Emerging antiangiogenic treatments for gliomas – efficacy and safety issues

Jörg Dietrich[a,b,c], Andrew D. Norden[a,b] and Patrick Y. Wen[a,b]

[a]Division of Neuro-Oncology, Department of Neurology, Brigham and Women's Hospital, Harvard Medical School, [b]Center for Neuro-Oncology, Department of Medical Oncology, Dana-Farber/Brigham and Women's Cancer Center and [c]Stephen E. and Catherine Pappas Center for Neuro-Oncology, Massachusetts General Hospital, Harvard Medical School, Boston, Massachusetts, USA

Correspondence to Patrick Y. Wen, MD, Center for Neuro-Oncology, Dana-Farber Cancer Institute, Shields Warren 430D, 44 Binney Street, Boston, MA 02115, USA
Tel: +1 617 632 2166; fax: +1 617 632 4773; e-mail: pwen@partners.org

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Purpose of review

To review the rationale and recent experience of angiogenesis inhibitors in malignant gliomas and to highlight both the promise and potential complications of these agents.

Recent findings

Several new agents targeting angiogenesis in malignant gliomas have become available and have been increasingly used to complement conventional chemotherapy. Specifically, bevacizumab, often in combination with irinotecan, has demonstrated favorable results in achieving significant radiographic responses and in prolonging progression-free survival in patients with recurrent malignant glioma.

Summary

Antiangiogenic drugs have been shown to have promising activity in recurrent malignant gliomas. Investigation of novel antiangiogenic compounds and future clinical trials will determine whether these drugs have a role in first-line therapy. This article reviews the rationale for targeting angiogenesis in malignant brain tumors and summarizes the results of recent clinical trials. In addition, this review will outline potential toxicities associated with angiogenesis inhibition in an attempt to provide practical guidance to physicians treating patients with malignant gliomas.

Keywords

adverse effects, angiogenesis, complications, glioblastoma, malignant glioma, treatment

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Introduction

Malignant gliomas are the most common type of malignant primary brain tumor and among the most challenging neoplasms to treat. Despite compelling advances in diagnostic imaging, surgery, radiation therapy, and the development of new antineoplastic agents, the survival rate for patients with malignant gliomas has improved only slightly in the last decade. With current standard therapy, median overall survival remains less than 15 months from time of diagnosis in patients with glioblastoma multiforme (GBM), the most frequent type of malignant gliomas [1••,2•,3]. Salvage therapies for recurrent tumors continue to be largely ineffective.

Among the reasons for this dismal prognosis are the highly invasive behavior of glioma cells into the brain parenchyma, preventing complete surgical resection, and impaired drug delivery across the blood–tumor barrier. Moreover, gliomas typically contain heterogenous cell populations, which differ with respect to phenotypic characteristics, proliferation, and resistance to current therapies. Recently, the findings that cancer stem cells in gliomas may play critical roles in tumor initiation and therapy resistance have added to the complexity of these devastating tumors [4•,5•].

As tumor growth is critically dependent on the formation of new blood vessels, inhibition of this process has offered an attractive strategy to complement standard therapies [6]. Endothelial proliferation is among the diagnostic hallmarks of glioblastomas, and angiogenesis plays a critical role in the progression and clinical behavior of theses tumors.

Although the process of angiogenesis and neovascularization is orchestrated by numerous factors and signaling cascades, vascular endothelial growth factor (VEGF) and its receptors have emerged as the most important mediators of neovascularization in gliomas. Encouraging results have come from initial studies using bevacizumab, a humanized monoclonal antibody targeting VEGF, in combination with conventional cytotoxic therapy. Additional studies targeting the VEGF-signaling pathway and other angiogenic factors in this patient population are also showing promise. Antiangiogenic therapies have been generally well tolerated, though some agents are associated with unique adverse effects. This review will summarize the current status of

Table 1 Selected angiogenesis inhibitors and their targets

Agent	Target
Anti-VEGF ligands	
Bevacizumab [Avastin (Genentech, South San Francisco, USA)]	VEGF-A
Aflibercept (VEGF-Trap)	VEGF-A/B, PLGF
Anti-HGF ligand	
AMG-102	HGF
Receptor tyrosine kinase inhibitors	
Cediranib (AZD2171)	VEGFR, PDGFR, c-Kit
Dasatinib [Sprycel (Bristol Myer Squibb, New York City, NewYork, USA)]	PDGFR, Src, Bcr-Abl
Pazopanib (GW786034)	VEGFR, PDGFR, c-kit
Sorafenib	VEGFR, PDGFR, c-Kit, Raf
Sunitinib [Sutent (Pfizer, Groton, Connecticut, USA]	VEGFR, PDGFR, c-Kit, FLT-3
Vandetanib (ZD6474)	VEGFR, EGFR, RET
Vatalanib (PTK787/ZK222584)	VEGFR, PDGFR, c-Kit
Tandutinib (MLN 518)	PDGFR, c-Kit, FLT-3
XL184	VEGFR, c-Met
Others	
Cilengitide (EMD 121974)	$\alpha v\beta 3$ and $\alpha v\beta 5$ integrins
Enzastaurin	PKC-β and Akt
Metronomic chemotherapy	Tumor endothelium
Thalidomide, Lenalidomide	Mutiple, FGF?

EGF, endothelial growth factor; FGF, fibroblast growth factor; FLT-3, fms-related tyrosine kinase 3; HGF, hepatocyte growth factor; PDGFR, platelet-derived growth factor receptor; PKC-b, protein kinase C beta; PLGF, placental growth factor; VEGFR, vascular endothelial growth factor receptor.

antiangiogenic therapies in malignant gliomas and discuss toxicities and emerging issues associated with these agents.

Angiogenesis and gliomas

Neovascularization is a complex process that involves tissue remodeling, destruction and growth, and results from activation of proangiogenic and inhibition of antiangiogenic factors. From a wide range of mediators identified, VEGF and its associated signaling cascade has been shown to be of paramount importance for the biology of malignant tumors, including glioblastomas [7,8]. Other key regulators in this process include fibroblast growth factor (FGF) [9], platelet-derived growth factor (PDGF) [10–12], hypoxia-inducible factor 1 alpha (HIF1-α [13,14]), hepatocyte growth factor/scatter factor (HGF/SF) [15], angiopoietins, interleukin (IL)-6 and IL-8 [16], angiostatin, endostatin, and thrombospondins [17,18]. Moreover, increased signaling through a number of growth factor receptors such as insulin-growth factor receptor (IGFR), stem cell factor receptor (c-Kit), and fibroblast growth factor receptor (FGFR) has also been shown to enhance VEGF activity [19–21].

Several VEGF family members and biologically active splice variants have been described so far, including VEGF-A to -D and placental growth factor. These act through receptor tyrosine kinases, among which at least three receptor subtypes have been identified (VEGFR-1, VEGFR-2, and VEGFR-3). Both endothelial cells and possibly glioma cells themselves may express and upregulate VEGF and its receptors, resulting in both paracrine and autocrine loops that drive endothelial cell proliferation, invasion, migration, and permeability [8,22]. The level of VEGF expression has been shown to correlate with the degree of malignancy and overall tumor prognosis [15,23].

Not surprisingly, there has been considerable interest in targeting VEGF signaling and other angiogenic pathways in gliomas (Table 1). This effort has been further stimulated by recent studies demonstrating that brain tumors may harbor small subpopulations of cancer stem cells that appear to be critically important in cancer initiation, progression, and resistance to treatment (for review, e.g. [4•,5•]). Interestingly, cancer stem cells appear to be directly involved in stimulating tumor angiogenesis through production of proangiogenic molecules such as VEGF [24]. Similar to neural stem cells that reside in neurovascular niches [25–29], cancer stem cells are thought to persist in close proximity to the tumor vasculature [30,31••]. Thus, inhibition of tumor angiogenesis may also potentially target the 'Achilles heel' of the tumor itself, the cancer stem cells, with the hope of achieving more durable clinical responses [32].

Targeting vascular endothelial growth factor in malignant gliomas

Due to its dominant role in tumor angiogenesis, targeting VEGF signaling has evolved into a promising therapeutic strategy. Bevacizumab, a humanized monoclonal antibody against VEGF-A, was among the first antiangiogenic drugs to be approved for the treatment of cancer when combined with cytotoxic agents [33,34].

In an initial evaluation of 29 patients with recurrent malignant glioma, the combination of bevacizumab (5 mg/kg every 2 weeks) with the topoisomerase I inhibitor

irinotecan (125 mg/m^2 weekly for 4 weeks, followed by 1–2 week breaks) resulted in a dramatic overall radiographic response rate of 66% [35]. This study demonstrated for the first time that bevacizumab was well tolerated in patients with malignant gliomas, with only one patient developing bowel perforation and one patient with an intracranial hemorrhage. A subsequent study [36] of 14 patients treated with bevacizumab and cytotoxic chemotherapy documented a radiographic response rate of 50%. A third retrospective study [37] of 44 patients with recurrent malignant glioma and treated with a comparable protocol reported an overall response rate of 34%. The treatment was well tolerated, though two patients were detected with asymptomatic intracranial hemorrhages on follow-up imaging. Of the 11 patients in this cohort, concurrently treated with anticoagulation and bevacizumab, only one developed a bleeding complication (mild epistaxis) [37]. The requirement for concurrent steroid treatment was significantly reduced in approximately 50% of patients.

In a phase II clinical trial, reported sequentially in two cohorts of patients [38••,39••], 35 recurrent glioblastoma and 33 recurrent grade III glioma patients were treated with bevacizumab and irinotecan. The initial cohort of 32 patients received bevacizumab (10 mg/kg) and irinotecan every 2 weeks of a 6-week cycle [38••]. The second cohort of 36 patients received bevacizumab (15 mg/kg) every 3 weeks and irinotecan on weeks 1,2, 4, and 5 of each 6-week cycle [39••]. Patients treated with cytochrome P-450 enzyme-inducing antiepileptic medications (EIAEDs), for example, phenytoin, received an irinotecan dose of 340 mg/m^2, whereas patients not on EIAEDs received a lower irinotecan dose of 125 mg/m^2. The overall radiographic response rate using modified Macdonald criteria [40] was 59% (65% in anaplastic gliomas; 53% in glioblastomas). In a recent survival update, 6-month progression-free survival (PFS6) was 43% for GBM patients and 59% for anaplastic gliomas patients; 2-year overall survival (OS) was 15% for GBM patients and 33% for anaplastic gliomas patients [41]. The regimen was overall well tolerated with acceptable toxicity. Adverse effects included thromboembolic complications (12%), intracranial hemorrhage (2%), fatigue (9%), proteinuria (6%), sepsis (2%), and nausea and emesis (6%) [38••,39••].

A subsequent randomized phase II trial compared a total of 167 patients with recurrent glioblastoma in first or second relapse treated with bevacizumab (10 mg/kg) every 2 weeks with or without irinotecan [42••]. Preliminary results reported a PFS6 of 43% and radiographic response rate of 28% in patients treated with bevacizumab alone, compared with a PFS6 of 50% and radiographic response rate of 38% in patients treated with the combination regimen [42••]. Median overall survival was 9.2 months in the group receiving bevacizumab alone and 8.7 months in the combination group. A steroid-sparing effect was noted in the majority of patients. Toxicity was relatively modest and only 2–3% of patients developed intracranial hemorrhage, most of which were asymptomatic.

Preliminary data suggests that the level of VEGF expression in gliomas may help to predict radiographic response to bevacizumab, but neither VEGF expression nor the expression of other angiogenic markers appears to predict survival [43]. There appears to be both VEGF-dependent and VEGF-independent pathways of edema production in gliomas; whether these pathways predict response to bevacizumab is unclear [44•].

There is increasing evidence of the fact that inhibition of angiogenesis may potentially enhance the effects of radiation therapy [45•]. Several trials combining bevacizumab with radiation therapy and temozolomide in newly diagnosed glioblastoma patients are underway. Preliminary data suggest that the regimen is safe, though wound healing may be impaired [46]. Many studies now defer treatment with bevacizumab until at least 4 weeks from surgery to avoid these problems with wound healing. Based on the encouraging results of bevacizumab in combination with irinotecan, there are now a large number of VEGF-inhibiting agents being evaluated in combination with cytotoxic agents or other targeted molecular therapies (Table 2).

Aflibercept (VEGF-Trap) is another VEGF-targeting agent that has been studied in patients with malignant glioma. Designed as a soluble decoy VEGF receptor that is fused to the constant region of IgG_1, aflibercept has several hundred times greater VEGF-binding affinity than bevacizumab and inhibits angiogenesis and tumor growth, and potentiates radiotherapy in preclinical glioma models [47,48]. Preliminary results from a single arm, phase II trial using aflibercept at a dose of 4 mg/kg every 2 weeks in patients with malignant glioma demonstrated a radiographic response rate of 50% in anaplastic gliomas and 30% in glioblastomas [49]. However, 25% of patients had to discontinue therapy because of toxicity, suggesting that the dose of 4 mg/kg every 2 weeks may be too high.

Targeting angiogenesis through receptor tyrosine kinase inhibition

In contrast to bevacizumab and aflibercept, which act through ligand sequestration, a large number of agents have been developed that act as competitive inhibitors of VEGF receptors and other receptor tyrosine kinases for various proangiogenic factors such as PDGF and stem cell factor (c-kit) (see Table 1). As reviewed in detail

Table 2 Selected ongoing clinical trials of vascular endothelial growth factor inhibitors

Agents	Phase	Diagnosis	Sponsor	Primary endpoint	Sites
Aflibercept	II	Recurrent MG	NCI	PFS6	NABTC
Aflibercept, TMZ + RT	I	New GBM; recurrent or stable MG	NCI	MTD	NABTC
Bev	II	Recurrent MG	NCI	PFS6	NCI
Bev and bortezomib	II	Recurrent GBM	Genentech, Millenium	PFS6	Duke
Bev and enzastaurin	II	Recurrent MG	NCI	PFS6	NCI
Bev and erlotinib	II	Recurrent MG	Genentech	PFS6	Duke
Bev and etoposide	II	Recurrent MG	Genentech	PFS6	Duke
Bev and LBH589	II	Recurrent MG	Novartis and Genentech	PFS6	DFCI and Northwestern
Bev and sorafenib	II	Recurrent GBM	NCI	PFS6	NCCTG
Bev and tandutinib	II	Recurrent MG	NCI	PFS6	NCI
Bev and metronomic TMZ	II	Recurrent GBM	Genentech, Schering-Plough	PFS	Duke
Bev and TMZ or etoposide	II	Recurrent GBM following bev and irinotecan	Genentech	PFS6	Duke
Bev and TMZ + RT	II	New GBM	Genentech	Survival	UCLA
Bev and TMZ post RT	II	New GBM	Genentech	PFS, RR	University of Chicago
Bev and TMZ + RT	III	New GBM	NCI	OS	RTOG
Bev and TMZ + RT	III	New GBM	Genentech	PFS, OS	Multiple sites (Europe)
Bev, TMZ, and erlotinib	II	Stable GBM following RT	NCI	OS, PFS	UCSF
Bev, TMZ + RT, followed by Bev, TMZ + irinotecan	II	New GBM	Genentech, Schering-Plough	OS	Duke

Bev, bevacizumab; DFCI, Dana-Farber Cancer Institute; GBM, glioblastoma multiforme; MDACC, M.D. Anderson Cancer Center; MG, malignant glioma; MTD, maximum tolerated dose; NABTC, North American Brain Tumor Coalition; NCI, National Cancer Institute; NCCTG, North Central Cancer Treatment Group; OS, overall survival; PFS, progression-free survival; RR, response rate; RT, radiation therapy; RTOG, Radiation Therapy Oncology Group; TMZ, temozolomide; UCLA, University of California – Los Angeles; UCSF, University of California – San Francisco.

elsewhere [50•], many of the newly developed receptor tyrosine kinase inhibitors that may influence angiogenesis and tumor growth through multiple targets are currently in various stages of preclinical development. Ongoing clinical trials of receptor tyrosine kinase inhibitors are summarized in Table 3.

Among other agents that are currently being evaluated in clinical trials are vatalanib, pazopanib, sorafenib, sunitinib, vandetanib, and XL-184. Only limited data are currently available regarding efficacy and toxicity in these agents. Encouraging results have come from a recent phase II clinical trial of cediranib (AZD2171), a potent pan-VEGF receptor inhibitor [51••]. In this study, 45 mg of cediranib was administered orally once daily to 31 patients with recurrent glioblastoma. The PFS6 was 26% and the overall radiographic response rate was approximately 56%. Similar to studies with bevacizumab, a significant steroid-sparing effect was observed. The regimen was associated with moderately severe toxicity,

Table 3 Selected ongoing clinical trials of vascular endothelial growth factor receptor inhibitors

Agents	Phase	Diagnosis	Sponsor	Primary endpoint	Sites
Cediranib and lomustine	I	Recurrent GBM	AZ	MTD	Multiple
Cediranib +/− lomustine	III	Recurrent GBM	AZ	PFS6, OS	Multiple
Cediranib, TMZ + RT	I/II	New GBM	NCI	MTD (phase I), PFS (phase II)	MGH, DFCI
Pazopanib	II	Recurrent GBM	NCI	PFS6	NABTC
Pazopanib and lapatinib	I/II	Recurrent MG	GSK	MTD (phase I), PFS6 (phase II)	Multiple
Sorafenib and bevacizumab	II	Recurrent GBM	NCI	PFS6	NCCTG
Sorafenib and erlotinib	II	Recurrent GBM	NCI	OS	NABTT
Sorafenib and erlotinib, tipifarnib, or temsirolimus	I/II	Recurrent GBM	NCI	MTD (phase I), PFS6 (phase II)	NABTC
Sorafenib and temsirolimus	I/II	Recurrent GBM	NCI	MTD (phase I), PFS6 (phase II)	NCCTG
Sorafenib and TMZ	II	Recurrent GBM	Bayer, S-P	PFS6	Duke
Sorafenib and TMZ	II	New GBM	Bayer	PFS	SCRI
Sunitinib	II	Recurrent MG	NCI	PFS6	Multiple
Sunitinib and irinotecan	I	Recurrent MG	Pfizer	MTD	Duke
Vandetanib	I/II	Recurrent glioma	NCI	MTD (phase I), PFS (phase II)	NCI
Vandetanib, imatinib, and hydroxyurea	I	Recurrent MG	Novartis, AZ	MTD	Duke
Vandetanib, TMZ + RT	I/II	New GBM	AZ	MTD (phase I), OS (phase II)	Multiple
XL184	II	Recurrent GBM	Exelixis	PFS6	DFCI, UCSF, MDACC

AZ, AstraZeneca; CNS, central nervous system; DFCI, Dana-Farber Cancer Institute; GBM, glioblastoma multiforme; GSK, GlaxoSmithKline; MDACC, M.D. Anderson Cancer Center; MGH, Massachusetts General Hospital; NABTC, North American Brain Tumor Consortium; NABTT, New Approaches to Brain Tumor Therapy; NCCTG, North Central Cancer Treatment Group; NCI, National Cancer Institute; PFS, progression-free survival; RT, radiation therapy; S-P, Schering-Plough; SCRI, Sarah Cannon Research Institute; TMZ, temozolomide; UCSF, University of California – San Francisco.

requiring temporary drug suspension in 69% of the initial 16 patients. Adverse effects included gastrointestinal toxicity, fatigue, and hypertension. In future studies, the single agent dose of cediranib will be reduced to 30 mg daily and should be better tolerated. Advanced MRI studies [51••] and a histopathological follow-up study [52] from a subset of these patients showed that decreased contrast enhancement was paralleled by reduction in blood vessel size, permeability, blood flow, and blood volume, supporting the concept of vascular normalization of abnormal tumor blood vessels [53•]. However, this effect appeared to be transient, and blood vessel size began to rebound by 8 weeks into treatment and after cessation of drug administration.

The phenomenon of vascular normalization appears to be one of the critical features of antiangiogenic therapies. This effect has also been observed with other antiangiogenic agents and may facilitate delivery of concurrently administered cytotoxic drugs and potentially improve the efficacy of radiation therapy [54]. The observation that vascular normalization is a transient phenomenon suggests that a specific therapeutic window exists during which chemotherapy and radiation may be most effective [51••,53•]. The mechanisms responsible for the re-establishment of pathological vascularization are poorly understood, but may be associated with upregulation of Tie-2 and alternate angiogenic factors such as basic fibroblast growth factor [51••].

Other antiangiogenic approaches

In addition to VEGF-related pathways, many other signaling pathways are involved in glioma angiogenesis. Platelet-derived growth factor receptors (PDGFRs) are present on pericytes and play a potentially important role [54,55]. Although initial trials of imatinib mesylate, which inhibits PDGFR and c-Kit, showed only minimal activity [56], newer PDGFR inhibitors such as tandutinib might potentially be more effective due to improved blood–brain barrier penetration.

Enzastaurin is an oral inhibitor of protein kinase C-β (PKCβ). Despite promising results in a phase II trial in recurrent malignant glioma [57], a randomized phase III trial comparing lomustine (CCNU) with enzastaurin in recurrent glioblastoma was stopped prematurely due to disappointing results on interim analysis [58]. It remains to be shown if the combination of enzastaurin with radiation therapy or other antiangiogenic therapies will result in a better outcome.

Celecoxib, a cyclooxygenase-2 (COX-2) inhibitor, shown to inhibit tumor angiogenesis and growth *in vitro*, has been evaluated in several studies [59,60] in recurrent malignant glioma with unclear benefit.

Cilengitide, an inhibitor of αvβ3 and αvβ5 integrins, has shown only modest single agent activity in recurrent glioblastoma, with a 9% response rate and PFS6 of 10–15% [61,62]. However, cilengitide may have greater activity combined with radiation therapy and temozolomide in patients with newly diagnosed glioblastomas with hypermethylation of the DNA repair enzyme O^6-methylguanine–DNA methyltransferase (MGMT) [63]. Overall survival at 15 months was 47% for unmethylated patients and 75% for the hypermethylated cohort. In light of these results, the European Organisation for Research and Treatment of Cancer (EORTC) is planning a phase III trial comparing cilengitide with standard therapy in newly diagnosed glioblastoma patients with MGMT promoter hypermethylation.

The antiangiogenic mechanism of thalidomide (Celgene, Summit, New Jersey, USA) and its analog lenalidomide (Celgene) are not fully understood, but likely involve FGF receptor blockade [64]. However, both thalidomide [65–67] and lenalidomide [68] have minimal efficacy in malignant gliomas.

Another antiangiogenic strategy involves the use of prolonged low-dose administration of chemotherapy (metronomic chemotherapy) to inhibit endothelial growth [69]. Although this approach may limit toxicity, available clinical data has been controversial in regard to efficacy in gliomas [70,71]. Nonetheless, metronomic chemotherapy remains an attractive strategy that may be more effective in patients with low disease burden, or when combined with potent angiogenesis inhibitors.

Toxicity of antiangiogenic treatment

The encouraging preliminary results with antiangiogenic agents have led to their widespread use. Although these agents are generally well tolerated, the emerging data suggest unique patterns of adverse effects that require careful patient selection [72•,73•].

There is an increased risk of thromboembolism in a patient population already at a significant risk of developing deep venous thrombosis, pulmonary embolism, and stroke [74,75]. Despite concerns of hemorrhage with the use of anticoagulants, preliminary data suggests that low molecular weight heparin is reasonably well tolerated in patients receiving bevacizumab [37,76]. In a small series of 21 patients with malignant gliomas with thromboembolism who received low molecular weight heparin and bevacizumab, there were no large hemorrhages; three patients had small intraparenchymal hemorrhages [76]. Hypertension is a common and dose-limiting toxicity of many anti-VEGF inhibitors, consistent with the physiological role of VEGF in regulating vasomotor tone and blood pressure [73•,77]. Thus, patients on antiangiogenesis therapy need

to be carefully monitored, and if necessary, rigorously treated for hypertension.

Due to the physiological role of VEGF in new blood vessel formation, most anti-VEGF/VEGFR agents are associated with an increased bleeding risk. Although the episodes of bleeding are usually relatively minor, life-threatening intracranial hemorrhages may occur in a small percentage of patients ($\leq$3%) [35,37,38••,39••,42••]. Because of concerns regarding bleeding, prior intratumoral hemorrhage has generally been considered to be a relative contraindication to antiangiogenic therapy. Other common systemic side effects of VEGF inhibitors are fatigue, proteinuria, epistaxis, impaired wound healing and rarely skin toxicity, and gastrointestinal perforation [73•]. Rare side effects involving the nervous system include reversible posterior leukoencephalopathy, seizures, disequilibrium, and ataxia [78–80]. The possible complications following VEGF-antiangiogenic and non-VEGF-antiangiogenic therapies are as follows:

(1) Hypertension
(2) Thromboembolic events (e.g. pulmonary embolism, deep venous thrombosis and stroke)
(3) Bleeding complications (e.g. intracranial or pulmonary hemorrhage)
(4) Gastrointestinal complications (including nausea, emesis, mucositis and diarrhea)
(5) Impaired wound healing
(6) Bowel perforation
(7) Fatigue
(8) Rash (including hand–foot syndrome)
(9) Headache
(10) Proteinuria
(11) Transaminitis
(12) Myelosuppression (e.g. anemia, neutropenia, thrombocytopenia)
(13) Reversible posterior leukoencephalopathy
(14) Rebound vasogenic edema.

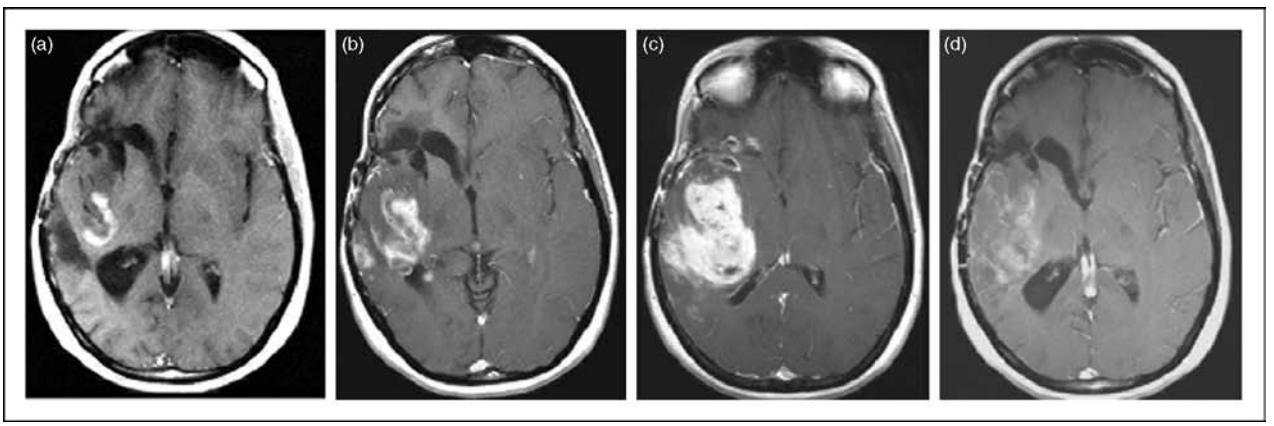
Other issues related to antiangiogenic therapy

Beside their unique toxicity patterns, there are other emerging issues that need to be considered in patients treated with bevacizumab or other antiangiogenic agents.

Although current response criteria have been based on the degree of contrast enhancement on MRI [40], this parameter may be misleading following the use of VEGF pathway inhibitors. The reduction in enhancement may simply reflect a decrease in vascular permeability of MRI contrast rather than real effect on the tumor, complicating the interpretation of radiologic responses with these agents. It will be important to incorporate advanced imaging modalities, such as dynamic contrast-enhanced MRI, perfusion imaging, magnetic resonance spectroscopy, or PET studies in future trials to assess antitumor effects.

Although angiogenesis inhibitors are frequently associated with a significant reduction in tumor size and desirable steroid sparing effect [42••,81], this effect may only be temporary due to upregulation of alternate and VEGF-independent mediators of angiogenesis (e.g. PDGF/PDGF-R, FGF, Tie-2) [51••,82•]. Furthermore, abrupt cessation of anti-VEGF treatment may result in rebound edema and clinical deterioration (Fig. 1). If discontinuation of anti-VEGF therapy is being considered, patients should be monitored closely and

Figure 1 Axial T1 contrast enhanced MRIs of a 41-year-old woman with a recurrent right temporal glioblastoma demonstrating increased enhancement and edema following discontinuation of anti-vascular endothelial growth factor (VEGF) therapy



(a) Baseline MRI following response to aflibercept (VEGF-Trap). (b) Progression after 4 months on therapy. Aflibercept was discontinued. (c) 4 weeks later, MRI shows significantly more enhancement. The patient began to experience headache. (d) MRI 4 weeks after starting bevacizumab showing marked reduction in contrast enhancement and mass effect. The rapid improvement in clinical and radiologic features suggests that at least some of the changes are an effect of bevacizumab on vascular permeability rather than an antitumor effect.

perhaps treated prophylactically with corticosteroids. The radiographic and clinical deterioration of patients who stop anti-VEGF therapy also potentially complicates the evaluation of subsequent therapeutic agents.

Another concern has come from studies [14,83,84] showing that blockade of VEGF-mediated angiogenesis may ultimately promote tumor infiltration by co-option of existing cerebral blood vessels. These changes are often poorly detected by conventional contrast-enhanced MRI, but appear as enlarging areas of increased T2/FLAIR signal [37,85–87]. Perhaps in part because of increased infiltrative growth, tumors that progress during antiangiogenic therapy become extremely challenging to treat, and rapid clinical deterioration and death are often the consequence [88]. Despite the overall encouraging results from current antiangiogenic trials in glioma patients, such therapies are ultimately ineffective and will need to be combined with other strategies, such as inhibitors of tumor cell invasion and migration.

One issue of concern has come from recent studies on the cell-biological analysis of cancer therapy associated neurotoxicity. Conventional cytotoxic agents have been shown to preferentially target neural progenitor cells critically important in maintenance of normal brain function and white matter integrity [5•,89,90]. Moreover, the physiological function of normal neural stem cells and progenitor cells is dependent on a number of factors, such as VEGF, FGF, EGF, and PDGF. By targeting those signaling pathways, long-term adverse effects, such as cognitive dysfunction, potentially may be encountered in survivors.

Summary

Antiangiogenic therapies, especially in combination with conventional cytotoxic drugs, have shown encouraging results in patients with malignant gliomas. In addition, anti-VEGF agents have a potent antiedema effect, commonly allowing steroid doses to be significantly reduced. In general, VEGF-pathway inhibitors are well tolerated. Among the most important toxicities are hypertension, thromboembolic complications, bleeding, and impaired wound healing. As experience with these agents increase, additional safety concerns may arise. Vulnerability of stem cell compartments comes with both the potential to target cancer stem cells and concerns of increased neurotoxicity. Despite encouraging preliminary results, current antiangiogenic therapies eventually result in tumor resistance and progression. Approaches combining antiangiogenic agents with cytotoxic therapies, agents that inhibit putative pathways of resistance such as FGFR, or agents that target tumor invasion may lead to improved outcomes.

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References and recommended reading

Papers of particular interest, published within the annual period of review, have been highlighted as:

- • of special interest
- •• of outstanding interest

Additional references related to this topic can also be found in the Current World Literature section in this issue (p. 770).

1 Furnari FB, Fenton T, Bachoo RM, *et al.* Malignant astrocytic glioma: genetics, •• biology, and paths to treatment. Genes Dev 2007; 21:2683–2710.
Comprehensive overview of the molecular genetics of malignant gliomas.

2 Wen PY, Kesari S. Malignant gliomas in adults. N Engl J Med 2008; • 359:492–507.
A recent overview of malignant gliomas.

3 Stupp R, Mason WP, van den Bent MJ, *et al.* Radiotherapy plus concomitant and adjuvant temozolomide for glioblastoma. N Engl J Med 2005; 352:987–996.

4 Dirks PB. Brain tumor stem cells: bringing order to the chaos of brain cancer. J • Clin Oncol 2008; 26:2916–2924.
A recent overview of brain tumor stem cells.

5 Dietrich J, Imitola J, Kesari S. Mechanisms of disease: the role of stem cells in • the biology and treatment of gliomas. Nat Clin Pract Oncol 2008; 5:393–404.
A recent review of the role of stem cells in gliomas.

6 Folkman J. Tumor angiogenesis: therapeutic implications. N Engl J Med 1971; 285:1182–1186.

7 Plate KH, Breier G, Weich HA, Risau W. Vascular endothelial growth factor is a potential tumour angiogenesis factor in human gliomas in vivo. Nature 1992; 359:845–848.

8 Ferrara N, Gerber HP, LeCouter J. The biology of VEGF and its receptors. Nat Med 2003; 9:669–676.

9 Stefanik DF, Rizkalla LR, Soi A, *et al.* Acidic and basic fibroblast growth factors are present in glioblastoma multiforme. Cancer Res 1991; 51:5760–5765.

10 Plate KH, Breier G, Farrell CL, Risau W. Platelet-derived growth factor receptor-beta is induced during tumor development and upregulated during tumor progression in endothelial cells in human gliomas. Lab Invest 1992; 67:529–534.

11 Guo P, Hu B, Gu W, *et al.* Platelet-derived growth factor-B enhances glioma angiogenesis by stimulating vascular endothelial growth factor expression in tumor endothelia and by promoting pericyte recruitment. Am J Pathol 2003; 162:1083–1093.

12 Dunn IF, Heese O, Black PM. Growth factors in glioma angiogenesis: FGFs, PDGF, EGF, and TGFs. J Neurooncol 2000; 50:121–137.

13 Zagzag D, Zhong H, Scalzitti JM, *et al.* Expression of hypoxia-inducible factor 1alpha in brain tumors: association with angiogenesis, invasion, and progression. Cancer 2000; 88:2606–2618.

14 Du R, Lu KV, Petritsch C, *et al.* HIF1alpha induces the recruitment of bone marrow-derived vascular modulatory cells to regulate tumor angiogenesis and invasion. Cancer Cell 2008; 13:206–220.

15 Schmidt NO, Westphal M, Hagel C, *et al.* Levels of vascular endothelial growth factor, hepatocyte growth factor/scatter factor and basic fibroblast growth factor in human gliomas and their relation to angiogenesis. Int J Cancer 1999; 84:10–18.

16 Brat DJ, Bellail AC, Van Meir EG. The role of interleukin-8 and its receptors in gliomagenesis and tumoral angiogenesis. Neuro Oncol 2005; 7:122–133.

17 Hanahan D, Folkman J. Patterns and emerging mechanisms of the angiogenic switch during tumorigenesis. Cell 1996; 86:353–364.

18 Tuettenberg J, Friedel C, Vajkoczy P. Angiogenesis in malignant glioma–a target for antitumor therapy? Crit Rev Oncol Hematol 2006; 59:181–193.

19 Trojan J, Cloix JF, Ardourel MY, *et al.* Insulin-like growth factor type I biology and targeting in malignant gliomas. Neuroscience 2007; 145:795–811.

20 Sun L, Hui AM, Su Q, *et al.* Neuronal and glioma-derived stem cell factor induces angiogenesis within the brain. Cancer Cell 2006; 9:287–300.

21 Lamszus K, Heese O, Westphal M. Angiogenesis-related growth factors in brain tumors. Cancer Treat Res 2004; 117:169–190.

22 Millauer B, Shawver LK, Plate KH, *et al.* Glioblastoma growth inhibited in vivo by a dominant-negative Flk-1 mutant. Nature 1994; 367:576–579.

23 Leon SP, Folkerth RD, Black PM. Microvessel density is a prognostic indicator for patients with astroglial brain tumors. Cancer 1996; 77:362–372.

24 Bao S, Wu Q, Sathornsumetee S, *et al.* Stem cell-like glioma cells promote tumor angiogenesis through vascular endothelial growth factor. Cancer Res 2006; 66:7843–7848.

25 Palmer TD, Willhoite AR, Gage FH. Vascular niche for adult hippocampal neurogenesis. J Comp Neurol 2000; 425:479–494.

26 Shen Q, Goderie SK, Jin L, *et al.* Endothelial cells stimulate self-renewal and expand neurogenesis of neural stem cells. Science 2004; 304:1338–1340.

27 Alvarez-Buylla A, Lim DA. For the long run: maintaining germinal niches in the adult brain. Neuron 2004; 41:683–686.

28 Sanai N, Tramontin AD, Quinones-Hinojosa A, *et al.* Unique astrocyte ribbon in adult human brain contains neural stem cells but lacks chain migration. Nature 2004; 427:740–744.

29 Quinones-Hinojosa A, Sanai N, Soriano-Navarro M, *et al.* Cellular composition and cytoarchitecture of the adult human subventricular zone: a niche of neural stem cells. J Comp Neurol 2006; 494:415–434.

30 Gilbertson RJ, Rich JN. Making a tumour's bed: glioblastoma stem cells and the vascular niche. Nat Rev Cancer 2007; 7:733–736.

31 •• Calabrese C, Poppleton H, Kocak M, *et al.* A perivascular niche for brain tumor stem cells. Cancer Cell 2007; 11:69–82.
Important study showing that brain tumor stem cells have a perivascular niche.

32 Folkins C, Man S, Xu P, *et al.* Anticancer therapies combining antiangiogenic and tumor cell cytotoxic effects reduce the tumor stem-like cell fraction in glioma xenograft tumors. Cancer Res 2007; 67:3560–3564.

33 Hurwitz H, Fehrenbacher L, Novotny W, *et al.* Bevacizumab plus irinotecan, fluorouracil, and leucovorin for metastatic colorectal cancer. N Engl J Med 2004; 350:2335–2342.

34 Jain RK, Duda DG, Clark JW, Loeffler JS. Lessons from phase III clinical trials on anti-VEGF therapy for cancer. Nat Clin Pract Oncol 2006; 3:24–40.

35 Stark-Vance V. Bevacizumab and CPT-11 in the treatment of relapsed malignant glioma. In: Sixth Meeting of the European Association for Neuro-Oncology: Neuro-Oncology, Edinburgh, Scotland: Duke University Press; 2005. p. 369.

36 Pope WB, Lai A, Nghiemphu P, *et al.* MRI in patients with high-grade gliomas treated with bevacizumab and chemotherapy. Neurology 2006; 66:1258–1260.

37 Norden AD, Young GS, Setayesh K, *et al.* Bevacizumab for recurrent malignant gliomas: efficacy, toxicity, and patterns of recurrence. Neurology 2008; 70:779–787.

38 •• Vredenburgh JJ, Desjardins A, Herndon JE 2nd, *et al.* Phase II trial of bevacizumab and irinotecan in recurrent malignant glioma. Clin Cancer Res 2007; 13:1253–1259.
First phase II study of bevacizumab and irinotecan in recurrent malignant gliomas.

39 •• Vredenburgh JJ, Desjardins A, Herndon JE 2nd, *et al.* Bevacizumab plus irinotecan in recurrent glioblastoma multiforme. J Clin Oncol 2007; 25:4722–4729.
Phase II study of bevacizumab and irinotecan in recurrent glioblastomas.

40 Macdonald DR, Cascino TL, Schold SC Jr, Cairncross JG. Response criteria for phase II studies of supratentorial malignant glioma. J Clin Oncol 1990; 8:1277–1280.

41 Wagner SA, Desjardins A, Reardon DA, *et al.* Update on survival from the original phase II trial of bevacizumab and irinotecan in recurrent malignant gliomas [abstract]. J Clin Oncol 2008; 26:2021.

42 •• Cloughesy TF, Prados MD, Wen PY, *et al.* A phase II, randozimed, noncomparative clinical trial of the effect of bevacizumab (BV) alone or in combination with irinotecan (CPT) on 6-month progresssion free survival (PFS6) in recurrent, treatment-refractory glioblastoma (GBM) [abstract]. J Clin Oncol 2008; 26:2010b.
Large randomized phase II study showing that bevacizumab increases radiographic response and PFS6 in recurrent malignant gliomas.

43 Sathornsumetee S, Cao Y, Marcello JE, *et al.* Tumor angiogenic and hypoxic profiles predict radiographic response and survival in malignant astrocytoma patients treated with bevacizumab and irinotecan. J Clin Oncol 2008; 26:271–278.

44 • Carlson MR, Pope WB, Horvath S, *et al.* Relationship between survival and edema in malignant gliomas: role of vascular endothelial growth factor and neuronal pentraxin 2. Clin Cancer Res 2007; 13:2592–2598.
Interesting study showing that edema may be regulated by VEGF-related and neuronal pentraxin 2 related pathways.

45 • Duda DG, Jain RK, Willett CG. Antiangiogenics: the potential role of integrating this novel treatment modality with chemoradiation for solid cancers. J Clin Oncol 2007; 25:4033–4042.
Good review discussing combination of anatiangiogenic therapy with chemoradiation.

46 Lai A, Filka E, McGibbon B, *et al.* Phase II pilot study of bevacizumab in combination with temozolomide and regional radiation therapy for up-front treatment of patients with newly diagnosed glioblastoma multiforme: interim analysis of safety and tolerability. Int J Radiat Oncol Biol Phys 2008; 71:1372–1380.

47 Holash J, Davis S, Papadopoulos N, *et al.* VEGF-Trap: a VEGF blocker with potent antitumor effects. Proc Natl Acad Sci U S A 2002; 99:11393–11398.

48 Wachsberger PR, Burd R, Cardi C, *et al.* VEGF trap in combination with radiotherapy improves tumor control in u87 glioblastoma. Int J Radiat Oncol Biol Phys 2007; 67:1526–1537.

49 De Groot JF, Wen PY, Lamborn K, *et al.* Phase II single arm trial of aflibercept in patients with recurrent temozolamide-resistant glioblastoma: NABTC 0601 [abstract]. J Clin Oncol 2008; 26:2020.

50 • Chi A, Norden AD, Wen PY. Inhibition of angiogenesis and invasion in malignant gliomas. Expert Rev Anticancer Ther 2007; 7:1537–1560.
Recent review discussing angiogenesis and invasion in malignant gliomas.

51 •• Batchelor TT, Sorensen AG, di Tomaso E, *et al.* AZD2171, a pan-VEGF receptor tyrosine kinase inhibitor, normalizes tumor vasculature and alleviates edema in glioblastoma patients. Cancer Cell 2007; 11:83–95.
Phase II study of pan-VEGFR inhibitor AZD2171 summarizing preliminary results. This study provides radiologic evidence supporting vessel normalization and suggests that at tumor recurrence angiogenic factors such as basic FGF may be upregulated.

52 Di Tomaso E, Frosch MP, Auluck PK, *et al.* Characterization of blood vessels in brain autopsies of GBM patients who received antiangiogenic treatment [abstract]. J Clin Oncol 2008; 26:2009.

53 • Jain RK, di Tomaso E, Duda DG, *et al.* Angiogenesis in brain tumours. Nat Rev Neurosci 2007; 8:610–622.
Good review of angiogenesis in brain tumors.

54 Zhou Q, Guo P, Gallo JM. Impact of angiogenesis inhibition by sunitinib on tumor distribution of temozolomide. Clin Cancer Res 2008; 14:1540–1549.

55 Song S, Ewald AJ, Stallcup W, *et al.* PDGFRbeta+ perivascular progenitor cells in tumours regulate pericyte differentiation and vascular survival. Nat Cell Biol 2005; 7:870–879.

56 Wen PY, Yung WK, Lamborn KR, *et al.* Phase I/II study of imatinib mesylate for recurrent malignant gliomas: North American Brain Tumor Consortium Study 99-08. Clin Cancer Res 2006; 12:4899–4907.

57 Fine HA, Kim L, Royce C, *et al.* Results from a phase II trial of enzastaurin (LY317615) in patients with recurrent high grade gliomas [abstract]. J Clin Oncol 2005; 23:1505.

58 Fine HA, Puduvalli VK, Chamberlain MC, *et al.* Enzastaurin (ENZ) versus lomustine (CCNU) in the treatment of recurrent, intracranial glioblastoma multiforme (GBM): a phase III study [abstract]. J Clin Oncol 2008; 26:2005.

59 Levin VA, Giglio P, Puduvalli VK, *et al.* Combination chemotherapy with 13-cis-retinoic acid and celecoxib in the treatment of glioblastoma multiforme. J Neurooncol 2006; 78:85–90.

60 Pannullo SC, Burton J, Serventi J, *et al.* Phase I/II trial of twice-daily temozolomide and celecoxib for treatment of relapsed malignant glioma: final data [abstract]. J Clin Oncol 2006; 24:1519.

61 Nabors LB, Mikklesen T, Rosenfeld SS, *et al.* Phase I and correlative biology study of cilengitide in patients with recurrent malignant glioma. J Clin Oncol 2007; 25:1651–1657.

62 Reardon DA, Fink K, Nabors B, *et al.* Phase IIa trial of cilengitide (EMD 121974) single-agent therapy in patients with recurrent glioblastoma: EMD 121974-009 [abstract]. J Clin Oncol 2007; 25:2002.

63 Stupp R, Goldbrunner R, Neyns B, *et al.* Phase I/IIa trial of cilengitide (EMD 121974) and temozolonide with concomitant radiotherapy, followed by temozolomide and cilenitide maintenance therapy in patients with newly diagnosed glioblastoma (GBM) [abstract]. J Clin Oncol 2007; 25:2000.

64 D'Amato RJ, Loughnan MS, Flynn E, Folkman J. Thalidomide is an inhibitor of angiogenesis. Proc Natl Acad Sci U S A 1994; 91:4082–4085.

65 Fine HA, Wen PY, Maher EA, *et al.* Phase II trial of thalidomide and carmustine for patients with recurrent high-grade gliomas. J Clin Oncol 2003; 21:2299–2304.

66 Fine HA, Figg WD, Jaeckle K, *et al.* Phase II trial of the antiangiogenic agent thalidomide in patients with recurrent high-grade gliomas. J Clin Oncol 2000; 18:708–715.

67 Groves MD, Puduvalli VK, Chang SM, *et al.* A North American brain tumor consortium (NABTC 99-04) phase II trial of temozolomide plus thalidomide for recurrent glioblastoma multiforme. J Neurooncol 2007; 81:271–277.

68 Fine HA, Kim L, Albert PS, *et al.* A phase I trial of lenalidomide in patients with recurrent primary central nervous system tumors. Clin Cancer Res 2007; 13:7101–7106.

69 Browder T, Butterfield CE, Kraling, *et al.* Antiangiogenic scheduling of chemotherapy improves efficacy against experimental drug-resistant cancer. Cancer Res 2000; 60:1878–1886.

70 Kesari S, Schiff D, Doherty L, *et al.* Phase II study of metronomic chemotherapy for recurrent malignant gliomas in adults. Neuro Oncol 2007; 9:354–363.

71 Kieran MW, Turner CD, Rubin JB, *et al.* A feasibility trial of antiangiogenic (metronomic) chemotherapy in pediatric patients with recurrent or progressive cancer. J Pediatr Hematol Oncol 2005; 27:573–581.

72 • Verheul HM, Pinedo HM. Possible molecular mechanisms involved in the toxicity of angiogenesis inhibition. Nat Rev Cancer 2007; 7:475–485.
Recent review of toxicities of antiangiogenic therapy.

73 • Eskens FA, Verweij J. The clinical toxicity profile of vascular endothelial growth factor (VEGF) and vascular endothelial growth factor receptor (VEGFR) targeting angiogenesis inhibitors; a review. Eur J Cancer 2006; 42:3127–3139.
Recent review of toxicities of anti-VEGF therapy.

74 Marras LC, Geerts WH, Perry JR. The risk of venous thromboembolism is increased throughout the course of malignant glioma: an evidence-based review. Cancer 2000; 89:640–646.

75 Semrad TJ, O'Donnell R, Wun T, *et al.* Epidemiology of venous thromboembolism in 9489 patients with malignant glioma. J Neurosurg 2007; 106:601–608.

76 Nghiemphu PL, Green RM, Pope WB, *et al.* Safety of anticoagulation use and bevacizumab in patients with glioma. Neuro Oncol 2008; 10:355–360.

77 van Heeckeren WJ, Ortiz J, Cooney MM, Remick SC. Hypertension, proteinuria, and antagonism of vascular endothelial growth factor signaling: clinical toxicity, therapeutic target, or novel biomarker? J Clin Oncol 2007; 25:2993–2995.

78 Allen JA, Adlakha A, Bergethon PR. Reversible posterior leukoencephalopathy syndrome after bevacizumab/FOLFIRI regimen for metastatic colon cancer. Arch Neurol 2006; 63:1475–1478.

79 El Maalouf G, Mitry E, Lacout A, *et al.* Isolated brainstem involvement in posterior reversible leukoencephalopathy induced by bevacizumab. J Neurol 2008; 255:295–296.

80 Glusker P, Recht L, Lane B. Reversible posterior leukoencephalopathy syndrome and bevacizumab. N Engl J Med 2006; 354:980–982.

81 Gonzalez J, Kumar AJ, Conrad CA, Levin VA. Effect of bevacizumab on radiation necrosis of the brain. Int J Radiat Oncol Biol Phys 2007; 67:323–326.

82 • Bergers G, Hanahan D. Modes of resistance to anti-angiogenic therapy. Nat Rev Cancer 2008; 8:592–603.
An excellent review of mechanisms of resistance to antiangiogenic therapies.

83 Holash J, Maisonpierre PC, Compton D, *et al.* Vessel cooption, regression, and growth in tumors mediated by angiopoietins and VEGF. Science 1999; 284:1994–1998.

84 Rubenstein JL, Kim J, Ozawa T, *et al.* Anti-VEGF antibody treatment of glioblastoma prolongs survival but results in increased vascular cooption. Neoplasia 2000; 2:306–314.

85 Lassman AB, Iwamoto FM, Gutin PH, Abrey LE. Patterns of relapse and prognosis after bevacizumab (BEV) failure in recurrent glioblastoma (GBM) [abstract]. J Clin Oncol 2008; 26:2028.

86 Zuniga RM, Torcuator R, Doyle T, *et al.* Retrospective analysis of patterns of recurrence seen on MRI in patients with recurrent glioblastoma multiforme treated with bevacizumab plus irinotecan [abstract]. J Clin Oncol 2008; 26:13013.

87 Narayana A, Raza S, Golfinos JG, *et al.* Bevacizumab therapy in recurrent high grade glioma: impact on local control and survival [abstract]. J Clin Oncol 2008; 26:13000.

88 Quant E, Norden AE, Drappatz J, *et al.* Role of a second chemotherapy in recurrent malignant glioma patients who progress on a bevacizumab-containing regimen [abstract]. J Clin Oncol 2008; 26:2008.

89 Dietrich J, Han R, Yang Y, *et al.* CNS progenitor cells and oligodendrocytes are targets of chemotherapeutic agents in vitro and in vivo. J Biol 2006; 5:22.

90 Han R, Yang YM, Dietrich J, *et al.* Systemic 5-fluorouracil treatment causes a syndrome of delayed myelin destruction in the central nervous system. J Biol 2008; 7:12.