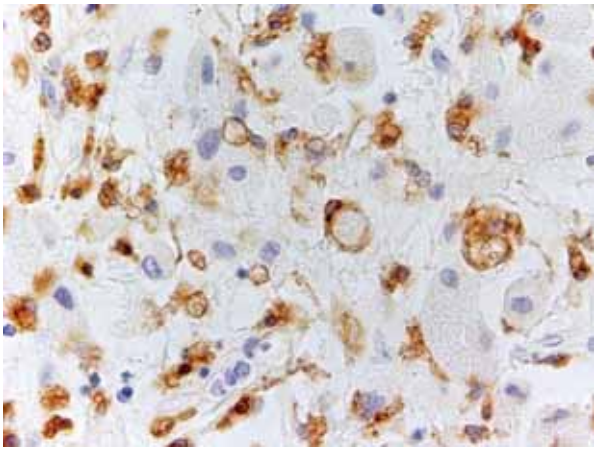

Fig. 14. Various immunolabelling for CD68 in large granular tumour cells, ×400

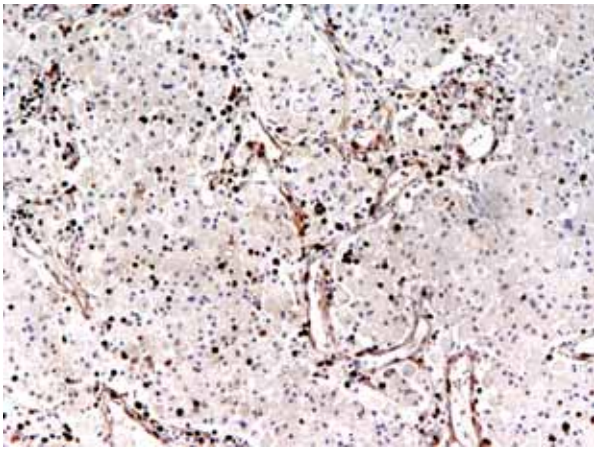

Fig. 15. Ki 67 labelling index low in tumour cells and higher in stromal cells, ×100

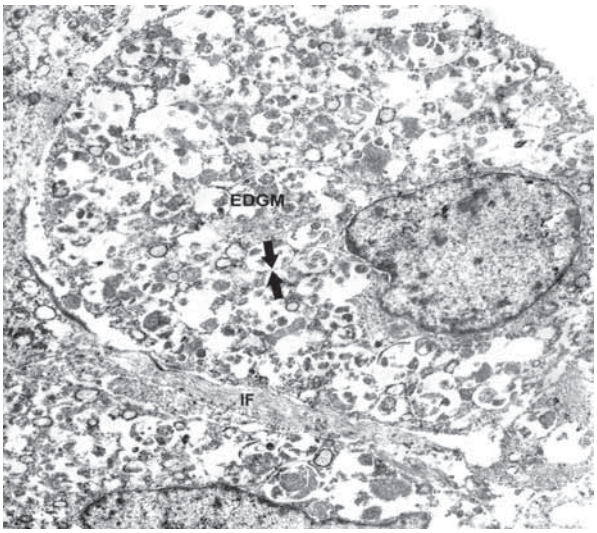

Fig. 16. Tumour cells with electron-dense granular material (EDGM) surrounded by single membrane (arrows). Between cells a bundle of intermediate filaments (IF). Orig. magn. ×4400

numerous condensed, electron-dense mitochondria with bizarre, altered shape, markedly dilated granular endoplasmic reticulum and non-numerous filaments (Fig. 21). We also observed processes filled with numerous filaments and vacuoles with electro-dense material. Macrophages with a large number of vacuoles and lamellar structures were visible among granular cells. Small blood vessels usually demonstrated normal morphology of endothelial cells although numerous collagen fibres were seen around vessels.

Discussion

Granular cell tumour (GCT) is a rare, usually benign tumour of heterogeneous origin. This lesion is most frequently encountered in the tongue, but can occur in various visceral and cutaneous sites. On histological examination, this tumour consists of typical large, rounded or polygonal cells containing an abundance of eosinophilic, PAS-positive cytoplasmic granules. On ultrastructural examination, osmiophilic granules of varied size could be observed in the cytoplasm of tumour cells.

The histogenesis of GCA has remained controversial. Most recently the tumour has been thought to be of Schwann cell origin on the basis of morphological and immunohistochemical studies [8,32]. Some authors now consider GCT as a hamartomatous lesion and others as a tumour of uncertain histogenesis.

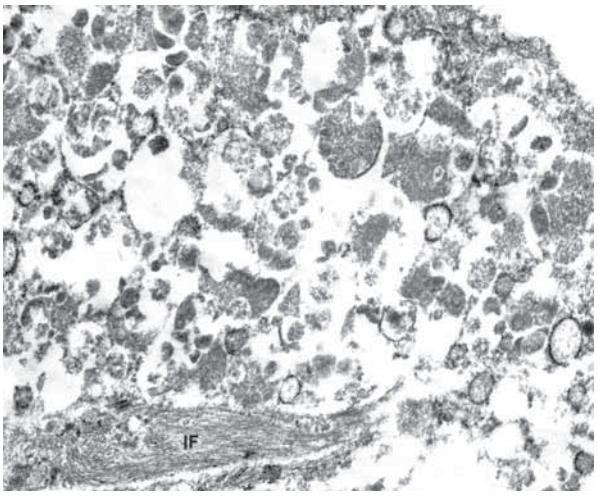

Fig. 17. Granular cell showing bundle of filaments (IF) in the cytoplasm. Orig. magn. ×7000

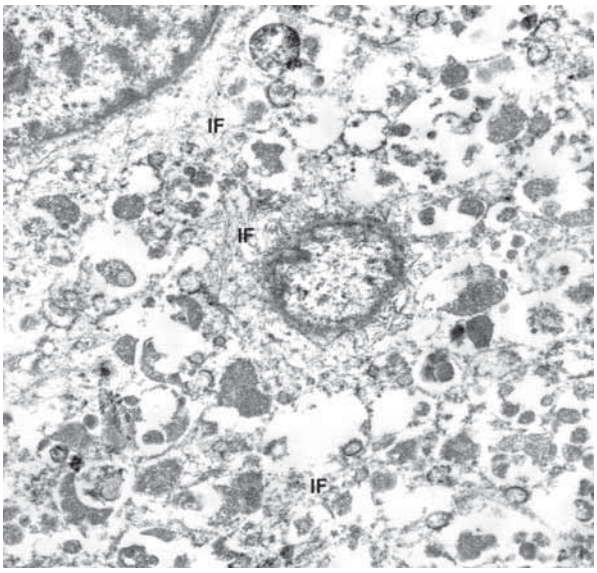

Fig. 18. Granular cell with network of filaments (IF) around the nucleus and a few filaments dispersed in the cytoplasm. Orig. magn. ×7000

It has been documented that tumour cells of intracranial and intraspinal GCTs exhibited distinct immunohistochemical and ultrastructural features that were sometimes site-depended [4,17-19,25,31]. Thus, granular cells in meningioma were immunoexpressed for epithelial membrane antigen (EMA). Ultrastructurally, desmosomes confirmed the diagnosis of meningioma [14,19]. Granular cell tumours, occurring in cranial nerves, were positive for S-100

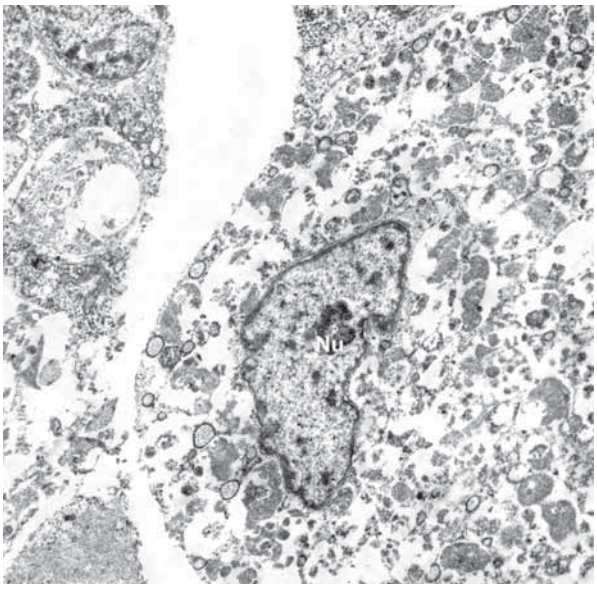

Fig. 19. Granular cell with nucleus exhibiting slightly condensed nuclear chromatin and prominent nucleoli (Nu).Orig. magn. ×4400

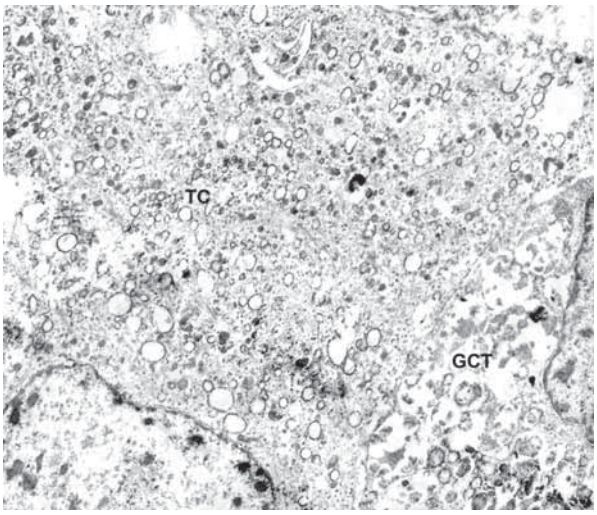

Fig. 20. Tumour cells with abundant cytoplasm (TC) and typical granular tumour cells (GCT). Orig. magn. ×4400

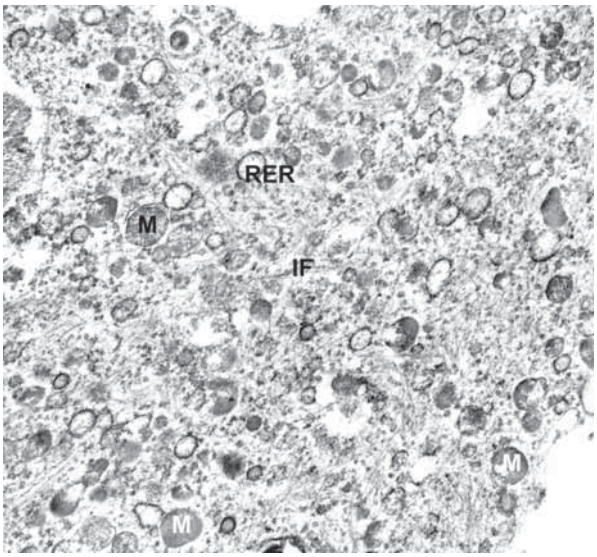

Fig. 21. Fragment of tumour cell cytoplasm showing intermediate filaments (IF), markedly dilated granular endoplasmic reticulum (RER) and mitochondria (M). Orig. magn. ×7000

protein, being among others a marker of Schwann cells [5,11]. Granular cells in medulloblastoma co-expressed neuronal immunohistochemical markers, synaptophysin and NeuN [24]. Granular cells in tumours of the pituitary stalk, hypophysis, sellar or suprasellar region and oligodendroglioma exhibited similar immunoreactivity and ultrastructural features [7,22,27,35]. Genetic alterations specific to GCA could not be found, except for a high frequency of allelic loss on 9p and 10q [6].

The present case was diagnosed as granular cell astrocytoma as the astrocytic origin of the predominant granular cell population was supported by both immunohistochemical and ultrastructural studies. The tumour demonstrated the morphological picture of granular cell tumour. It was characterized by nests of plump cells with eosinophilic granular cytoplasm, positive for PAS stain and immunoreactive for GFAP, vimentin, ubiquitin and also positive for EMA, similarly to all previously reported cases of GCA.

The Ki-67 labelling index in granular cells was low (<2). Granular cells were surrounded by blood vessels and stromal cells. Tumour necrosis was not present, but focal endothelial proliferation was observed.

The cases of GCT reported in the literature were mostly benign, regarding their histopathology and biological behaviour. It has been suggested that granular cell astrocytoma might be of any grade according to the WHO grading system. However, some GCA are biologically more aggressive than the same grade astrocytoma lacking granular cell features [13,24,30].

Ultrastructurally, the majority of tumour cells exhibited typical morphology of granular cells. Their cytoplasm was filled with numerous vacuoles with granular, electron-dense material and sometimes

with autophagosomes. Intermediate filaments corresponding with GFAP-immunoreactive glial filaments were observed in the cytoplasm of tumour cells.

References

1. Abrikossoff A. Uber Myome ausgehend von der quergestreifter willkurlichen Musculature. Virchows Arch A Pathol Anat 1926; 260: 215-233.
2. Boyce R, Beadles CF. A further contribution to the study of the pathology of the hypophysis cerebri. J Pathol Bacteriol 1893; 1: 359-383.
3. Brat DJ, Scheithauer BW, Medina-Flores R, Rosenblum MK, Burger PC. Infiltrative astrocytomas with granular cell features (Granular Cell Astrocytomas): A study of histopathologic features, grading, and outcome. Am J Surg Pathol 2002; 26: 750-757.
4. Burton BJ, Kumar VG, Bradford R. Granular cell tumour of the spinal cord in a patient with Rubenstein-Taybi syndrome. Br J Neurosurg 1997; 11: 257-259.
5. Carvalho GA, Lindeke A, Tatagiba M, Ostertag H, Samii M. Cranial granular-cell tumour of the trigeminal nerve. Case report. J Neurosurg 1994; 81: 795-798.
6. Castellano-Sanchez AA, Ohgaki H, Yokoo H, Scheithauer BW, Burger PC, Hamilton RL, Finkelstein SD, Brat DJ. Granular cell astrocytomas show a high frequency of allelic loss but are not a genetically defined subset. Brain Pathol 2003; 13: 185-194.
7. Cohen-Gadol AA, Pichelmann MA, Link MJ, Scheithauer BW, Krecke KN, Young WF, Hardy J, Giannini C. Granular cell tumor of the sellar and suprasellar region: clinicopathologic study of 11 cases and literature review. Mayo Clin Proc 2003; 78: 567-573.
8. Fisher ER, Wechsler H. Granular cell myoblastoma; a misnomer. Cancer 1962; 15: 936-954.
9. Giangaspero F, Cenacchi G. Oncocytic and granular cell neoplasms of the central nervous system and pituitary gland. Semin Diagn Pathol 1999; 16: 91-97.
10. Higgins RJ, LeCouteur RA, Vernau KM, Sturges BK, Obradovich JE, Bollen AW. Granular cell tumor of the canine central nervous system: two cases. Vet Pathol 2001; 38: 620-627.
11. Inci S, Gülşen S, Söylemezoglu F, Kansu T, Ozgen T. Intracavernous granular cell tumor. Surg Neurol 2004; 61: 384-390.
12. Jarosz B, Papierz W, Siezieniewska-Skowrońska Z, Gil K, Korobowicz E, Trojanowski T. Granular cell astrocytoma – a case report. Pol J Pathol 2004; 55 (Suppl.): 21-22.
13. Kinjo S, Yokoo H, Hirato J, Nakazato Y. Anaplastic astrocytoma with eosinophilic granular cells. Neuropathology 2007; 27: 457-462.
14. Lach B, Kanaan I. Granular cell meningioma. A case report. Folia Neuropathol 2007; 45: 19-22.
15. Lack EE, Worsham GF, Callihan MD, Crawford BE, Klappenbach S, Rowden G, Chun B. Granular cell tumor: a clinicopathologic study of 110 patiens. J Surg Oncol 1980; 13: 301-316.
16. Lee D, Suh YL, Nam do H. Cerebral granular cell tumor. Neuropathology 2008; 28: 417-421.
17. Nishio S, Takeshita I, Yoshimoto K, Yamaguchi T. Granular cell tumor of the pituitary stalk. Clin Neurol Neurosurg 1998; 100: 144-147.
18. Pasquier B, Pasquier D, Gasnier F, Lachard A, N'Golet A, Panh MH, Couderc P. [Primary intracerebral granular cell tumor of astrocytic origin. Case report and literature review.] Arch Anat Cytol Pathol 1981; 29: 215-220.
19. Pérez V, Vidal E, González N, Benavides J, Ferreras MC, Villagrasa M, Pumarola M. Orbital meningioma with a granular cell component in a dog, with extracranial metastasis. J Comp Pathol 2005; 133: 212-217.
20. Quist CF, Rivera A, Latimer KS, Goldade S, Dein FJ. Granular cell tumor in an Endangered Puerto Rican Amazon Parrot (Amazona vittata). http://www.vet.uga.edu/vpp/ivcvm/1998/quist/index.php.
21. Qureshi NA, Tahir M, Carmichael AR. Granular cell tumour of the soft tissues: a case report and literature review. Int Semin Surg Oncol 2006; 3: 21.
22. Rickert CH, Kuchelmeister K, Gullotta F. Morphological and immunohistochemical characterization of granular cells in non-hypophyseal tumours of the central nervous system. Histopathology 1997; 30: 464-471.
23. Rodriguez y Baena R, Di Ieva A, Colombo P, Collini P, Navarria P, Scorsetti M, Gaetani P, Santoro A. Intramedullary astrocytoma with granular cell differentiation. Neurosurg Rev 2007; 30: 339-343.
24. Rodriguez FJ, Scheithauer BW. Anaplastic medulloblastoma with granular cell change. Acta Neuropathol 2007; 113: 95-99.
25. Saad A, Mo J, Miles L, Witte D. Granular cell astrocytoma of the cerebellum: report of the first case. Am J Clin Pathol 2006; 126: 602-607.
26. Sahn EE, Dunlavey ES, Parsons JL. Multiple cutaneous granular cell tumors in a child with possible neurofibromatosis. J Am Acad Dermatol 1997; 36: 327-330.
27. Sakurama N, Matsukado Y, Marubayashi T, Kodama T. Granular cell tumour of the brain and cellular identity. Acta Neurochir (Wien) 1981; 56: 81-94.
28. Schulster PL, Khan FA, Azueta V. Asymptomatic pulmonary granular cell tumor presenting as a coin lesion. Chest 1975; 68: 256-258.
29. Shin E, Ki Chung C, Park SH. Granular cell astrocytoma. Path Res Pract 2007; 203: 57-62.
30. Simsir A, Osborne BM, Greenebaum E. Malignat granular cell tumor: A case report and review of the recent literature. Hum Pathol 1996; 27: 853-858.
31. Snipes GJ, Horoupian DS, Shuer LM, Silverberg GD. Pleomorphic granular cell astrocytoma of the pineal gland. Cancer 1992; 70: 2159-2165.
32. Stefansson K, Wollmann RL. S-100 protein in granular cell tumors (granular cell myoblastomas). Cancer 1982; 49: 1834-1838.
33. Weber CO. Anatomische Untersuchung einer hypertrophischen Zunge nebst Bemerkungen uber die Neubildung quergestreifter Muskelfasem. Virchows Arch A Pathol Anat 1854; 7: 115-125.
34. Yokoyama H, Kontani K, Komiyama I, Yoneyama T, Nakazawa K. Granular cell tumor of the urethra. Int J Urol 2007; 14: 461-462.
35. Yoshida T, Nakazato Y. Characterization of refractile eosinophilic granular cells in oligodendroglial tumors. Acta Neuropathol 2001; 102: 11-19.