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REVIEW

New Directions in Anti-Angiogenic Therapy for Glioblastoma

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Abstract Anti-angiogenic therapy has become an important component in the treatment of many solid tumors given the importance of adequate blood supply for tumor growth and metastasis. Despite promising preclinical data and early clinical trials, anti-angiogenic agents have failed to show a survival benefit in randomized controlled trials of patients with glioblastoma. In particular, agents targeting vascular endothelial growth factor (VEGF) appear to prolong progression free survival, possibly improve quality of life, and decrease steroid usage, yet the trials to date have demonstrated no extension of overall survival. In order to improve duration of response and convey a survival benefit, additional research is still needed to explore alternative pro-angiogenic pathways, mechanisms of resistance, combination strategies, and biomarkers to predict therapeutic response.

Keywords Angiogenesis · Glioblastoma · Glioma · Bevacizumab · VEGF

Introduction

Angiogenesis is a hallmark of glioblastoma and remains an important therapeutic target in its treatment. Despite a multimodality approach consisting of surgery in addition to radiation therapy with concurrent and adjuvant temozolomide, the prognosis for glioblastoma remains poor with a median survival of 14−16 months [1, 2]. Because glioblastomas are histologically characterized by microvascular proliferation and express high levels of pro-angiogenic factors such as vascular endothelial growth factor (VEGF), agents that target the VEGF-pathway have been investigated in hopes of expanding currently limited treatment options (Table 1).

Bevacizumab is a recombinant, humanized, monoclonal antibody targeting the VEGF ligand A that was first approved for the treatment of metastatic colorectal cancer by the United States Food and Drug Administration (FDA) in 2004. It was subsequently approved for the treatment of non-small cell lung cancer in 2006, followed by renal cell carcinoma and recurrent glioblastoma in 2009 [12–15]. Despite its approval, multiple large phase 3 clinical trials have since failed to show survival benefit in patients with glioblastoma. This review will focus on the rationale behind anti-angiogenic therapy, data regarding the use of bevacizumab in newly diagnosed and recurrent glioblastoma, mechanisms of resistance to anti-VEGF therapy, and emerging areas of research to define optimal use and maximize clinical benefit.

Angiogenesis

Tumors acquire blood supply through multiple mechanisms of vessel recruitment: (1) angiogenesis, where new vessels sprout from existing vessels; (2) migration and growth of tumor cells around pre-existing vessels through a process known as vessel co-option; (3) intussusception, or dilation and bifurcation of existing vessels; (4) vascular mimicry, whereby tumor cells incorporate into the endothelial lining;

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Table 1 Landmark clinical trials of antiangiogenic agents for glioblastoma

Trial	Phase	Disease type	Patients (n)	Arms	Median PFS (mo)	PFS-6 (%)	Median OS (mo)	Reference
BRAIN	2	rGBM	167	BEV	4.2	42.6	9.2	[3]
				BEV + irinotecan	5.6	50.3	8.7	
NCI	2	rGBM	48	BEV	4.0	29.0	7.8	[4]
BELOB	2	rGBM	153	BEV	3.0	16.0	8.0	[5]
				Lomustine	1.0	13.0	8.0	
				BEV + lomustine	4.0	42.0	12.0	
EORTC 26101	3	rGBM	437	BEV + lomustine	4.2	NR	9.1	[6]
				Lomustine	1.5	NR	8.6	
REGAL	3	rGBM	325	Cediranib	92 days	16.0	8.0	[7]
				Cediranib + lomustine	125 days	35.0	9.4	
				Lomustine + placebo	82 days	25.0	9.8	
Enzastaurin	3	rGBM	266	Enzastaurin	1.5	11.1	6.6	[8]
				Lomustine	1.6	19	7.1	
RTOG 0825	3	nGBM	637	BEV + TMZ/XRT	10.7	NR	15.7	[9]
				TMZ/XRT	7.3	NR	16.1	
AVAGlio	3	nGBM	921	BEV + TMZ/XRT	10.6	NR	16.9	[10]
				TMZ/XRT	6.2	NR	16.8	
GLARIUS	2	nGBM (MGMT unmethylated)	170	BEV + irinotecan/XRT	9.7	71.1	16.6	[11]
				TMZ/XRT	5.9	26.2	17.3	

Abbreviations: *rGBM* recurrent glioblastoma, *nGBM* newly diagnosed glioblastoma, *BEV* bevacizumab, *TMZ* temozolomide, *XRT* radiation therapy, *NR* not reported

(5) recruitment of endothelial progenitor cells; and (6) differentiation of cancer stem-like cells into endothelial cells [16–18]. Nonsprouting mechanisms of vessel recruitment may be important for development of resistance to anti-angiogenic therapy (Fig. 1).

Glioblastoma vasculature is functionally and structurally abnormal, characterized by uneven vessel diameter, permeability, tortuosity, and thickened basement membranes. This leads to hypoxic regions observed histopathologically as pseudopalisading necrosis, another hallmark of glioblastoma. Hypoxia and angiogenesis are intricately tied to tumor growth and invasion. Hypoxia results in upregulation of hypoxia-inducible factor-1α (HIF-1α), which subsequently leads to upregulation of VEGF [20]. It also promotes cancer cell invasion, genetic instability, stem-like phenotype, epithelial to mesenchymal transition, altered metabolism, and creation of an immunosuppressive environment [21]. In addition to VEGF, other pro-angiogenic factors upregulated in glioblastomas include hepatocyte growth factor (HGF), fibroblast growth factor (FGF), platelet-derived growth factor (PDGF), angiopoietins, and interleukin-8 [22–25]. Angiogenesis is also constitutively activated through non-hypoxia dependent pathways such as Ras/mitogen-activated protein kinase (MAPK) and phosphoinositide-3-kinase (PI3K) [26]. The many contributing pathways involved in angiogenesis create multiple opportunities for therapeutic targeting.

Targeting Angiogenesis and VEGF

Anti-angiogenic strategies have primarily focused on VEGF signaling by using antibodies to bind VEGF, blocking VEGF receptor (VEGFR) activation via small molecule tyrosine kinase inhibitors (TKIs), and directly blocking VEGF binding via engineered peptides or monoclonal antibodies. Many TKIs also have activity against other kinases involved in signaling in endothelial cells and pericytes, such as PDGFR, FGFR, EGFR, KIT, RAF, and RET, and even some types of cancer cells.

Early trials on anti-angiogenic agents in glioblastoma included thalidomide, a weak inhibitor of FGF and VEGF-mediated angiogenesis. It demonstrated modest activity when used alone or in combination with carmustine for recurrent glioblastoma, however there was no benefit observed when it was combined with the DNA alkylating agent temozolomide and radiation in the newly diagnosed setting [27–31]. Other potential angiogenesis inhibitors studied with no clear benefit included agents such as the thalidomide analogue

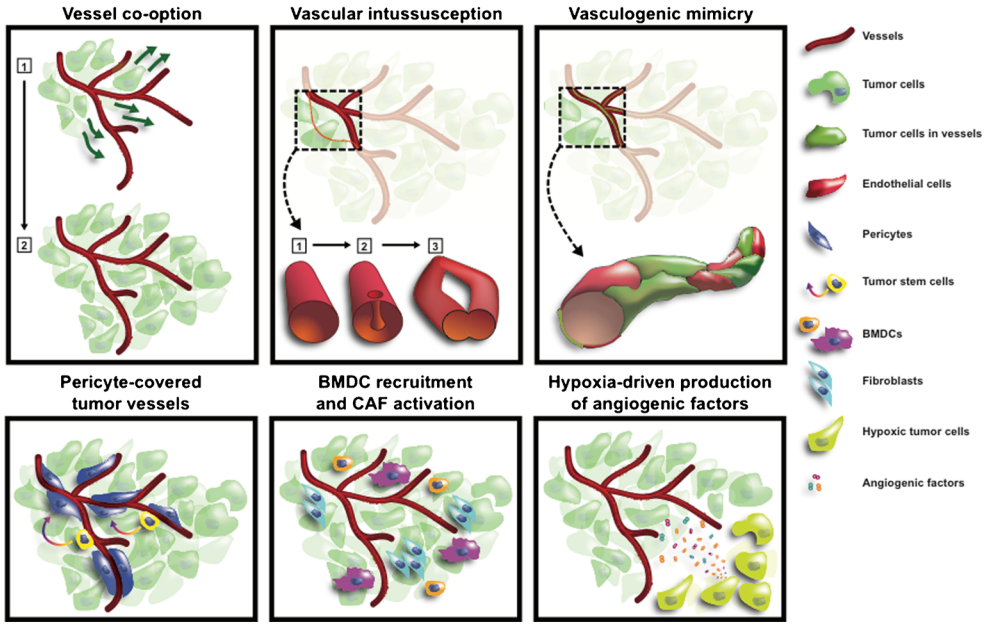

Fig. 1 Mechanisms of angiogenesis and resistance to anti-VEGF therapy. Resistance to anti-VEGF therapy is thought to occur through the following mechanisms: (1) Vessel co-option: tumor cell migration and growth along native vasculature without new blood vessel formation. (2) Vascular intussusception: enlargement and bifurcation of existing vessels. (3) Vascular mimicry: incorporation of tumor cells into the endothelial lining, possibly through endothelial differentiation of tumor stem cells. (4) Peri-cyte covered vessels may be inherently more resistant to VEGF signaling. (5) Recruitment of bone marrow-derived cells and cancer associated fibroblasts which provide paracrine support. (6) Hypoxia-driven release of alternate angiogenic factors. Reprinted with permission from Lu-Emerson et al, J Clin Oncol 2015 [19]

lenalidomide, carboxyamidrotriazole, and penicillamine [32–35].

Recent clinical trials have focused on more potent inhibitors of angiogenesis, particularly bevacizumab, a humanized monoclonal antibody that binds VEGF-A with high affinity and specificity. Bevacizumab inhibits angiogenesis and tumor growth in preclinical models of glioblastoma [36–39]. The initial proposed mechanism of action is through decreased tumor perfusion, thereby depriving the tumor of nutrients and oxygen [40]. However, more recent studies have suggested that in the initial stages of treatment and at low doses, anti-angiogenic agents such as bevacizumab normalize tumor blood vessels, thereby improving vessel function and reducing tumor-associated edema [21]. Figure 2 demonstrates the frequently observed partial response with decrease in T2 hyperintensity and enhancement seen after bevacizumab initiation. Unfortunately, this usually transient phenomenon, known as pseudoresponse, does not confer a survival benefit [41, 42]. Increased perfusion is also observed in a subset of glioblastoma patients after bevacizumab and may sensitize the tumor to radiation and chemotherapy [43, 44].

Numerous receptor tyrosine kinase inhibitors of VEGF and other pro-angiogenic pathways have been tested in clinical trials. However, with the exception of cediranib and enzastaurin, an oral serine/threonine kinase inhibitor, none have progressed beyond phase 2 clinical trials. Despite data showing promising radiographic response rates and progression free survival (PFS) at 6 months, a phase 3 trial

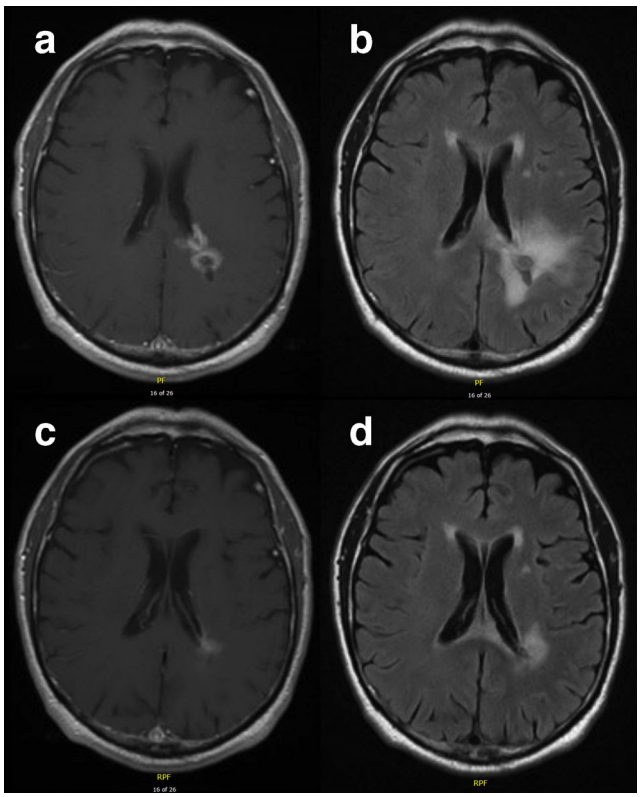

Fig. 2 Radiographic response to bevacizumab. Magnetic resonance imaging of a patient with left parietal glioblastoma before (*A, B*) and after (*C, D*) bevacizumab. There is decreased enhancement (*A, C*) and decreased T2/FLAIR hyperintensity (*B, D*) two months after initiating treatment

demonstrated no difference in PFS or overall survival (OS) in recurrent glioblastoma patients treated with cediranib monotherapy or cediranib with lomustine, an alkylating nitrosourea, versus lomustine alone [45,]. Enzastaurin, which targets the protein kinase C and PI3K/AKT pathways, showed an objective radiographic response rate of 22% when combined with bevacizumab in a phase 2 study in patients with recurrent glioblastoma [46]. However, a randomized, phase 3 trial of enzastaurin in recurrent glioblastoma showed no improvement in PFS and OS compared to lomustine [8].

In addition to VEGF, other proangiogenic targets include integrins [47–50], transforming growth factor beta (TGF-β) [51, 52], and matrix metalloproteinases (MMPs) [53–55]. MMPs are important for vascular remodeling, recruitment of bone marrow derived cells, and release of cytokines such as integrin [56, 57]. Integrins mediate cell adhesion, proliferation, migration, and invasion and also play a role in activation of TGF-β, which additionally is critical for tumor growth, invasion, and immune regulation [58]. The angiopoietin-2 (Ang-2)/TIE2 pathway has been targeted in systemic malignancies and preclinical models of glioblastoma given its interactions with VEGF and role in vessel stabilization as well as recruitment and reprograming of tumor-associated macrophages [59]. Other pathways under investigation include Delta-like ligand 4 (DLL4)/Notch, important for angiogenic sprouting [60, 61]; HGF/c-Met, important for tumor growth and angiogenesis [62, 63]; and Wnt/β -catenin, thought to promote glioblastoma stem cells, cell invasion and migration, and treatment resistance [64].

As bevacizumab has been the best-studied anti-angiogenic agent with the most promising results, the following sections will focus on the data regarding its use for recurrent and newly diagnosed glioblastoma.

Recurrent Glioblastoma

Two prospective, phase 2 studies led to accelerated FDA approval of bevacizumab as monotherapy for recurrent glioblastoma in 2009. The BRAIN study, completed in 2007, compared bevacizumab to bevacizumab plus irinotecan, an inhibitor of topoisomerase I. Radiographic response was assessed by the WHO Response Evaluation Criteria, which is based on the longest unidimensional measurement of a target enhancing lesion [65], although non-enhancing lesions and steroid dosage were also considered in the study. The overall response rates (ORR) were 28.2% and 37.8% with PFS-6 of 42.6% and 50.3%, respectively [66]. However, the trial was not designed as a superiority trial and allowed for crossover from single agent bevacizumab to the combination arm, potentially confounding the results. A second, single-arm study evaluated 48 recurrent glioblastoma patients treated with bevacizumab at the National Cancer Institute (NCI) and found an ORR of 35% and PFS-6 of 29% [67]. Both the Levin and the MacDonald criteria were used, with the Levin criteria qualitatively assessing enhancement, edema, and mass effect, and the MacDonald criteria focusing on objective bidimensional measurements of an enhancing target lesion [66, 67]. While the FDA approved the use of bevacizumab monotherapy in recurrent GBM based on these two trials, the European Medicines Agency declined approval due to the lack of a non-bevacizumab control arm, modest improvement in OS, and challenges with radiographic response assessment [68].

Multiple studies have also evaluated the use of bevacizumab in combination regimens. The BELOB trial was a randomized phase 2 study of 148 patients with recurrent glioblastoma randomized to lomustine, bevacizumab, or both. Combination therapy resulted in a PFS-6 of 41% compared to 11% and 18% with OS at 9 months of 59% compared to 43% and 38% for lomustine and bevacizumab alone, respectively [69]. Based on these results, a phase 3 study (EORTC 26101) was conducted to compare lomustine versus lomustine plus bevacizumab. There was no significant difference in OS for combination treatment versus lomustine alone, although median PFS was increased from 1.5 to 4.2 months for combination therapy [70].

Additional phase 2 trials have evaluated bevacizumab in combination with irinotecan, cetuximab, carboplatin, etoposide, fotemustine, sorafenib, temozolomide, erlotinib, panobinostat, and temsirolimus [66, 69–88]. There have also been trials evaluating bevacizumab and re-irradiation [89–91]. Unfortunately, none of these trials have demonstrated outcomes superior to historical controls treated with bevacizumab alone.

Newly Diagnosed Glioblastoma

Several early, single-arm phase 2 studies evaluating the use of bevacizumab with temozolomide and radiation showed near doubling of median PFS to 13−14 months compared to historical controls. However, only a modest improvement in median OS to 10−21 months was observed [92–94].

Two randomized, placebo-controlled, phase 3 trials investigated the addition of bevacizumab to standard temozolomide plus radiation (chemoradiation) in patients with newly diagnosed glioblastoma. The AVAglio study compared patients randomized to bevacizumab versus placebo in combination with standard chemoradiation. The PFS was significantly prolonged at 10.6 months in the bevacizumab group compared to 6.2 months in the standard therapy group [95]. The RTOG 0825 study also compared bevacizumab to placebo in combination with standard chemoradiation and demonstrated an improvement in PFS to 10.7 months versus 7.3 months with placebo, although this did not meet the predefined significance level of $P = 0.004$ [96]. Unfortunately, both studies failed to demonstrate a benefit in OS. Both studies also had crossover rates of 30 to 50%. Thus, the true impact on OS may

have been obscured by a large number of patients in the placebo arm who subsequently were treated with bevacizumab at the time of disease progression.

The AVAglio and RTOG 0825 studies also differed in significant ways. While AVAglio used the revised Response Assessment in Neuro-oncology (RANO) criteria to assess disease progression, RTOG 0825 used the traditional Macdonald criteria [67, 95, 96]. Unlike the RANO criteria, the MacDonald criteria do not account for non-enhancing tumor volume, which may be important given the alteration of contrast permeability with bevacizumab. Both trials also attempted to assess other measures of clinical benefit such as performance status, corticosteroid requirement, and quality of life measures. Interestingly, the studies had some divergent findings. While the AVAglio trial showed that bevacizumab prolonged maintenance of performance status, decreased steroid utilization, and prolonged time to deterioration in pre-specified cognitive domains, RTOG 0825 found that bevacizumab led to worsened cognitive function. The cause of the differences is unclear, but possible explanations include different radiographic response criteria, substantial dropout in the RTOG trial, and differences in statistical modeling.

Combination therapies with bevacizumab in newly diagnosed glioblastoma have also been assessed. The randomized, phase 2 GLAIRUS study compared standard of care chemoradiation with temozolomide versus radiation with bevacizumab and irinotecan in patients whose tumors expressed the DNA repair enzyme O6-methyl guanine DNA methyltransferase (MGMT). Loss of MGMT function through gene promoter methylation has been shown to confer increased sensitivity to therapy with temozolomide in glioblastoma [97]. The GLARIUS trial found that PFS was significantly prolonged at 9.7 months in the bevacizumab plus irinotecan arm compared to 5.99 months in the control arm. However, the OS did not significantly differ with OS of 17.5 months in the control arm compared to 16.6 months in the experimental arm. Neither therapy regimen was superior in delaying the time to deterioration in pre-specified dimensions of quality of life [11].

Mechanisms of Resistance

Despite impressive radiographic responses and improved PFS, response to anti-angiogenic therapy is unfortunately not durable. Alternative mechanisms of vessel recruitment are ultimately utilized (Fig. 1). Local hypoxia may trigger alternative pro-angiogenic factors such as HGF, FGF, Ang-2, SDF1α, and interleukin-8 [98–102]. Preclinical studies have shown that dual targeting of VEGF and Ang-2 may overcome this form of resistance to anti-VEGF monotherapy [103–105]. Vessel co-option, the process whereby tumors utilize native brain vessels to recruit blood supply, is also under investigation as an escape mechanism to anti-angiogenic therapy [21]. The molecular mechanisms of vessel co-option are poorly understood and may yield novel therapeutic approaches once the pathways are identified.

Some tumor vessel subtypes are thought to have inherent insensitivity to VEGF inhibition due to decreased sensitivity of pericytes [106, 107]. There is also preclinical data that anti-angiogenic therapy induces transformation from a proneural to a more invasive mesenchymal phenotype, including upregulation and increased phosphorylation of the receptor tyrosine kinase c-Met [108–110]. Increased invasion may also be mediated by MMPs [111]. Retrospective data in patients found that treatment with bevacizumab was associated with non-enhancing, diffuse or distant recurrence (Fig. 3) [70].

Given the potential disease progression via non-enhancing, infiltrative or invasive disease with anti-angiogenic therapy, combination therapy with an agent that targets invasion may also be a promising strategy to overcome resistance. For example, inhibition and knockdown of c-Met inhibit tumor growth and prolong survival in GBM mouse models [108–110]. Interim analysis from a completed phase 2 trial of the monovalent MET inhibitor onartuzumab or placebo with bevacizumab showed no difference in PFS, OS, or

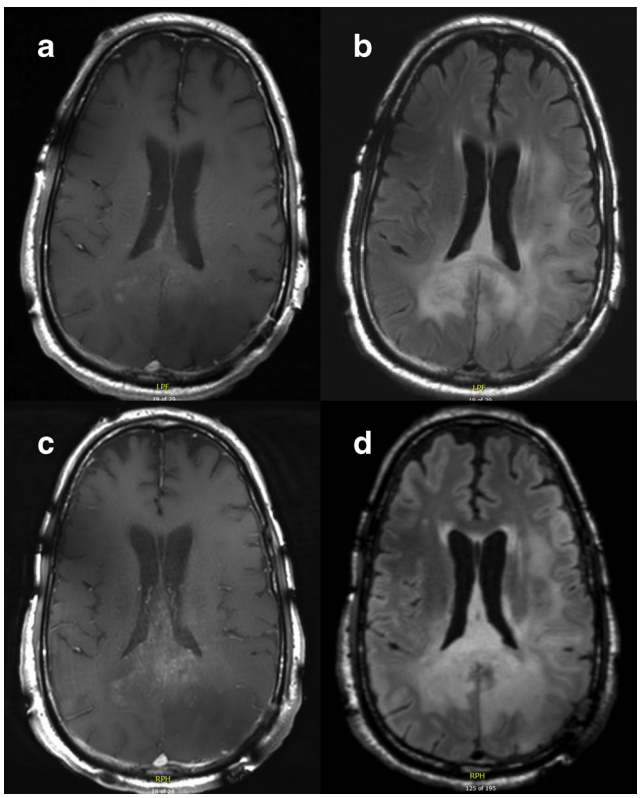

Fig 3 Disease progression on bevacizumab. Magnetic resonance imaging from a patient with glioblastoma who progressed on bevacizumab. *Panels A* and *B* show T1 post-contrast and T2/FLAIR sequences, respectively, of a patient with glioblastoma involving the splenium of the corpus callosum. *Panels C* and *D* show post-contrast and T2/FLAIR sequences after one month with notable increase in patchy enhancement and corresponding increase in T2 hyperintensity

ORR, however a phase 1 trial of another c-Met inhibitor INC280 is ongoing (NCT02386826).

Anti-angiogenesis and the Immune System

Angiogenesis plays an important role in tumor immunity, and the immune composition of the tumor microenvironment changes with vascular normalization and decreased local hypoxia. Preclinical data in extracranial tumors suggest that anti-angiogenic therapies increase tumor delivery of activated T cells, rendering the tumor more susceptible to immune attack [112]. Anti-angiogenic therapy also facilitates the recruitment of bone marrow-derived cells and polarization of tumor associated macrophages to immune stimulatory M1-skewed macrophages (Fig. 4) [20, 113, 114]. Furthermore, increased circulating levels of VEGF inhibit T cell immune response by suppressing maturation of dendritic cell precursors and promoting proliferation of regulatory T cells [112]. However, studies have also suggested that immune activation may play a role in resistance to anti-angiogenic therapy. Peripheral mobilization of myeloid cells via granulocyte colony stimulating factor (G-CSF) has been associated with refractoriness to anti-angiogenic therapy [114, 115]. Pro-angiogenic M2-polarized macrophages may also be important, and strategies to reprogram or inhibit the M2 phenotype, such as through inhibition of Ang-2 or proinflammatory cytokines, have been shown to augment anti-angiogenic therapy in animal models [103, 116, 117].

Despite the conventional notion that the central nervous system is immune-privileged due to the blood brain barrier, the immunomodulatory effect of anti-VEGF has led to recent clinical trials combining bevacizumab with immunotherapy in hopes of a synergistic effect in facilitating anti-tumor immunity. Preliminary results from a phase 2 trial of standard of care chemoradiation versus standard of care plus the dendritic cell vaccine AV0113 in patients with newly diagnosed glioblastoma showed no difference in PFS or OS; however, in the subgroup of 22 patients that received the vaccine as second-line therapy with bevacizumab, there was an improvement in OS compared to the control arm (535 ± 155 days versus 406 ± 224 days) [118]. Preliminary results from the phase 2 study of patients with EGFRvIII mutant recurrent glioblastomas (approximately 20–30% of all primary glioblastomas) demonstrate that the combination of rindopepimut, a peptide vaccine against EGFRvIII, with bevacizumab prolonged median OS from 8.8 months in the control arm (bevacizumab plus keyhole limpet hemocyanin) to 12 months in the experimental arm [54]. PFS-6 was also significantly increased from 11% to 27%. However, the phase 3 ACT IV trial of standard of care with temozolomide versus temozolomide plus rindopepimut was recently stopped after interim analysis showed no difference in median OS (20.4 months in the rindopepimut group versus 21.1 months in the control group, HR 0.99) [119]. The question remains whether rindopepimut may have a more robust effect on survival when combined with an anti-angiogenic therapy such as bevacizumab. In addition to vaccine strategies, adoptive cell transfer (ACT) combined with anti-angiogenic therapy in a mouse model of B16 melanoma showed a synergistic effect with increased infiltration of transferred cells and prolonged survival compared to ACT alone [120].

Ongoing clinical trials for glioblastoma patients are evaluating the use of anti-angiogenic therapy in combination with immune checkpoint inhibitors (pembrolizumab NCT02337491 and durvalumab NCT02336165) and vaccines (SL-701 NCT02078648 and heat shock protein peptide complexes NCT01814813).

Searching for Biomarkers

Unlike other targeted therapies, no established biomarkers currently exist to help predict response to anti-angiogenic

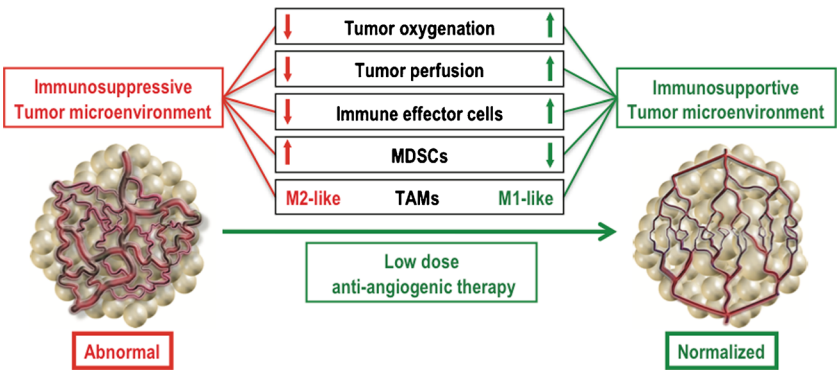

Fig. 4 Anti-angiogenic therapy and the immune microenvironment. Abnormal tumor vasculature creates a hypoxic tumor microenvironment, thereby decreasing tumor oxygenation, tumor perfusion, reducing the number of immune effector cells in the microenvironment, increasing myeloid-derived stem cells, and polarizing tumor associated macrophages to the immune inhibitory M2-like phenotype. Low dose anti-VEGF therapy is thought to normalize blood vessels and have the opposite effect. Reprinted with permission from Huang et al, Cancer Res [112]

therapy. Tissue, serologic, and imaging markers are all under active investigation, although none have yet been validated for use in clinical practice.

Tumor Tissue

Tumor tissue biomarkers have been evaluated mostly in the setting of bevacizumab. In the RTOG-0825 randomized trial, a 10-gene panel assessed the degree of mesenchymal gene enrichment and was significantly associated with worse PFS and OS in response to bevacizumab [121]. Other tissue markers include levels of VEGF, carbonic anhydrase 9 (a marker of hypoxia), and the number of CD68+, CD11+ tumor-associated macrophages [122, 123]. Markers that have not been found to predict response include MGMT methylation status, IDH1, EGFR, PDGFR-α, and c-KIT [124–126].

Blood Biomarkers

Circulating blood biomarkers are particularly important for glioblastomas as repeat surgery for tissue sampling is often not feasible. Candidate biomarkers include VEGF and sVEGFR2, SDF-1α, PIGF, and MMPs [95, 113, 127–129]. Some studies have shown associations between changes in biomarker levels and outcomes. For example, elevated levels of MMP-9, sVEGFR-1 (a negative regulator of VEGF), and SDF-1α have been associated with decreased survival in patients receiving anti-angiogenic agents such as cediranib and aflibercept [45, 130]. On the other hand, increased collagen IV levels have been associated with improved PFS in recurrent glioblastoma [131]. The identification of validated serum biomarkers will aid in drug development as well as noninvasive monitoring and treatment selection in patients.

Imaging Biomarkers

In 2010, the Response Assessment in Neuro-Oncology Working Group proposed the RANO criteria to account for rapid reduction of contrast enhancement after anti-angiogenic therapy as well as transient increase in tumor enhancement shortly after chemoradiation (pseudoprogression) [95]. The criteria include the assessment of non-enhancing lesions and also offers guidelines for distinguishing pseudoprogression from progression. Quantifiable radiographic changes after treatment have led to investigation of various imaging markers to help predict tumor response, including apparent diffusion coefficient [132], restriction spectrum imaging [133], dynamic contrast enhanced (DCE) and dynamic susceptibility-contrast (DSC) techniques [134, 135], vessel architectural imaging [136], and dopamine and positron emission tomography [137, 138]. Consistent with the vascular normalization hypothesis, data suggest that improved perfusion is correlated with increased OS in both newly-diagnosed and recurrent glioblastoma patients treated with the pan-VEGF inhibitor cediranib. In one study, patients with recurrent glioblastoma treated with cediranib, an oral pan-VEGF inhibitor, who had sustained increase in perfusion longer than one month had increased OS of 348 days compared to patients with decreased or stable tumor perfusion (213 days and 169 days, respectively, $P < 0.01$) [139]. Another study of cediranib in newly diagnosed glioblastoma patients found that patients with durably increased perfusion had a mean OS of 26.3 months compared to 17 months in those with stable or decreased perfusion ($P < 0.05$) [126]. Thus, early imaging changes in response to anti-angiogenic therapy may help identify patients more likely to benefit from anti-VEGF therapy.

Future Directions

While bevacizumab continues to be a mainstay in the treatment of recurrent glioblastoma, the lack of survival benefit in clinical trials has prompted investigation of biomarkers that can help determine the optimum patient population and predict radiographic response. Although the RANO criteria does account for nonenhancing disease and decreased enhancement after anti-angiogenic therapy, more research is needed to distinguish treatment effect from disease progression and further clarify radiographic response patterns in the setting of altered vascular permeability.

The lack of survival benefit despite increase in progression free survival also highlights the potential importance of secondary endpoints such as quality of life measures and steroid usage. Given the vital location, slight radiographic progression of a glioblastoma or increased edema can translate into a disproportionate impact on functional status. While clinical trials and new therapies should continue to focus on prolonging survival, there remains a need to better characterize the impact on day-to-day challenges patients face.

Another area of investigation includes optimal dosing and timing of bevacizumab or other anti-angiogenic therapies. Retrospective data suggest that the treatment of patients with high-grade glioma with low doses of bevacizumab (5 mg/kg per week or 7.5 mg/kg every 3−4 weeks) may be superior to standard dosing, potentially due to vascular normalization at lower doses [140, 141]. A recent randomized phase 2 trial comparing low dose bevacizumab plus lomustine to standard dose bevacizumab monotherapy in recurrent GBM showed no difference in the primary endpoint of PFS (4.34 months for the combination arm versus 4.11 months for bevacizumab monotherapy, $p = 0.19$) [142]. However, there was a trend toward improved median PFS in patients with first recurrence. Due to the potential of rebound cerebral edema after discontinuation of bevacizumab, salvage therapy has been difficult, leading many patients and oncologists to delay bevacizumab as long as possible. However, it is unclear to what extent this delay is

warranted as additional retrospective data in systemic malignancies indicate that there may be benefit to continuing anti-angiogenic therapy past progression [143–146].

While optimizing bevacizumab administration is important, the multiple, complex pathways that promote angiogenesis also support the use of combination strategies. Improved orthotopic animal models from patient-derived tumors will play an important role in the evaluation of new therapies, treatment combinations, and resistance mechanisms, although these models will have limited utility in the evaluation of immunotherapies. It is notable that the only randomized clinical trial that appeared to confer some improvement in survival was in patients with recurrent glioblastoma treated with bevacizumab and chemotherapy. Thus, targeting several pathways or combining anti-angiogenic agents with other classes of drugs such as immunotherapy may prevent the development of treatment resistance and maximize survival benefit.

Required Author Forms Disclosure forms provided by the authors are available with the online version of this article.

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