CASE REPORT

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An autopsy case of primary gliosarcoma with multiple extracranial metastases: pathology after administration of bevacizumab and genetic profile

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ABSTRACT

Gliosarcoma (GS), a morphological variant of glioblastoma, pathologically shows a biphasic pattern with gliomatous and sarcomatous components. It has been reported that GS has much higher metastatic capacity than glioblastoma. A few reports on the pathology of the extracranial metastasis of GS have shown that metastatic lesions had a sarcomatous component alone or a mixture of gliomatous and sarcomatous ones. Therefore, it is considered that GS tends to disseminate hematogenously due to its mesenchymal sarcomatous component. Herein, we report an autopsy case of GS with multiple extracranial metastases treated by craniotomy, radiotherapy, and bevacizumab. In this case, metastatic lesions at autopsy contained a gliomatous component alone, but no sarcomatous component. In addition, the sarcomatous component disappeared from the intracranial lesion at autopsy after the administration of bevacizumab. In this report, we discuss the clinical course and pathological findings at the initial state, recurrence, and autopsy, including the results of whole-genome analysis.

Keywords: gliosarcoma, metastasis, bevacizumab, pathology, whole-genome analysis

Abbreviations:
GB: glioblastoma
GS: gliosarcoma
MRI: magnetic resonance imaging

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INTRODUCTION

Gliosarcoma (GS) is a rare primary malignant brain tumor corresponding to World Health Organization grade 4.[1] It accounts for less than 0.5% of all intracranial tumors and is estimated to comprise 1.8%–2.4% of glioblastoma, IDH-wild-type (GB) cases.[2-4] In terms of the histopathology of GS, as one of the morphological variants of GB, it features a biphasic pattern with gliomatous and sarcomatous components.[4] Metastasis of GS has been reported to occur at a rate as high as 11%, which is much higher than that of GB.[3,5,6] A few reports on the pathology of the extracranial metastasis of GS have shown that metastatic lesions had a sarcomatous component alone or a mixture of gliomatous and sarcomatous ones.[5,6] Therefore, it is considered that GS tends to disseminate hematogenously due to its mesenchymal sarcomatous component.

GS has been treated in the same manner as GB.[3-5] Bevacizumab is frequently used for the recurrence of GB. However, the efficacy of bevacizumab for GS or its metastatic lesions is still poorly understood. The pathology after the administration of bevacizumab is also unclear, especially in metastatic specimens.

Recent genetic studies have demonstrated that GS shares gene alterations that also function as drivers of GB. It has been reported that somatic mutations, such as in *TP53* and *PTEN*, are found in both gliomatous and sarcomatous components.[7-9] Germline mutations associated with GB, such as in *TP53* and *NF1*, have been reported,[10] but few with GS.

In this report, we review an autopsy case of GS with multiple extracranial metastases treated by bevacizumab, including the results of genetic analysis. This study was approved by the ethics committee of Shizuoka Cancer Center (J2021-56-2021-2-3).

CASE PRESENTATION

A 77-year-old woman with a history of progressively worsening left hemiparesis visited our hospital. Past medical history and occupational history were unremarkable. Magnetic resonance imaging (MRI) demonstrated a 5×4.5 cm ring-enhancing lesion in the right frontal lobe with irregular margins and peritumoral edema, which adhered to the dura mater (Fig. 1A). One week after the first visit, the patient underwent a right frontal craniotomy with gross total resection of the tumor (Fig. 1B). The tumor adhered tightly to the dura. The histopathological diagnosis was GS, showing a biphasic pattern with areas of gliomatous and sarcomatous components (Fig. 2A). In the gliomatous component (Fig. 2B), immunohistochemistry showed positivity for glial fibrillary acid protein (GFAP) (Fig. 2C) and MGMT (O-6-methylguanine-DNA methyltransferase), and negativity for IDH1-R132H, Olig2, and p53. ATRX expression was retained in the nucleus. In the sarcomatous component, tumor cells were spindle-shaped and proliferated with fibrosis (Fig. 2D). Silver impregnation staining showed that the tumor cells were surrounded by reticular fibers (Fig. 2E). Ki-67 expression was found in about 50% of cells. Two weeks after the craniotomy, the patient underwent hypofractionated radiotherapy with a total dose of 40 Gy in 15 fractions. We did not administer concurrent temozolomide treatment due to the poor performance status of the patient. She was discharged home 1 month after the craniotomy. MRI 8 months after the craniotomy revealed local recurrence of the tumor (Fig. 1C). The tumor had infiltrated into the falx cerebri and recurred in the right and left frontal lobes. We performed craniotomy for these recurrent tumors. The recurrence in the right frontal lobe was completely resected, but that in the left frontal lobe remained (Fig. 1D). The histological diagnosis was GS, the same as at the first craniotomy (Fig. 2F, G). One week after the second craniotomy, stereotactic radiotherapy with a total dose of 25 Gy in five fractions was performed for the residual tumor. Three weeks

later, MRI revealed enlargement of the enhancing area in the left frontal lobe (Fig. 1E) without neurological symptoms. We thus started the administration of bevacizumab. Two months after completion of the administration of bevacizumab, we confirmed the disappearance of enhancement in the left frontal lobe (Fig. 1F).

However, a cough gradually appeared and concurrent chest computed tomography showed the appearance of new neoplastic lesions in the right lung (Fig. 3). After six courses of bevacizumab, the patient's left hemiparesis worsened and bevacizumab was discontinued. She was readmitted to our hospital and received palliative treatment. Approximately 1 and a half years after the first craniotomy, the patient died of respiratory failure due to multiple lung tumors and pleural effusion. An autopsy was performed 6 hours after death. We prepared specimens containing as many viable cells as possible, and made a diagnosis based on the results of morphology, special staining, and immunostaining. Autopsy specimens of extracranial metastases such as those to lung, tracheal lymph nodes, and bone marrow contained a gliomatous component alone, but not a sarcomatous component (Fig. 2H, I). Similarly, autopsy specimens of brain contained a gliomatous component alone (Fig. 2J–L). Viable tumor cells had infiltrated through the corpus callosum to the left frontal lobe. Immunohistochemistry showed positivity for GFAP (Fig. 2I, L) and Olig2, and negativity for IDH1-R132H, p53, and MGMT. The expression of ATRX was retained in the nucleus. Ki-67 expression was found in about 10% of cells. We also performed whole-genome analysis with specimens of the brain and blood from both the first and second craniotomies, as part of the HOPE project.[11] *PTEN* and *TP53* somatic mutations were detected as Tier 1 and Tier 2 driver mutations (Tier 1 means pathogenic mutation and Tier 2 means mutation suspected of being pathogenic[12]). We detected a missense mutation in *PTEN* and a frameshift mutation in

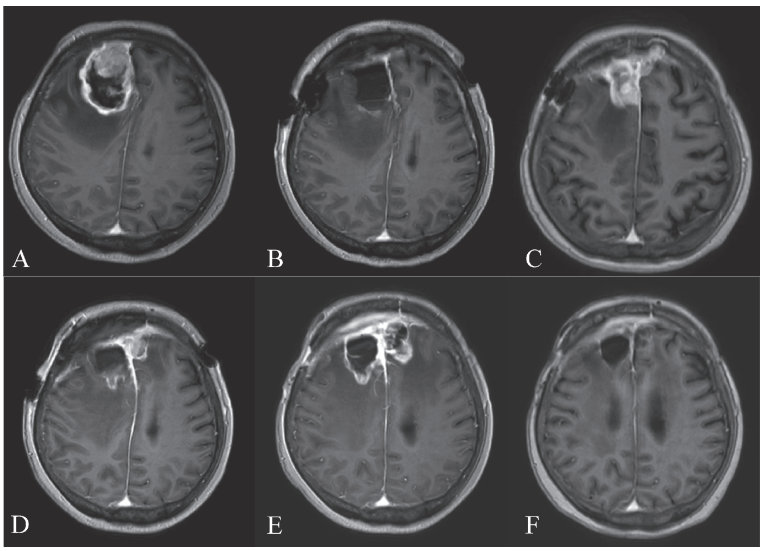

Fig. 1 Brain MR images

Fig. 1A: T1-weighted magnetic resonance imaging (MRI) with gadolinium demonstrating a 5×4.5 cm ring-enhancing lesion in the right frontal lobe with irregular margins and peritumoral edema.

Fig. 1B: Postoperative MRI showing gross total resection of the tumor.

Fig. 1C: MRI 8 months after the first craniotomy.

Fig. 1D: MRI after the second craniotomy.

Fig. 1E: MRI 3 weeks after the radiotherapy.

Fig. 1F: MRI 2 months after the administration of bevacizumab.

TP53. Furthermore, germline mutation in *TP53* was detected as a member of the Tier 2 driver mutation class (Fig. 4A). Copy number alterations including gain of chromosome 7 and loss of chromosomes 10 and 13 were detected in the specimens from the first craniotomy. Meanwhile, in those from the second craniotomy, the number of copies of chromosome 7 was amplified, but the number of copies of chromosome 10 was normal (Fig. 4B).

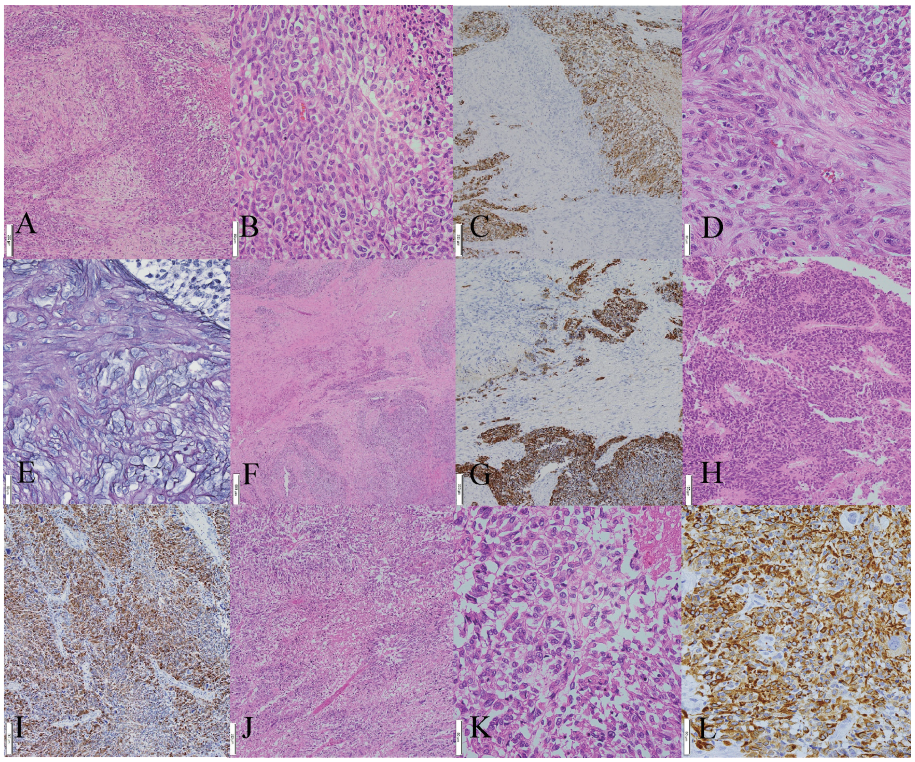

Fig. 2 The histopathology in the first craniotomy (A–E) and the second craniotomy (F, G). Autopsy specimens of lung (H, I) and brain (J–L).

Fig. 2A: Hematoxylin and eosin (H&E) staining (×100); scale bar, 200 μm.
Fig. 2B: H&E staining (×400); scale bar, 50 μm.
Fig. 2C: Glial fibrillary acidic protein (GFAP) staining showing positivity in the gliomatous component of GS (×100); scale bar, 200 μm.
Fig. 2D: H&E staining (×400); scale bar, 50 μm.
Fig. 2E: Silver impregnation staining showing sarcomatoid findings of spindle cell type (×400); scale bar, 50 μm.
Fig. 2F: H&E staining (×40); scale bar, 500 μm.
Fig. 2G: GFAP staining showing positivity in the gliomatous component of GS (×100); scale bar, 200 μm.
Fig. 2H: H&E staining of an autopsy specimen of the lung (×200); scale bar, 100 μm.
Fig. 2I: GFAP staining showing strong positivity in an autopsy specimen of the lung (×100); scale bar, 200 μm.
Fig. 2J: H&E staining of an autopsy specimen of the brain (×100); scale bar, 200 μm.
Fig. 2K: H&E staining of an autopsy specimen of the brain (×400); scale bar, 50 μm.
Fig. 2L: GFAP staining showing positivity in an autopsy specimen of the brain (×400); scale bar, 50 μm.

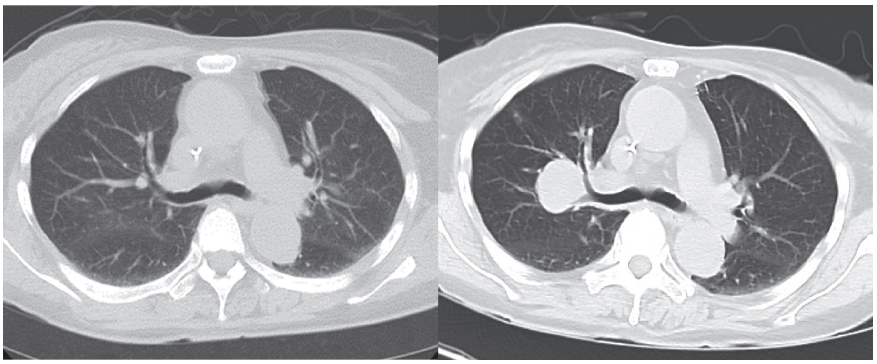

Fig. 3 Chest CT scan images
Left: Chest computed tomography (CT) showing no lesions before the first craniotomy.
Right: CT showing the appearance of new neoplastic lesions in the right lung.

A

Molecular alteration	Gene symbol	Driver mutation class	Coding DNA change	Mutation	VAF (Primary)	VAF (Recurrence)
Somatic mutation	*PTEN*	Tier1	c.388C>G	missense	85.45	20.78
	TP53	Tier2	c.499_500delCA	frameshift	93.90	41.84
Germline mutation	*TP53*	Tier2	c.145G>C	missense	48.30	41.20

B

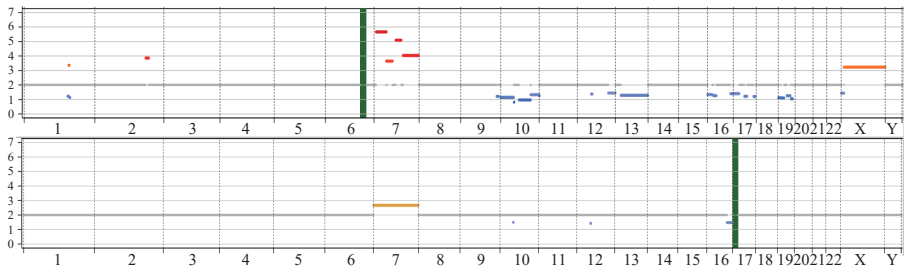

Fig. 4 Genetic alterations

Fig. 4A: *PTEN* and *TP53* somatic mutations were detected as Tier 1 and Tier 2 driver mutations (Tier 1 means pathogenic mutation and Tier 2 means mutation suspected of being pathogenic[12]). A missense mutation in *PTEN* and a frameshift mutation in *TP53* were detected. *TP53* germline mutation was detected as a Tier 2 driver mutation. VAF indicates variant allele fraction.

Fig. 4B: In copy number variation analysis, gain of chromosome 7 and loss of chromosomes 10 and 13 were detected in the specimens from the first craniotomy. Meanwhile, in those from the second craniotomy, the number of copies of chromosome 7 was amplified, but the number of copies of chromosome 10 was normal (upper row represents the first craniotomy, lower row represents the second craniotomy).

DISCUSSION

In our case of GS, metastatic lesions at autopsy contained a gliomatous component alone, but no sarcomatous component. GS has a propensity to manifest as extracranial metastases, most commonly involving the lungs (72%), liver (41%), and lymph nodes (18%).[2] However, only a few reports on the pathology of extracranial metastases of GS have been published. Ramos et al reported 31 cases of metastatic GS, the metastasis of all of which showed sarcomatous or mixed components histologically.[6] In contrast, Georgescu et al reported a case of metastatic GS that contained only a gliomatous component in the pathological specimens of metastases.[13] However, because this diagnosis was achieved only by biopsy, the tumor tissue may not have been collected correctly. As far as we know, no reports demonstrating the pathology of extracranial metastases of primary gliosarcoma after the administration of bevacizumab have been published in English.

A few reports have mentioned significant benefits of bevacizumab for GS.[4,14] Bevacizumab is a human monoclonal antibody directed specifically against vascular endothelial growth factor (VEGF), which reduces the number of intratumoral blood vessels. It is used for many tumors including GB and sarcoma, in which VEGF is expressed at a high level.[15] Especially in sarcoma, VEGF expression correlates with stage, grade, and prognosis.[16] In recent years, bevacizumab has been proven to be effective for certain types of sarcomas, including angiosarcoma, uterine leiomyosarcoma, and HIV-associated Kaposi's sarcoma.[17-19] In our case, the administration of bevacizumab may have played a role in the disappearance of the sarcomatous component in autopsy specimens of brain, since bevacizumab was the only treatment applied from the second craniotomy until death. Bevacizumab may also have been involved in suppressing the sarcomatous components in extracranial metastases in our case.

In the present case, we performed whole-genome analysis with specimens of the brain and blood from both the first and second craniotomies. We detected somatic mutations in *TP53* and *PTEN* (Fig. 4A). *TP53* somatic frame shift mutation in the same codon as this case was not identified in gliomas using a public database, COSMIC v96 (Catalogue of Somatic Mutations in Cancer) (https://cancer.sanger.ac.uk/cosmic). Biernat et al first demonstrated somatic *p53* mutations in gliomatous and sarcomatous components.[7] In addition, Reis et al demonstrated somatic *PTEN* mutations, *p53* nuclear accumulation, *p16* deletion, and *CDK4* amplifications in both components.[8] Recent molecular genetic studies have suggested that the sarcomatous component develops from neoplastic glial cells that have acquired a sarcomatous phenotype during tumor progression.[2,5,8] This monoclonal hypothesis, which is strongly supported by recent genetic analysis,[9] would mean that GS shares gene alterations that function as drivers of GB. Many reports suggesting the presence of *TP53* and *PTEN* somatic mutations in both sarcomatous and gliomatous components have been published.[7-9] Our results are similar to these previous findings.

As for the analysis of copy number variation, gain of chromosome 7 and loss of chromosomes 10 and 13, which are relatively common in GB, were detected in the specimens from the first craniotomy. Recent studies have shown similar results,[9] and these copy number variations are also common in GS. Meanwhile, in the specimens from the second craniotomy, the number of copies of chromosome 7 was amplified, but the number of copies of chromosome 10 was normal. Glioma infiltrates normal brain tissue, so the specimens may have contained normal brain tissue. In fact, the VAF for gene mutations of TP53 and PTEN differed between the primary and recurrent tumors (Fig. 4A). Since the gliomatous and sarcomatous components were not analyzed separately in this study, the genetic alterations specific to each component could not be assessed. To obtain accurate results, it may be necessary to compare the results of genetic analysis with the histopathological findings.

To the best of our knowledge, only two reports have shown germline mutations in GS.

Georgescu et al demonstrated *FANCD2* germline mutation in GS,[13] while Abdulla et al reported a case of GS with *TP53* germline mutation.[20] In our case, genetic analysis showed germline mutation in *TP53*. Generally, *TP53* germline mutation has been described in Li-Fraumeni syndrome (LFS). However, our case did not meet the diagnostic criteria for LFS.

In summary, we experienced an autopsy case of GS with multiple extracranial metastases presenting no sarcomatous component. Bevacizumab was effective for GS, particularly for the sarcomatous component. Further investigation to reveal the genetic profile of GS is warranted.

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CONFLICTS OF INTEREST

The authors declare no conflicts of interest.

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