ABSTRACT

Background: It is generally accepted that gliomas rarely metastasize outside the central nervous system (CNS). However, the issue that anaplastic oligodendrogliomas is prone to spread outside CNS is being disputed again. Methods: We described in detail the clinical case of one patient with recurrent anaplastic oligodendroglioma, which metastasized to right pelvis bones 5 years after second neurosurgical procedures. Genetic analyses included detection of 1p and 19q loss of heterozygosity (LOH) and IDH-1 mutations. A review of literature was conducted on metastatic anaplastic oligodendrogliomas. Results: A puncture biopsy of the right ischium bone in this patient revealed that high grade glioma cells had infiltrated the right ischium marrow. Moreover, the diagnosis of multiple-bone metastases of glioma was confirmed by the whole-body [18F] fluorodeoxyglucose positron emission tomography computed tomography (PET/CT) scan. Immunohistochemistry stain analyses showed that the right ischium metastasis glioma cells originated from right parietal lobe anaplastic oligodendrogliomas. The 1p and 19q LOH and the R132H site mutation of IDH-1 were detected in the samples of the recurrence brain tumor in 2009. Conclusions: The incidence of extracranial metastases in anaplastic oligodendrogliomas is very low but they do occur; bone metastases may be the most common site. The molecular markers such as the 1p/19q LOH and IDH-1 mutations not only help prove the pathologic diagnosis of oligodendrogliomas, but also help elucidate a subtype of anaplastic oligodendrogliomas which has a good prognosis even with extracranial metastases.

Keywords: gliomas, anaplastic oligodendrogliomas, extracranial metastasis, 1p/19q LOH, IDH-1 mutation

INTRODUCTION

The development of extracranial metastasis of primary intracranial gliomas occurs very rarely [1–3]. However, extracranial metastases of gliomas have increased in frequency due to longer survival of patients as a result of recent advancements in the treatment of gliomas [4–5].

Oligodendroglioma (OGD) is an uncommon diffuse glial tumor of central neuroepithelial origin; it is a rare tumor with an incidence of 2%–5% of
all primary brain tumors [6]. Five- and ten-year survivals are reported to be 62% and 38%, respectively [7]. It has been mostly identified in adults, with the highest incidence occurring in the fifth and sixth decades of life, although it has also been reported in children and adolescents. Various forms of combination therapy administered as comprehensive treatment have significantly improved the survival of patients with oligodendrogliomas or mixed oligoastrocytomas [6-7].

Recent investigations into the molecular and genetic make-up of oligodendrogliomas have provided a new insight into overall survival and responses to therapy. The 1p and 19q loss of heterozygosity (LOH) is strongly related to oligodendroglial morphology, prolonged overall patient survival, and enhanced responses to both alkylating chemotherapeutic agents (such as temozolomide) and radiation [8-10]. An interesting dilemma is that the same genetic signatures are associated with prolonged survival or may also identify a glioma subtype with greater metastatic potential. To date, only few reports have evaluated the molecular and genetic alterations in anaplastic oligodendrogliomas that have metastasized beyond the central nervous system [11]. Herein, we report a rare case of anaplastic oligodendroglioma of right parietal lobe metastatic to the pelvis bones appearing 5 years after the second resection of the intracranial tumor in 2009.

**CASE PRESENTATION**

A 64-year-old male who initially presented with a short history of headache and vomiting was admitted to a local country hospital in September 2005. No focal neurological deficit was found on admission. On 22 September 2005, he underwent a right parietal lobe tumor craniotomy with gross total resection. The pathological diagnosis was right parietal lobe “astrocytomas” (WHO grade II) by HE stain. Subsequently, the patient had gamma ray radiotherapy on the original tumor site (50% dose: 12Gy, Max dose: 24Gy) again, and sequential chemotherapy was given by 6 cycles of temozolomide [10] of 150 mg/m2 d-1 for 5 days, repeated every 28 days. The patient experienced no significant hematological toxicity, and had no difficulties in understanding and remembering.

In October 2014, 5 years after the second right parietal lobe tumor resection, he presented with 7-month history of gradually worsened right pelvis pain, and was hospitalized in November 2014. A brain MRI showed no evident of recurrence in the original intracranial site (Figure 1a, 1b, 1c). Subsequent whole-body [18F] fluorodeoxyglucose positron emission tomography computed tomography (PET/CT) scans revealed significantly higher glucose metabolism in multiple right pelvis bones, including right acetabulum, corpus ossis ischii, pubis superior and inferior ramus and tuber ischiadicum osteoblastic lesion (Figure 2). In order to determine the nature of the pelvis bone lesion, we executed the aspiration on the right corpus ossis ischii. A puncture biopsy of the right corpus ossis ischii of this patient revealed the replacement of the patient’s marrow by malignant cells, which exhibited nuclear pleomorphism (Figure 3). It was considered that the pelvis bone metastasis might originate from the intracranial glioma. The immunohistochemical staining of the right corpus ossis ischii samples showed that the CD99, AE1/AE3, CD117, NF, NSE, and Vimintin marker are negative, but the glial fibrillary acidic protein (GFAP) and S-100 marker are positive (Figures 4 and 5). This confirmed that the metastatic right corpus ossis ischii was originated from intracranial glioma. In addition, the metastatic right corpus ossis ischii was immunoreactive Ki67 protein (50%-75% positive); it implied the metastatic bone lesion was highly proliferative (Figure 6).
Figure 2: The whole-body [18F] fluorodeoxyglucose positron emission tomography computed tomography (PET/CT) scans revealed significantly higher glucose metabolism in multiple right pelvis bones, including right acetabulum, corpus ossis ischia, pubis superior and inferior ramus and tuber ischiadicum osteoblastic lesion.

Figure 3: A puncture biopsy of the right corpus ossis ischia of this patient revealed the replacement of the patient’s marrow by malignant cells, which exhibited nuclear pleomorphism.

Figure 4: The immunohistochemical staining of the right corpus ossis ischia samples showed that the glial fibrillary acidic protein (GFAP) positive.

Figure 5: The immunohistochemical staining of the right corpus ossis ischii samples showed that the S-100 marker positive.

Figure 6: The immunohistochemical staining of the right corpus ossis ischii samples showed that Ki67 protein 50%-75% positive.

These results indicated the necessity to reconfirm the pathological diagnosis in the previous intracranial glioma samples made by the local country hospital in 2005 and 2009, because low grade astrocytoma metastases outside the central nervous system are extremely rare, compared to anaplastic astrocytomas, anaplastic oligodendrogliomas and glioblastomas (GBM). We checked the paraffin samples in 2005 and in 2009 by HE stain and immunohistochemical staining. The final pathology diagnosis of this patient was anaplastic oligodendroglioma in the right parietal lobe in 2005 and in 2009 (Figures 7 and 8). Photomicrographs of the tumor showed that there were higher cell densities, densely packed round cells with perinuclear haloes, microscopically round-to-oblong cells with hyperchromatism and pleomorphism, and clusters of capillary or plexiform capillaries. In addition, the irregular mitosis densities were higher (Figures 7 and 8). They were strongly positive for Ki-67, with proliferation index >15% (Figure 9). They were...
also all positive for the marker oligodendrocyte transcription factor (Oligo-2; Figure 10). Further findings were negative for O6–methylguanine-DNA methyltransferase (MGMT) (Figure 11). Three state hospital pathology centers confirmed our diagnosis of anaplastic oligodendroglioma.

Figure 7: The final pathology diagnosis of this patient was anaplastic oligodendroglioma in the right parietal lobe in 2005. Photomicrographs of the tumor showed that there were higher cell densities, densely packed round cells with perinuclear haloes, microscopically round-to-oblong cells with hyperchromatism and pleomorphism, and clusters of capillary or plexiform capillaries.

Figure 8: The final pathology diagnosis of this patient was anaplastic oligodendroglioma in the right parietal lobe in 2009 by HE stain.

Figure 9: The anaplastic oligodendroglioma in the right parietal lobe in 2009 stained by immunohistochemical staining. They were strongly positive for Ki-67, with proliferation index > 15%.

Figure 10: The anaplastic oligodendroglioma in the right parietal lobe in 2009 stained by immunohistochemical staining. They were positive for the marker oligodendrocyte transcription factor (Oligo-2).

Figure 11: The anaplastic oligodendroglioma in the right parietal lobe in 2009 stained by immunohistochemical staining. They were negative for O6-methylguanine-DNA methyltransferase (MGMT).

The diagnostic molecular markers, such as 1p and 19q LOH in original brain tumor samples in 2009 were tested. Fluorescent in situ hybridization (FISH) assay on samples from the patient was evaluated with routine dual-color FISH assay as previously published [11]. The FISH result of specimens in 2009 showed 1p and 19q loss of heterozygosity (LOH) (Figures 12a and 12b). Briefly, paired probes for LSI 1p36 (red)/1q25 (green) and 19q13 (red)/19p12 (green) (Pathvysion) were prepared. Green and red fluorescent signals were enumerated under a Leica Microscope with appropriate filters. For each hybridization slice, a minimum of 40 non-overlapping nuclei was assessed for numbers of green and red signals, counted separately by two individuals. DNA sequencing of IDH-1 mutation was performed on the paraffin samples in 2009. The result showed that samples in 2009 had IDH-1 R132H mutation (Figure 13).
Figure 12: The fluorescent in situ hybridization (FISH) assay on samples from the patient was evaluated with routine dual-color FISH assay. The FISH result of specimens in 2009 showed 1p and 19q loss of heterozygosity (LOH) (Figure 12a, 12b, respectively). Briefly, paired probes for LSI 1p36 (red)/1q25 (green) and 19q13 (red)/19p12 (green) (Pathvysion) were prepared. Green and red fluorescent signals were enumerated under a Leica Microscope with appropriate filters. For each hybridization slice, a minimum of 40 non-overlapping nuclei was assessed for numbers of green and red signals, counted separately by two individuals.

Figure 13: DNA sequencing of IDH-1 mutation was performed on the paraffin samples in 2009. The result showed that samples in 2009 had IDH-1 R132H mutation.

After conversation with the patient and his family, he was given radiation therapy to the right pelvis bone lesion (52Gy/20f, 6MV-X ray, Varian 21EX). A significant pain alleviation was observed 1 month after radiation. Three and six months after radiation, the KPS (Karnofsky score) for the patient was 100 with no further pelvis pain until the last follow-up.

DISCUSSION

Extracranial metastasis of original glioma was extremely rare. However, it was reported that glioma could exhibit multiple and systematic metastases [2-3]. Following conditions are defined for a metastasizing glioma: (1) the presence of a single histologically characteristic tumor of the CNS must have been proved; (2) the clinical history must indicate that initial symptoms are due to this tumor; (3) an autopsy must have been performed and reported in sufficient detail to rule out the possibility of any other primary site; and (4) the morphology of the glioma and of the distant metastases must have been identical with due allowance for differences in degree of heteromorphism [1-11]. Extracranial metastases from high grade glioma have been reported [1-3, 8-10]. These reports document that higher incidence of malignant tumors result in higher incidences of metastases, especial glioblastoma (GBM). Metastatic disease of glioma usually occurs within the neuraxis but extracranial spread to many organs and regions, including the pleura, small bowel and pancreas [12], lung [13, 14, 18], intradural extramedullary, leptomeninges [14, 15, 16] and spine [16, 17], lymph nodes [17, 18], cervical muscles and epidural space [18], scalp [14, 19, 20], bone [17, 21], and liver [22]. Although the reported incidence of extracranial glioma is very low (<1%), this phenomenon may not be as rare as we believed, primarily because we do not systematically look for metastases outside the brain. Extracranial metastasis is considered correlated with multiple craniotomies, shunt surgery, and long-term survival [23-26]. Literature reviews indicate that post-transmission metastases of intracranial GBM occur rarely following organ transplantation, with some concerns on the safety for transplant of organs from affected donors [24-25].

The diagnosis of metastatic disease is based on the high histopathological similarity to the original tumor. Sometimes the tumor becomes undifferentiated and morphological diagnosis can be difficult, requiring immunostaining by glial fibrillary acid protein (GFAP) and S100 protein [10]. In our case, radiologic studies and autopsy did suggest secondary metastasis from CNS primary tumor. In addition, a biopsy specimen from the right iliac bone was immunoreactive positive for GFAP and S100 protein. Consequently, we diagnosed this patient with multiple pelvis bone metastases from glioma. Although the first and second diagnoses were astrocytoma (WHO grade II) by HE (hematoxylin and eosin) stain without immunohistochemistry in 2005 and in 2009 at the local country hospital, the paraffin sample by HE stain and immohistochemistry in our center confirmed the pathology diagnosis of first and second resection samples in this patient as anaplastic oligodendroglioma, which was also confirmed by three state pathology centers.

Liwicz and Rubinstein reported that the presence of metastasis was 5.25% in oligodendrogliomas [11]. Oligodendrogliomas are rare neoplasms with an incidence of 2%-5% of all primary brain tumors. Oligodendrogliomas have been reported to metastasize to the neck lymph nodes, bones or bone marrow, spinal dura mater, thymus gland and chest wall but bones and bone marrow are the most common sites [26-29]. The high predilection for the bone and bone marrow may be linked to the presence of the neural cell
adhesion molecule, which seems to be fundamental for the function of the bone marrow microenvironment and hematopoiesis [29]. A literature review yielded 61 reports of metastatic anaplastic oligodendrogliomas from 1951 to the present [26]. In our present case, a systematic examination found multiple right pelvis bone metastases, including the right acetabulum, corpus ossis ischii, pubis superior and inferior ramus and tuber ischiadicum osteoblastic metastases 9 years and 5 years after the first and second resection, respectively. Metastases in these sites suggested that anaplastic oligodendroglioma cells were delivered via the blood vessels. Most cases of extracranial metastasis occurred after craniotomy [26-29]. Although craniotomy, usually at least as a diagnostic procedure in patients with brain tumors, is also considered the main responsible factor, one should take into account the possibility of a rare event, namely spontaneous distant metastases of these tumors.

The 1p/19q LOH is strongly associated with classic oligodendroglioma morphology, prolonged overall patient survival, and enhanced responses to both alkylating chemotherapeutic agents and radiation. Among gliomas, 1p/19q LOH is associated with tumors with oligodendroglial components. Combined deletions of both arms have been observed in up to 70% of oligodendrogliomas. Oligodendrogliomas is characterized by 1p19q LOH, which is unique to oligodendrogliomas and not seen in low- and high-grade astrocytomas [30]. 1p/19q LOH may be closely connected to classic oligodendroglioma morphology, longer survival, and sensitivity to alkylating chemotherapeutic agents and radiation [31, 32]. The presence of 1p19q LOH may provide a proof of oligodendrogliomas, as 1p19q LOH had been reported to occur in over 85% [33] and 68% of oligodendrogliomas [34], suggesting the presence of 1p19q LOH is strongly correlated with the histological diagnosis corresponding to oligodendroglioma. The 1p/19q co-deletion is mediated by an unbalanced translocation of 19p to 1q:der (1;19) (p10;q10), which occurs during the tumorigenesis of oligodendroglioma [35]. In our case, the 1p/19q LOH was found in samples obtained in 2009. This further proved that the final pathologic diagnosis should be oligodendrogliomas, not astrocytomas.

Several researches found that patients with 1p/19q LOH had longer survival time and longer time to first recurrence [10, 29]. The longer survival and good prognosis may be due, firstly, to the presence of 1p/19q, as combined deletion of the 1p and 19q chromosomal arms (LOH) is expected in oligodendrogliomas [6, 8-11, 26-29]. This increased survival in patients with 1p/19q codeleted tumors remained significant when adjustments were made for age, tumor grade, type of surgical procedure, and treatment with radiation or chemotherapy. It suggests that in classic oligodendrogliomas: 1p/19q tumor status is a powerful predictor of patient survival, even after recurrence [36]. Further, deletions of 1p and 19q have been associated with a prolonged survival in patients with anaplastic oligodendrogliomas using retrospectively collected cohorts. The prognostic relevance of the deletions has been validated by prospective clinical trials. Oligodendroglial neoplasms show a demonstrable radiographic response to chemotherapeutic agents, especially PCV (procarbazine/CCNU/ vincristine), but more recently temozolamide as well, even in metastatic deposits of anaplastic oligodendroglioma [20]. Tumors with such a co-deletion are sensitive to comprehensive therapy, with 90%-100% of patients responding [9, 32]. Both retrospective and prospective data suggest that 1p/19q LOH predict the responsiveness of anaplastic oligodendroglial tumors to combined radiation therapy and chemotherapy. In short, molecular studies have revealed that deletions of chromosome 1p and 19q are not only tumor marker of oligodendroglial tumors, but also usually associated with longer survival in OGD, as well as a better response to irradiation and chemotherapy.

Fallon et al. found that co-deletions of 1p/19q were conserved in 100% of oligodendroglial tumors at diagnosis and recurrence [36]. Campbell et al. investigated 24 patients with oligodendroglial neoplasms and 53 tumor specimens and also reported that 100% of those with 1p/19q co-deletions demonstrated persistent 1p/19q co-deletions in progressive disease and recurrences [37]. Thus, progression of these tumors does not appear to be due to a proliferating subpopulation of treatment resistant cells [29]. The prolonged survival of oligodendroglioma patients associated with their enhanced chemosensitivity may be a contributing factor to the recently increased numbers of metastatic oligodendrogliomas reported in the literature [38-41]. Investigations into the molecular and genetic make-up of oligodendrogliomas have provided new insight into overall survival and responses to therapy.

On the other hand, the prolonged survival resulting from concomitant radiotherapy and chemotherapy, which could significantly alter the immune status of patients with primary brain tumors, will probably increase the possibility of the development of undetected metastases [26, 29-32]. It is important to bear this in mind, particularly in cases when...
the history of primary brain tumor is unavailable. For our patient, the first tumors from the brain lesions, as well as the secondary lesions, all had 1p/19q LOH. Thus the 1p/19q LOH had led to a longer survival time (more than 9 years) under gamma ray radiotherapy and chemotherapy, and then bone metastasis occurred.

Isocitrate dehydrogenase (IDH) catalyzes the oxidative decarboxylation of isocitrate (ICT) to produce α-ketoglutarate (α-KG) [42]. Mutated IDH1 consumed rather than produced NADPH, thus likely lowering NADPH levels even further. The low NADPH levels could sensitize glioma to radiation and chemotherapy, thus explaining the prolonged survival of patients with IDH1 mutated glioma [43-45]. After assessing IDH1 mutations in 321 gliomas of various histological types and biological behaviors, the fact that high frequencies of IDH1 mutations were found in oligodendrogliomas (79%), and there were no cases in which an IDH1 mutation occurred after the acquisition of either a TP53 mutation or 1p/19q LOH suggested IDH1 mutations were very early events in glioma genesis and might affect a common glial precursor cell population [46]. The IDH1 and IDH2 mutations are relevant to the progression of gliomas, the prognosis and treatment of the patients with gliomas harboring the mutation [47]. Expression of the R132H IDH1 mutant, rather than wild-type IDH1, strongly induced the expression of HIF-1α target genes, such as glucose transporter 1 (Glut1), vascular endothelial growth factor (VEGF), and phosphoglycerate kinase (PGK1). IDH1 and IDH2 mutations may have significant applications for the diagnosis, prognosis, and treatment of patients with these tumors. Presence of IDH1 mutations in anaplastic oligodendrogial tumors was shown to be associated with a significantly better outcome [47]. Novel IDH1 sequencing and staining techniques have allowed this marker to play an increasingly important role in the histological determination of brain tumor specimens [48-49]. Together, IDH-1 mutation might lead to a longer survivor and cell metastasis potential increase, and there might be other genes such as HIF-1, VEGF, etc. that are involved to trigger other pathways [47-49]. For our patient, the samples from the brain lesions in 2009 had IDH-1 R132H mutation. Thus the R132H mutation combined with 1p/19q LOH may have led to a longer survival time under comprehensive therapy. The right pelvis metastasis had higher Ki67 than the original brain tumor, indicating that metastatic lesions might have more malignant potential than the original brain tumors after a long time survive. Further studies are needed to determine which pattern of enzyme expression or molecular genetic finding may identify patients with primary brain tumors at risk of developing extraneural metastases.

CONCLUSIONS

In summary, we would like to stress the importance of being aware of the possibility of oligodendrogial tumors to metastasize extracranially. We should alert the treating clinician to the features that are common in ODG metastases, because of lessons to be learned from the combination of all these reported oligodendrogliomas cases. The molecular markers such as the 1p/19q LOH and IDH-1 mutations not only help prove the pathologic diagnosis of oligodendrogliomas, but also help elucidate a subtype of oligodendrogliomas, which have a good prognosis even with extracranial metastases. The relationship and mechanism of 1p19q LOH and IDH-1mutation to extracranial metastasis requires further research in the future.

CONFLICT OF INTEREST

All authors declared to disclose any actual or potential conflict of interest including any financial, personal or other relationships with other people or organizations within three years of beginning the submitted work that could inappropriately influence, or be perceived to influence, their work.

REFERENCES


