Case Report

Analysis of a synchronous gliosarcoma and meningioma with long survival: A case report and review of the literature

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INTRODUCTION

Gliosarcoma is a glioblastoma variant characterized by a biphasic tissue pattern with alternate areas displaying glial and mesenchymal differentiation.\textsuperscript{[17]} It was originally described in 1895 by Stroebe \textit{et al.} and comprises approximately 2\% of all glioblastoma.\textsuperscript{[17,28]} The sarcomatous areas commonly resemble fibrosarcoma, but may show a variety of lines of mesenchymal differentiation, such as osteogenic, chondrogenic,
adipogenic, smooth, and skeletal muscle.\textsuperscript{[2-24]} The occurrence of similar genetic alterations in both glial and mesenchymal components supports the concept of a monoclonal origin of the metaplastic mesenchymal differentiation from the astrocytic component.\textsuperscript{[11,12,28,29]} However, the molecular mechanisms governing this mesenchymal differentiation are still unclear. Interestingly, a recent study reported the isolation of gliosarcoma stem cells, which were able to further undergo glial and mesenchymal differentiation.\textsuperscript{[9]} Meningiomas are the most common extra-axial neoplasms and the second most common primary tumors of the central nervous system, accounting for 24-30% of all brain tumors.\textsuperscript{[17,31]}

The occurrence of simultaneous brain tumors of different histological natures in the absence of hereditary syndromes or prior exposure to ionizing radiotherapy is rare.\textsuperscript{[8]} Nevertheless, several reports described the concurrent association of meningioma and gliomas, mainly glioblastomas.\textsuperscript{[7,10,23,33,36]} Herein, we report a case of a long survival gliosarcoma with a synchronous meningioma. Due to the exceptionality of the case, we performed an immunohistochemical and molecular characterization of the lesions, in order to better understand their biology.

**CASE REPORT**

A previously healthy 51-year-old woman, with no family history of cancer, was admitted in another institution in October 2003 with a history of dysarthria and left hemibody paresthesias followed by generalized tonic-clonic seizure. On physical examination the patient was full awake with mild dysarthria and a grade 4 left hemiparesis with brachiofacial predominance. She had no signs of intracranial hypertension. The Karnofsky performance status (KPS) was 70. A computed tomography (CT) scan was performed showing a right frontal cortico-subcortical hypodense area resembling a secondary lesion in nature. Primary neoplasm investigation was negative and the patient was referred to our institution (Hospital S. João, Porto; Portugal). A magnetic resonance imaging (MRI) was done and revealed a right frontal parasagittal and well-demarcated hyperintense lesion with homogenous contrast-enhancement and a second lesion in the posterior right frontal lobe with poorly demarcated borders and heterogeneous contrast-enhancement. The patient underwent a right frontal craniotomy with gross total removal of the two lesions in December 2003. There was no complication and the patient was discharged with a grade 4 hemiparesis and a complete recovery of the dysarthria. The KPS at discharge was 80. The pathological examination revealed two distinct lesions being the anterior a meningioma and the posterior a gliosarcoma [Figure 1]. The patient received postoperative radiotherapy with a total dose of 60 Gy given in 30 fractions with margin of 1 cm in the area of the gliosarcoma. The patient remained asymptomatic until September 2005 when she developed dysarthria and worsening of the hemiparesis. The MRI showed a regrowth of the initial gliosarcoma and the patient was re-operated with macroscopically complete removal of the recurrent gliosarcoma [Figure 1]. The patient recovered again from the neurological deficits and started temozolomide treatment with 150 mg/m\textsuperscript{2} followed by 200 mg/m\textsuperscript{2}. In November 2005, the patient showed progressive neurological deterioration with headaches, disorientation episodes, and paresis worsening with left arm plegia. The MRI showed an early new regrowth with no indication for surgical removal and the patient underwent palliation with dexamethasone. In June 2006 a new MRI showed a growth of the right frontal lesion with involvement of the basal ganglia and corpus callosum, crossing the midline and development of hydrocephalus. The patient died in October 2006 after a progressive neurological deterioration. The overall survival was 36 months. No postmortem examination was performed.

**Pathological findings**

Histological examination of the initial anterior lobular frontal and posterior lobular frontal lesions revealed two clear distinct tumors: A meningioma (M) depicting positivity for epithelial membrane antigen (EMA) staining and a gliosarcoma (GS) exhibiting typical features of intermingled GFAP and reticuline neoplastic regions. The analysis of recurrent lesion showed the presence of a gliosarcoma (GS-R) with a more prominent glial component.

In order, to determine the presence of mutations in the TP53 (exons 5-8) and BRAF (exon 11 and 15) genes, DNA was isolated from the formalin-fixed and paraffin-embedded tissues of all three lesions. The screening of mutations was carried out by polymerase chain reaction (PCR)-single-strand conformational polymorphism (PCR-SSCP), followed by direct DNA sequencing, as previously described.\textsuperscript{[27,29]} None of lesions showed TP53 or BRAF mutations [Table 1].

We further investigated for the presence of MGMT protein expression by immunohistochemistry, using the mouse anti-MGMT monoclonal antibody (dilution 1:400; clone MT3.1, Chemicon International) and correlated with MGMT gene promoter hypermethylation, assessed by methylation-specific PCR (MSP) as previously described.\textsuperscript{[6,21]} As illustrated in Figure 2, MGMT methylation was found in both primary and recurrent gliosarcoma. The meningioma lesion was negative for MGMT methylation [Table 1 and Figure 2]. In concordance with methylation status, MGMT expression was only present in the meningioma and absent in both primary and recurrent gliosarcoma [Table 1 and Figure 3].
Next, we analyzed the presence of EGFR alterations at both gene and protein levels by CISH (EGFR spot-Light amplification probes from Zymed® Laboratories Inc., South San Francisco, CA, USA) and immunohistochemistry (anti-EGFR polyclonal antibody, dilution 1:100, clone 31G7, Zymed® Laboratories Inc., San Francisco, CA, USA), respectively. The meningioma and primary gliosarcoma were negative for EGFR protein expression (Table 1 and Figure 3). EGFR immunohistochemistry showed a focal region with strong immunoreactivity in the recurrent gliosarcoma. In both meningioma and primary gliosarcoma, it was observed the presence of 2 signals per neoplastic cell, representing absence of EGFR gene copy number alterations (Table 1 and Figure 3). EGFR gene amplification, represented by clusters of more than 5 signals per neoplastic cell, was observed only in the recurrent gliosarcoma, in the same region with strong focal positivity for EGFR protein (Table 1 and Figure 3).

Finally, we studied the expression of COX-2 by immunohistochemistry, using the anti-COX-2 monoclonal antibody (dilution 1:50, clone SP21, Neomarkers, Fremont, CA, USA). We observed positivity only in meningioma lesion (Figure 3). Both gliosarcomas (primary and recurrent) were negative for COX-2 expression (Table 1 and Figure 3).

**DISCUSSION**

Gliosarcomas are rare tumors of the central nervous system. In general, the epidemiology and natural history of gliosarcoma appears to be similar to the glioblastoma, mainly primary glioblastoma. Both show a propensity to affect elderly patients, with a median age of diagnosis over 60 years. With the current standard treatment, which includes adjuvant radiotherapy with concomitant temozolomide, gliosarcoma patients have a median survival time of approximately 14 months. Parameters such as age of presentation, extension of tumor resection and adjuvant radiotherapy (RT) are also significantly associated with gliosarcoma patient’s survival. They tend to occur in the temporal lobe and some of them are well-circumscribed.

It is not frequent the concurrent occurrence of central nervous system neoplasms outside familial tumor syndromes or previous to radiotherapy. Several

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**Table 1: Molecular and immunohistochemical analysis of the reported case**

| Case   | Molecular analysis | Immunohistochemistry analysis |
|--------|--------------------|--------------------------------|
|        | BRAF mutations | TP53 mutations | MGMT methylation | EGFR amplification | EGFR | Cox2 | MGMT |
| M | Normal | Normal | Unmethylated | Normal | Negative | Positive | Positive |
| P | Normal | Normal | Methylated | Normal | Negative | Negative | Negative |
| R | Normal | Normal | Methylated | Amplified | Positive | Negative | Negative |
authors have previously reported synchronous glioma and meningioma. Literature review shows that glioblastoma is the most frequent glioma subtype associated.\cite{7,8,15,20,23,32,37,38} The occurrence of gliosarcoma and meningioma was rarely reported.\cite{13,25,40} The explanation of this simultaneous occurrence of two primary distinct brain tumors is not clear. Some authors suggested that this occurrence is most likely a casual coincidence,\cite{5,15,36,37} whereas others hypothesized that they can be the result of common etiological route.\cite{38} In the present study, the observation of distinct molecular characteristics displayed by the two tumors, namely MGMT, COX-2 and EGFR profile, suggest that both tumors evolved through distinct pathways.

It is known that gliosarcomas can present with different imaging features. The lesions can be similar to glioblastoma with a central necrotic area surrounded by a ring of heterogeneous contrast enhancement, as occurred in our patient, or as homogeneous hyperdense lesions resembling meningiomas. Those that resemble meningiomas tend to have better prognosis.\cite{19,34} In our case, the presence of distinct histological and molecular features in both meningioma and primary gliosarcoma, exclude the possibility of monoclonal origin of both tumors.

MGMT is a DNA repair enzyme that removes promutagenic methyl groups from the O-6 position of guanine induced by alkylating agents such as temozolomide.\cite{6,26} Regulation of MGMT expression is an epigenetic event directly dependent on MGMT promoter methylation status. Although not completely consensual, MGMT promoter methylation has been associated with better survival in glioblastoma patients and has prognostic value.\cite{6,26} Our case has an unusual good behavior with a 36 months survival. The methylation of MGMT is present in both primary and recurrent gliosarcoma, thus potentially contributing to the long survival of the patient.

Some studies suggested the involvement of the p53 pathway in the development of meningiomas and gliosarcomas.\cite{1,3,28} In our case we did not find any mutation in the TP53 gene. EGFR alterations, namely gene amplification, are reported to be infrequent in gliosarcomas and meningiomas.\cite{18,28} In other glioma subtypes, mainly glioblastomas it can be associated with worse clinical outcome.\cite{16} Interestingly, in the present case, EGFR amplification and overexpression was only present in the recurrent tumor and this change could explain the aggressive behavior of the recurrent gliosarcoma. Meningiomas are strongly positive for COX-2, that can be a predictor of shortest outcome.\cite{22,35} In gliomas COX-2 overexpression can also occur, however, this is usually less common than in meningiomas.\cite{35} Interestingly, in the present case, COX-2 expression was only present in the meningioma lesion.
CONCLUSION

We report for the first time the occurrence of a synchronous meningioma and gliosarcoma with a long survival. We observed the absence of mutations in TP53 and BRAF genes. MGMT loss of expression/function by promoter methylation was only found in both primary and recurrent gliosarcoma, which could in part explain the good prognosis of the patient. EGFR gene overexpression/amplification was present only in the recurrent gliosarcoma supporting its association with tumor aggressiveness. Overall, immunohistochemistry and molecular data suggest the distinct clonal origin of meningioma and gliosarcoma lesions.

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