

Subependymal giant cell astrocytomas with atypical histological features mimicking malignant gliomas

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Abstract

Subependymal giant cell astrocytoma (SEGA) is a rare, benign brain tumour developing in patients with tuberous sclerosis complex (TSC). Typical histopathological findings of this neoplasm are solid sheets and perivascular pseudorosettes of large, gemistocytic, polygonal and occasionally ganglion-like cells within a fibrillated background, accompanied by spindle-shaped cells creating broad fascicles. Rich vascular stroma, and numerous calcifications are common. Mitoses, focal necrosis, and endothelial proliferation are rarely encountered.

In this study we report 3 cases of SEGA out of 29 TSC patients, operated on in the Department of Neurosurgery, Children's Memorial Health Institute, from 1990 to 2011 and retrospectively reviewed. These 3 cases exhibited distinct anaplastic features that might mimic malignant glioma. Histologically, the tumours were composed of pleomorphic, gemistocytic, polygonal, ganglion-like or multinucleated cells arranged in sheets or forming perivascular pseudorosettes. Numerous foci of necrosis, microvascular proliferation and high mitotic activity with atypical mitotic figures were documented. The Ki67 labelling index was about 15-20%. These tumours might be confused with high-grade gliomas and such a misleading diagnosis might result in aggressive radio- or chemotherapy. Despite the common statement that morphological features of anaplasia, i.e. pleomorphism, necrosis, microvascular proliferation and increased mitotic activity, are not of prognostic value, the tumour behaviour in our cases seems to be more aggressive and requires longer follow-up studies. We suggest recognizing such cases of subependymal giant cell astrocytoma as atypical SEGAs.

Key words: *subependymal giant cell astrocytoma (SEGA), tuberous sclerosis complex, histopathology, anaplastic features, atypical SEGA.*

Introduction

Subependymal giant cell astrocytoma (SEGA) is a rare, low-grade intraventricular tumour represent-

ing about 1-2% of all paediatric brain tumours [5,6,25]. SEGAs occur almost exclusively in patients with tuberous sclerosis complex (TSC) [8,9,12]. TSC is a neurocutaneous disorder occurring in 1 : 6000 live births. It

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is characterized by the development of benign, highly vascular, hamartomatous tumours in various tissues and organs, including brain, kidneys, heart, liver, lungs, retina, and skin. Tuberous sclerosis complex is caused by inactivating mutations in either of two genes: the *TSC1* or *TSC2* gene [6]. *TSC1* is located on 9q34 and encodes a protein called hamartin whereas *TSC2* is located on 16p13 and encodes tuberin [23]. Both proteins form a heterodimer that blocks the activity of the main mammalian regulator of cell growth and proliferation, mTOR kinase. If either tuberin or hamartin is lost, as in cases of TSC, the activity of mTOR is high, enabling cell growth and proliferation [3,23]. Loss of heterozygosity in the *TSC1* or *TSC2* gene occurs in most angiomyolipomas, rhabdomyomas, and SEGAs from TSC patients [14,15].

The most frequent tumours in TSC include SEGAs, facial angiofibromas, cardiac rhabdomyomas, and renal angiomyolipomas. Subependymal giant cell astrocytomas present the major cause of morbidity and mortality among children and adolescents with TSC. They are considered to be histologically benign, typically arising in the wall of the lateral ventricles, but tend to grow and may obstruct cerebrospinal fluid

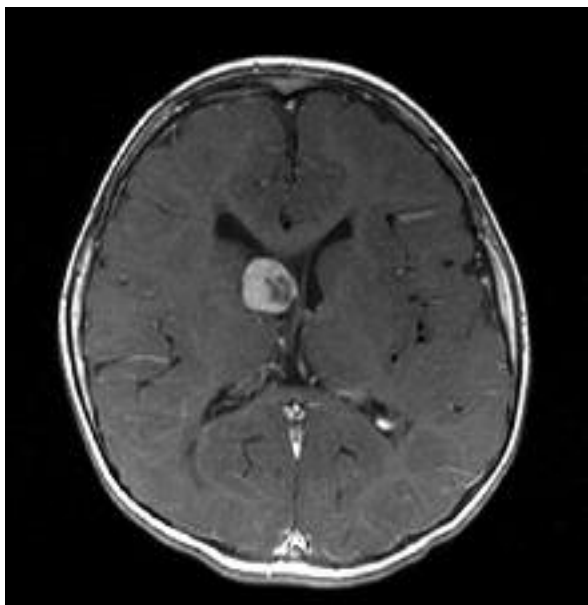


Fig. 1 Axial post-contrast T1-weighted image with a large, well-demarcated, intensely enhanced mass protruding into the left lateral ventricle. The tumour is causing ventricular enlargement. Secondary small nodules are observed in the right lateral ventricle.

pathways, causing hydrocephalus [20]. SEGAs correspond to WHO grade I [25] with a low proliferative labelling index [31]. They usually develop in the first two decades of life, and can be found even in fetuses and newborns [26,27,30,36]. SEGAs can be revealed on neuroimaging as tumours located on the surface of lateral or rarely the third ventricle [28].

In this study we report 3 patients with SEGA out of 29 TSC patients, operated on in the Department of Neurosurgery, Children's Memorial Health Institute, from 1990 to 2011 and retrospectively reviewed. These 3 cases exhibited histological anaplastic features that mimic malignant gliomas.

Clinical presentation

Case 1

The patient at the age of 4 months was admitted to the Neurosurgical Department. The first brain MRI revealed multiple cortical and subcortical tubers and subependymal nodules, including an intraventricular tumour mass with a maximum diameter of 2.5 cm. The multiple cardiac tumours and multiple hypomelanotic macules confirmed the diagnosis of TSC. After 2 years follow-up, the brain MRI showed enlargement of the intraventricular tumour to 3.8 cm in diameter, with no features of hydrocephalus. One year later the tumour increased to 4.8 cm in longest diameter, leading to hydrocephalus (Fig. 1). The child underwent total resection of the tumour. Histopathological examination indicated SEGA.

Case 2

In the second patient at the age of 3 years, the first brain MRI revealed cortical and subcortical tubers and an intraventricular tumour of diameter 2 cm. The child was diagnosed as having TSC as he had multiple cardiac tumours and multiple cysts in the kidneys. His father suffered from TSC. Follow-up brain MRI performed at the age of 7 years showed an intraventricular tumour of maximum diameter 5.5 cm and hydrocephalus (Fig. 2A-B). Total tumour removal was performed with the diagnosis of SEGA.

Case 3

The patient was diagnosed prenatally as having TSC, based on prenatal MRI showing multiple cardiac tumours and a large intraventricular brain tumour.

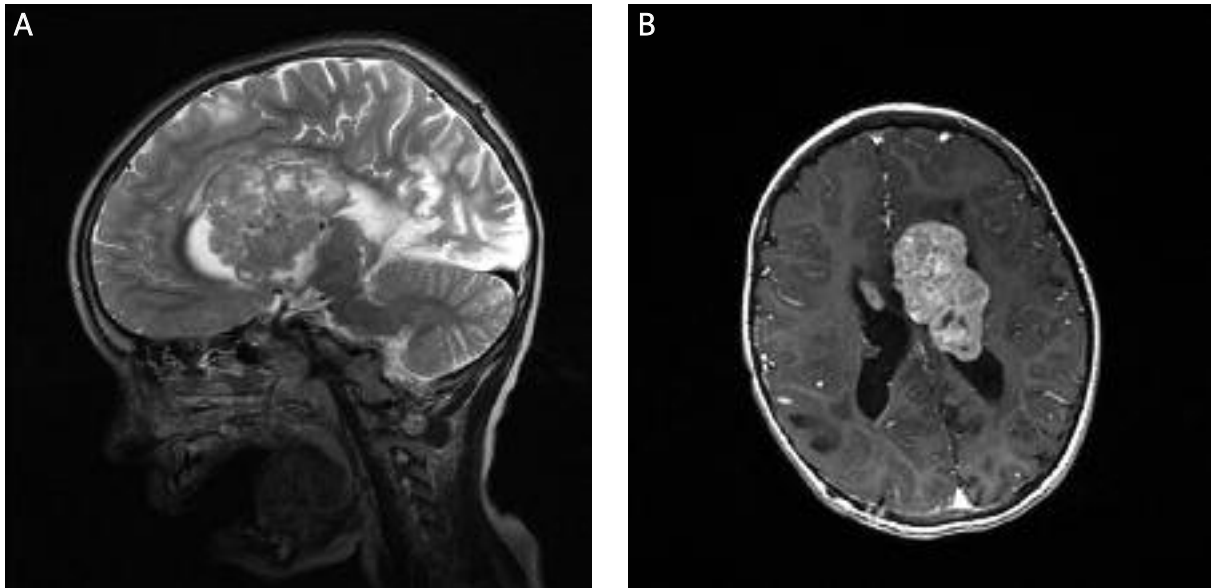


Fig. 2. **A)** Sagittal T2-weighted image shows a huge heterogeneous tumour at the foramen of Monroe extending into the left ventricle. **B)** Axial T1-weighted image after contrast administration. Nearly homogeneously enhancing tumour at the right foramen of Monroe is seen.

The longest diameter of the brain tumour was 3 cm. One week after birth, the child had shunt implantation due to hydrocephalus. When the patient was 7 months old, follow-up MRI examination revealed enlargement of the intraventricular tumour and cortical/subcortical tubers (Fig. 3). The longest diameter of the tumour was 4 cm. The child underwent total resection of the tumour and SEGAs diagnosis was established.

Material and methods

The retrospective analysis of biopsy material diagnosed as SEGAs from 29 cases of TSC patients was performed. Formalin-fixed and paraffin-embedded tumour tissue was routinely stained with haematoxylin and eosin (H&E). To determine the phenotype of tumour cells, immunohistochemical studies were performed according to the labelled avidin-biotin complex (ABC) method with 3-3'-diaminobenzidine (DAB) as a chromogen, using antibody to: glial fibrillary acidic protein (GFAP, polyclonal, dilution 1 : 5000), synaptophysin (dilution 1 : 200), neurofilament proteins (dilution 1 : 100), S-100 protein (polyclonal; 1 : 800) and Ki67 antigen (dilution 1 : 100). All antibodies were from Dako, Glostrup, Denmark.

Neuropathological and immunohistochemical findings

Histopathologically, all 26 cases of SEGAs revealed typical morphological features with solid sheets and

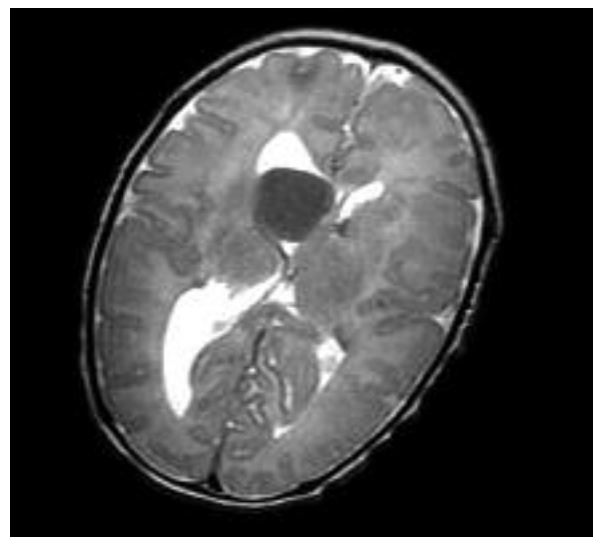


Fig. 3. Axial T2-weighted image shows a large hypointense mass located at the right foramen of Monroe. A small hypointense lesion at the left foramen of Monroe is seen. A cortical/subcortical tuber in the right frontal lobe is also visible.

perivascular pseudorosettes composed of large, gemistocytic, and polygonal cells with abundant glassy eosinophilic cytoplasm and fibrillary background. The spindle-shaped cells of the tumour sometimes were arranged in broad fascicles. No mitotic figures or necrosis were found. The Ki67 proliferative index was low, about 1-2%.

Three cases of SEGAs exhibited an atypical histopathological pattern with anaplastic features. The neoplastic tissue was composed of large, gemistocytic, polygonal, sometimes multinucleated cells arranged in sheets (Fig. 4A) or perivascular pseudorosettes. The population of spindle-shaped cells often created broad fascicles. Some neoplastic cells displayed abundant cytoplasm with eccentrically located nucleus/nuclei with prominent nucleoli, often resembling ganglion-like cells with mitotic figures (Fig. 4B). The tissue revealed numerous small foci and large areas of necrosis (Fig. 4C-D), occasionally with pseudopalisading of neoplastic cells (Fig. 4E). Mitotic figures were common, sometimes up to 2-3/1HPF, including atypical mitoses (Fig. 4B). Additionally, the proliferation of microvessels could be detected (Fig. 4F). The majority of SEGA cells were strongly positive for glial fibrillary acid protein (GFAP) and S-100 protein (Fig. 5A-B). They also displayed immunoreactivity of neuronal markers such as neurofilament proteins and synaptophysin (Fig. 5C-D). The proliferative Ki67 labelling index was high, focally about 15-20% (Fig. 5E-F).

Discussion

Brain lesions in TSC include cortical tubers, subependymal nodules (SEN) and subependymal giant cell astrocytomas [5,6,8,9,22,24,25,32]. SEGAs are slowly growing tumours arising in the ependymal layer lining the ventricular walls. They have a tendency to grow near the foramen of Monro and obstruct the flow of cerebrospinal fluid, leading to hydrocephalus. These tumours are pathognomonic for TSC, but sometimes they occur in patients without stigmata of tuberous sclerosis complex [13,17,18,33].

Recent studies of TSC showed several protein cascades that might be involved in the pathogenesis of the disease. Proteins encoded by genes *TSC1* and *TSC2*, hamartin and tuberin, respectively, form a heterodimer which suppress the mammalian target of rapamycin (mTOR), a major cell growth and proliferation controller. The mechanism of tumourigenesis in TSC is inactivation of the hamartin-tuberin complex

after phosphorylation by various kinases, such as extracellular signal-regulated kinase (ERK), which was found in subependymal giant cell astrocytomas and dysplasia of Taylor's balloon cell type [10]. Some other genes implicated in tumourigenesis and nervous system development, i.e. *ANXA1*, *GPNMB*, *LTF*, *RND3*, and *NPTX1*, are likely to be mTOR effector genes in SEGA, as their expression was modulated by the mTOR inhibitor rapamycin in SEGA-derived cells [21,37]. The discovery of mTOR pathway upregulation in tuberous-sclerosis-associated tumours presents new possibilities for strategies of treatment.

Histologically, SEGAs are composed of large cells with abundant cytoplasm. The neoplastic cells can also be polygonal, epithelioid and spindle shaped or ganglion-like [1,2]. They were arranged in sheets, clusters, or pseudorosettes. The characteristic feature of SEGAs is rich vascular stroma [19] and parenchymal or vascular calcifications. The cytoplasm of SEGA cells is usually strongly positive for glial fibrillary acid protein (GFAP) and S-100 protein and numerous cells are also positive for neuronal markers, i.e. neurofilament proteins (NF) and synaptophysin [2,16,29,34,38]. Because of the glioneuronal character of these tumours, the term "subependymal giant cell tumours" (SEGTs) has been suggested by some authors. This may reflect their postnatal origin from neural progenitors that are normally resident in the subependymal zone [7].

The majority of SEGAs are histologically benign and cellular pleomorphism, mitoses and areas of necrosis are seen only sporadically. In the 3 presented cases from 29 SEGAs in our cohort we have found distinct features of anaplasia. In these patients, SEGAs developed in the first years of life and were accompanied by other clinical manifestations of TSC. In 1 case the tumour was discovered on MRI examination prenatally. The brain tumours were large, with the longest diameter ranging from 4 to 5.5 cm. The patients experienced symptoms of acute hydrocephalus before the age of 7 years that acquired surgical tumour resection. In these 3 cases, the neoplastic tissue of SEGA displayed numerous foci of necrosis with pseudopalisading, frequent mitotic figures and microvascular proliferation. In contrast to the majority of SEGAs, our cases revealed a high Ki67 labelling index in the range of 15-20%. Such anaplastic features might be a cause of misdiagnosis with malignant gliomas, especially upon intraoperative examination. However, in the majority of cases reported so

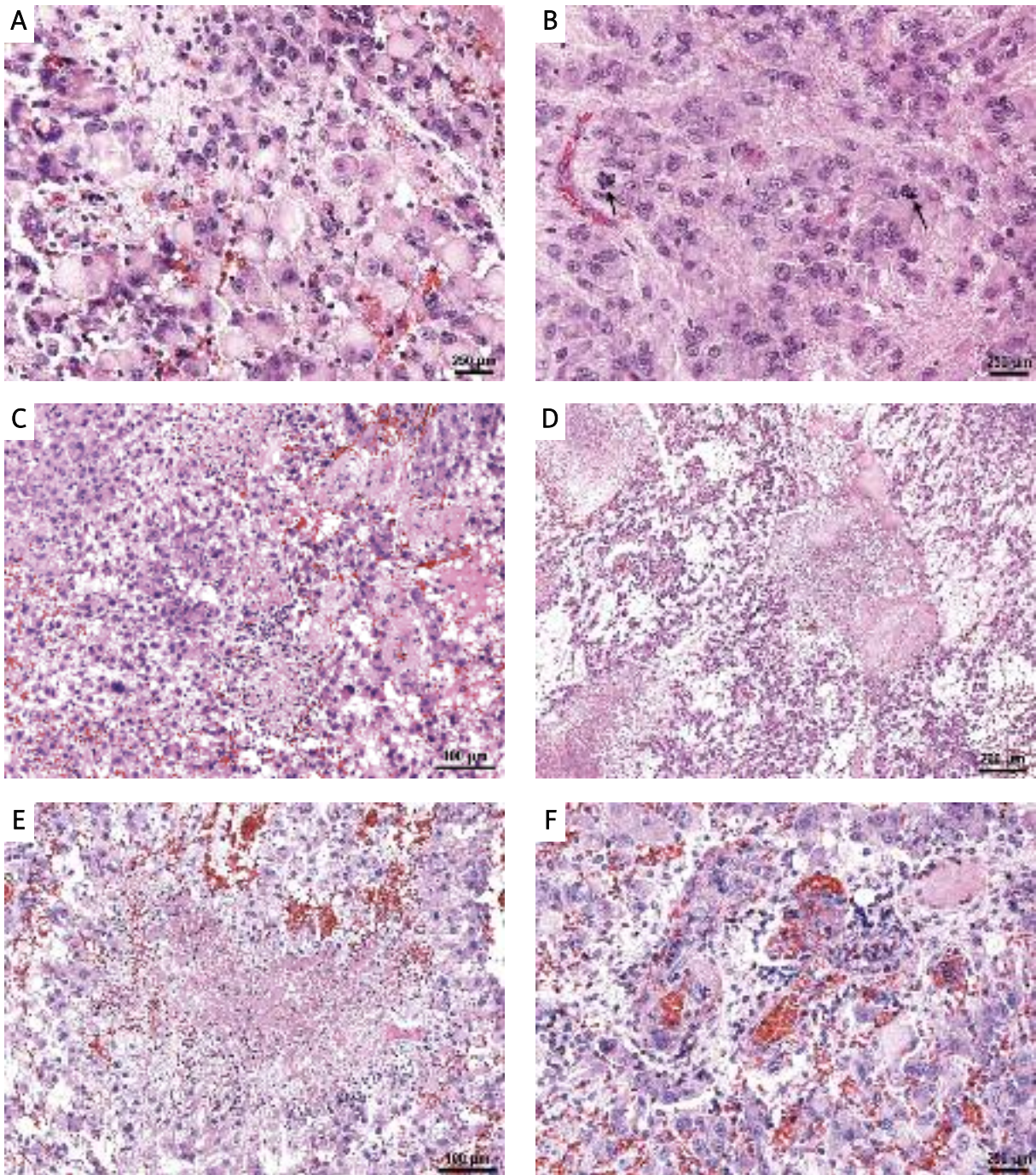


Fig. 4. Histopathology of SEGAs. **A)** Tumour composed of gemistocytic and ganglion-like cells, H&E. **B)** Sheets of polygonal cells with abundant eosinophilic cytoplasm, eccentrically located nucleus and mitotic figures (arrows), H&E. **C)** Small foci of necrosis, H&E. **D)** Large areas of necrosis, H&E. **E)** Focus of necrosis with pseudopalisading arrangement of neoplastic cells, H&E. **F)** Area with prominent vascular proliferation, H&E.

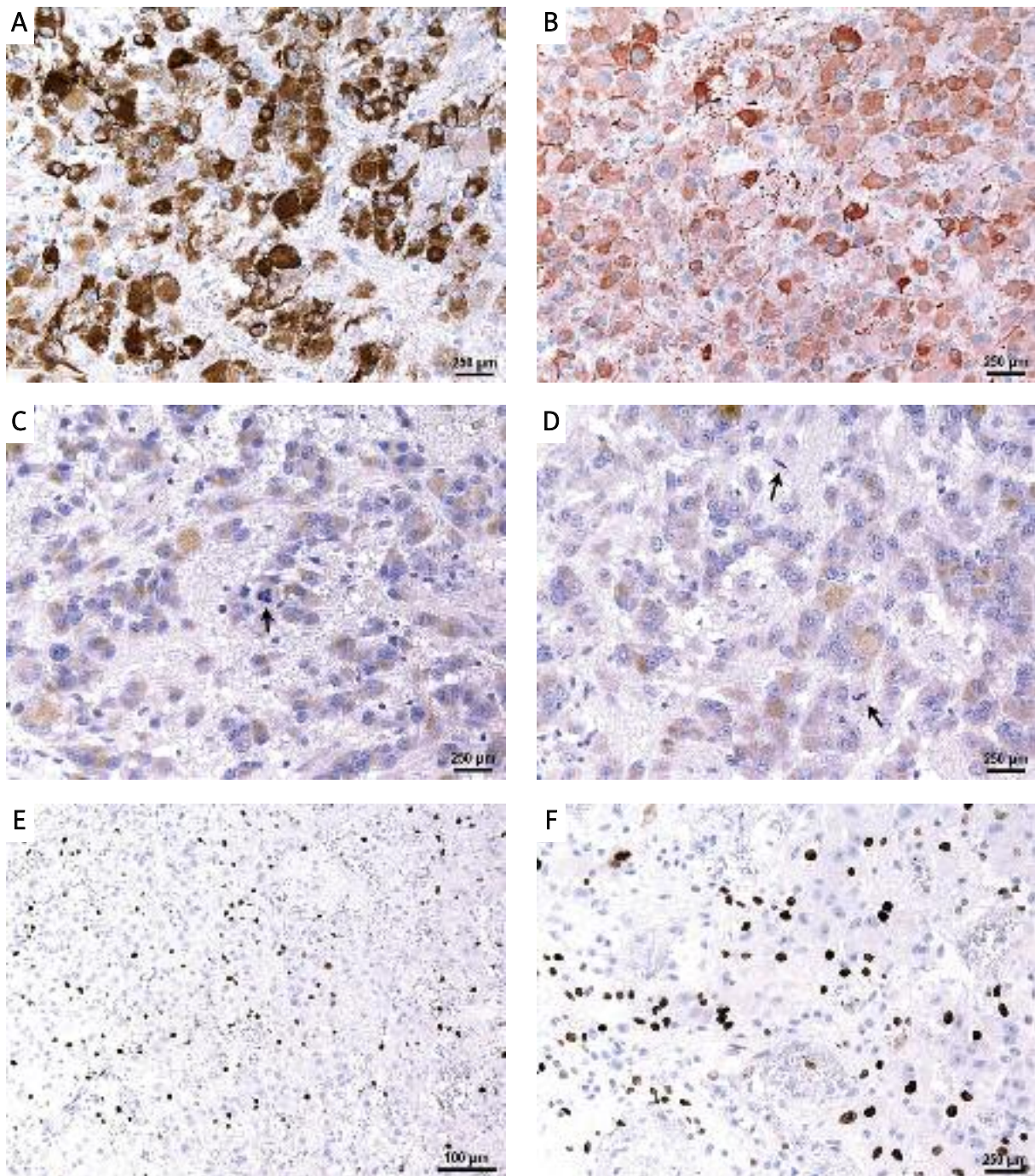


Fig. 5. Immunohistochemistry of SEGA. **A)** Numerous neoplastic cells with strong GFAP immunostaining. **B)** Diffuse GFAP immunoreactivity. **C)** Tumour cells immunopositive for synaptophysin with atypical mitotic figure (arrow). **D)** Weak synaptophysin immunoreactivity and two mitotic figures (arrows). **E)** Ki67 nuclear immunopositivity in numerous neoplastic cells. **F)** Focus with increased Ki67 labelling index.

far, the anaplastic features of SEGAs have no prognostic value, although in some cases the neoplastic growth is rapid and causes hydrocephalus, visual loss, and death [11,20,35]

The correlation between clinical course and histopathological features of SEGAs is not fully established [4]. It is well known that SEGAs usually develops between the first and second decade of life. Very few prenatally diagnosed SEGAs have been reported and in most cases their outcome was poor. It is also known that SEGAs grow slowly and the prognosis is good. Follow-up brain MRI examinations are recommended every 2 years in patients with TSC2 mutation and every 3 years in patients with TSC1 mutation. In our cases, the tumours grew more rapidly but longer follow-up ought be established to confirm their more aggressive nature.

In conclusion, our cases emphasize the occasional occurrence of anaplastic features in SEGAs, which might cause some diagnostic confusion with malignant gliomas. We suggest that cases with such unique morphological characteristics might be termed atypical SEGAs.

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