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ORIGINAL REPORT

MGMT Promoter Methylation Status Can Predict the Incidence and Outcome of Pseudoprogression After Concomitant Radiochemotherapy in Newly Diagnosed Glioblastoma Patients

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ABSTRACT

Purpose
Standard therapy for glioblastoma (GBM) is temozolomide (TMZ) administration, initially concurrent with radiotherapy (RT), and subsequently as maintenance therapy. The radiologic images obtained in this setting can be difficult to interpret since they may show radiation-induced pseudoprogression (psPD) rather than disease progression.

Methods
Patients with histologically confirmed GBM underwent radiotherapy plus continuous daily temozolomide (75 mg/m^2/d), followed by 12 maintenance temozolomide cycles (150 to 200 mg/m^2 for 5 days every 28 days) if magnetic resonance imaging (MRI) showed no enhancement suggesting a tumor; otherwise, chemotherapy was delivered until complete response or unequivocal progression. The first MRI scan was performed 1 month after completing combined chemoradiotherapy.

Results
In 103 patients (mean age, 52 years [range 20 to 73 years]), total resection, subtotal resection, and biopsy were obtained in 51, 51, and 1 cases, respectively. *MGMT* promoter was methylated in 36 patients (35%) and unmethylated in 67 patients (65%). Lesion enlargement, evidenced at the first MRI scan in 50 of 103 patients, was subsequently classified as psPD in 32 patients and early disease progression in 18 patients. PsPD was recorded in 21 (91%) of 23 methylated *MGMT* promoter and 11 (41%) of 27 unmethylated *MGMT* promoter ($P = .0002$) patients. *MGMT* status ($P = .001$) and psPD detection ($P = .045$) significantly influenced survival.

Conclusion
PsPD has a clinical impact on chemotherapy-treated GBM, as it may express the glioma killing effects of treatment and is significantly correlated with MGMT status. Improvement in the early recognition of psPD patterns and knowledge of mechanisms underlying this phenomenon are crucial to eliminating biases in evaluating the results of clinical trials and guaranteeing effective treatment.

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INTRODUCTION

Data recently reported in the randomized EORTC 22981/26981–NCIC CE.3 (European Organisation for Research and Treatement of Cancer/National Cancer Institute of Canada) phase III trial on newly diagnosed patients with glioblastoma (GBM) patients[1] given temozolomide (TMZ) plus radiotherapy (RT) have provided a new standard of care. A small, but significant, progression-free survival (PFS) advantage (5 months *v* 6.9 months) has been achieved with this approach, whereas a marked and significant benefit has been obtained in 2-year overall survival (11% *v* 27%). This type of effect is not frequent in medical oncology, where significant PFS advantages do not often translate into an overall survival advantage. The conversion of a small PFS advantage into a consistent survival benefit may depend on the overestimation of disease progression in the temozolomide-radiotherapy arm. Classically, response evaluation in neuro-oncology is based on planimetric variations in enhanced lesions, but it is also based on corticosteroids dosage and variations in neurological conditions.[2] However, the brief time

interval from the end of radiotherapy and its combination with a sensitizing agent such as TMZ could create biases in neuroradiological imaging evaluation. Radiation injury to the CNS may, in fact, depend on increased capillary permeability induced by radiotherapy, leading to fluid transudation into the interstitial space and consequent brain edema. Furthermore, if capillary permeability is altered, damage from chemotherapy may occur earlier and be more severe; radiotherapy may enhance the efficacy of chemotherapy by maximizing drug uptake either at the cell membrane, through a disruption of the blood-brain barrier, and/or through an alteration in cell metabolism.[3-5] This can lead to the observation of an early radiological increase in contrast enhancement at magnetic resonance imaging (MRI) consequent to alterations in the blood-brain barrier, thus falsely suggesting tumor progression. This phenomenon (also called therapy-induced necrosis or pseudoprogression [psPD], which may be the expression of treatment-induced necrosis) leads to the rupture of the hematoencephalic barrier and the passage of contrast medium. Although this phenomenon has long been known,[3,4,6] its real incidence has not yet been reported in a large series of patients given concomitant radiotherapy and TMZ treatment; nor has the potential impact of O^6-methylguanine–DNA methyltransferase (*MGMT*) promoter methylation status been described in this patient category. A retrospective analysis was therefore made of newly diagnosed GBM patients, with assessable *MGMT* promoter methylation status treated prospectively with radiotherapy plus continuous daily temozolomide (75 mg/m^2/d), followed by 12 maintenance temozolomide cycles (150 to 200 mg/m^2 for 5 days every 28 days).

METHODS

Patient Eligibility

Adult patients with newly diagnosed GBM were prospectively enrolled onto the trial if they had a WHO performance status of 2 or less and adequate hematological, renal, and hepatic function (absolute neutrophil count, $\geq$ 1,500/mm^3; platelet count, $\geq$ 100,000/mm^3; serum creatinine level, $\leq$ 1.5 times the upper limit of the normal laboratory range; total serum bilirubin level, $\leq$ 1.5 times the upper limit of the normal range; liver-function values, $<$ 3 times the upper limit of the normal laboratory range). Patients were treated with radiotherapy plus continuous daily temozolomide (75 mg/m^2/d) followed by 12 cycles of maintenance temozolomide (150 to 200 mg/m^2 for 5 days every 28 days). Treatment was suspended after 12 cycles only if the MRI showed no enhancement suggesting presence of tumor; otherwise, chemotherapy was delivered until complete response or clear disease progression. The first MRI was planned 4 weeks after the end of concurrent chemoradiotherapy. If patients presented stable disease or had no evident lesion, they were considered as having nonprogressive disease (non-PD) and TMZ was continued. In cases of lesion growth, which may be due to potential early-delayed reactions after RT,[7] altering immediate post-RT neuroradiological imaging, another two cycles were delivered followed by another MRI. At this point, the lesions were considered psPD if they were stable or had improved; otherwise they were registered as early disease progression (ePD) and TMZ was suspended. TMZ was suspended at anytime if the MRI image evidenced a new lesion outside the radiotherapy field. All patients with psPD and non-PD could experience a PD in the course of the disease. Moreover, patients were evaluated taking into account clinical and neurological examinations (performed monthly before each cycle) according to MacDonald's criteria[2] by a multidisciplinary team consisting of an oncologist and a neuroradiologist. Neurological status was assessed by considering signs and symptoms possibly correlated with progression with respect to the previous examination; each variation in daily corticosteroids dosage was recorded.

A retrospective analysis was made to correlate *MGMT* promoter methylation status and type of progression. All patients signed a form giving their fully informed consent to take part in the prospective study on the duration of maintenance chemotherapy; they also gave their consent in writing for research tests to be conducted on the tissue blocks obtained from them in any future research projects approved by ethical committee and aiming to improve on the understanding and treatment of brain tumors.

The study, approved by the institutional review board of Padova Azienda Ospedaliera (Padova, Italy), was conducted according to the principles of the Declaration of Helsinki and the rules of Good Clinical Practice.

MGMT *Status Assessment*

MGMT status was evaluated with the methylation specific polymerase chain reaction (MSP) after a nested-polymerase chain reaction protocol,[8] using methods and assessment criteria described elsewhere.[9] Because the quality of DNA obtained from formalin-fixed, paraffin-embedded tumor tissue affects the success rate of MSP, in some cases *MGMT* methylation status was determined using a different nested MSP approach, with a first pair of primers to obtain smaller amplicons (129 base pairs), for which forward and reverse primers have been described.[8,10] The results obtained in the present study were verified using a second step of both modification and nested polymerase chain reaction; the entire process was repeated in triplicate in some cases.

Statistical Analysis

Tumor progression was defined according to MacDonald's Criteria[2] as a 25% increase in tumor size, the appearance of new lesions, or an increased need of corticosteroids. Time-to-progression (TTP) and overall survival (OS) were measured from the time of surgery to disease progression or death, respectively, or date of last follow-up, and analyzed using the Kaplan-Meier method; 95% CIs were calculated using the associated estimated SEs. The log-rank test was employed to compare *MGMT* promoter methylation status, methylated versus unmethylated and psPD versus ePD and to test the significance of the following prognostic variables: age, extent of surgery, and performance status.[11] Multivariate analysis was performed using the Cox proportional hazards model. Significance level was set at $P < .05$.

RESULTS

Between September 2001 and January 2007, 208 patients with newly diagnosed GBM were treated with concurrent RT/TMZ followed by 12 cycles of maintenance chemotherapy according to the above-described protocol. An analysis was made of all patients (n = 103) for whom *MGMT* promoter methylation status was assessable. The median follow-up of patients included in the analysis was 18.93 months (range, 6.6 to 62 months). The patients' baseline characteristics are presented in Table 1. The median number of maintenance TMZ cycles was 6 (range, 0 to 30 cycles). One patient had rapid disease progression after completion of concomitant treatment, and it was impossible to administer the first cycle of maintenance chemotherapy; this patient was therefore considered ePD, and his data was included in the analysis.

Toxicity

During the concomitant therapy phase, grade 4 neutropenia occurred in one patient (1%), and grade 3 to 4 thrombocytopenia in four patients (3.9%). Grade 1 to 2 lymphocytopenia occurred in 10 patients (9.7%). One patient had pneumonia with normal WBCs. During the maintenance therapy phase, grade 3 to 4 neutropenia and thrombocytopenia occurred in 2% and 5% of patients, respectively. Two patients discontinued treatment in the maintenance phase of therapy: one during the third cycle due to prolonged grade 4 thrombocytopenia, and one after the fifth cycle due to prolonged grade 2 thrombocytopenia.

Table 1. Baseline Characteristics of Patients

Characteristic	Patients No.	Patients %
Age, years		
Median	53	
Range	20-73	
Sex		
Male	68	66
Female	35	34
WHO performance status		
0	18	17
1	77	75
2	8	8
Extent of surgery		
Biopsy	1	1
Subtotal resection	51	49.5
Total resection	51	49.5
MGMT promoter status		
Methylated	36	35
Unmethylated	67	65

Abbreviation: *MGMT*, O^6-methylguanine–DNA methyltransferase.

Evaluation at First MRI After Concomitant Radiochemotherapy and Correlation With Mgmt Status

At the first MRI scan, performed 1 month after concurrent RT/TMZ, lesion enlargement was recorded in 50 patients (48.5%), while 53 patients were non-PD. The findings were psPDs in 32 (64%) of 50 patients and ePDs in 18 (36%) of 50 patients. *MGMT* promoter was methylated in 21 (66%) of the 32 psPD patients and in two (11%) of the 18 ePD patients ($P = .0002$). Thirteen (25%) of the 53 non-PD patients had *MGMT* promoter methylated and the other 40 patients had *MGMT* promoter unmethylated status (Fig 1). A significant difference was found between the non-PD and the psPD group ($P = .0002$), but not between non-PD and ePD groups ($P = .23$), for *MGMT* promoter status. Clinical deterioration was found in 21 (42%) of 50 patients with enlarged lesion images, being present in 10 (55.6%) of 18 with ePD, and in 11 (34%) of 32 patients with psPD ($P = .14$). All patients with psPD and clinical deterioration had a recovery of clinical function at a median time of 7 months (range 1.2 to 18 months). *MGMT* promoter status predicted psPD in 91.3% of methylated cases (95% CI, 72% to 99%), but predicted ePD in only 59% of unmethylated cases (95% CI, 38% to 76%).

TTP

In the present study, *MGMT* status significantly influenced overall median TTP, which was 11.7 months (95% CI, 8.9 to 14.5 months), being 21.9 months (95% CI, 12.9 to 30.8 months) in *MGMT* promoter methylated patients and 9.2 months (95% CI, 8.5 to 9.8 months) in *MGMT* promoter unmethylated patients ($P < .0001$). Extent of surgery ($P = .44$), age ($P = .69$) and performance status ($P = .86$) were not significantly correlated with TTP. In 85 patients (32 psPD; 53 non-PD), the psPD patients had a significantly longer mTTP than the non-PD patients (20.7 *v* 11.4 months; $P = .001$; Table 2). In the subgroup of patients with psPD, the median time interval between recording psPD and subsequent real progression was 16.2 months: 21 months in *MGMT* promoter methylated patients and 15.3 months in *MGMT* promoter unmethylated patients ($P = .41$). Subsequent disease progression was recorded in 21 (65.6%) psPD and in 46 (86.8%) non-PD patients ($P = .02$).

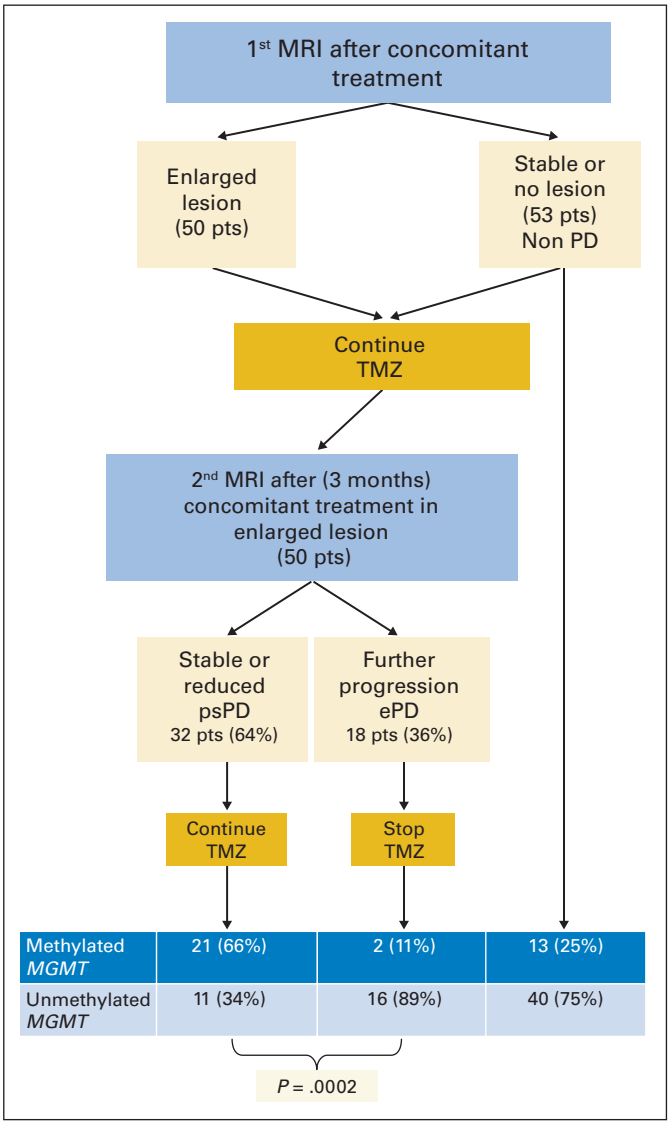

Fig 1. MRI findings, outcomes and MGMT status of patients. MRI, magnetic resonance imaging; PD, disease progression; TMZ, temozolomide; psPD, pseudoprogression; ePD, early disease progression; *MGMT*, O^6-methylguanine–DNA methyltransferase.

Overall Survival

A median survival of 20.7 months was achieved (95% CI, 17.3 to 24 months): 43.6 months (95% CI, 25.5 to 61.7 months) and 16.8 months (95% CI, 14.1 to 19.6 months) in methylated *MGMT* promoter and in unmethylated *MGMT* promoter patients, respectively ($P < .0001$; Fig 2). In 53 patients without images of lesion increase after combined chemoradiotherapy, censored patients were significantly higher in methylated *MGMT* promoter subgroup (nine of 13 patients; 69%) with respect to the unmethylated *MGMT* promoter subgroup (nine of 40 patients; 22.5%; $P = .002$) despite the median follow-up for methylated *MGMT* promoter being significantly higher than in unmethylated *MGMT* promoter patients (21.6 and 17.8

Table 2. Effects of *MGMT* Promoter Methylation Status and First MRI Findings

Characteristic	TTP (months)	OS (months)
MGMT promoter status		
Median	11.7	20.7
Methylated	21.9*	43.6*
Unmethylated	9.2	16.8
MRI findings		
psPD	20.7*	38*
ePD	5.7	10.2
No PD images	11.4	20.2

Abbreviations: *MGMT*, O^6-methylguanine–DNA methyltransferase; TTP, time to diease progression; OS, overall survival; psPD, pseudoprogression; ePD, early disease progression; PD, disease progression.
*P = significant

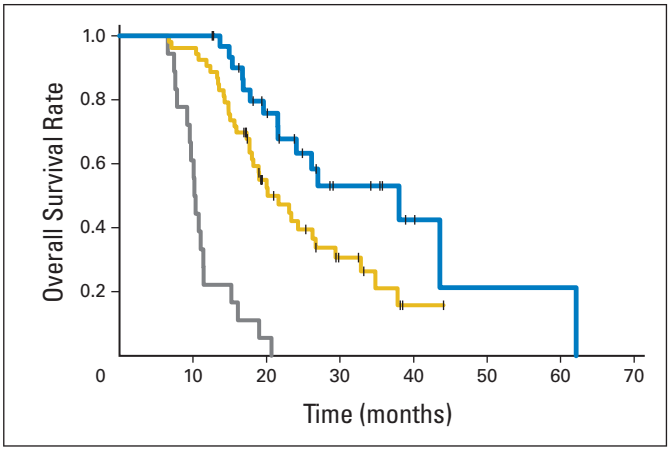

Fig 3. Overall survival: blue line, patients with pseudoprogression; gray line, patients with early disease progression; yellow line, patients with neither pseudoprogression nor early disease progression.

months, respectively; $P < .05$). Extent of surgery ($P = .10$), second surgery ($P = .12$), age ($P = .65$), and performance status ($P = .36$) were not significant prognostic factors. Median survival was significantly influenced by psPD, with a value of 38 months in this group, 10.2 months in patients with ePD, and 20.2 months in patients with non-PD ($P < .0001$; Fig 3), and by the number of TMZ cycles administered (< 6 cycles $v \geq 6$ cycles; 13.7 and 34.8 months; $P < .0001$). At multivariate analysis, survival was significantly influenced by *MGMT* promoter methylation status ($P < .001$) and by the detection of psPD ($P = .045$). As the number of TMZ is not an independent variable, it was not evaluated in the regression model.

DISCUSSION

psPD was recently described in neuro-oncology as a transient blood-brain barrier alteration with neuroradiological images frequently indistinguishable from disease progression. Some authors have underlined the problems linked to this entity[12-15] (Table 3). De Wit et al,[13] who showed that transient neuroradiological enhancement simulating progression can appear within 3 months after the end of radiotherapy, focused on the potential risk of including patients in clinical trials on recurrent disease that is not really in progression but in psPD.

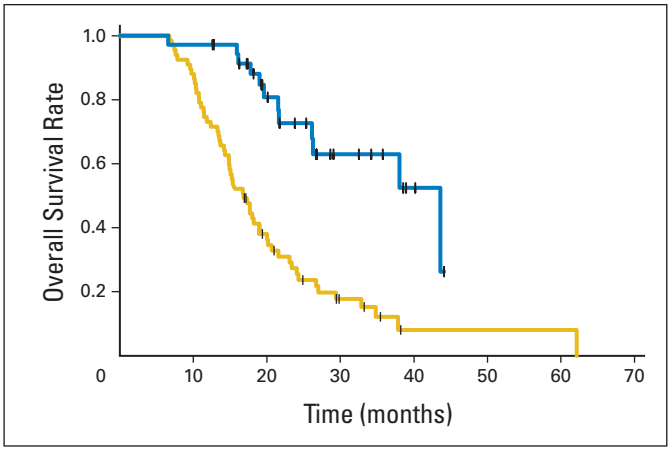

Fig 2. Overall survival by presence of O^6-methylguanine–DNA methyltransferase (*MGMT*) promoter methylation status. Blue line, patients with methylated *MGMT* promoter; yellow line, patients with unmethylated *MGMT* promoter status.

Chamberlain et al[12] evaluated 65 GBM patients treated with concurrent RT and TMZ and reported that seven of 15 (46%) of those who underwent surgical resection for suspected recurrence had histologically confirmed psPD with patterns of radiation-induced necrosis.

Using proton MR spectroscopic imaging, specific changes were reported in cases of radiation injury including a reduction in *N*-acetylaspartate (NAA) and various changes in choline and creatine levels and/or alterations in choline/NAA and choline/creatine ratios, described elsewhere.[16,17] Moreover, perfusion MRI is considered a useful tool in the diagnosis of recurrence and necrosis; changes in cerebral tumor blood volume occurring during the early radiotherapy course can also be predictive of survival. Some authors have observed that apparent diffusion coefficient values are useful in distinguishing between high-grade glioma and normal tissue, though they do not allow a differentiation between a high-grade glioma and the surrounding edema.[18,19] Nevertheless, to date, the only available way of distinguishing between psPD and PD by conducting a follow-up on patients with early enlarged images, as standard MRI is not sufficient, nor have alternative neuroradiological techniques been validated in prospective trials. Furthermore, the real impact of this entity has not yet been established due to the absence of prospective studies on large series consisting exclusively of patients who have been treated with concomitant radio-chemotherapy. The findings made in the present study show, for the first time, that the real incidence of psPD in GBM patients treated with concomitant TMZ and RT is 30%. Moreover, in approximately 50% of patients, the first MRI scan images after combined RT/TMZ were doubtful for progression, but only 36% of these patients were subsequently evaluated as true ePD; the other 64% had a psPD. Therefore, the next step for clinical research should be a priori identification of patients with psPD. We found that there is a 91.3% (95% CI, 72% to 99%) probability of psPDs in patients with methylated *MGMT* promoter tumors and a 59% (95% CI, 38% to 76%) probability of early PD in unmethylated *MGMT* promoter tumors. If the probability of methylated *MGMT* promoter patients having psPD is high, it is almost equally probable that unmethylated *MGMT* promoter patients will have psPD or ePD if the first MRI images reveal

Table 3. Studies on psPD in GBM Patients Treated With Concurrent Chemoradiation

Study	No. of Patients	% of psPD	*MGMT* Promoter Status
Chamberlain et al, 2006[12]	65	46.7*	Not reported
Jefferies, 2007[15]	15	20	Not reported
Taal et al, 2007[14]	85	21	Not reported
Present study	103	31	Reported

Abbreviations: GBM, glioblastoma; psPD, pseudoprogression; *MGMT*, O^6-methylguanine–DNA methyltransferase.
*Calculated in patients undergoing resection for images of lesion enlargement.

lesion enlargement. New prospective studies testing new neuroradiological techniques on larger patient populations are therefore required in order to obtain sounder findings, and to study alternative psPD predictors. The higher rates of methylated *MGMT* promoter found in patients with psPD is probably correlated with the efficacy of concurrent RT/TMZ treatment on the residual tumor burden; in this setting the neuroradiological image of psPD may represent not only a treatment-induced blood brain barrier disruption, but also reflect the efficacy of therapy, since the OS of patients with, was significantly higher than in those without psPD.

It has not yet been demonstrated that maintenance chemotherapy prolongs the survival of patients with solid tumors. However, prolonged TMZ therapy can substantially deplete *MGMT*,[20] thus providing the rationale for continuous treatment. In the present study, patients who received less than six TMZ cycles had an OS of 13.7 months, while those who received $\geq$ six TMZ cycles had an OS of 34.8 months ($P < .0001$). However, in view of the presence of several factors that may have influenced the duration of maintenance therapy, we decided not to perform a multivariate analysis of this datum, also in view of the nonrandomized nature of our trial.

More information is required for a better understanding of the nature of psPD in order to distinguish it from real early PD, thus obviating biases in the evaluation of results from clinical trials, and preventing patients from being denied effective treatment. Trials should also be conducted to evaluate the predictive value of novel neuroradiogical techniques, such as the impact of prolongation of maintenance TMZ and intensified schedules; the ongoing RTOG 0525/EORTC 26052-22053 trial is investigating this issue. Moreover, as vascular damage may play a role in the pathogenesis of this radiological pattern or therapy-induced effect, the evaluation of angiogenetic pathways and correlations with *MGMT* status in GBM will be the backbone for future research.

AUTHORS' DISCLOSURES OF POTENTIAL CONFLICTS OF INTEREST

The author(s) indicated no potential conflicts of interest.

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REFERENCES

1. Stupp R, Mason WP, van den Bent MJ, et al: Radiotherapy plus concomitant and adjuvant temozolomide for glioblastoma. N Engl J Med 352:987-996, 2005
2. Macdonald DR, Cascino TL, Schold SC Jr, et al: Response criteria for phase II studies of supratentorial malignant glioma. J Clin Oncol 8:1277-1280, 1990
3. Brandes AA, Rigon A, Zampieri P, et al: Carboplatin and teniposide concurrent with radiotherapy in patients with glioblastoma multiforme: A phase II study. Cancer 82:355-361, 1998
4. DeAngelis LM, Delattre JY, Posner JB: Radiation-induced dementia in patients cured of brain metastases. Neurology 39:789-796, 1989
5. Stupp R, Dietrich PY, Ostermann Kraljevic S, et al: Promising survival for patients with newly diagnosed glioblastoma multiforme treated with concomitant radiation plus temozolomide followed by adjuvant temozolomide. J Clin Oncol 20:1375-1382, 2002
6. Kumar AJ, Leeds NE, Fuller GN, et al: Malignant gliomas: MR imaging spectrum of radiation therapy- and chemotherapy-induced necrosis of the brain after treatment. Radiology 217:377-384, 2000
7. Sheline GE, Wara WM, Smith V: Therapeutic irradiation and brain injury. Int J Radiat Oncol Biol Phys 6:1215-1228, 1980
8. Palmisano WA, Divine KK, Saccomanno G, et al: Predicting lung cancer by detecting aberrant promoter methylation in sputum. Cancer Res 60:5954-5958, 2000
9. Brandes AA, Tosoni A, Cavallo G, et al: Correlations between O^6-methylguanine–DNA methyltransferase promoter methylation status, 1p and 19q deletions, and response to temozolomide in anaplastic and recurrent oligodendroglioma: A prospective GICNO study. J Clin Oncol 24:4746-4753, 2006
10. van Engeland M, Weijenberg MP, Roemen GM, et al: Effects of dietary folate and alcohol intake on promoter methylation in sporadic colorectal cancer: The Netherlands cohort study on diet and cancer. Cancer Res 63:3133-3137, 2003
11. Mirimanoff RO, Gorlia T, Mason W, et al: Radiotherapy and temozolomide for newly diagnosed glioblastoma: Recursive partitioning analysis of the EORTC 26981/22981-NCIC CE3 phase III randomized trial. J Clin Oncol 24:2563-2569, 2006
12. Chamberlain MC, Glantz MJ, Chalmers L, et al: Early necrosis following concurrent Temodar and radiotherapy in patients with glioblastoma. J Neurooncol 82:8-83, 2006
13. de Wit MC, de Bruin HG, Eijkenboom W, et al: Immediate post-radiotherapy changes in malignant

glioma can mimic tumor progression. Neurology 63:535-537, 2004

14. Taal W, Brandsma D, de Bruin HG, et al: The incidence of pseudo-progression in a cohort of malignant glioma patients treated with chemo-radiation with temozolomide. J Clin Oncol 25:18s, 2007 (suppl; abstr 2009)

15. Jefferies S, Burton K, Jones P, et al: Interpretation of early imaging after concurrent radiotherapy and temozolomide for glioblastoma. Clin Oncol (R Coll Radiol) 19:S33, 2007

16. Schlemmer HP, Bachert P, Henze M, et al: Differentiation of radiation necrosis from tumor progression using proton magnetic resonance spectroscopy. Neuroradiology 44:216-222, 2002

17. Schlemmer HP, Bachert P, Herfarth KK, et al: Proton MR spectroscopic evaluation of suspicious brain lesions after stereotactic radiotherapy. AJNR Am J Neuroradiol 22:1316-1324, 2001

18. Catalaa I, Henry R, Dillon WP, et al: Perfusion, diffusion and spectroscopy values in newly diagnosed cerebral gliomas. NMR Biomed 19:463-475, 2006

19. Castillo M, Smith JK, Kwock L, et al: Apparent diffusion coefficients in the evaluation of high-grade cerebral gliomas. AJNR Am J Neuroradiol 22:60-64, 2001

20. Brandes AA, Tosoni A, Cavallo G, et al: Temozolomide 3 weeks on and 1 week off as first-line therapy for recurrent glioblastoma: Phase II study from gruppo italiano cooperativo di neuro-oncologia (GICNO). Br J Cancer 95:1155-1160, 2006

