Advances in Treatment Options for High-grade Glioma—Current Status and Future Perspectives

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Abstract

High-grade gliomas, including glioblastoma, anaplastic astrocytoma, anaplatic oligodendroglioma, and anaplastic oligoastrocytoma, account for the majority of malignant primary brain tumors diagnosed in adults. The prognosis for these tumors is poor despite multimodality therapy with surgery, radiation, and/or chemotherapy. This article summarizes treatment options for high-grade glioma, including standard regimens, targeted agents, and novel therapies.

Keywords

High-grade glioma, radiation therapy, chemotherapy, targeted molecular therapies

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High-grade glioma (HGG) is the most common type of primary brain tumor in adults and accounts for >75% of the estimated 22,070 newly diagnosed malignant primary brain tumors in the US each year.[1] More than half of HGGs are glioblastoma (GBM), the most aggressive subtype. The remainder include anaplastic gliomas (AGs),[1,2] such as anaplastic astrocytoma (AA), anaplastic oligodendroglioma (AO), and anaplastic oligoastrocytoma (AOA), and rarer subtypes. HGG is incurable and is responsible for a disproportionate share of cancer-related morbidity and mortality.[3] With optimal treatment, median survival is only 12–18 months for GBM and two to five years for AG. There have been recent advances in elucidating the molecular pathogenesis of HGG, which may provide additional prognostic information and lead to more effective treatments.[4–10] This article summarizes the standard treatment of adult HGG with a particular focus on recent therapeutic advances.

Standard Treatment Options for High-grade Glioma

Surgery for High-grade Glioma

Maximal surgical resection is recommended in all newly diagnosed HGG patients. Although a surgical cure is impossible, benefits of resection include improvement of symptoms related to mass effect, reduction of tumor volume,[11] and removal of the necrotic tumor core, which may be resistant to radiation therapy and poorly accessible to circulating chemotherapy. Mounting evidence suggests that a near gross total resection confers a modest survival benefit compared with biopsy or subtotal resection.[12–14] Surgery may be considered in recurrent HGG patients with good performance status when the tumor is accessible, symptomatic, and distant from eloquent areas. Surgical resection in the recurrent setting may improve quality of life and allow time for additional therapy, but the impact on overall survival is negligible.

Radiation Therapy for High-grade Glioma

Radiation therapy (RT) has the biggest impact on overall survival for HGG of all standard treatment modalities. The addition of RT to surgery for GBM increases median survival from three to four months to approximately 12 months.[15,16]

Many variations of standard RT have been investigated in an attempt to increase efficacy, including using doses >60Gy, altered fractionation schemes, brachytherapy, stereotactic radiosurgery (SRS), and the use of radiosensitizing agents. None of these has demonstrated additional benefit over standard fractionated RT.[17,18] Newer approaches including chemotherapy,[19] targeted molecular agents,[20] and anti-angiogenic agents[21] may potentially work synergistically with RT and improve outcomes.

Additional involved-field RT is rarely offered to patients with recurrent HGG, as doses >60Gy offer marginal benefit and an increased risk for radiation necrosis.[22] Small non-randomized studies have demonstrated a survival benefit for HGG patients treated with SRS at recurrence.[23] However, many of the data are subject to selection bias, and this

Table 1: Summary of Therapeutic Options for High-grade Glioma

Setting	Histology	Recommended Treatment Options
Newly Diagnosed Tumor*		
	Glioblastoma	• RT with concomitant and adjuvant TMZ • Clinical trial enrollment
	Anaplastic astrocytoma**	• RT with concomitant and adjuvant TMZ • RT with adjuvant TMZ • Clinical trial enrollment
	Anaplastic oligodendroglioma or oligoastrocytoma**	• RT alone • RT with concomitant and adjuvant TMZ • RT with adjuvant TMZ or PCV only • TMZ or PCV alone • Clinical trial enrollment
Recurrent Tumor**		
	Any	• Clinical trial enrollment*** • Surgical resection, re-irradiation or SRS for selected candidates • Carmustine wafers • Chemotherapy (TMZ, carmustine, lomustine, others) • Bevacizumab with or without chemotherapy (irinotecan, others)

**Treatment should always begin with maximal surgical resection when possible.*
***No standard of care has been defined. ***Clinical trial enrollment should be offered to recurrent malignant glioma patients whenever possible.*
PCV = procarbazine, lomustine (CCNU), and vincristine; RT = radiation therapy; SRS = stereotactic radiosurgery; TMZ = temozolomide.
Source: Wen and Kesari, 2008.[3]

approach is not routinely utilized. Fractionated stereotactic RT has also been evaluated for treatment of recurrent HGG, but its efficacy is also unproven.[24]

Chemotherapy for Glioblastoma

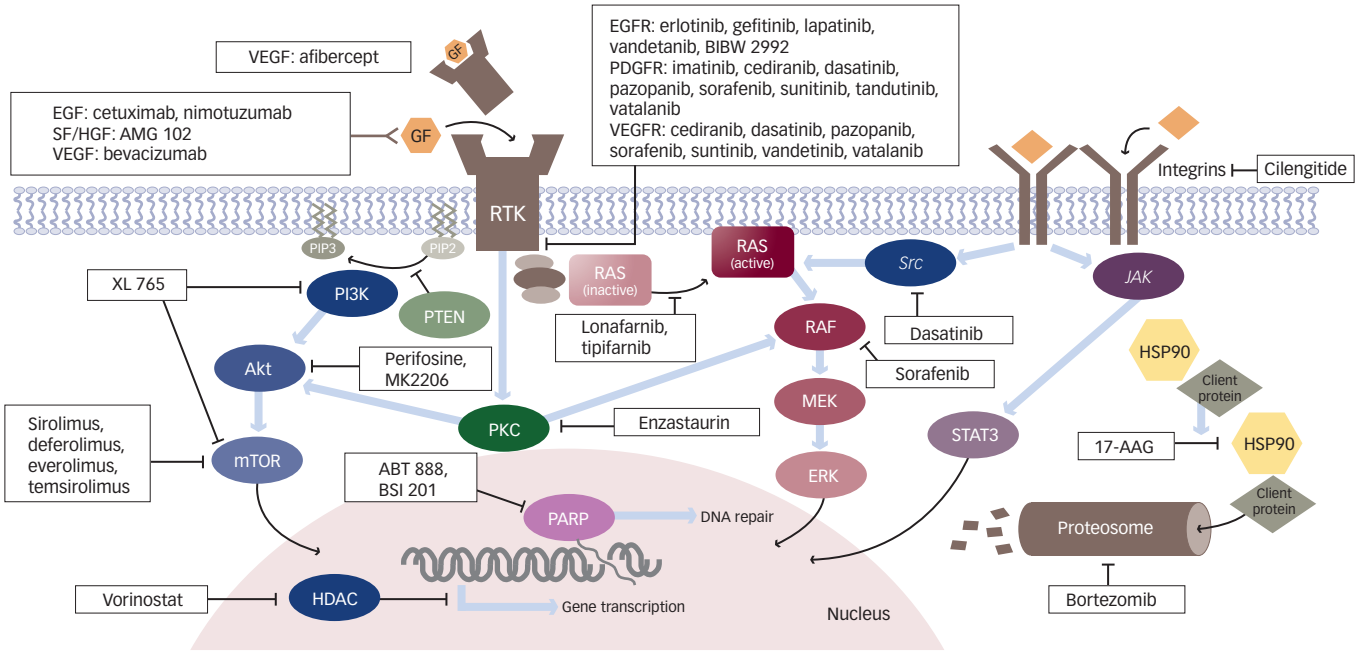
Temozolomide has replaced nitrosureas as the standard of care for treatment of newly diagnosed GBM, based on the results of a phase III study conducted by the European Organization for Research and Treatment of Cancer (EORTC) and the National Cancer Institute of Canada (NCIC) in newly diagnosed GBM comparing RT alone (60Gy over six weeks) with RT and concomitant daily temozolomide (75mg/m^2/day), followed by adjuvant temozolomide therapy (150–200mg/m^2/day for five consecutive days every 28-day cycle, for six cycles).[16] The addition of temozolomide to RT increased median survival compared with RT alone (14.6 versus 12.1 months; p<0.0001). Recently, updated results from this study showed that the added survival benefit with temozolomide was maintained, even at five years.[25]

An established mechanism of temozolomide resistance is based on DNA repair through O-6-methylguanine-DNA methyltransferase (*MGMT*), an endogenous DNA-repair enzyme that removes alkyl groups from DNA and thus confers resistance to temozolomide and other alkylating agents. *MGMT* promoter methylation has been shown to predict temozolomide sensitivity in GBM.[6,26] In a companion study to the EORTC/NCIC, tumor specimens were evaluated for methylation status of the *MGMT* gene promoter.[6] As predicted, the benefit of temozolomide was significantly increased in patients with *MGMT* promoter methylation. Among GBM patients with *MGMT* promoter methylation who were treated with temozolomide, median survival was 21.7 months and two-year survival 46%. Temozolomide-treated patients with unmethylated *MGMT* promoters had a significantly shorter median survival of only 12.7 months and a two-year survival of 13.8%.[6] Because this study was conducted retrospectively in a relatively small sample of patients, temozolomide remains the standard of care for newly diagnosed GBM patients, regardless of *MGMT* promoter methylation status. A randomized phase III trial sponsored by the Radiation Therapy Oncology Group (RTOG 0525) will definitively evaluate the utility of *MGMT* promoter methylation in determining temozolomide sensitivity. In the future, patients whose tumors have unmethylated *MGMT* promoters may be offered alternatives to the standard temozolomide regimen. Investigational approaches to overcome *MGMT* activity include dose-intense temozolomide regimens[27,28] or continuous dosing,[29] which may deplete the enzyme,[30] and combination therapy with O6-benzylguanine or other *MGMT* inhibitors.[31–33]

An alternative to systemic chemotherapy involves the surgical implantation of carmustine-containing biodegradable wafers (Gliadel) into the resection cavity following tumor debulking. A double-blind, randomized phase III trial demonstrated a modest benefit in patients with newly diagnosed GBM. Those patients who received radiation and placebo had a median survival of only 11.6 months compared with 13.9 months for patients who received radiation and carmustine wafers, with median overall survival of 11.6 and 13.9 months, respectively (p=0.03),[34] resulting in approval of this therapy by the US Food and Drug Administration (FDA). The benefits of traditional cytotoxic chemotherapy have been modest in the treatment of recurrent GBM. Phase II trials of temozolomide for recurrent GBM demonstrated radiographic response rates (RR) of only 5% and six-month progression-free survival (PFS6) of about 21%.[35,36] However, the recently published RESCUE study showed that continuous dosing of temozolomide at 50mg/m^2 daily rather than the conventional 5/28 schedule had favorable efficacy and was well-tolerated as a second-line agent.[29] Other agents, such as carmustine, carboplatin, etoposide, irinotecan and procarbazine, lomustine (CCNU) and vincristine (PCV), produce low response rates and no significant survival benefit.[37] In selected patients with recurrent GBM who can undergo resection, carmustine implants produce a modest survival advantage of approximately eight weeks.[38] In light of the limited data, treatment decisions for patients with recurrent GBM must be made on an individual basis. Factors to consider include tumor histology, prior therapy, time to relapse, and performance status. In general, patients with recurrent disease should be enrolled in clinical trials whenever possible.

More recently, clinical trials in recurrent GBM have focused on agents targeting important pathways involved in gliomagenesis and progression. Most notably, angiogenesis inhibitors have changed the treatment of recurrent GBM and will be discussed in greater detail below. Bevacizumab is a monoclonal antibody that selectively binds vascular endothelial growth factor (VEGF), an important mediator of angiogenesis. Favorable initial results of bevacizumab in recurrent GBM led to two phase II trials containing bevacizumab monotherapy

Figure 1: Aberrant Pathways in High-grade Glioma and Selected Targeted Agents



EGFR = epidermal growth factor receptor; ERK = extracellular signal-regulated kinase; HDAC = histone deacetylase; HGF = hepatocyte growth factor; HSP90 = heat shock protein 90; MEK = methyl ethyl ketone; mTOR = mammalian target of rapamycin; PARP = poly (ADP-ribose) polymerase; PDGFR = platelet-derived growth factor receptor; PI3K = phosphatidylinositol 3-kinase; PKC = protein kinase C; PTEN = phosphatase and tensin homolog; RTK = receptor tyrosine kinases; SF = scatter factor; VEGFR = vascular endothelial growth factor receptor.
Source: Quant EC, Wen PY, Neuroimaging Clin N Am*, 2010;20(3): in press.*

arms, which demonstrated an RR of 28–35% and PFS6 of 29–42%.[39,40] Bevacizumab monotherapy was well-tolerated with a low incidence of intracranial hemorrhage (0–2.4%) and thromboembolism (8.4–12.5%). Based on the results of these trials, bevacizumab was granted accelerated FDA approval in May 2009 for recurrent GBM. Although several phase II studies have demonstrated improved PFS with bevacizumab for recurrent GBM, its impact on overall survival remains unknown.

The previous practice of combining other cytotoxic agents, such as lomustine, carboplatin, and etoposide, with bevacizumab for recurrent GBM that progresses despite bevacizumab and irinotecan has recently been challenged by a study that showed that these regimens have marginal efficacy.[41] *Table 1* summarizes the standard therapeutic options for GBM.

Chemotherapy for Anaplastic Glioma

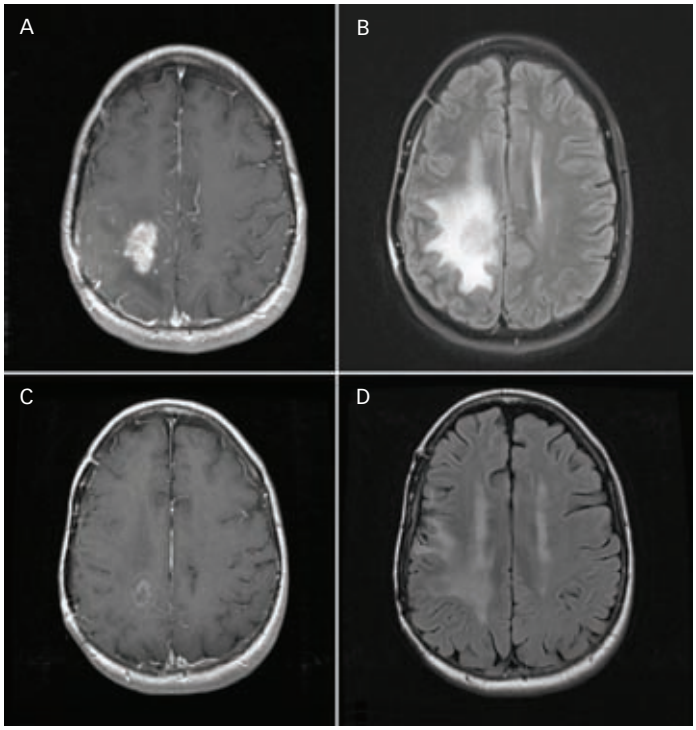
Due to the paucity of randomized clinical trials, there is no consensus in terms of treatment of newly diagnosed AG. The recent Randomized Phase III Study of Sequential Radio-chemotherapy of Anaplastic Glioma With PCV or Temozolomide (NOA-04) that randomized patients with AG to initial radiation followed by chemotherapy (temozolomide or PCV) at progression or initial chemotherapy followed by radiation at progression showed no difference in PFS between the two groups, regardless of histology.[42] Commonly used adjuvant regimens for AA following biopsy or surgery include RT with temozolomide (using a similar regimen to GBM) or RT with adjuvant temozolomide only. An ongoing randomized phase III trial of radiation versus radiation plus temozolomide in non-1p/19q co-deleted AG patients may provide further guidance on management of AA.

Tumors with oligodendroglial components, including AO and AOAs, are less common than AA. However, they have a better prognosis than pure astrocytic tumors and may have increased sensitivity to treatment.[43] The majority of AOs and 14–20% of AOAs have deletions of chromosomes 1p and 19q[43] due to an unbalanced translocation of 19p to 1q.[44] Tumors with 1p/19q co-deletion are particularly sensitive to PCV chemotherapy[4,45] and likely have sensitivity to temozolomide, with an increase in response rate from 34 to 59% in one study.[26] The value of PCV chemotherapy in combination with RT for newly diagnosed AO/AOA has been evaluated in two large phase III trials.[46,47]

Although neither study showed an overall survival benefit, patients treated with both RT and PCV chemotherapy had 10–12 months of additional PFS compared with RT alone. As previously mentioned, the NOA-04 trial that randomized AG patients to initial radiation or to initial chemotherapy (PCV or temozolomide) did not demonstrate a difference in PFS, regardless of treatment.[42] In all of these studies, 1p/19q co-deletion and *MGMT* status were associated with marked survival prolongation.

As most published studies in AO/AOA were initiated prior to 2005, the majority of available data involve PCV chemotherapy. Although the NOA-04 study was not powered to directly compare PCV and temozolomide, no PFS difference was observed between patients randomized to initial PCV versus initial temozolomide,[42] although temozolomide may be associated with less toxicity than PCV.[43] Several large intergroup trials are under way evaluating the optimal combination of RT and temozolomide in patients with newly diagnosed AO/AOA.

Figure 2: 56-year-old Woman with Left Parietal Glioblastoma Showing Response to Therapy with XL184



XL184 is a vascular endothelial growth factor receptor and Met inhibitor. A: Axial T_1 with contrast before therapy; B: Axial fluid-attenuated inverse recovery (FLAIR) before therapy; C: Axial T_1 with contrast four weeks after therapy showing partial response; D: Axial FLAIR four weeks after therapy showing significant reduction in peritumoral edema.

Experimental Therapies

Targeted Molecular Therapies

With improved understanding of the pathways that drive gliomagenesis, targeted molecular therapy has emerged as an important treatment paradigm in HGG in the upfront and recurrent setting. Many investigational drugs target signal transduction pathways involved in cell proliferation, growth, survival, adhesion, motility and differentiation.[48] Targets of particular importance include receptor tyrosine kinases (RTK) such as vascular endothelial growth factor receptor (VEGFR), integrins, epidermal growth factor receptor (EGFR), platelet-derived growth factor receptor (PDGFR), and cMet. RTKs may be inhibited extracellularly by monoclonal antibodies (mAb) and intracellularly by tyrosine kinase inhibitors (TKIs). Inhibitors of intracellular signaling molecules are also being developed against downstream signaling targets such as phosphatidylinositol 3-kinase (PI3K), Akt, mammalian target of rapamycin (mTOR), Raf, and methyl ethyl ketone (MEK). *Figure 1* is a schematic of these pathways.

Anti-angiogenic Therapies

Vascular Endothelial Growth Factor Pathway Inhibitors

Angiogenesis is important to the growth and proliferation of HGG and is mediated through several pathways, most notably VEGF.[49–51] Higher levels of VEGF expression are observed in more malignant tumors.

Targeting VEGF and VEGFR has been the focus of many recent clinical trials. As noted above, bevacizumab has shown promising activity in recurrent GBM and is now FDA-approved for this indication.[40,52,53] Phase II studies of bevacizumab plus irinotecan have also demonstrated efficacy in AG with an RR of 55–66% and PFS6 of 56–61%.[54,55] Bevacizumab is generally well-tolerated, with the most common side effects being fatigue, hypertension, and proteinuria. Less common serious side effects include thromboembolism, hemorrhage, and bowel perforation.

There is emerging evidence that inhibitors of angiogenesis may work synergistically with RT.[21] Two large multicenter trials evaluating the efficacy of adding bevacizumab to RT and temozolomide in newly diagnosed GBM patients are under way. Phase II studies of the regimen appear to be safe despite a possible increase in wound-healing complications.[56] Treatment of HGG with bevacizumab combined with a variety of targeted molecular agents is being studied as well.[52]

Another VEGF-pathway inhibitor currently in clinical trials for HGG is aflibercept (a VEGF decoy receptor that consists of a VEGF receptor fused to an immunoglobulin constant region).[57] In addition to inhibitors of VEGF such as bevacizumab and aflibercept, there are many small-molecule TKIs directed against VEGFR. Cediranib is an oral pan-VEGFR inhibitor that also has activity against platelet-derived growth factor (PDGFR) and c-Kit. In a phase II clinical trial for recurrent GBM, cediranib achieved a promising RR of 27% and PFS6 of 26%.[58] As had been noted in the bevacizumab studies, there was a striking steroid-sparing effect, and the drug was well-tolerated. Other multitargeted VEGFR agents include vandetanib (VEGFR and EGFR), sorafenib (VEGFR, Raf, c-Kit), sunitinib (VEGFR-2, PDFR, c-kit and Flt-3), pazopanib (VEGFR, PDGFR, c-Kit), XL184 (VEGFR and c-Met), and CT322.[59]

Unfortunately, the benefits of anti-angiogenesis therapy are transitory, and it has been suggested that the impressive radiographic responses observed in patients treated with bevacizumab may be the result of decreased permeability of the vasculature rather than a true antitumor effect (see *Figure 2*). Mechanisms of resistance to anti-angiogenic therapy are beginning to be elucidated.[60,61] Some pre-clinical data suggest that blockade of VEGF-mediated angiogenesis may promote tumor infiltration by co-option of native vessels.[62–65] In recurrent HGG patients who are treated with anti-angiogenesis agents, tumor progression is occasionally radiographically observed as an increase in non-enhancing hyperintensity on T_2-weighted or fluid-attenuated inverse recovery (FLAIR) magnetic resonance imaging (MRI). Some hypothesize that this may represent infiltrative tumor growth.[66–70]

Due to the lack of assessment of non-enhancing tumor and other limitations with the standard Macdonald et al. criteria, the multi-disciplinary Response Assessment in Neuro-Oncology (RANO) Working Group recently proposed updated response criteria for HGG.[71] In addition, levels of basic fibroblast growth factor (bFGF) and stromal-derived growth factor 1 alpha (SDF-1α) increased in GBM patients when tumors escaped treatment with cediranib.[72] These findings imply that one may overcome resistance to anti-angiogenic agents by combining anti-VEGF/VEGFR therapy with agents that target tumor invasion, non-VEGF-pro-angiogenic signaling pathways such as the FGF pathway, or vasculogenic pathways such as the SDF-1α pathway.

Integrins

The αvβ3 and αvβ5 integrins are cell-surface receptors that promote endothelial cell migration and survival during angiogenesis.[73] Cilengitide (EMD121974) competitively inhibits αvβ3 and αvβ5. Phase II trials showed a PFS6 of 15% and a median OS of 9.9 months when cilengitide was added to RT and temozolomide. Patients with methylated *MGMT* promoter had better responses.[74] Based on the favorable results of this trial, a multicenter phase III trial is under way using cilengitide in patients with newly diagnosed GBM with methylated *MGMT* promoter.

Receptor Tyrosine Kinases

Epidermal Growth Factor Receptor Inhibitors

EGFR is the most commonly altered RTK in HGG.[75] Approximately 20–30% of GBM have a constitutively active EGFR mutant known as EGFRvIII, and all of these EGFRvIII-expressing tumors also exhibit EGFR amplification or overexpression.[76] Signaling through these and other growth factor receptors activates fundamental signal transduction pathways such as the Ras/mitogen-activated protein kinase (MAPK) pathway and the PI3K/Akt/mTOR pathway, both of which promote cell proliferation.[10] Additionally, many of these pathways upregulate VEGF.[49,77]

While subsets of GBM patients have sustained responses to reversible TKIs that target EGFR, to date the studies have been largely disappointing. Studies of erlotinib (EGFR), gefitinib (EGFR), and lapatinib (ErbB2/HER2, EGFR) have failed to demonstrate any significant survival benefit compared with historical controls.[78–86] The combination of EGFR inhibitors with other therapies is discussed later in this article.

Potential reasons for lack of response include poor blood–brain barrier penetration, insufficient local tumor concentrations, coactivation of multiple TKIs,[87] redundant signaling pathways, and resistance. Irreversible EGFR inhibitors, such as BIBW 2992 and PF-00299804, could have better efficacy in GBM than gefitinib or erlotinib due to increased potency and better brain concentration. This newer class of EGFR inhibitors has been shown to circumvent mechanisms of response to gefitinib or erlotinib in non-small-cell lung cancer cells.[88–93]

mAb and vaccines that target EGFR are currently under investigation in GBM. Both nimotuzumab and cetuximab, a chimeric anti-EGFR human–mouse mAb, are now being studied in combination with RT and temozolomide as upfront GBM therapies. Preliminary results from a phase II clinical trial suggest that the addition of CDX-110, a peptide-based EGFRvIII vaccine, to standard therapy prolongs survival in patients with newly diagnosed GBM.[94] However, since patients were required to have gross total resections and EGFRvIII mutation in order to be eligible for the trial, they represent a highly selected group with good prognosis.

Platelet-derived Growth Factor Inhibitors

Platelet-derived growth factors (PDGF) are a pleiotropic family of peptides that signal through PDGFR to stimulate cellular functions including growth, proliferation, and differentiation.[95] Imatinib mesylate (Gleevec), an inhibitor of PDGFR-α and β, Bcr-Abl, c-Fms, and c-Kit tyrosine kinases, demonstrated activity in pre-clinical models of glioma.[96] However, in clinical trials neither imatinib monotherapy[97,98] nor imatinib in combination with hydroxyurea (a ribonucleoside diphosphate reductase inhibitor)[99] has demonstrated clinically useful activity in GBM. One explanation for the lack of efficacy is that imatinib is a substrate for the

Table 2: Selected Novel Therapies

Type	Therapy
Surgical	Convection-enhanced delivery (e.g. cintredekin besudotox, anti-TGF-β, antisense AP 12009, PRX321 (IL-4 linked to *Pseudomonas* exotoxin)
Overcoming resistance to TMZ	Dose-dense TMZ *MGMT* inhibitors (e.g. O^6-benzylguanine, lomeguatrib) PARP inhibitors (e.g. BSI-201, ABT-888)
Novel chemotherapies	e.g. ANG1005, RTA744
Anti-angiogenic therapy	Anti-αvβ5 integrins (e.g. cilengitide) Anti-hepatocyte growth factor (e.g. AMG-102) Anti-VEGF (e.g. bevacizumab, aflibercept [VEGF-Trap]) Anti-VEGFR (e.g. cediranib, vandetinib, pazopanib sorafenib, sunitinib, XL184, CT-322, IMC-1121B) Others (e.g. thalidomide)
Targeted molecular therapy	Akt (e.g. perifosine, MK2206) Bcl2 (AT101) EGFR inhibitors (e.g erlotinib, gefitinib, lapatinib, BIBW 29992, PF00299804, nimotuzumab, cetuximab) FTI inhibitors (e.g. tipifarnib and lornafarnib) HDAC inhibitors (e.g. vorinostat, depsipeptide, LBH589) HSP90 inhibitors (e.g. 17AAG, IPI504) Insulin-like growth factor receptor (OSI906) Met (e.g. XL184) mTOR inhibitors (e.g. everolimus, sirolimus, temsirolimus, AP23573) PI3K inhibitors (XL765) PKCβ (e.g. enzastaurin) PDGFR inhibitors (e.g. dasatinib, imatinib, tandutinib, IMC3G3 (Mab against PDGFR-alpha) Proteosome (e.g bortezomib) Raf (e.g. sorafenib) Src (e.g dasatinib, bosutinib [SK606]) TGF-β (e.g. AP12009)
	Combination therapies: Erlotinib + temsirolimus Gefitinib + everolimus Gefitinib + sirolimus Sorafenib + temsirolimus, erlotinib, or tipifarnib Pazopanib + lapatinib Bortezomib + vorinostat Vandetinib + sirolimus Cediranib + cilengitide
Immunotherapy	Dendritic cell and EGFRvIII peptide vaccines, monoclonal antibodies (e.g ^{74}I-anti-tenascin antibody) (CDX110)
Gene therapy	Delta-24-RGD-4C Cerepro
Therapy directed against stem cells	Notch inhibitors (MRK0752, R4929097) Sonic hedgehog inhibitor (GDC-4409)
Miscellaneous	^{74}I-TM-601

EGFR = epidermal growth factor receptor; FTI = farnesylytransferase; HDAC = histone deacetylase; HSP90 = heat shock protein 90; MGMT *= methylguanine-DNA-methyl transferase; mTOR = mammalian target of rapamycin; PARP = poly (ADP-ribose) polymerase; PDGFR = platelet-derived growth-factor inhibitor; PI3K = phosphatidylinositol 3-kinase; PKC = protein kinase C; RT = radiotherapy; TMZ = temozolomide; TGF= transforming growth factor; TMZ = temozolomide; VEGFR = vascular endothelial growth factor receptor.*

P-glycoprotein efflux pump that limits its intracranial distribution.[100] Tandutinib and dasatinib, second-generation PDGFR inhibitors with improved CNS penetration, are in clinical trials for recurrent HGG.

c-Met Inhibitors

Scatter factor/hepatocyte growth factor (SF/HGF) and its TKR c-Met play a role in cell growth, cell motility, morphogenesis, and angiogenesis.[101] AMG 102 is a fully human monoclonal antibody that selectively targets SF/HGF. A phase II study of AMG 102 in recurrent GBM was recently completed but failed to produce any benefit.[102] A recent study suggests that the combination of EGFR inhibitors and c-Met inhibitors may be more effective than either agent alone in phosphatase and tensin homolog (PTEN) null GBM.[103] Trials of c-Met TKIs such as XL184 are under way in GBM.

Intracellular Signaling Kinase Inhibitors

As activation of several RTKs, including EGFR and PDGFR, converges at the Ras/MAPK and PI3K/Akt pathways, inhibiting these downstream molecules may be more efficacious than targeting individual RTKs.

In the Ras/MAPK pathway, potential targets include Raf, MEK, and farnesyltransferase. An early step in activation of the Ras/MAPK pathway is localization of Ras to the cell membrane, which depends on Ras farnesylation by the enzyme farnesyltransferase. Farnesyltransferase inhibitors (FTIs) such as tipifarnib[104] showed modest activity as monotherapy in recurrent HGG and are now being studied in combination with temozolomide for HGG.

The Raf serine/threonine kinases are the main downstream effectors of Ras in the MAPK pathway. Sorafenib is an inhibitor of c-Raf kinase, but also inhibits pro-angiogenic RTKs including VEGFR-2, VGFRR-3, PDGFR-β, Flt-3, c-Kit, and FGFR-1. Several trials of sorafenib in HGG are under way, although the preliminary results have been disappointing.

Several PI3K and Akt inhibitors are in development or early clinical trials. XL765, an inhibitor of PI3K and mTOR, is currently in a phase I clinical trial in combination with temozolomide for HGG. Studies of XL147 and BKM120 are planned. Akt inhibitors undergoing evaluation in HGG include perifosine and MK2206.

mTOR, a downstream molecule in the PI3K/Akt pathway, is also an attractive target for therapy.[105] The mTOR inhibitor sirolimus (rapamycin) and its analogs temsirolimus, everolimus, and ridaforolimus are the most clinically advanced PI3K/Akt pathway inhibitors. Despite promising results from pre-clinical studies, temsirolimus monotherapy was not clinically active in recurrent GBM in two multicenter phase II clinical trials,[106,107] possibly because inhibition of only the TORC1 component may result in the activation of Akt. An Akt inhibitor, such as perifosine or MK2206, or a combined PI3K/mTOR inhibitor, such as XL765, may ultimately prove more effective. Studies combining mTOR inhibitors with other targeted agents are discussed below.

New Molecular Targets

Histone Deacetylase Inhibitors

Histone deacetylase (HDAC) inhibitors cause the growth arrest, differentiation, or apoptosis of many transformed cells by altering transcription of various genes.[108] Vorinostat is a small-molecule inhibitor of most human class I and class II HDACs. Vorinostat demonstrated moderate clinical activity in a phase II study of patients with recurrent GBM with a PFS6 of 15.2%.[109] The Adult Brain Tumor Consortium and the North Central Cancer Treatment Group are now jointly conducting a trial of vorinostat with RT and temozolomide in patients with newly diagnosed HGG. Clinical trials combining HDAC inhibitors with other agents such as bortezomib, a proteosome inhibitor, or bevacizumab are currently under way in recurrent GBM. A more potent HDAC inhibitor, LBH589, is entering phase II studies in recurrent GBM.

DNA Repair

Poly(ADP-ribose) polymerase (PARP) is a nuclear enzyme that signals the presence of DNA breaks and facilitates DNA repair by engaging mechanisms such as base excision repair (BER).[110] As PARP inhibitors disrupt BER, an important mediator of TMZ resistance, these agents may enhance the antitumor effects of temozolomide against HGG. Two PARP inhibitors, BSI-201 and ABT-888, are being tested in combination with radiation and temozolomide for newly diagnosed GBM.

Glioma Stem Cells

Glioma stem cells (GSCs) are believed to represent a subpopulation of cells in the tumor with the ability to self-renew, proliferate, and give rise to progeny of multiple neuroepithelial lineages.[111] They may contribute to treatment resistance in HGG.[112,113] Stem cells are predicted to be difficult treatment targets because they transition slowly through the cell cycle, express high levels of drug-export proteins, and may not express oncoproteins that are targeted by newer chemotherapeutic drugs.[112] As a result, there is significant interest in molecular therapies affecting stem-cell pathways, such as notch (e.g. MRK0752 and R4929097), sonic hedgehog (e.g. GDC4409),[111,112,114] and hypoxia-inducible factors 1 and 2α.[113]

Overcoming Resistance to Targeted Molecular Therapy

Monotherapy with most targeted molecular agents (except for anti-VEGFR agents) has shown modest activity at best. These results are not surprising when one considers that most HGG have co-activation of multiple tyrosine kinases[87] and highly redundant signaling pathways. Approaches now under evaluation in clinical trials include the combination of a targeted agent with radiotherapy and chemotherapy, the combination of several targeted agents and agents that hit multiple relevant targets at once.[10,20,115,116] For example, the EGFR inhibitor erlotinib has been studied in combination with mTOR inhibitors, such as sirolimus[117] and temsirolimus.[118] Although preliminary results from these erlotinib combination studies suggest only modest efficacy due to poor tolerability,[118,119] other combinations may be better tolerated and are in clinical trials. There is also continued interest in clinical trial designs that incorporate tissue specimens to identify biomarkers to predict tumor response to target inhibition.[120,121] This may allow identification of patients who are more likely to respond to specific therapies. Advances in molecular profiling of tumor tissue may lead to more selective use of targeted molecular agents and tailoring of therapy to individual patients. A recent study demonstrated that GBM can be subdivided by genomic profiling into four subtypes, each of which demonstrates unique molecular alterations.[122]

A prominent mechanism of resistance to targeted molecular therapy is inadequate drug delivery across the blood–brain barrier. Increasingly, trials of novel targeted agents include a surgical component to evaluate the ability of the drug to reach therapeutic concentrations in the tumor and inhibit the putative target. Patients with recurrent HGG were administered the agent prior to planned surgery and the tumor is obtained for drug concentration and evidence of pathway inhibition.[123] If drug concentration and target inhibition in the tumor is poor, further evaluation of that agent in HGG is probably not warranted.[120]

Other Therapeutic Modalities

A large number of therapeutic modalities are being explored for HGG. Examples include inhibitors of the ubiquitin-proteosome system such as bortezomib,[124–126] heat-shock protein inhibitors,[127,128] cytokines,[129] gene therapy,[130] synthetic chlorotoxins (TM-601),[131] chemotherapeutic agents with enhanced ability to penetrate into tumor tissue, and convection-enhanced delivery (CED) of drugs and toxins.[132] Intracavitary TM-601, the synthetic version of a chlorotoxin found in the venom of the giant yellow Israeli scorpion, is under evaluation in a phase II study. Agents administered directly into HGG via CED that have been studied in phase III clinical trials include interleukin-13 (IL-13), *Pseudomonas aeruginosa* exotoxin, and transferrin-C diphtheriae toxin. Unfortunately, both trials were terminated for futility after interim analysis.[133] By contrast, studies of trabedersen (AP1007), a phosphorothioate antisense oligonucleotide against transforming growth factor β2, appears to have activity in recurrent AG and is being evaluated in a phase III study. Therapy involves the insertion or modification of genes in a patient's cell to treat a disease.[134] Transfer of 'suicidal' genes via viral vectors such as herpes simple virus thymidine kinase gene (HSV-tk) has demonstrated only limited survival benefit in several clinical trials for recurrent GBM.[134] Viral vectors can also deliver pro-apoptotic cytokines such as tumor necrosis factor-related apoptosis-inducing ligand (TRAIL) and p53 as well as cytokines such as IL-2 and interferon beta (IFN-β). Other methods of delivery under investigation include cell-based transfer and synthetic vectors.

Antitumor vaccines based on peptide antigens, dendritic cells, or whole tumor cells represent another major avenue of investigation. Among the many promising vaccines in addition to the previously described CDX-110 are GVAX, which involves administration of irradiated autologous tumor cells mixed with granulocyte macrophage colony-stimulating factor (GM-CSF)-producing cells[135] and vaccines against HSP90.[136] In small phase II studies, these vaccines appear to be well-tolerated and show promising efficacy compared with historic controls. However, larger prospective controlled studies will be required to confirm any clinical benefit. *Table 2* summarizes selected novel therapies for HGG.

Conclusions

Despite progress in recent years, the prognosis for most patients with HGG remains poor. The introduction of temozolomide to radiation treatment was an important advance, and anti-angiogenic therapy has now emerged as a critical component of treatment for recurrent tumors. Thus far, the potential of targeted molecular drug therapy has not been fully realized. Future approaches include the use of treatment regimens that inhibit complementary targets, combinations of targeted molecular drugs with RT, chemotherapy, and anti-angiogenic therapies, and novel agents directed at tumor stem cells. Additionally, the understanding of glioma biology and treatment resistance is evolving at a rapid pace. Genome-wide association studies are just beginning to uncover mutations that will lead to better characterization of HGG. Therapeutic strategies for circumventing treatment resistance mediated by *MGMT* and PARP as well as the intrinsic resistance of glioma stem cells are beginning to be developed. ■

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1. CBTRUS, Statistical Report: Primary Brain Tumours in the United Status, 2000–2004. In: *Central Brain Tumour Registry of the United States*, 2008.
2. Louis DN, Ohgaki H, Wiestler OD, et al., *The 2007 WHO classification of tumours of the central nervous system*, Lyon, France: IARC Press; 2007.
3. Wen PY, Kesari S, Malignant gliomas in adults, *N Engl J Med*, 2008;359(5):492–507.
4. Ino Y, Betensky RA, Zlatescu MC, et al., Molecular subtypes of anaplastic oligodendroglioma: implications for patient management at diagnosis, *Clin Cancer Res*, 2001;7(4):839–45.
5. Colman H, Aldape K, Molecular predictors in glioblastoma: toward personalized therapy, *Arch Neurol*, 2008;65(7):877–83.
6. Hegi ME, Diserens AC, Gorlia T, et al., *MGMT* gene silencing and benefit from temozolomide in glioblastoma, *N Engl J Med*, 2005;352(10):997–1003.
7. Phillips HS, Kharbanda S, Chen R, et al., Molecular subclasses of high-grade glioma predict prognosis, delineate a pattern of disease progression, and resemble stages in neurogenesis, *Cancer Cell*, 2006;9(3):157–73.
8. Mclendon R, Freidman H, Bigner D, et al., Comprehensive genomic characterization defines human glioblastoma genes and core pathways, *Nature*, 2008;455(7216):1061–8.
9. Parsons DW, Jones S, Zhang X, et al., An integrated genomic analysis of human glioblastoma multiforme, *Science*, 2008;321(5897):1807–12.
10. Furnari FB, Fenton T, Bachoo RM, et al., Malignant astrocytic glioma: genetics, biology, and paths to treatment, *Genes Dev*, 2007;21(21):2683–2710.
11. Keles GE, Lamborn KR, Chang SM, et al., Volume of residual disease as a predictor of outcome in adult patients with recurrent supratentorial glioblastomas multiforme who are undergoing chemotherapy, *J Neurosurg*, 2004;100:41–6.
12. Sanai N, Berger MS, Glioma extent of resection and its impact on patient outcome, *Neurosurgery*, 2008;62(4):753–64, discussion 264–6.

13. Stummer W, Reulen HJ, Meinel T, et al., Extent of resection and survival in glioblastoma multiforme: identification of and adjustment for bias, *Neurosurgery*, 2008;62(3):564–76.
14. Lacroix M, Abi-Said D, Fourney DR, et al., A multivariate analysis of 416 patients with glioblastoma multiforme: prognosis, extent of resection, and survival, *J Neurosurg*, 2001;95(2):190–98.
15. Walker MD, Alexander E Jr, Hunt WE, et al., Evaluation of BCNU and/or radiotherapy in the treatment of anaplastic gliomas. A cooperative clinical trial, *J Neurosurg*, 1978;49(3):333–43.
16. Stupp R, Mason WP, van den Bent MJ, et al., Radiotherapy plus concomitant and adjuvant temozolomide for glioblastoma, *N Engl J Med*, 2005;352(10):987–96.
17. Lee SW, Fraass BA, Marsh LH, et al., Patterns of failure following high-dose 3-D conformal radiotherapy for high-grade astrocytomas: a quantitative dosimetric study, *Int J Radiat Oncol Biol Phys*, 1999;43(1):79–88.
18. Fiveash JB, Spencer SA, Role of radiation therapy and radiosurgery in glioblastoma multiforme, *Cancer J*, 2003;9:222–9.
19. Stupp R, Hegi ME, Gilbert MR, Chakravarti A, Chemoradiotherapy in malignant glioma: standard of care and future directions, *J Clin Oncol*, 2007;25(26): 4127–36.
20. Chi AS, Wen PY, Inhibiting kinases in malignant gliomas, *Expert Opin Ther Targets*, 2007;11(4):473–96.
21. 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(26):4033–42.
22. Stieber VW, Mehta MP, Advances in radiation therapy for brain tumours, *Neurol Clin*, 2007;25(4):1005–33.
23. Tsao MN, Mehta MP, Whelan TJ, et al., The American Society for Therapeutic Radiology and Oncology (ASTRO) evidence-based review of the role of radiosurgery for malignant glioma, *Int J Radiat Oncol Biol Phys*, 2005;63(1): 47–55.
24. Combs SE, Thilmann C, Edler L, et al., Efficacy of fractionated stereotactic reirradiation in recurrent gliomas: long-term results in 172 patients treated in a single institution, *J Clin Oncol*, 2005;23(34):8863–9.
25. Stupp R, Hegi ME, Mason WP, et al., Effects of radiotherapy with concomitant and adjuvant temozolomide versus radiotherapy alone on survival in glioblastoma in a randomised phase III study: 5-year analysis of the EORTC-NCIC trial, *Lancet Oncol*, 2009;10(5):459–66.
26. Brandes AA, Tosoni A, Cavallo G, et al., Correlations between O6-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*, 2006;24(29):4746–53.
27. Brandes A, 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*, 2006;95(9):1155–60.
28. Wick A, Felsberg J, Steinbach JP, et al., Efficacy and tolerability of temozolomide in an alternating weekly regimen in patients with recurrent glioma, *J Clin Oncol*, 2007;25(22):3357–61.
29. Perry JR, Belanger K, Mason WP, et al., Phase II trial of continuous dose-intense temozolomide in recurrent malignant glioma: RESCUE study, *J Clin Oncol*, 2010;28(12):2051–7.
30. Tolcher AW, Gerson SL, Denis L, et al., Marked inactivation of O6-alkylguanine-DNA alkyltransferase activity with protracted temozolomide schedules, *Br J Cancer*, 2003;88(7):1004–11.
31. Broniscer A, Gururangan S, MacDonald TJ, et al., Phase I trial of single-dose temozolomide and continuous administration of o6-benzylguanine in children with brain tumours: a pediatric brain tumour consortium report, *Clin Cancer Res*, 2007;13(22 Pt 1):6712–18.
32. Quinn JA, Jiang SX, Reardon DA, et al., Phase 1 trial of temozolomide plus irinotecan plus O6-benzylguanine in adults with recurrent malignant glioma, *Cancer*, 2009;115(13):2964–70.
33. Quinn JA, Jiang SX, Reardon DA, et al., Phase II trial of temozolomide plus o6-benzylguanine in adults with recurrent, temozolomide-resistant malignant glioma, *J Clin Oncol*, 2009;27(8):1262–7.
34. Westphal M, Hilt DC, Bortey E, et al., A phase 3 trial of local chemotherapy with biodegradable carmustine (BCNU) wafers (Gliadel wafers) in patients with primary malignant glioma, *Neuro Oncol*, 2003;5(2):79–88.
35. Brandes AA, Ermani M, Basso U, et al., Temozolomide as a second-line systemic regimen in recurrent high-grade glioma: a phase II study, *Ann Oncol*, 2001;12(2): 255–7.
36. Yung W, Albright R, Olson J, et al., A phase II study of temozolomide vs. procarbazine in patients with glioblastoma multiforme at first relapse, *Br J Cancer*, 2000;83(5):588–93.
37. Wong ET, Hess KR, Gleason MJ, et al., Outcomes and prognostic factors in recurrent glioma patients enrolled onto phase II clinical trials, *J Clin Oncol*, 1999;17(8): 2572–8.
38. Brem H, Piantadosi S, Burger PC, et al., Placebo-controlled trial of safety and efficacy of intraoperative controlled delivery by biodegradable polymers of chemotherapy for recurrent malignant gliomas: The Polymer-brain Tumour Treatment Group, *Lancet*, 1999;345:1008–12.
39. Kreisl TN, Kim L, Moore K, et al., Phase II trial of single-agent bevacizumab followed by bevacizumab plus irinotecan at tumour progression in recurrent glioblastoma, *J Clin Oncol*, 2009;27(5):740–45.
40. Friedman HS, Prados MD, Wen PY, et al., Bevacizumab alone and in combination with irinotecan in recurrent glioblastoma, *J Clin Oncol*, 2009;27(28):4733–40.
41. Quant EC, Norden AD, Drappatz J, et al., Role of a second chemotherapy in recurrent malignant glioma patients who progress on bevacizumab, *Neuro Oncol*, 2009;11(5):550–55.
42. Wick W, Hartmann C, Engel C, et al., NOA-04 randomized phase III trial of sequential radiochemotherapy of anaplastic glioma with procarbazine, lomustine, and vincristine or temozolomide, *J Clin Oncol*, 2009;27(35): 5874–80.
43. van den Bent MJ, Anaplastic oligodendroglioma and oligoastrocytoma, *Neurol Clin*, 2007;25(4):1089–1109.
44. Jenkins RB, Blair H, Ballman KV, et al., A t(1;19)(q10;p10) mediates the combined deletions of 1p and 19q and predicts a better prognosis of patients with oligodendroglioma, *Cancer Res*, 2006;66(20):9852–61.
45. Cairncross JG, Ueki K, Zlatescu MC, et al., Specific genetic predictors of chemotherapeutic response and survival in patients with anaplastic oligodendrogliomas, *J Natl Cancer Inst*, 1998;90(19):1473–9.
46. Cairncross G, Berkey B, Shaw E, et al., Phase III trial of chemotherapy plus radiotherapy compared with radiotherapy alone for pure and mixed anaplastic oligodendroglioma: Intergroup Radiation Therapy Oncology Group Trial 9402, *J Clin Oncol*, 2006;24(18): 2707–14.
47. van den Bent MJ, Carpentier AF, Brandes AA, et al., Adjuvant procarbazine, lomustine, and vincristine improves progression-free survival but not overall survival in newly diagnosed anaplastic oligodendrogliomas and oligoastrocytomas: a randomized European Organisation for Research and Treatment of Cancer phase III trial, *J Clin Oncol*, 2006;24(18):2715–22.
48. Van Meir EG HC, Norden AD, Shu HK, Wen PY, Olson JJ. Exciting new advances in neuro-oncology: the avenue to a cure for malignant glioma, *CA Cancer J Clin*, 2010; 60(3):166–93.
49. Jain RK, di Tomaso E, Duda DG, et al., Angiogenesis in brain tumours, *Nat Rev Neurosci*, 2007;8(8):610–22.
50. Folkman J, Tumour angiogenesis: therapeutic implications, *N Engl J Med*, 1971;285(21):1182–6.
51. Folkman J, Angiogenesis, *Ann Rev Med*, 2006;57:1–18.
52. Norden AD, Drappatz J, Wen PY, Antiangiogenic therapy in malignant gliomas, *Curr Opin Oncol*, 2008;20(6):652–61.
53. Vredenburgh JJ, Desjardins A, Herndon JE II, et al., Bevacizumab plus irinotecan in recurrent glioblastoma multiforme, *J Clin Oncol*, 2007;25(30):4722–9.
54. Vredenburgh JJ, Desjardins A, Herndon JE II, et al., Phase II trial of bevacizumab and irinotecan in recurrent malignant glioma, *Clin Cancer Res*, 2007;13(4):1253–9.
55. Desjardins A, Reardon DA, Herndon JE II, et al., Bevacizumab plus irinotecan in recurrent WHO grade 3 malignant gliomas, *Clin Cancer Res*, 2008;14(21):7068–73.
56. 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(5):1372–80.
57. Holash J, Davis S, Papadopoulos N, et al., VEGF-Trap: a VEGF blocker with potent antitumour effects, *Proc Natl Acad Sci U S A*, 2002;99(17):11393–8.
58. Batchelor TT, Duda DG, di Tomaso E, et al., Phase II Study of Cediranib, an Oral Pan-Vascular Endothelial Growth Factor Receptor Tyrosine Kinase Inhibitor, in Patients With Recurrent Glioblastoma, *J Clin Oncol*, 2010;28(17):2817–23
59. Norden AD, Drappatz J, Wen PY, Antiangiogenic therapies for high-grade glioma, *Nat Rev Neurol*, 2009;5(11):610–20.
60. Bergers G, Hanahan D. Modes of resistance to anti-angiogenic therapy, *Nat Rev Cancer*, 2008;8(8):592–603.
61. Ellis LM, Hicklin DJ, Pathways mediating resistance to vascular endothelial growth factor-targeted therapy, *Clin Cancer Res*, 2008;14(20):6371–5.
62. Holash J, Maisonpierre P, Compton D, et al., Vessel cooption, regression, and growth in tumours mediated by angiopoietins and VEGF, *Science*, 1999;284(5422): 1994–8.
63. Lamszus K, Kunkel P, Westphal M, Invasion as limitation to anti-angiogenic glioma therapy, *Acta Neurochir Suppl*, 2003;88:169–77.
64. 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(4):306–14.
65. Paez-Ribes M, Allen E, Hudock J, et al., Antiangiogenic therapy elicits malignant progression of tumours to increased local invasion and distant metastasis, *Cancer Cell*, 2009;15(3):220–31.
66. Norden AD, Young GS, Setayesh K, et al., Bevacizumab for recurrent malignant gliomas: efficacy, toxicity, and patterns of recurrence, *Neurology*, 2008;70(10):779–87.
67. Iwamoto FM, Abrey LE, Beal K, et al., Patterns of relapse and prognosis after bevacizumab failure in recurrent glioblastoma, *Neurology*, 2009;73(15): 1200–1206.
68. Zuniga RM, Torcuator R, Jain R, et al., Efficacy, safety and patterns of response and recurrence in patients with recurrent high-grade gliomas treated with bevacizumab plus irinotecan, *J Neurooncol*, 2009;91(3): 329–36.
69. Gerstner ER, Chen PJ, Wen PY, et al., Infiltrative patterns of glioblastoma spread detected via diffusion MRI after treatment with cediranib, *Neuro Oncol*, 2010;12(5):466–72.
70. de Groot J, Fuller G, Kumar AJ, et al., Tumour invasion after treatment of glioblastoma with bevacizumab:radiographic and pathologic correlation in humans and mice, *Neuro Oncol*, 2010;12(3):233–42.
71. Wen PY, Macdonald DR, Reardon DA, et al., Updated response assessment criteria for high-grade gliomas: response assessment in neuro-oncology working group, *J Clin Oncol*, 2010;28(11):1963–72.
72. Batchelor T, Sorensen A, di Tomaso E, et al., AZD2171, a Pan-VEGF Receptor Tyrosine Kinase Inhibitor, Normalizes Tumour Vasculature and Alleviates Edema in Glioblastoma Patients, *Cancer Cell*, 2007;11(1):83–95.
73. Avraamides CJ, Garmy-Susini B, Varner JA. Integrins in angiogenesis and lymphangiogenesis, *Nat Rev Cancer*, 2008;8(8):604–17.
74. Stupp R, Goldbrunner R, Neyns B, et al., Mature results of a phase I/IIa trial of the integrin inhibitor cilengitide (EMD121974) added to standard concomitant and adjuvant temozolomide and radiotherapy for newly diagnosed glioblastoma, *Neuro Oncol*, 2007;9(4):517.
75. Maher E, Furnari F, Bachoo R, et al., Malignant glioma:

genetics and biology of a grave matter, *Genes Dev*, 2001;15(11):1311–33.
76. Aldape KD, Ballman K, Furth A, et al., Immunohistochemical detection of EGFRvIII in high malignancy grade astrocytomas and evaluation of prognostic significance, *J Neuropathol Exp Neurol*, 2004;63(7):700–707.
77. Guo P, Hu B, Gu W, et al., Platelet-derived growth factor-B enhances glioma angiogenesis by stimulating vascular endothelial growth factor expression in tumour endothelia and by promoting pericyte recruitment, *Am J Pathol*, 2003;162(4):1083–93.
78. Brown PD, Krishnan S, Sarkaria JN, et al., Phase I/II trial of erlotinib and temozolomide with radiation therapy in the treatment of newly diagnosed glioblastoma multiforme: North Central Cancer Treatment Group Study N0177, *J Clin Oncol*, 2008;26(34):5603–9.
79. Chakravarti A, Berkey B, Robins HI, et al., An update of phase II results from RTOG 0211: A phase I /II study of gefitinib with radiotherapy in newly-diagnosed glioblastoma multiforme, *J Clin Oncol*, 2006;24:1527.
80. Franceschi E, Cavallo G, Lonardi S, et al., Gefitinib in patients with progressive high-grade gliomas: a multicentre phase II study by Gruppo Italiano Cooperativo di Neuro-Oncologia (GICNO), *Br J Cancer*, 2007;96(7): 1047–51.
81. Rich JN, Reardon DA, Peery T, et al., Phase II trial of gefitinib in recurrent glioblastoma, *J Clin Oncol*, 2004; 22(1):133–42.
82. Lieberman F, Cloughesy T, DeAngelis LM, al. e. Phase I-II study of ZD-1839 for recurrent malignant gliomas and meningiomas progressing after radiation therapy, *J Clin Oncol*, (Meeting Abstracts) 2004;22:1510.
83. Cloughesy T, Yung A, Vrendenberg J, et al., Phase II study of erlotinib in recurrent GBM: Molecular predictors of outcome, *J Clin Oncol*, (Meeting Abstracts) 2005;23: 1507.
84. Vogelbaum MA, Peereboom D, Stevens G, et al., Phase II study of erlotinib single agent therapy in recurrent glioblastoma multiforme, *Eur J Cancer*, 2005;3(2):135S.
85. Raizer JJ, Abrey LE, Lassman AB, et al., A phase II trial of erlotinib in patients with recurrent malignant gliomas and nonprogressive glioblastoma multiforme postradiation therapy, *Neuro Oncol*, 2010;12(1):95–103.
86. Thiessen B, Stewart C, Tsao M, et al., A phase I/II trial of GW572016 (lapatinib) in recurrent glioblastoma multiforme: clinical outcomes, pharmacokinetics and molecular correlation, *Cancer Chemother Pharmacol*, 2009 (Epub ahead of print).
87. Stommel JM, Kimmelman AC, Ying H, et al., Coactivation of receptor tyrosine kinases affects the response of tumour cells to targeted therapies, *Science*, 2007;318(5848):287–90.
88. Greulich H, Chen TH, Feng W, et al., Oncogenic transformation by inhibitor-sensitive and -resistant EGFR mutants, *PLoS Med*, 2005;2(11):e313.
89. Kobayashi S, Boggon T, Dayaram T, et al., EGFR mutation and resistance of non-small-cell lung cancer to gefitinib, *N Engl J Med*, 2005;352(8):786–92.
90. Kobayashi S, Ji H, Yuza Y, et al., An alternative inhibitor overcomes resistance caused by a mutation of the epidermal growth factor receptor, *Cancer Res*, 2005; 65(16):7096–7101.
91. Kwak EL, Sordella R, Bell DW, et al., Irreversible inhibitors of the EGF receptor may circumvent acquired resistance to gefitinib, *Proc Natl Acad Sci U S A*, 2005;102(21):7665–70.
92. Li D, Ambrogio L, Shimamura T, et al., BIBW2992, an irreversible EGFR/HER2 inhibitor highly effective in preclinical lung cancer models, *Oncogene*, 2008;27(34): 4702–11.
93. Shimamura T, Ji H, Minami Y, et al., Non-small-cell lung cancer and Ba/F3 transformed cells harboring the ERBB2 G776insV_G/C mutation are sensitive to the dual-specific epidermal growth factor receptor and ERBB2 inhibitor HKI-272, *Cancer Res*, 2006;66(13): 6487–91.
94. Sampson JH, Archer GE, Bigner DD, et al., Effect of EGFRvIII-targeted vaccine (CDX-110) on immune response and TTP when given with simultaneous standard and continuous temozolomide in patients with GBM, *J Clin Oncol*, (Meeting Abstracts) 2008;26:2011.
95. George D, Targeting PDGF receptors in cancer – rationales and proof of concept clinical trials, *Adv Exp Med Biol*, 2003;532:141–51.
96. Kilic T, Alberta J, Zdunek P, et al., Intracranial inhibition of platelet-derived growth factor-mediated glioblastoma cell growth by an orally active kinase inhibitor of the 2-phenylaminopyrimidine class, *Cancer Res*, 2000;60: 5143–50.
97. Wen PY, Yung WK, Lamborn KR, et al., Phase I/II study of imatinib mesylate for recurrent malignant gliomas: North American Brain Tumour Consortium Study 99-08, *Clin Cancer Res*, 2006;12(16):4899–4907.
98. Raymond E, Brandes AA, Dittrich C, et al., Phase II study of imatinib in patients with recurrent gliomas of various histologies: a European Organisation for Research and Treatment of Cancer Brain Tumour Group Study, *J Clin Oncol*, 2008;26(28):4659–65.
99. Dresemann G, Weller M, Bogdahn U, et al., Imatinib plus hydroxyurea versus hydroxyurea monotherapy in progressive glibolastoma – an international multicenter, open-label, randomized phase III study (AMBROSIA-STUDY), *Neuro Oncol*, (Meeting Abstracts) 2008;10(5):820.
100. Dai H, Marbach P, Lemaire M, et al., Distribution of STI-571 to the brain is limited by P-glycoprotein-mediated efflux, *J Pharmacol Exp Ther*, 2003;304:1085–92.
101. Matsumoto K, Nakamura T. Emerging multipotent aspects of hepatocyte growth factor, *J Biochem*, 1996;119(4): 591–600.
102. Reardon DA, Cloughsey TF, Raizer JJ, et al., Phase II study of AMG 102, a fully human neutralizing antibody against hepatocyte growth factor/scatter factor, in patients with recurrent glioblastoma multiforme, *J Clin Oncol*, 2008; 26:2051.
103. Lal B, Goodwin CR, Sang Y, et al., EGFRvIII and c-Met pathway inhibitors synergize against PTEN-null/EGFRvIII+ glioblastoma xenografts, *Mol Cancer Ther*, 2009;8(7): 1751–60.
104. Cloughesy TF, Wen PY, Robins HI, et al., Phase II trial of tipifarnib in patients with recurrent malignant glioma either receiving or not receiving enzyme-inducing antiepileptic drugs: a North American Brain Tumour Consortium Study, *J Clin Oncol*, 2006;24(22):3651–6.
105. Chiang GG, Abraham RT, Targeting the mTOR signaling network in cancer, *Trends Mol Med*, 2007;13(10):433–42.
106. Chang SM, Wen P, Cloughesy T, et al., Phase II study of CCI-779 in patients with recurrent glioblastoma multiforme, *Invest New Drugs*, 2005;23(4):357–61.
107. Galanis E, Buckner J, Maurer M, et al., Phase II trial of temsirolimus (CCI-779) in recurrent glioblastoma multiforme: a North Central Cancer Treatment Group Study, *J Clin Oncol*, 2005;23(23):5294–5304.
108. Jones PA, Baylin SB, The fundamental role of epigenetic events in cancer, *Nat Rev Genet*, 2002;3(6):415–28.
109. Galanis E, Jaeckle KA, Maurer MJ, et al., Phase II trial of vorinostat in recurrent glioblastoma multiforme: a north central cancer treatment group study, *J Clin Oncol*, 2009;27(12):2052–8.
110. Ma WW, Adjei AA, Novel agents on the horizon for cancer therapy, *CA Cancer J Clin*, 2009;59(2):111–37.
111. Das S, Srikanth M, Kessler JA, Cancer stem cells and glioma, *Nat Clin Pract Neurol*, 2008;4(8):427–35.
112. Stiles CD, Rowitch DH, Glioma stem cells: a midterm exam, *Neuron*, 2008;58(6):832–46.
113. Dirks PB, Brain tumour stem cells: bringing order to the chaos of brain cancer, *J Clin Oncol*, 2008;26(17):2916–24.
114. Bao S, Wu Q, McLendon RE, et al., Glioma stem cells promote radioresistance by preferential activation of the DNA damage response, *Nature*, 2006;444(7120): 756–60.
115. Sathornsumetee S, Rich JN, Reardon DA, Diagnosis and Treatment of High-Grade Astrocytoma, *Neurol Clin*, 2007;25(4):1111–39.
116. Wen PY, New developments in targeted molecular therapies for glioblastoma, *Expert Rev Anticancer Ther*, 2009;9(1):7–10.
117. Friedman HS, Desjardins A, Vredenburgh JJ, et al., Phase II trial of erlotinib plus sirolimus for recurrent glioblastoma multiforme (GBM), *J Clin Oncol*, (Meeting Abstracts), 2008;26:2062.
118. Wen P, Kuhn J, Chang S, et al., Phase I/II Study Of Erlotinib And Temsirolimus (CCI-779) for Patients with Recurrent Malignant Gliomas (NABTC 04-02), *Neuro Oncol*, (Meeting Abstracts), 2008;10(5):824.
119. Prados M GM, Kuhn J, et al., Phase I/II study of sorefenib and erlotinib for patients with recurrent glioblastoma (GBM) (NABTC 05-02), *J Clin Oncol*, (Meeting Abstracts), 2009;27:2005.
120. Chang SM, Lamborn KR, Kuhn JG, et al., Neurooncology clinical trial design for targeted therapies: lessons learned from the North American Brain Tumour Consortium, *Neuro Oncol*, 2008;10(4):631–42.
121. Brennan C, Momota H, Hambardzumyan D, et al., Glioblastoma subclasses can be defined by activity among signal transduction pathways and associated genomic alterations, *PLoS One*, 2009;4(11):e7752.
122. Verhaak RG, Hoadley KA, Purdom E, et al., Integrated genomic analysis identifies clinically relevant subtypes of glioblastoma characterized by abnormalities in PDGFRA, IDH1, EGFR, and NF1, *Cancer Cell*, 2010;17(1): 98–110.
123. Lassman AB, Rossi MR, Raizer JJ, et al., Molecular study of malignant gliomas treated with epidermal growth factor receptor inhibitors: tissue analysis from North American Brain Tumour Consortium Trials 01-03 and 00-01, *Clin Cancer Res*, 2005;11(21):7841–50.
124. Kubicek GJ, Werner-Wasik M, Machtay M, et al., Phase I Trial Using Proteasome Inhibitor Bortezomib and Concurrent Temozolomide and Radiotherapy for Central Nervous System Malignancies, *Int J Radiat Oncol Biol Phys*, 2009;74(2):433–9.
125. Phuphanich S, Supko J, Carson KA, et al., Phase I trial of bortezomib in adults with recurrent malignant glioma, *J Clin Oncol*, 2006;24(18S):1567.
126. Kubicek GJ, Werner-Wasik M, Machtay M, et al., Phase I trial using proteasome inhibitor bortezomib and concurrent temozolomide and radiotherapy for central nervous system malignancies, *Int J Radiat Oncol Biol Phys*, 2009;74(2):433–9.
127. Sauvageot CM, Weatherbee JL, Kesari S, et al., Efficacy of the HSP90 inhibitor 17-AAG in human glioma cell lines and tumourigenic glioma stem cells, *Neuro Oncol*, 2009;11(2):109–21.
128. Garcia-Morales P, Carrasco-Garcia E, Ruiz-Rico P, et al., Inhibition of Hsp90 function by ansamycins causes downregulation of cdc2 and cdc25c and G(2)/M arrest in glioblastoma cell lines, *Oncogene*, 2007;26(51): 7185–93.
129. Hau P, Bogdahn U, Olyushin V, et al., Results of a phase IIb study in recurrent or refractory glioblastoma patients with the TGF-beta-2 inhibitor AP 12009, *J Clin Oncol*, (Meeting Abstracts), 2007;25:12521.
130. Fulci G, Chiocca EA, The status of gene therapy for brain tumours, *Expert Opin Biol Ther*, 2007;7(2):197–208.
131. Mamelak A, Rosenfeld S, Bucholz R, et al., Phase I single-dose study of intracavitary-administered iodine-131-TM-601 in adults with recurrent high-grade glioma, *J Clin Oncol*, 2006;24(22):3644–50.
132. Ferguson S, Lesniak MS. Convection enhanced drug delivery of novel therapeutic agents to malignant brain tumours, *Curr Drug Deliv*, 2007;4(2):169–80.
133. Celtic Pharma terminates TransMID trial KSB311R/CIII/001. Press release: Celtic Pharma, 2 February 2009.
134. Germano IM, Binello E, Gene therapy as an adjuvant treatment for malignant gliomas: from bench to bedside, *J Neurooncol*, 2009;93(1):79–87.
135. Nemunaitis J, Jahan T, Ross H, et al., Phase 1/2 trial of autologous tumour mixed with an allogeneic GVAX vaccine in advanced-stage non-small-cell lung cancer, *Cancer Gene Ther*, 2006;13(6):555–62.
136. Parsa A CC, Butkowski N, et al., Autologous heat shock protein vaccine for recurrent glioma:updated results of a phase I clinical trial, *Neuro Oncol*, (Meeting Abstracts), 2008;10(5):841.