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Original Contribution

Analysis of 24 cases of epithelioid glioblastoma: Experience from a tertiary centre of North India

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ABSTRACT

Background: Epithelioid glioblastoma (eGB) is a recently recognized and a rare variant of glioblastoma. This study aimed to describe the clinical, histological and immunohistochemical spectrum and outcome of eGB from a tertiary care hospital in north India.

Materials and methods: Twenty four cases of eGB diagnosed over past 10 years were reviewed with detailed morphological and immunohistochemical analysis (GFAP, EMA, Vimentin, Myogenin, INI-1, Cytokeratin, Synaptophysin, CD99, S100, MelanA, IDH1, ATRX, p16, EZH2, Ki-67, and BRAF V600E mutant antibody).

Result: The mean age was 29.9 years (3–54 years), with equal male and female patients. All had supratentorial tumor. All cases showed epithelioid cells in sheets; however, focal spindling (7 cases, 29.2%), grouping/nesting (6 cases, 25%) and papillary configuration (5 cases, 20.8%) were also noted. All showed microvascular proliferation (MVP) and all except one demonstrated areas of necrosis. INI1 was retained in all cases, while 2 showed patchy loss. EZH2 overexpression (>25%) was observed in 4 cases, while 5 cases showed loss of p16 expression. BRAF V600E mutant protein expression was seen in 12/23 (52.2%) cases. Outcome was available in 8 cases, out of which 6 (75%) experienced recurrence. The median survival was 25.5 months. Cases with tumor infiltrating lymphocytes had a better outcome.

Conclusion: eGB is a distinct variant of glioblastoma which has predilection towards younger age group. It shows high percentage of BRAF V600E mutation and a subset of it shows longer survival. Cases with presence of tumor infiltrating lymphocytes are associated with better outcome.

1. Introduction

Glioblastoma (GB) is the commonest brain tumor worldwide. It is grade IV tumor according to World health organization (WHO) classification of central nervous system (CNS) tumors [1]. Glioblastoma carries uniformly poor prognosis despite various forms of therapy. Histologically GB is known to show wide variation in morphology and heterogeneous growth pattern. Currently, three morphological variants of GB have been recognized, i.e- giant cell glioblastoma, gliosarcoma and epithelioid glioblastoma (eGB) [1]. These variants have characteristic histological features and show molecular signature different from conventional GB.

Epithelioid glioblastoma (eGB) is a relatively recently recognized and a rare variant of GB [2,3]. The average age of onset is lower than the conventional GB [4]. It may be multifocal and shows sharp demarcation on neuroimaging. Histologically, this tumor is characterized by cohesive sheets of medium to large size, round to polygonal, epithelioid cells with eosinophilic cytoplasm. The cells are non-lipidized and usually lack cytoplasmic process. Histological features of grade IV tumor like frequent mitosis, necrosis and endovascular proliferation are commonly found in eGB [1]. As this tumor shows a cohesive growth pattern and is composed of epithelioid cells, it can be confused with other neoplasms including metastatic carcinoma. The correct diagnosis of eGB depends on demonstration of glial fibrillar acid protein (GFAP) within the tumor cells by immunohistochemistry (IHC) and excluding other histologic mimics.

eGB is characterized by *BRAF V600E* mutation (1799 T>A) as first demonstrated in 2013 [5]. This mutation is found in approximately 50%

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of the eGB cases. Previously eGB was considered a morphological pattern of GB. As eGB shows distinct clinical, histological and molecular features, it is no more considered a morphological pattern of GB, rather a variant [1]. Demonstration of BRAF mutant protein expression by immunohistochemistry can be useful marker to diagnose eGB [6].

Being a rare tumor, the histological and immunohistochemical features of eGB are not widely described. The available published information is mostly in the form of single case report or a short series of cases [5-9]. A wide range of immunohistochemistry is mandatory to establish the diagnosis and differentiate it from its histological mimics. This study aimed to describe the clinical, histological, immunohistochemical spectrum and outcome of eGB from a tertiary care hospital in north India.

2. Materials and methods

In this retrospective study, all cases diagnosed as glioblastoma over past 10 years were retrieved from the database of Department of Histopathology, PGIMER, Chandigarh and reviewed by two neuropathologists. Cases of GB with dominant (>50%) epithelioid cell component and which met other diagnostic criteria of eGB were included in this study. Slides were thoroughly evaluated to exclude a diagnosis of anaplastic pleomorphic xanthoastrocytoma (APXA). APXA was considered if any of the following feature was present: presence of classical low grade PXA areas, reticulin rich stroma, presence of foam cells and presence of multiple refractile granular eosinophilic bodies [10]. The clinical details including age, location of the tumor, demographic profile were noted from the patient's file. Cases where slides/paraffin blocks were unavailable were excluded from the study.

All the slides were examined with detailed morphological evaluation. The biopsies were assessed for the pattern of growth, percentage of epithelioid cells, small cell component, presence of giant cells, necrosis, microvascular proliferation (MVP), other morphological patterns (papillae, spindling, nesting/grouping), perivascular lymphoid infiltrate and tumor infiltrating lymphocytes (TILs). TILs were classified as absent (no TILs), non-brisk (focal presence of TILs) and brisk (diffuse presence of TILs with focal aggregation) as used for melanoma on semi-quantitative assessment [11]. Reticulin stain was performed in all cases (using Gomori's modification of Gordon- Sweet technique).

Immunohistochemistry (IHC) was carried out on at least one representative tissue block. The following panel of antibodies was used: glial fibrillar acid protein (GFAP, DAKO, dilution 1:50), Epithelial membrane antigen (EMA, Cell Marque, 1:300), Vimentin (Cell Marque, 1:300), Myogenin (Dako, 1:50), INI-1(Cell Marque, 1:100), Cytokeratin (Cell Marque, 1:300), Synaptophysin (SNP, OSB, dilution 1:300), Smooth muscle actin (SMA, Dako, 1:200), CD99 (Dako, 1:100), S100 (Cell Marque, 1:200), MelanA (Dako, 1:50), IDH1 R132H (Dianova, dilution 1:40), ATRX (Sigma, dilution 1:300), p16 (Ventana, ready to use), EZH2 (Cell Marque, 1:50), Ki-67 (OSB, dilution 1:300) and BRAF mutant antibody (VE1 clone, Ventana). All IHCs were performed on Ventana autoimmunostainer.

Cases showing cytoplasmic positivity for IDH1 in >10% tumor cells were considered positive. Ki-67 proliferation index was determined by counting 10 high power fields (400× magnification) at the highest proliferating area and was expressed in percentage. A case of papillary thyroid carcinoma with known *BRAF* V600E mutation was used as external control for BRAF immunohistochemistry. For EZH2, nuclear staining was graded as negative (<5%), weak expression (5–25%) and strong expression (>25%) [7]. For p16, less than 1% cells expressing p16 was considered as loss of expression [12].

Follow up information of the patients were obtained from the department of Radiotherapy. Treatment detail, course of disease and outcome in terms of recurrence or death were noted for the available patients.

3. Results

Within this time period, 24 cases met the diagnostic criteria of eGB out of total 1900 glioblastoma diagnosed over this period (1.3%).

3.1. Clinical detail

The average age was 29.9 years (range 3 to 54 years). Eight cases (33.3%) were of pediatric age group (<18 years). There were equal number of male and female (12 each) patients (M:F ration 1:1). All the cases were located supratentorially. Twenty one cases were cortical-subcortical in origin (Fig. 1), two cases involved corpus callosum and one involved thalamus (Table 1). All patients underwent gross total excision of the tumor.

3.2. Histological features (Figs. 2 and 3, Table 1)

Most of the cases showed diffuse sheets of malignant astrocytic cells arranged in a discohesive manner (Fig. 2A). All cases showed variable number of epithelioid astrocytes (range 50–90%). These epithelioid astrocytes were large, round to polygonal cells with abundant eosinophilic cytoplasm. These cells showed moderate nuclear pleomorphism, coarse nuclear chromatin and frequent mitosis. Tumor cells in one case focally showed clear cytoplasm. There was no intracytoplasmic pigment. No case showed presence of lipidized or foam cells. Giant cells were seen in 14 cases (58.3%), which contributed 2–10% of the tumor cellularity. Seven cases (29.2%) also showed foci of undifferentiated small cells with high N:C ratio (Fig. 2B), which occupied <10% of the total tumor volume. Focal papillary pattern of growth was identified in five (20.8%) cases. Presence of cohesive grouping or nesting of the tumor cells was observed in 6 cases (25%) and one case each showed cord and alveolar pattern of growth. Focal (<10%) spindling of tumor cells was observed in 7 cases (29.2%), which didn't fulfill the criteria of gliosarcoma. Myxoid change in the stroma was noted in two cases (Figs. 2C–F and 3A). In two cases, rhabdoid cells with presence of intracytoplasmic inclusion were identified focally (<5%). Microcystic changes (3 cases) and calcification (2 cases) were infrequent observations.

All cases showed microvascular proliferation (MVP) and all except one showed areas of necrosis. Twenty cases showed large geographic areas of necrosis, while it was focal in 3 cases and one case didn't show necrosis. The tumor was infiltrating the adjacent brain parenchyma as cohesive sheets around the Virchow-robin spaces (Fig. 3B). Perivascular lymphocytic infiltrate was seen in 12 cases (50%). TILs were observed in 6 cases (25%), among which 4 were non-brisk and 2 were brisk (Fig. 3C–D).

3.3. Immunohistochemistry (Fig. 4A–H)

All cases except one were positive for GFAP. GFAP positivity was strong, but the extent of staining was ranging from focal to diffuse. One case was completely negative for GFAP. All cases showed diffuse and strong S100 positivity. Vimentin positivity was noted in 12 (50%), synaptophysin in 4 (16.6%), CD99 in 7 cases (29%, diffuse and membranous). All cases except two showed complete INI1 retention. These two cases had small cell component which showed patchy loss of INI1 expression, while it was retained in their epithelioid cell component. All cases were negative for IDH1 and ATRX was retained in all. All cases were negative for cytokeratin, SMA and Melan A. EMA positivity was seen in eight (33.3%) cases as patchy membranous staining. BRAF V600E mutant protein cytoplasmic positivity was seen in 40–100% of cells in 12/23 (52.2%) cases. The staining was faint granular in 3 cases and dense granular in 9 cases (Fig. 4).

EZH2 and p16 IHC was performed in 17 cases. EZH2 strong expression was observed in 4 cases (23.5%). The staining intensity was weak to moderate, only one case showed strong nuclear positivity. Loss of p16 expression was observed in 5 cases (29.4%). There was no

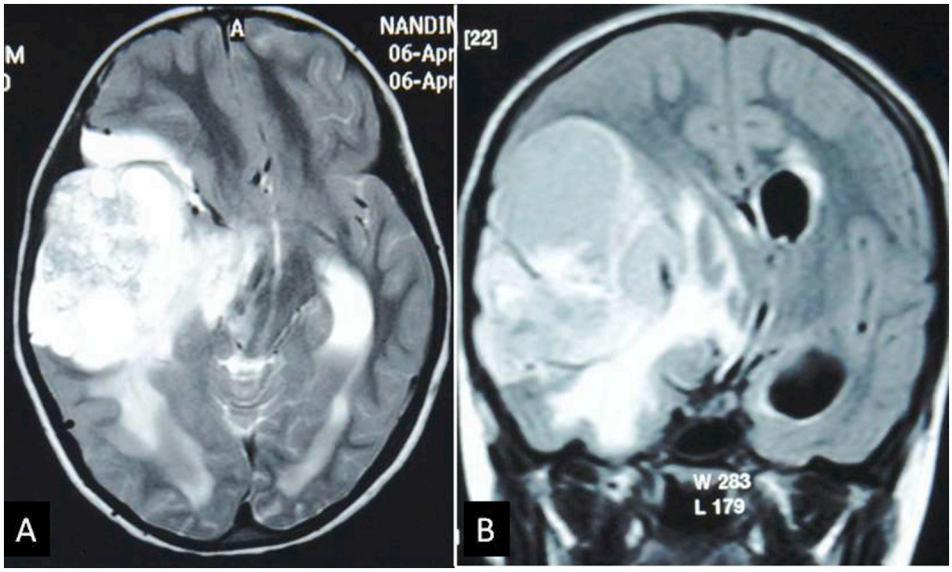

Fig. 1. (A and B) Magnetic resonance imaging showing a large heterogeneously enhancing mass involving parietal and temporal lobes.

Table 1
Clinical and histological features of patients of epithelioid glioblastoma:

Case no.	Age (years)	Gender	Location	% epithelioid cells	Papillae	Grouping/nesting	Spindling	Necrosis	MVP
1	15	Female	Cerebral cortex	80	No	Yes	No	Present	Present
2	14	Male	Left parietal	50	No	Yes	No	Present	Present
3	15	Female	Temporal	80	Yes	No	No	Focal	Present
4	10	Female	Occipital horn of lateral ventricle	50	Yes	No	Yes	Extensive	Present
5	11	Male	Left frontal	50	No	No	No	Absent	Present
6	24	Female	Left temporo-parietal	70	No	No	No	Present	Present
7	45	Male	Right thalamic	50	No	No	No	Present	Present
8	40	Male	Corpus callosum	80	No	No	No	Present	Present
9	34	Male	Left frontal	60	No	Yes	Yes	Present	Present
10	21	Male	Cerebral cortex	50	No	No	No	Extensive	Present
11	33	Female	Temporal	70	Yes	Yes	No	Focal	Present
12	47	Male	Left temporo-parietal	80	No	Yes	No	Present	Present
13	25	Male	Left parietal	70	Yes	No	No	Focal	Present
14	10	Female	Right temporal	80	Yes	No	No	Extensive	Present
15	46	Male	Left frontal	80	No	No	Yes	Extensive	Present
16	3	Female	Left parietal	90	No	No	No	Present	Present
17	54	Female	Corpus callosum	70	No	No	No	Extensive	Present
18	52	Male	Right temporal	80	No	No	Yes	Extensive	Present
19	41	Female	Right parieto-occipital	60	No	No	Yes	Extensive	Present
20	47	Female	Right parieto-occipital	70	No	Yes	No	Present	Present
21	36	Male	Right medial temporal with insular extension	80	No	No	No	Extensive	Present
22	10	Female	Frontal	80	No	No	No	Extensive	Present
23	41	Male	Left parietal	80	No	No	Yes	Extensive	Present
24	44	Female	Left temporal	80	No	No	Yes	Present	Present

MVP - microvascular proliferation.

morphological difference between cases with or without p16 expression. There was no correlation between EZH2 strong expression and loss of p16 expression.

3.4. Outcome

Treatment detail and outcome was available in 8 cases (Table 2). Rest of the cases were either lost to follow up or had moved to other institutions for further treatment. The follow up period ranged from 5 to 41 months. All patients received post-operative radiotherapy. Six patients (75%) experienced recurrence. The mean interval of recurrence was 19.8 months. Five patients died and three were alive at the time of last follow up. The mean overall survival (OS) was 24.5 months, and median OS was 25.5 months. There was no significant effect of BRAF expression, EZH2 strong expression, loss of p16 expression or necrosis on survival. However, cases with TILs had a higher OS (33.3 months) than cases without TIL (19.2 months) but due to small number of cases a statistical inference could not be drawn.

4. Discussion

Epithelioid GB is a rare variant of glioblastoma, which is now recognized as a separate clinicopathological entity. It is incorporated in 2016 WHO classification of brain tumors as a subtype of glioblastoma [1]. It is characterized by the presence of large epithelioid cells with abundant voluminous cytoplasm and prominent nucleoli. There is no cut off of epithelioid cells for diagnosis of eGB [1]. Many studies have used variable minimum percentage of epithelioid cells for diagnosis of eGB, ranging from 30 to 50% [5,8,13]. According to WHO classification, epithelioid cells should be predominant cell population in eGB [1]. In our experience, small percentage of epithelioid cells are not uncommonly encountered in glioblastomas. We have included only those cases

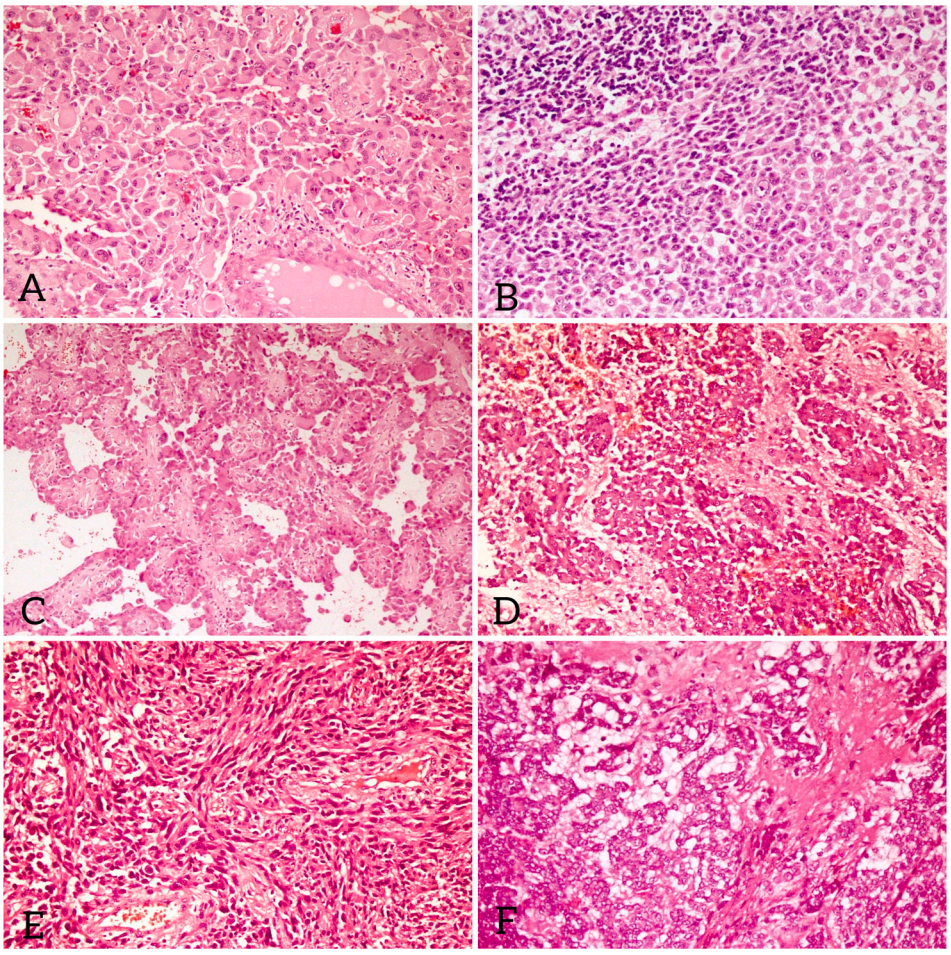

Fig. 2. (A) Photomicrograph showing diffuse sheets of epithelioid cells in a discohesive manner (hematoxylin and eosin, ×200).
(B) One case showing epithelioid cells (right half) admixed with undifferentiated small, round cells (left upper part) (hematoxylin and eosin, ×200).
(C) Epithelioid glioblastoma showing papillary architecture (hematoxylin and eosin, ×200).
(D) Epithelioid glioblastoma infiltrating the adjacent brain in tight clusters (hematoxylin and eosin, ×200).
(E) Spindling of tumor cells in a case of eGB (hematoxylin and eosin, ×200).
(F) Case of eGB showing tumor cells arranged in cords and trabeculae in a myxoid background resembling adenoid GB (HE, ×200).

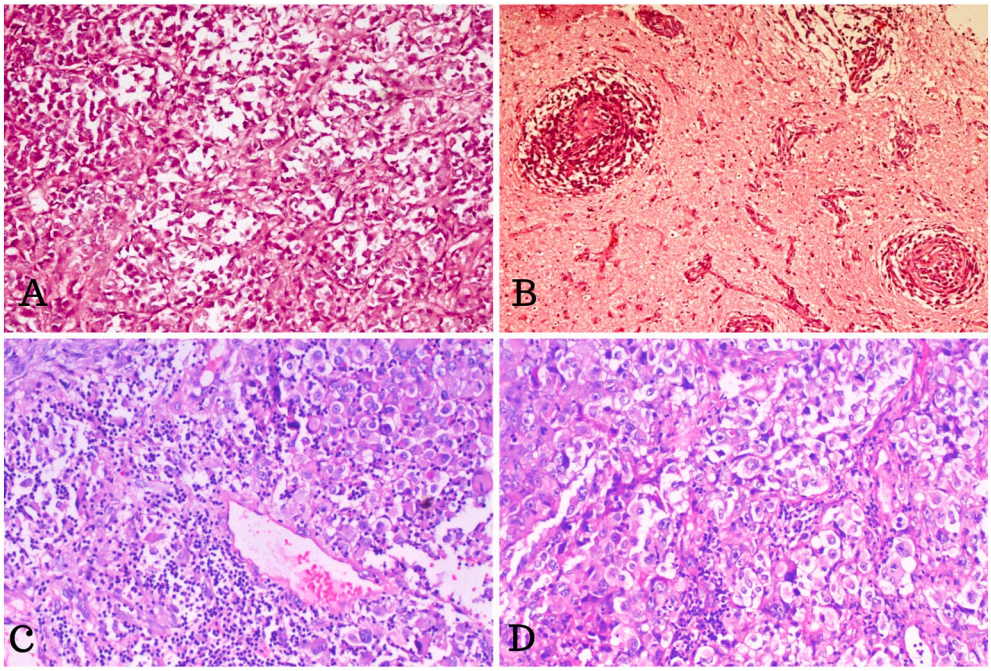

Fig. 3. (A) One case of eGB showing predominant alveolar pattern of arrangement (hematoxylin and eosin, ×200).
(B) eGB spreading along Virchow Robin spaces (hematoxylin and eosin, ×100).
(C) Moderately dense (brisk) tumor infiltrating lymphocytes in eGB (hematoxylin and eosin, ×200).
(D) Focal, non brisk tumor infiltrating lymphocytes in a case of eGB. The tumor cells show cytoplasmic clearing (hematoxylin and eosin, ×200).

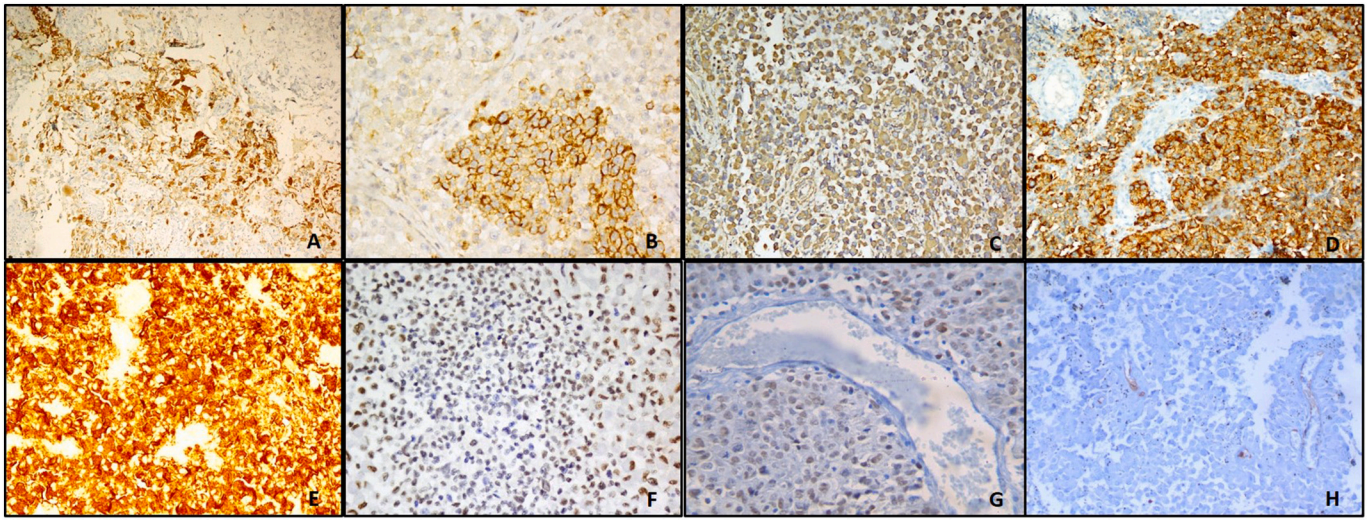

Fig. 4. Immunohistochemistry in epithelioid glioblastoma (eGB): (A) eGB showing patchy, but strong GFAP positivity (Immunohistochemistry, ×200).
(B) Patchy membranous EMA positivity in tumor cells (Immunohistochemistry, ×400).
(C) eGB showing diffuse cytoplasmic positivity for vimentin (Immunohistochemistry, ×200).
(D) eGB showing diffuse and strong S100 expression (Immunohistochemistry, ×200).
(E) A case of eGB showing loss of INI1 expression in the undifferentiated small cell component (Immunohistochemistry, ×200).
(F) eGB showing diffuse strong cytoplasmic positivity for BRAF (Immunohistochemistry, ×400).
(G) eGB showing strong nuclear expression of EZH2 (Immunohistochemistry, ×400).
(H) A case of eGB with loss of p16 (Immunohistochemistry, ×400).

Table 2
Treatment details and outcome in cases of epithelioid glioblastoma:

Case no	Age/ sex	Necrosis	TIL	BRAF V600E	Treatment and outcome	Recurrence	Interval of recurrence (months)	Alive or dead at last follow up	Overall survival (in months)
3	15/f	Focal	Absent	Positive	Received craniospinal RT, recurrence after 11 months, surgery done, received RT and 6 cycles of TMZ, expired after 6 months of recurrence	Yes	11	Dead	17
4	10/f	Extensive	Absent	Negative	Received RT, expired after 8 months	No	–	Dead	8
5	11/m	Extensive	Absent	Positive	Received craniospinal RT and 6 cycles of adjuvant chemotherapy, recurrence after 25 months, expired 3 months after recurrence	Yes	25	Dead	28
13	25/m	Focal	Heavy	Negative	Received Post op radiotherapy 60 Gray in 30 fractions followed by 12 cycles of adjuvant temozolomide (TMZ). No evidence of disease on MRI after 17 months of surgery, asymptomatic on last follow up	No	–	Alive	41
14	10/f	Extensive	Absent	Positive	Received Post op radiotherapy 60 Gray in 30 fractions followed by 4 cycles of adjuvant temozolomide, recurrence after 36 months	Yes	36	Alive	38
15	46/m	Extensive	Non heavy	Negative	Received Post op radiotherapy 60 Gray in 30 fractions followed by 12 cycles of adjuvant temozolomide. Recurrence after 26 months, treated with RT, asymptomatic on last follow up	Yes	26	Alive	36
18	52/m	Extensive	Absent	Negative	Received Post op radiotherapy 60 Gray in 30 fractions, recurrence after 3 months—2nd surgery—received Post op RT 25Gy in 5 fractions with concurrent TMZ—expired 10 days after RT	Yes	3	Dead	5
23	41/m	Extensive	Non heavy	Positive	Received Post op radiotherapy 60 Gray in 30 fractions, recurrence after 18 months of completion of RT, expired after 4 months	Yes	18	Dead	23

TIL - tumor infiltrating lymphocytes, RT - radiotherapy, CT - chemotherapy.

with more than 50% epithelioid cells as eGB in this series.

Due to rarity of this entity, there is limited available literature on eGB. The largest series published on eGB till date comprised of 20 cases and our series on eGB consists of 24 cases [6]. Although glioblastoma is a disease of adults, eGB shows predilection for younger patients. A significant proportion of eGB cases occurs in pediatric age group (<18 years age) [10,14]. In a meta-analysis of 59 published eGBs from 28 studies by Lu et al., the mean age of eGB patients was found to be 30 years with 46% patients being female [15]. These observations match with the demographic findings of our study.

Most of the eGB cases arise in the supratentorial location. In our study, all cases were supratentorial. Huang et al. described the detailed radiological features of eGB [16]. It shows iso-hyperintensive signal on the T2-weighted image and iso-hypointensive signal on the T1-weighted image. Dural tail sign and multifocality were observed in some cases. Radiologically, eGB may be easily misdiagnosed as meningioma, metastasis or lymphoma [16]. Due to retrospective nature of our study, we could not access the radiological details of most cases.

The detailed histological features of eGB has been described by Kleinschmidt-DeMasters et al. [3] Similar to our study, they also found sheets of epithelioid cells in most of their cases. Different studies have also described features such as papillary growth pattern, spindling, nesting, myxoid changes variably in eGB [5,6,8]. Most of the cases of eGB shows large geographic necrosis, although some cases may show focal necrosis or rarely no necrosis [5]. Nakajima et al. reported 10 out of 14 eGB cases with areas showing low grade morphology, however, this appears to be an exceptional finding as none of the other studies have reported such observation [9]. Although we observed microcystic changes and calcification in 3 and 2 cases respectively, none of our cases showed areas with low grade morphology. In adults, the commonest differential diagnoses are anaplastic pleomorphic xanthoastrocytoma (APXA), metastatic carcinoma and metastatic amelanotic melanoma. The differential diagnosis in children include atypical teratoid/rhabdoid tumor (AT/RT) [5]. Thus, IHC is necessary to confirm the diagnosis in intracranial tumors with epithelioid morphology. eGB shows patchy and variable GFAP, strong and diffuse S100 and patchy EMA expression [5,6,13]. They are negative for other epithelial (CK) and melanocytic markers. Most of the studies have reported universal retention of INI1 in eGB. Kleinschmidt-DeMasters et al. (2010) reported INI1 is an useful marker to differentiate between rhabdoid GB and eGBs, as its expression is lost in rhabdoid GBMs and retained in eGBs [3]. However, most of the subsequent studies have used rhabdoid/epithelioid GBMs interchangeably and found INI1 retention almost universally [5,14]. We also found INI1 retention in all cases except 2, which showed focal loss of expression of INI1 in the small, undifferentiated cell component. Thus, INI1 is useful to differentiate AT/RT from eGB, as in the former INI1 expression is almost universally lost. As APXA contains predominantly epithelioid cells, it closely resembles eGB both morphologically and on molecular aspect. Morphologically, APXA shows at least some areas resembling classical grade II PXA (as discussed in the materials and methods). We evaluated each case thoroughly to exclude a diagnosis of APXA. Absence of classical low grade PXA areas, reticulin rich stroma, foam cells and multiple refractile granular eosinophilic bodies excluded the diagnosis of APXA [10]. However, considerable overlap is known to occur between the two entities and cases diagnosed as eGB may later prove to be APXA at recurrence or on additional sampling or on global methylation profiling. Thus, it is still possible that some of our cases could represent APXA. Tanaka et al. reported a case of eGB arising in the background of PXA [17]. Few studies have tried to explore relationship between eGB and APXA. Alexandrescu et al. (2016) concluded that eGB and APXA share clinical, histological and molecular features and suggested that they are closely related i.e. either a same entity or first cousins [10]. Enhancer of zesty homolog 2 (EZH2) is the catalytic subunit of catalytic subunit of the Polycomb repressive complex 2 (PRC2) which causes silencing of the genes involved in histone methylation. Few studies have reported EZH2 overexpression in gliomas, including eGBs [7,18]. Wang et al. found EZH2 overexpression in 69.2% eGB cases, compared to 23.5% cases in our study [7]. Homozygous deletion of *CDKN2A/B* is a frequent genetic event (73%) in epithelioid glioblastoma [9]. Loss of p16 expression is considered to a surrogate marker of homozygous deletion of *CDNK2A/B* in brain tumors. However, loss of p16 expression doesn't correlate accurately with homozygous deletion of *CDNK2A/B* as it may be seen frequently in cases without homozygous deletion of *CDNK2A/B* [12]. We found loss of p16 expression in 29.4% cases, however, couldn't perform fluorescent in situ hybridization (FISH) for *CDNK2A/B* deletion because of technical constrains. No previous study has reported p16 status in eGB. In a recent study by Wang et al. it was found that overexpression of EZH2 and CDNK2A homozygous deletion was more common in eGB compared to APXA [7]. We didn't find any prognostic significance of EZH2 overexpression or loss of p16 expression in the limited follow up data available.

Several authors have evaluated the molecular features of eGB. The most explored molecular alteration is BRAF mutation. Approximately 50% of eGBs show *BRAF V600E* mutation [5]. We found BRAF mutant protein expression in 52.2% of our cases. The reported frequency of BRAF mutation in eGB is variable (33.3% to 80%). depending on method employed [7,19] Although direct gene sequencing is the gold standard for detecting *BRAF V600E* mutation, an antibody is currently commercially available against BRAF V600E mutant protein (VE1 clone), which can detect this mutation with high sensitivity and specificity [20,21]. In our own experience and in other studies, 100% correlation between IHC and molecular methods for detecting BRAF mutation has been documented [6,22]. Thus IHC is a simple and reliable method to detect BRAF mutation in eGB, which is rarely found in other variants of glioblastomas. In a previous study from our institute, none of the 25 GBMs showed *BRAF* mutation by sequencing [23]. Targeted inhibitor of BRAF and its downstream MEK protein has been used for treatment of BRAF mutant eGBs with variable success [24,25]. Epithelioid GBs are almost universally IDH wild type [14]. Other molecular alterations are also reported in eGB. Khanna et al. found TERT promoter mutation and EGFR amplification in 40% and 14% eGBs respectively [13]. Furata et al. and Nakajima et al. reported TERT promoter mutation in 12.5% and 50% of eGBs, respectively [9,19]. These findings indicate that apart from *BRAF* mutation, other molecular alterations are inconsistently reported in eGB. It may be assumed that more molecular information is required to accurately define this special entity.

eGB is classically believed to be a highly aggressive variant of GBM with poor prognosis [1]. Three out of 8 patients in our series were alive at the time of last follow up with mean and median OS 24.5. and 25.5 months respectively. Recurrence was seen in 6/8 (75%) patients. Although few patients died early, some had long survival. A thorough literature search also revealed similar findings. Out of 13 cases included in a series by Kleinschmidt-DeMasters et al., 8 were alive till last follow up with four being alive more than 3 years (maximum 328 weeks) [5]. Alexandrescu et al. reported that six out of 11 eGBs (54.5%) recurred; the median interval to recurrence was 12 months. However, most of the patients in their study remained alive at last follow-up with survival times already exceeding 2 years in three eGB cases (33, 38 and 65 months at last follow-up) [10]. These findings suggest that long survival times are seen in a subset of eGB. Zeng et al. suggested extensive necrosis, MGMT promoter unmethylation, EZH2 overexpression, and a lack of adjuvant chemo-radiotherapy may indicate a poor prognosis [8]. Wang et al. observed that extensive necrosis and BRAF mutation are associated with poorer outcome in eGB [7]. However, we didn't observe any such relationship. In a meta-analysis by Lu et al., median OS and PFS in eGB were estimated to be 11.0 months (95% confidence interval, 6.5–13.0) and 7.0 months (95% confidence interval, 3.0–10.0), respectively. Surgical extent of resection, radiation therapy, and chemotherapy all predicted superior OS and PFS on multivariate analysis ($P < 0.05$). Although no biomarkers is known to prognosticate survival [15] we have observed that cases with TILs had a longer survival than the cases without TILs. However, outcome information was available only in 8 cases in our series and this observation needs validation in larger future series.

In conclusion, eGB which is a distinct variant of glioblastoma with predilection towards pediatric age group should be recognized histologically. It has characteristic morphology and shows high percentage of BRAF V600E mutation. Although it is an aggressive tumor, a subset of it shows longer survival. Cases with presence of tumor infiltrating lymphocytes are associated with better outcome.

Declaration of competing interest

None.

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Nil.

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