Date Submitted: ***[10/25/17]***

Alert Page

DF/HCC Protocol #: **[17-037]**

Protocol Clarifications (non-drug related e.g. eligibility criteria, study assessments)

Protocol Section 9.2.1.3: At appropriate time points, 50 cc of fresh whole blood will be collected in K2 EDTA 10 ml Blood Collection tubes, delivered immediately after collection at room temperature (no ice) to Dr. Curry's laboratory (3rd Floor Simches Research Building, Room CPZN 3) for isolation and storage of peripheral blood mononuclear cells (PBMC's). Label subject ID, date and time of collection. Contact Leland Richardson (LRICHARDSON7@mgh.harvard.edu) to arrange delivery (or phone 732-600-6514) Monday through Friday 8:00am - 4:00 pm.

NCI Protocol #: Not Applicable

DF/HCC Protocol #: 17-037

TITLE: Phase II clinical trial of bavituximab with radiation and temozolomide for patients with newly diagnosed glioblastoma

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NCI-Supplied Agent(s): NA
Other Agent(s): bavituximab, Oncologie, Inc.

IND #: 134862
IND Sponsor: Elizabeth Gerstner

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SCHEMA

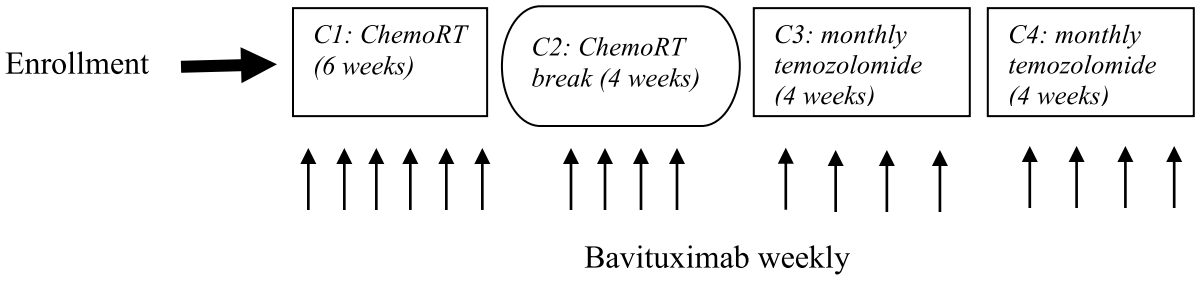
Enrollment
C1: ChemoRT (6 weeks)
C2: ChemoRT break (4 weeks)
C3: monthly temozolomide (4 weeks)
C4: monthly temozolomide (4 weeks)
Bavituximab weekly

TABLE OF CONTENTS

1. OBJECTIVES

1.1 Study Design

This is an open-label, single-arm, phase II study of bavituximab in combination with standard of care radiation and temozolomide in adult patients with newly diagnosed GBM. All patients will be treated with bavituximab weekly until 3 months after completion of chemoradiation or until there is radiographic or clinical evidence of disease progression or unacceptable toxicity.

1.2 Primary Objectives

The primary objective will be to determine the proportion of patients alive at 12 months (OS12).

1.3 Secondary Objectives

Secondary Objectives will include progression free survival and toxicity of bavituximab in this patient population.

2. BACKGROUND

2.1 Study Disease(s)

An estimated 51,410 primary brain tumors were diagnosed in 2007, and 19% of these tumors were glioblastomas (GBM) (CBTRUS 2008). GBM is the most common malignant primary brain tumor and is a uniformly fatal disease with 5-year survival rates less than 4% despite aggressive treatment with surgery, radiation and chemotherapy. Consequently, new therapies for this patient population are desperately needed.

2.2 IND Agent(s)

Oncologie, Inc. (hereinafter referred to as Oncologie, is developing bavituximab as an immunotherapeutic that is being investigated in the treatment of cancer. Bavituximab is a genetically engineered immunoglobulin gamma 1 (IgG1) chimeric (human/mouse) monoclonal antibody containing the variable region sequences of the murine phosphatidylserine (PS)-targeting mouse antibody 3g4 and human IgG1 κ constant region sequences. Bavituximab binds β2-glycoprotein-1 (β2-GP1) to form the complex of β2-GP1 with PS. PS is a highly immunosuppressive molecule typically expressed inside the membrane of normal cells, but "flips" and becomes exposed on the outside of cells that line tumor blood vessels, tumor cells, and exosomes, creating a specific target for anticancer treatments.

Bavituximab is a viable immunotherapeutic approach because its primary mechanism of action involves modulating the tumor microenvironment from a primarily immunosuppressive, angiogenesis-promoting state (with infiltrating myeloid-derived suppressor cells [MDSCs] and m2-macrophages) to an immune-activating state (with long term tumor-specific immunity

facilitated by m1 macrophages, mature dendritic cells, and activated t lymphocytes). This process occurs via fc-mediated signaling and direct cell killing (antibody-dependent cellular cytotoxicity). In essence, expression of PS on the external surface of the cell acts as an immunosuppressive signal in normal cell death and immune phagocytic clearance. Tumor PS exposure in the microenvironment is immunosuppressive in malignant transformation and progression. Thus bavituximab has been shown to overcome immune suppression and stimulate antitumor immunity in nonclinical models (DeRose 2011; Yin 2013).

Nonclinical Experience

Data suggest that PS is primarily responsible for expansion of MDSCs and M2-like tumor-associated macrophages (TAMs) in tumors, and that bavituximab could reverse this process and reactivate antitumor immunity. Treatment of tumor-bearing mice with docetaxel in combination with 2aG4 (a PS-targeting antibody and the murine precursor to bavituximab), has been shown to potently suppress the growth and progression of prostate tumors and increase the presence of M1-like TAMs and mature dendritic cells in the tumor microenvironment (Yin 2013).

The tumor and tumor vasculature localization of PS-targeting antibodies is followed by tumor immunity enhancing effects, tumor vessel damage, tumor vascularity reduction, tumor necrosis, and tumor growth retardation in multiple models (Ran 2005). The synergistic antitumor activity of antibodies targeting PS in combination with cytotoxic chemotherapy has been demonstrated in animal models, including but not limited to the Pan02 pancreatic adenocarcinoma cell line with gemcitabine and the MDA MB 435 cell line with docetaxel (Beck 2006, Huang 2005).

In toxicology studies, no target organs of toxicity were identified in rats (up to 20 mg/kg) and monkeys (up to 10 mg/kg) receiving weekly doses for 8 weeks of bavituximab, and minimal histological findings in heart and lung were noted when administered to monkeys at 100 mg/kg. Detailed information about comprehensive nonclinical safety, immunology, and pharmacokinetics (PK) is provided in the current Investigator's Brochure for bavituximab.

Clinical Experience

Bavituximab has been evaluated by Oncologie in clinical studies in over 800 patients (as of July 2016), most of whom were treated with combination therapy. These clinical trials have included patients with a number of tumor types, including breast, lung, pancreatic and hepatocellular carcinoma, and in chronic HCV. A dose of 3 mg/kg bavituximab given intravenously (IV) was determined and selected for further clinical study based on Phase 1 single-agent and combination therapy studies.

In a Phase II, multicenter, breast cancer trial (PPHM 0702), bavituximab (3 mg/kg), given weekly until progression, was combined with paclitaxel (100 mg/m2) and carboplatin (area under the concentration-time curve = 2), given on Days 1, 8 and 15 of planned 4-week cycles for up to 6 cycles, to 46 patients with metastatic disease, unrestricted by hormone or HER2 status. Objective response per Response Evaluation Criteria in Solid Tumors (RECIST) occurred in 34 of 46 patients (73.9%) with a median duration of response (DOR) of 3.7 months (95%

confidence interval [CI]: 3.1, 5.8) and median PFS of 6.9 months (95% CI: 5.6, 7.7) (Jain 2010). The most common Grade 4 treatment-emergent adverse event (TEAE) was neutropenia (12 patients, 26.1%), and the most common Grade 3 TEAEs were leukopenia (11 patients, 23.9%), neutropenia (9 patients, 19.6%), and anemia (5 patients, 10.9%).

In a Phase II, multicenter, lung cancer trial (PPHM 0704), bavituximab (3 mg/kg), given weekly until progression, was combined with docetaxel (35 mg/m2), given on Days 1, 8, and 15 of planned 4-week cycles for up to 6 cycles, to 46 patients with MBC (with any hormone or HER2 status). Objective response occurred in 28 of 46 patients (60.9%) with median DOR of 6.1 months (95% CI: 5.7, 7.5) and median PFS of 7.4 (95% CI: 6.1, 9.1) months (Tabagari 2010). Of the most common TEAEs reported, only fatigue, headache, back pain, and hypertension were Grade ≥3. In another Phase II, multicenter trial in patients with advanced nonsquamous non–small-cell lung cancer patients were randomized 1:1:1 to docetaxel 75 mg/m 2 every 21 days for up to 6 cycles combined with weekly, blinded infusions of placebo, bavituximab 1 mg/kg, or bavituximab 3 mg/kg until disease progression or unacceptable toxicity (Gerber 2016). Efficacy end points in the bavituximab 3 mg/kg group (n = 41) and in the placebo/bavituximab 1 mg/kg group (n = 80), respectively, were as follows: ORR, 17.1% (95% confidence interval [CI], 5.6%-28.6%) and ORR, 11.3% (95% CI, 4.3%-18.2%); median progression-free survival 4.5 and 3.3 months (hazard ratio [HR], 0.74 [95% CI, 0.45-1.21]; P = .24); median overall survival 11.7 and 7.3 months (HR, 0.66 [95% CI, 0.40-1.10]; P = .11). Toxicities were manageable and similar between arms.

In a single-center, investigator sponsored study, breast cancer patients received bavituximab 3 mg/kg weekly in combination with paclitaxel (80 mg/m2) given on Days 1, 8, and 15 in 4-week cycles to 14 patients with HER2-negative MBC. Treatment resulted in an ORR of 85% (2 patients had complete responses) and a median PFS of 7.3 months (95% CI: 2.8, 10.8) (Chalasani 2015). Bone pain, fatigue, headache, and neutropenia were the most common AEs. Infusion-related reactions were the most common AE related to bavituximab.

A Phase II, single institution study of bavituximab and sorafenib in advanced hepatocellular carcinoma (HCC) was also conducted. Patients received 3 mg/kg IV weekly of bavituximab and 400 mg PO BID of sorafenib until radiologic progression. Secondary endpoints included overall survival, disease specific survival, 4 month progression free survival, safety, and response rate. The study accrued 38 patients. Median OS was 6.2 months. Two patients achieved partial response and four month PFS was 61%. There were no grade 4 or 5 adverse events recorded. Most common all grade events were diarrhea (32%), fatigue (26%), and anorexia (24%) (Yopp 2015). These results demonstrated that bavituximab and sorafenib were well tolerated in patients with advanced HCC.

Overall, results from Phase I and Phase II studies have demonstrated a clinically meaningful treatment effect of bavituximab, and results were consistent among the studies. The overall safety profile of bavituximab was acceptable, and the safety data were consistent with those observed in other clinical studies. Combination therapy did not substantially increase the risks of side effects. Thus the potential additional benefits of combination therapy are likely to outweigh the risk of AEs experienced during bavituximab treatment. Refer to the Investigator's Brochure for additional details.

2.3 *Other Agent(s)*

Temozolomide will be given as per standard of care.

2.4 Rationale

Bavituximab is a chimeric (human/mouse) monoclonal antibody (mAb) derived from murine mAb 3G4 that targets phosphatidylserine (PS) after binding to β2-glycoprotein 1 (β2-GP1). PS is expressed on GBM cells and targeting PS in GBM xenografts models has shown efficacy in killing tumor cells (Blanco 2014; Blanco 2015). In addition to immune activation, targeting PS has also been shown to have anti-angiogenic effects in GBM xenograft models as well as other models – an additional potential mechanism of tumor cell kill in a tumor characterized by neovascularization (Wojton 2013; Ran 2005). PS appears to be very specific for GBM cells so has also been suggested as a diagnostic imaging agent to identify tumor (Blanco 2014; Blanco 2015).

Radiation therapy increases PS expression in GBM cell lines suggesting a strong rationale for combining radiation with a drug like bavituximab that targets PS (Nida 2015). Temozolomide, the chemotherapy used in conjunction with radiation for newly diagnosed GBM, is an apoptosis inducing agent and works synergistically with radiation to induce tumor cell kill. Prior studies using SapC-DOPS, nanovesicles formed by coupling sphingolipid activating protein saposin C (SapC) and dioleoylphosphatidylserine (DOPS), interact with PS and when used in conjunction with temozolomide, there was an increase in tumor cell kill (Wojton 2014). Consequently, the triple punch combination of radiation, temozolomide, and an agent such as bavituximab appears to hold great therapeutic potential in newly diagnosed GBM.

PS is also a highly immunosuppressive membrane phospholipid and acts as an upstream immune checkpoint. In normal non-tumorigenic cells, PS is segregated to the inner leaflet of the plasma membrane but becomes externalized to the outer leaflet of the plasma membrane in cells in the tumor microenvironment (tumor cells, exosomes, and vascular endothelial cells). Phosphatidylserine is recognized and bound by PS receptors on immune cells where it induces and maintains immune suppression. In the absence of a tumor microenvironment and PS-exosome mediated signaling, this process occurs under conditions of normal cell death and immune phagocytic cell clearance (Birge 2016).

Phosphatidylserine-targeting agents block PS-mediated immunosuppression by multifocal reprograming of the immune cells in the tumor microenvironment to support immune activation (Yin 2013). Antibody-mediated PS blockade reduces the levels of myeloid-derived suppressor cells (MDSC), transforming growth factor-beta (TGF-β), and interleukin (IL) -10, and increases the levels of tumor necrosis factor-alpha (TNF-α) and IL-12. Phosphatidylserine blockade also repolarizes tumor-associated macrophages (TAMs) from predominant M2 to predominant M1 phenotype, promotes the maturation of dendritic cells (DCs), and induces potent adaptive antitumor T-cell immunity (Burrows 1994, Denekamp 1993).

Experimental studies have shown that bavituximab binds to PS in the presence of β2-GP1 as a high affinity complex, modulating β2-GP1 binding to PS from 1 μM to 1 nM. In experimental cancer models, bavituximab treatment reduced tumor growth and prolonged survival, where the anti-tumor effects are enhanced by co-administration of chemotherapy or radiation (conditions that increase expression of PS).

Thus, blocking PS with bavituximab may reverse the tumor immunosuppressive environment (Yin 2013). In GBM, tumor associated macrophages (TAMs) are the predominate infiltrating immune cell and most of these are M2 cells with an anti-inflammatory phenotype (Li 2012). PS appears to play a role in maintaining this M2 phenotype in cancer and ultimately promoting T cell tolerance (Yin 2013). Blocking PS may shift the immunosuppressive phenotype driven by TAMs in GBM to a more active immunesurveillance phenotype. Furthermore, combining antiphosphatidylserine antibody with radiation induces GBM tumor immunity (He 2009). Therefore, targeting PS and reversing immunesuppression in newly diagnosed GBM with bavituximab is a novel therapeutic avenue for this devastating disease. Our clinical trial will include correlative studies that will evaluate the immune system, vascular biology and tumor microenvironment to understand the impact bavituximab is having in GBM patients.

In sum, these data support evaluation of bavituximab in GBM because:
1. PS is highly expressed on GBM tumor endothelial cells and tumor cells (Blanco 2014; Blanco 2015; Nida 2015)
2. PS is very specific for tumor as evidenced by interest in using PS as a target in diagnostic imaging as well as a target for local delivery of chemotherapy (Zhang 2014; Blanco 2014; Blanco 2015; Winter 2015).
3. Targeting PS can have anti-angiogenic properties in GBM which is attractive in a tumor characterized by vascular proliferation (Wojton 2013).
4. Targeting PS has shown tumor cell kill in GBM models (Blanco 2014; Blanco 2015; Wojton 2013; Woljton 2014).
5. There is synergism between targeting PS and temozolomide in GBM (Wojton 2014).
6. There is synergism between targeting PS and radiation in GBM xenograft models (He 2009; Nida 2015).
7. Targeting PS can relieve tumor immunosuppression and GBM is a cancer characterized by immunosuppression (Yin 2013; He 2009)
8. Bavituximab has been testing with other cytotoxic chemotherapies without increase in toxicity (see Section 2.2)

Unfortunately, there are limited options for GBM with no new drugs FDA approved since temozolomide was approved in 2005 so we desperately need better therapies. Patients in this trial will receive current standard of care treatment so do not miss out on the therapeutic benefit of standard of care but will also receive bavituximab in the hopes of augmenting the limited response to standard of care.

2.5 Correlative Studies Background

Hypothesis

GBM is considered a relatively low mutational load tumor so we hypothesis that radiation will increase antigen presentation and improve outcomes when combined with bavituximab in newly diagnosed GBM. The underlying mechanisms for this improved response can be measured using blood and MRI markers of immune status, vascular biology, and the tumor microenvironment.

Based on this hypothesis, we propose evaluating the following correlative markers:

1. Correlate baseline and change in circulating immune cell biomarkers and cells within the resected tumors (i.e. from blood as well as histological samples) with radiographic response (RR), progression free survival (PFS), and overall survival (OS).
2. Determine if changes in MRI parameters (specifically changes in perfusion, vascular permeability, or tissue cellularity/metabolism) will shed light on the mechanism of action of bavituximab in GBM.
3. Determine if changes in MRI parameters (specifically changes in perfusion, vascular permeability, or tissue cellularity/metabolism) will help distinguish responders from nonresponders early in the course of bavituximab therapy.
4. Determine if changes in MRI parameters (specifically changes in perfusion, vascular permeability, or tissue cellularity/metabolism) will help distinguish a positive inflammatory response from active tumor.
5. Determine changes in PS exosomes and serum B2GP1 during treatment with bavituximab.

Blood will be analyzed for circulating immune cells to help distinguish inflammation from active tumor by tracking T cell receptor (TCR) repertoire and its correlation with TCRs that expand in the tumor. In addition, myeloid cells will be analyzed to detect changes with therapy.

Tumor tissue will be analyzed for myeloid cells and T cells using single cell RNA-seq (fresh tissue) or population RNA-seq (fixed tissue), which will provide information about changes in myeloid cells with treatment as well as TCRs present in infiltrating T cells. If patient requires further surgery, tissue will be analyzed again for changes in myeloid cells and TCRs with therapy. This will allow for pre- and post-therapy comparison.

Preliminary data

MRI

We have conducted several clinical trials that incorporate advanced imaging to shed light on tumor response to treatment (Batchelor 2013, Gerstner 2016, Gerstner 2015). Most relevant to this proposal, we have looked at MRI changes in newly diagnosed GBM patients receiving standard radiation and temozolomide with or without cediranib, a tyrosine kinase inhibitor of VEGF receptors (Batchelor 2015). These data demonstrate our ability to analyze complex imaging datasets but, more importantly shed light on how anti-angiogenic therapy might be helping during chemoradiation. We found that increased tumor perfusion was associated with improved survival with cediranib therapy suggesting a vascular normalization effect in a subset of patients leading to improved delivery of chemotherapy and oxygen needed for radiation to be effective. In our patients treated with standard chemoradiation, we did not see this change in

perfusion. Therefore, our MRI techniques were able to shed light on the physiological impact the drug was having on GBM. Both of these datasets will be available to compare to data derived from this proposal so we will be able to determine how immunotherapy impacts tumor differently from standard therapy or anti-angiogenic therapy. Furthermore, there is a complex interaction between tumor vasculature and the immune system and our MRI, which captures vessel size and permeability as well as perfusion and blood volume, will be able to shed light on this complexity by measuring structural and functional changes in tumor vasculature longitudinally.

Vessel architectural imaging (VAI), derived from the MR perfusion acquisition, can distinguish between responders and non-responders treated with immunotherapy by measuring changes in tissue oxygenation status (SO2). Patients with metastatic melanoma to the brain treated with stereotactic radiosurgery +/- ipilumimab experienced changes in vascular structure and function. Shorter surviving patients experienced worsening in SO2 relative to healthy tissue suggesting that the immunotherapy was not effectively modulating vasculature or ultimately killing the tumor itself (Digenes 2016). Hypoxia is a negative prognostic marker in cancer so a decrease in the tissue oxygenation status (i.e. decrease in SO2) suggests that the tumor is actively growing – a fact supported by the shorter survival in these patients. The improvement in SO2 suggests vascular remodeling in response to therapy. These findings are similar to our experience in glioblastoma where a decrease in SO2 compared to healthy tissue was associated with shorter survival. We will include monitoring of tissue oxygenation, perfusion, and vascular permeability in this clinical trial to help assess tumor response.

Immunomonitoring

The Hacohen lab has analyzed myeloid cells in both blood and tumor tissue in depth using flow cytometry and single cell RNA sequencing (scRNA-seq). For example, they can identify 6 types of dendritic cells and 4 types of monocytes using scRNA-seq, allowing an unbiased analysis of the myeloid compartment. In addition, both myeloid and T cells can be seen by scRNA-seq of tumors pre- and post immune therapy. They have also analyzed TCR repertoire using scRNA-seq and identified enriched clones expressing a specific pair of TCRα/β□ in patient tumors. For FFPE samples, the Hacohen lab has published methods to analyze the T cell responses using bulk RNA-seq (Rooney 2015).

3. PARTICIPANT SELECTION

3.1 Eligibility Criteria

3.1.1 Participants must have histologically confirmed newly diagnosed glioblastoma or glioblastoma variant (ex. gliosarcoma), including documentation of unmutated isocitrate dehydrogenase (IDH) by immunohistochemistry or sequencing.

3.1.2 No prior immunotherapy allowed or prior alkylating agents or prior radiation to the brain.

3.1.3 Age >17 years since adult GBM is biologically different from pediatric GBM and there is no data for bavituximab in pediatric populations.

3.1.4 Karnofsky ≥60%, see Appendix A

3.1.5 Life expectancy of greater than 6 months.

3.1.6 Participants must have normal organ and marrow function as defined below:

– leukocytes	≥3,000/mcL
– absolute neutrophil count	≥1,500/mcL
– platelets	≥100,000/mcL
– total bilirubin	within normal institutional limits (unless patient has Gilbert's syndrome in which total bilirubin should be ≤ 2xULN)
– AST(SGOT)/ALT(SGPT)	≤2.5 × institutional upper limit of normal
– creatinine	within normal institutional limits OR
– creatinine clearance	≥60 mL/min/1.73 m^2 for participants with creatinine levels above institutional normal(using Cockcroft Gault Formula)
– negative serum pregnancy test in WOCBP	
– INR/PT	- ≤1.5 x institutional ULN unless subject is receiving anticoagulant therapy as long as PT or INR is within therapeutic range of intended use of anticoagulants
– aPTT	≤1.5 x institutional ULN unless subject is receiving anticoagulant therapy as long as PTT is within therapeutic range of intended use of anticoagulants

3.1.7 < 4 mg dexamethasone daily (or equivalent if on another corticosteroid) at time of start of therapy. Patients on a steroid taper post-surgery and are anticipated to be on <4 mg at time of chemoradiation initiation will be eligible to consent but to initiate treatment on trial, the participant must be on <4 mg or equivalent of steroids otherwise participate will be deemed a screen fail and be replaced.

3.1.8 The effects of bavituximab on the developing human fetus are unknown. For this reason, women of child-bearing potential and men must agree to use adequate contraception (hormonal or barrier method of birth control; abstinence) prior to study entry and for the duration of study participation. Should a woman become pregnant or suspect she is pregnant while she or her partner is participating in this study, she should inform her treating physician immediately. Men treated or enrolled on this protocol must also agree to use adequate contraception prior to the study, for the duration of study participation, and 4 months after completion of bavituximab administration.

3.1.9 Able to undergo an MRI scan and receive gadolinium-based contrast.

3.1.10 1 cm3 of available tissue for N=5 patients for correlative tissue studies (Section 9.2.2). Patients can enroll regardless of their tissue availability being checked beforehand but at least 5 patients will need to have sufficient tissue by study accrual completion. Tissue availability will be checked after patients are successfully enrolled.

3.1.11 Ability to understand and the willingness to sign a written informed consent document.

3.2 Exclusion Criteria

3.2.1 Participants who are receiving any other investigational agents.

3.2.2 History of allergic reactions attributed to compounds of similar chemical or biologic composition to bavituximab.

3.2.3 Participants receiving any medications or substances that are moderate and/or potent enzyme inducers or inhibitors which may have an effect on the metabolism of bavituximab. As part of the enrollment/informed consent procedures, the patient will be counseled on the risk of interactions with other agents, and what to do if new medications need to be prescribed or if the patient is considering a new over-the-counter medicine or herbal product (Appendix C for partial list).

3.2.4 Uncontrolled intercurrent illness including, but not limited to, ongoing or active infection, symptomatic congestive heart failure, unstable angina pectoris, cardiac arrhythmia, or psychiatric illness/social situations that would limit compliance with study requirements.

3.2.5 Pregnant women are excluded from this study because bavituximab is an immunotherapy agent with the potential for teratogenic or abortifacient effects. Because there is an unknown but potential risk for adverse events in nursing infants secondary to treatment of the mother with bavituximab breastfeeding should be discontinued if the mother is treated with bavituximab. These potential risks may also apply to other agents used in this study.

3.2.6 HIV-positive participants on combination antiretroviral therapy are ineligible because of the potential for pharmacokinetic interactions with bavituximab. In addition, these participants are at increased risk of lethal infections when treated with marrow-suppressive therapy.

3.2.7 Participants with other active malignancy in the past 3 years excluding in situ tumors.

3.2.8 Participants must meet the following windows from procedures (there is no window required for port placement since there is no anticipated impact on wound healing with bavituximab):

- Major surgery (ex. craniotomy) within 3 weeks of initiation of treatment.
- Brain biopsy within 2 weeks

3.2.9 Participants with active bleeding disorder/coagulopathy.

3.2.10 Participants with active chronic or acute hepatitis C or B infection.

3.3 Inclusion of Women and Minorities

Both men and women of all races and ethnic groups are eligible for this trial.

4. REGISTRATION PROCEDURES

4.1 General Guidelines for DF/HCC Institutions

Institutions will register eligible participants in the Clinical Trials Management System (CTMS) OnCore. Registrations must occur prior to the initiation of protocol therapy. Any participant not registered to the protocol before protocol therapy begins will be considered ineligible and registration will be denied.

An investigator will confirm eligibility criteria and a member of the study team will complete the protocol-specific eligibility checklist.

Following registration, participants may begin protocol therapy. Issues that would cause treatment delays should be discussed with the Overall Principal Investigator (PI). If a participant does not receive protocol therapy following registration, the participant's registration on the study must be canceled. Registration cancellations must be made in OnCore as soon as possible.

4.2 Registration Process for DF/HCC Institutions

DF/HCC Standard Operating Procedure for Human Subject Research Titled *Subject Protocol Registration* (SOP #: REGIST-101) must be followed.

4.3 General Guidelines for Other Investigative Sites

Not applicable

4.4 Registration Process for Other Investigative Sites

Not applicable

5. TREATMENT PLAN

5.1 Treatment Regimen

Bavituximab will be administered weekly for 18 weeks starting the first week of chemoradiation. Patients may continue beyond 18 weeks at the discretion of the treating physician if the patient is deriving clinical benefit. Cycle 1 will encompass the 6 weeks of chemoradiation. Cycles 2 onward will be 4 weeks in length. Treatment will be administered on an outpatient basis. Reported adverse events and potential risks are described in Section 7. No investigational or commercial agents or therapies other than those described below may be administered with the intent to treat the participant's malignancy with the exception of NovoTTF.

Cycle 1: Chemoradiation

Regimen Description					
Agent	***Premedications; Precautions***	***Dose***	***Route***	***Schedule***	***Cycle Length***
Bavituximab	Optional: Premedicate with 250 mg hydrocortisone IV and 50 mg diphenhydramine IV administered ideally 30 minutes prior to infusion	3 mg/kg	First IV infusion must be administered over approximately 90 (±10) minutes. If the first IV infusion is tolerated, subsequent infusions may be administered over 60 (±10) minutes	Weekly starting with the 5th day of radiation (window = after 4th dose but before 6th dose). Administration can be at any time of the day that fits the patient's schedule regardless of RT or temozolom	Cycle 1: 6 weeks

				ide administration.	
Temozolomide	Zofran 4-8 mg or other anti-emetic as needed	75mg/m2	PO	Daily for up to 42 days	

Cycle 2: Post Radiation

Regimen Description					
Agent	***Premedications; Precautions***	***Dose***	***Route***	***Schedule***	***Cycle Length***
Bavituximab	Optional: Premedicate with 250 mg hydrocortisone IV and 50 mg diphenhydramine IV administered ideally 30 minutes prior to infusion	3 mg/kg	IV infusion can be administered over approximately 60 (±10) minutes if the first IV infusion was tolerated. Otherwise continue at approximately 90 (±10) minutes.	Weekly	4 weeks

Cycle 3 onward: With Monthly Temozolomide

Regimen Description					
Agent	***Premedications; Precautions***	***Dose***	***Route***	***Schedule***	***Cycle Length***
Bavituximab	Optional: Premedicate with 250 mg hydrocortisone IV and 50 mg diphenhydramine IV administered ideally 30 minutes prior to infusion	3 mg/kg	IV infusion can be administered over approximately 60 (±10) minutes if the first IV infusion was tolerated. Otherwise continue at	Weekly	4 weeks

			approximately 90 (±10) minutes.		
Temozolomide	Zofran 4-8 mg or other anti-emetic as needed	150-200mg/m2	PO	Daily for 5 consecutive days in cycles 3-4	

5.2 Pre-Treatment Criteria

5.2.1 Cycle 1, Day 1

Prior to administration of the first dose of bavituximab on day 1 of cycle 1, participants must meet the following criteria:

- Signed informed consent
- Demographic information, medical history and concurrent medication list
- Physical examination including mini-mental status examination, vital signs, height, weight, and Karnofsky performance status
- Laboratory work meeting the eligibility criteria must be obtained.

5.2.2 Subsequent Days (Chemoradiation)

Prior to administration of the subsequent doses of bavituximab during chemoradiation participants must meet the following criteria. Patients can continue bavituximab if temozolomide is held for temozolomide related toxicity:

- Platelet count ≥ 75,000/mcL
- WBC ≥ 2,500/mcL
- No other bavituximab treatment related toxicities CTCAE grade > 2 (except for not maximally treated nausea and vomiting)
- If a bavituximab cannot be administered because of toxicity, there is a +3 day window to treat, otherwise the dose will be skipped and the patient resume dosing the next week if criteria met.

5.2.3 Subsequent Cycles of temozolomide (Post-chemoradiation cycles)

Prior to administration of the subsequent doses of temozolomide participants must meet the following criteria within the windows outlined in the Study Calendar in Section 10:

- Resolution of all grade 3 non-hematologic AEs (except for not maximally treated nausea and vomiting) to grade ≤ 1
- Absolute neutrophil count (ANC) < grade 2
- Platelet count ≥ 100,000/mcL
- Hemoglobin ≥ 8.0 g/dL
- White blood cell (WBC) count ≥ 2,500/mcL

- AST/SGOT and ALT/SGPT ≤ 2.5 x upper limit of normal (ULN)
- Serum total bilirubin ≤ 1.5 x ULN (unless patient has Gilbert's syndrome in which total bilirubin should be ≤ 2xULN)
- Serum creatinine ≤ 1.5 x ULN or 24-hour creatinine clearance ≥ 60 ml/min
- No other temozolomide related toxicities CTCAE grade >2
- If AEs persist, the start of the cycle may be delayed up to 30 consecutive days. If all AEs have not resolved to grade ≤ 1 after a 30-day delay, any further study treatment will be discontinued.
- For C3 and C4 when monthly temozolomide starts, bavituximab should be administered within 5 days of Day 1 of the temozolomide

5.2.4 Subsequent Cycles of bavituximab (Post-chemoradiation cycles)

Prior to administration of the subsequent doses of bavituximab participants must meet the following criteria within the windows outlined in the Study Calendar in Section 10. Patients can continue bavituximab if temozolomide is held for temozolomide related toxicity:

- Resolution of all grade 3 non-hematologic AEs (except for not maximally treated nausea and vomiting) to grade ≤ 2 thought related to bavituximab
- Platelet count ≥ 75,000/mcL
- Hemoglobin ≥ 8.0 g/dL
- White blood cell (WBC) count ≥ 2,500/mcL
- AST/SGOT and ALT/SGPT ≤ 2.5 x upper limit of normal (ULN)
- Serum total bilirubin ≤ 1.5 x ULN (unless patient has Gilbert's syndrome in which total bilirubin should be ≤ 2xULN)
- Serum creatinine ≤ 1.5 x ULN or 24-hour creatinine clearance ≥ 60 ml/min
- No other bavituximab related toxicities CTCAE grade > 2
- If AEs persist, the start of the cycle may be delayed up to 30 consecutive days. If all AEs have not resolved to grade < 2 after a 30-day delay, any further study treatment will be discontinued.
- For C3 onwards when monthly temozolomide starts, bavituximab should be administered within 7 days of Day 1 of the temozolomide

5.3 Agent Administration

5.3.1 *CTEP and/or CIP IND Agent(s), or other IND agent: Bavituximab*

Administration: Bavituximab will be administered IV.

Dosing: Bavituximab dose is 3 mg/kg body weight. The total dose is only required to be recalculated if there is a ≥10% change in weight from Day 1 or per institutional policy. There will otherwise be no modification of dose level or schedule of bavituximab treatment. Bavituximab will be given weekly +/- 3 days.

If infusion is not administered within this window, it is considered skipped and dosing

will resume at next scheduled time.

Infusion Reactions: Administer standard hypersensitivity reaction medications per institution guidelines. In addition, all patients should be pre-medicated as outlined in Section 5.

Clinically mild reactions (eg, generalized rash or itching, hives) are treated as soon as possible with Benadryl® (diphenhydramine hydrochloride) 25 to 50 mg, orally or IV at the investigator's discretion. The period of observation is extended beyond 3 hours, as necessary, until symptoms and signs have resolved or stabilized. Patients who have experienced a clinically mild reaction may continue to have study drug administered.

Clinically moderate reactions (eg, hypotension, shortness of breath, presyncope, facial edema) are treated immediately and supportive care measures instituted as medically indicated (eg, IV fluids, corticosteroids, vasopressors, oxygen, bronchodilators, diphenhydramine, and acetaminophen). Vital signs are monitored at 10-minute intervals until they have normalized. The period of observation is extended beyond 3 hours, if necessary, until symptoms and signs have resolved. In the event of a clinically moderate reaction, the patient should receive no further treatment with study drug.

For grade 3-4 reactions, halt infusion and discuss with principle investigator if rechallenge is reasonable or if the patient should go off treatment.

5.3.2 *Other Agent(s)*

Temozolomide will be taken per standard of care. Guidelines provided below on dosing/schedule.

Temozolomide During Concomitant Radiation Therapy

Temozolomide will be administered continuously from day 1 of radiotherapy to the last day of radiotherapy at a daily oral dose of 75 mg/m^2 for a maximum of 42 days. The drug will be administered orally 1-3 hours before each session of radiotherapy during weekdays (Monday through Friday). During weekends without radiotherapy (Saturday and Sunday), the drug will be taken in the morning. The dose will be determined using actual body surface area (BSA) as calculated in square meters at the beginning of the concomitant treatment.

Patients will be instructed to swallow the capsules whole, in rapid succession, without chewing them. If vomiting occurs during the course of treatment, no re-dosing of the patient is allowed before the next scheduled dose. The capsules should be taken on an empty stomach, therefore a minimum of 2 hours after eating and with no food consumption for at least 1 hour after temozolomide administration. Although, nightly administration just before bedtime has been reported to improve tolerance, the low daily dose administered during radiation is very well tolerated and administration in the morning before radiation dosing is required for this protocol. Administration of the higher dosing regimen during the adjuvant phase of the protocol should be performed using the nightly administration.

Antiemetic prophylaxis is usually not required for the continuous daily dosing schedule (during radiation). However, prophylaxis with a 5-HT3 antagonist is recommended 30-60 minutes prior to administration of the first few temozolomide doses. Most patients report optimal nausea control with the use of a 5-HT3 antagonist. Routine use of antiemetics is recommended during the adjuvant phase of treatment. Additionally, pneumocystis carinii prophylaxis is at the discretion of the Investigator during the radiation phase.

Post-Radiation Temozolomide Dosing

As part of this protocol, patients will receive up to 2 cycles of adjuvant temozolomide at a dose of 150-200 mg/m2 days 1-5 of a 28-day cycle. Patients will be treated for up to 6 months of post-RT temozolomide (Stupp 2005). If patients experience grade 3 or higher toxicity attributable to the temozolomide that does not resolve to a grade 2 or less within 60 days of the last temozolomide dose, temozolomide should be discontinued. If the toxicity returns to a grade 2 or less within 30 days of the last dose, temozolomide can be dose reduced to 150mg/m2. The active period of this clinical trial will be approximately 18 weeks as long as there are no delays or missed doses that need to be made up.

As part of this protocol, temozolomide will be administered orally once per day for 5 consecutive days (days 1-5) of a 28-day cycle. The starting dose for the first cycle will be 150 mg/m^2/day, with a single dose escalation to 200 mg/m^2/day in subsequent cycles if no treatment-related adverse events > Grade 2 are noted. Patients can be increased to 200 mg/m2 at any time during the maintenance phase if deemed clinically appropriate by the treating physician.

The start of the cycle 2 will be scheduled 28 days ± 7 days after the last day of radiotherapy. The start of the subsequent cycles (Cycles 3-4) will be scheduled in 28 days ± 7 days) after the first daily dose of temozolomide of the preceding cycle.

The dose will be determined using the BSA per institutional standard.

Patients will be instructed to fast at least 2 hours before and 1 hour after temozolomide administration. Water is allowed during the fast period. Patients will be instructed to swallow the capsules whole, in rapid succession, without chewing them. Treatment can be given at night if patient prefers.

If vomiting occurs during the course of treatment, no re-dosing of the patient is allowed before the next scheduled dose.

If a patient misses the dose of temozolomide, it made be made up within 8 hours, otherwise the dose should be delayed until the next time it is due. Missed doses can be added on to the end of the 5 day dosing (i.e. taken on day 6) during the maintenance phase (i.e. 5/28 day cycles).

Antiemetic prophylaxis with a 5-HT3 antagonist is recommended and should be administered 30 to 60 minutes before temozolomide administration. Temozolomide should be stored at room temperature.

5.3.3 *Other Modality(ies) or Procedures*

Radiation will be delivered per standard of care guidelines which includes 60 GY of involved field radiation delivered in up to 2 Gy fractions per day over 6 weeks.

5.4 General Concomitant Medication and Supportive Care Guidelines

The case report form must capture the concurrent use of all other drugs, over-the-counter medications, or alternative therapies. There are no known drugs that have an interaction with bavituximab. Full dose anticoagulation is permitted although LMWH is preferred.

5.5 Criteria for Taking a Participant Off Protocol Therapy

Duration of therapy will depend on individual response, evidence of disease progression and tolerance. In the absence of treatment delays due to adverse event(s), treatment may continue for 18 weeks or until one of the following criteria applies:

- Disease progression
- Intercurrent illness that prevents further administration of treatment
- Unacceptable adverse event(s)
- Participant demonstrates an inability or unwillingness to comply with the oral medication regimen and/or documentation requirements
- Participant decides to withdraw from the protocol therapy
- General or specific changes in the participant's condition render the participant unacceptable for further treatment in the judgment of the treating investigator
- Completion of the 18 weeks of treatment and all study related MRI scans. However, patients may continue beyond 18 weeks at the discretion of the treating physician if the patient is deriving clinical benefit.

Participants will be removed from the protocol therapy when any of these criteria apply. The reason for removal from protocol therapy, and the date the participant was removed, must be documented in the case report form (CRF) and in OnCore. Alternative care options will be discussed with the participant.

In the event of unusual or life-threatening complications, treating investigators must immediately notify the Overall PI, Dr. Elizabeth Gerstner at 617-724-8770.

5.6 Duration of Follow Up

Participants will be followed every 3 months for 5 years from date of registration or until death, whichever occurs first. Participants removed from protocol therapy for unacceptable adverse event(s) will be followed until resolution or stabilization of the adverse event. Patients who do not continue to follow up in clinic will be reached by phone call to determine status. During this follow-up period, specific attention will be paid to any late effects of radiation which will be specifically captured on the case report forms.

5.7 Criteria for Taking a Participant Off Study

Participants will be removed from study when any of the following criteria apply:

- Lost to follow-up
- Withdrawal of consent for data submission
- Death

The reason for taking a participant off study, and the date the participant was removed, must be documented in the case report form (CRF). The research team should also update the relevant Off Treatment/Off Study information in OnCore.

6. DOSING DELAYS/DOSE MODIFICATIONS

Dose delays and modifications will be made as indicated in the following table(s). The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 4.0 will be utilized for dose delays and dose modifications. A copy of the CTCAE version 4.0 can be downloaded from the CTEP website http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm.

If temozolomide is held, bavituximab can continue if the adverse event is deemed to be unlikely related to bavituximab. There will be no change in dosing schedule or assessment schedule if either drug is held.

Nausea	**Management/Next Dose for temozolomide**
≤ Grade 1	No change in dose
Grade 2	Hold until ≤ Grade 1. Resume at same dose level.
Grade 3	Hold* until < Grade 2. Resume at one dose level lower, if indicated.**

Nausea	**Management/Next Dose for temozolomide**
Grade 4	Off protocol therapy
*Participants requiring a delay of >30 days should go off protocol therapy. **Participants requiring > two dose reductions should go off protocol therapy.	
Recommended management: antiemetics.	

Vomiting	**Management/Next Dose for temozolomide**
≤ Grade 1	No change in dose
Grade 2	Hold until ≤ Grade 1. Resume at same dose level.
Grade 3	Hold* until < Grade 2. Resume at one dose level lower, if indicated.**
Grade 4	Off protocol therapy
*Participants requiring a delay of >30 days should go off protocol therapy. **Participants requiring > two dose reductions should go off protocol therapy.	
Recommended management: anti-emetics.	

Neutropenia	**Management/Next Dose for temozolomide**
≤ Grade 1	No change in dose
Grade 2	Hold until ≤ Grade 1. Resume at same dose level.
Grade 3	Hold* until < Grade 2. Resume at one dose level lower, if indicated.**
Grade 4	Off protocol therapy
*Participants requiring a delay of >30 days should go off protocol therapy. **Participants requiring > two dose reductions should go off protocol therapy.	

Thrombocytopenia	**Management/Next Dose for temozolomide**
≤ Grade 1	No change in dose
Grade 2	Hold until ≤ Grade 1. Resume at same dose level.
Grade 3	Hold* until < Grade 2. Resume at one dose level lower, if indicated.**
Grade 4	Off protocol therapy
*Participants requiring a delay of >30 days should go off protocol therapy. **Participants requiring > two dose reductions should go off protocol therapy.	

Dose reductions of temozolomide during the adjuvant period should be made according to the prescribing information, as summarized below. No dose reductions will be made during the chemoradiation phase. If an adverse event related to temozolomide occurs during chemoradiation, dosing will be held and missed doses will not be made up.

Table: Temozolomide Dose Levels for Cycles 3 and 4

Dose Level	Dose (mg/m2/day)	Remarks
-1	100	Reduction for prior toxicity
0	150	Dose during Cycle 1
+1	200	Dose during Cycle 2 in absence of toxicity

AE's attributed to bavituximab will be managed with maximal supportive care but since temozolomide is the more likely culprit for toxicity, a decrease in temozolomide should be tried prior to stopping bavituximab.

If a patient develops a blood clot, low molecular weight heparin can be used. The bavituximab does not need to be held but may be held at the treating physician's discretion.

7. ADVERSE EVENTS: LIST AND REPORTING REQUIREMENTS

Adverse event (AE) monitoring and reporting is a routine part of every clinical trial. The following list of reported and/or potential AEs (Section 7.1) and the characteristics of an observed AE (Section 7.2) will determine whether the event requires expedited reporting **in addition** to routine reporting.

7.1 Expected Toxicities

7.1.1 Adverse Events List(s)

7.1.1.1 Adverse Event List(s) for bavituximab

Table 24: Treatment-emergent Adverse Events Occurring in More Than 5% of Patients Treated with Bavituximab and Chemotherapy by Preferred Term and Dose Level for all Oncology Studies Safety Population

	Number of Patients(%)		
	Oncology[a]		Total
Preferred Term	1 mg/kg[b] (N=79)	3 mg/kg (N=570)	Total (N=649)
Alopecia	14 (17.7%)	174 (30.5%)	188 (29.0%)
Nausea	21 (26.6%)	167 (29.3%)	188 (29.0%)
Fatigue	21 (26.6%)	162 (28.4%)	183 (28.2%)
Diarrhoea	15 (19.0%)	166 (29.1%)	181 (27.9%)
Neutropenia	18 (22.8%)	149 (26.1%)	167 (25.7%)
Anaemia	14 (17.7%)	145 (25.4%)	159 (24.5%)
Asthenia	13 (16.5%)	121 (21.2%)	134 (20.6%)
Cough	17 (21.5%)	103 (18.1%)	120 (18.5%)
Dyspnoea	11 (13.9%)	106 (18.6%)	117 (18.0%)
Pyrexia	13 (16.5%)	101 (17.7%)	114 (17.6%)
Vomiting	9 (11.4%)	103 (18.1%)	112 (17.3%)
Constipation	12 (15.2%)	95 (16.7%)	107 (16.5%)
Leukopenia	12 (15.2%)	85 (14.9%)	97 (14.9%)
Decreased appetite	5 (6.3%)	84 (14.7%)	89 (13.7%)
Oedema peripheral	16 (20.3%)	66 (11.6%)	82 (12.6%)
Arthralgia	8 (10.1%)	71 (12.5%)	79 (12.2%)
Back pain	8 (10.1%)	70 (12.3%)	78 (12.0%)
Headache	8 (10.1%)	61 (10.7%)	69 (10.6%)
Pain in extremity	8 (10.1%)	61 (10.7%)	69 (10.6%)
Anorexia	12 (15.2%)	48 (8.4%)	60 (9.2%)
Stomatitis	4 (5.1%)	55 (9.6%)	59 (9.1%)

7.1.1.2 Adverse Event List(s) for Other Agent(s)

Please see package insert for adverse events for temozolomide.

7.2 Adverse Event Characteristics

- **CTCAE term (AE description) and grade:** The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 4.0 will be utilized for AE reporting. All appropriate treatment areas should have access to a copy of the CTCAE version 4.0. A copy of the CTCAE version 4.0 can be downloaded from the CTEP web site http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm.

- **For expedited reporting purposes only:**
  - AEs for the agent(s) that are listed above should be reported only if the adverse event varies in nature, intensity or frequency from the expected toxicity information which is provided.
  - Other AEs for the protocol that do not require expedited reporting are outlined in the next section (Expedited Adverse Event Reporting) under the sub-heading of Protocol-Specific Expedited Adverse Event Reporting Exclusions.

- **Attribution** of the AE:

- Definite – The AE *is clearly related* to the study treatment.
- Probable – The AE *is likely related* to the study treatment.
- Possible – The AE *may be related* to the study treatment.
- Unlikely – The AE *is doubtfully related* to the study treatment.
- Unrelated – The AE *is clearly NOT related* to the study treatment.

7.3 Expedited Adverse Event Reporting

7.3.1 Investigators **must** report to the Overall PI any serious adverse event (SAE) that occurs after the initial dose of study treatment, during treatment, or within 30 days of the last dose of treatment on the local institutional SAE form.

7.3.2 Expedited Adverse Event Reporting to Company Supplying Drug

Investigators are required to report to Oncologie Drug Safety any SAE (Serious Adverse Event) as soon as possible.

A SAE is any sign, symptom or medical condition that emerges during bavituximab treatment or during a post-treatment follow-up period that (1) was not present at the start of bavituximab treatment and it is not a chronic condition that is part of the patient's medical history, OR (2) was present at the start of bavituximab treatment or as part of the patient's medical history but worsened in severity and/or frequency during therapy, AND that meets any of the following regulatory serious criteria:

- Results in death
- Is life-threatening
- Requires or prolongs inpatient hospitalization
- Is disabling
- Is a congenital anomaly/birth defect
- Is medically significant or requires medical or surgical intervention to prevent one of the outcomes listed above.

All SAEs should be recorded on a MedWatch 3500A Form and emailed to:

Oncologie Drug Safety (c/o MedAssessment Pharmacovigilance)
E-mail: oncologiesafety@medassessment.com

AND:

NCCN: E-mail: ORPreports@nccn.org or faxed to 215-358-7699

MedWatch 3500A Reporting Guidelines:

In addition to completing appropriate patient demographic and suspect medication information, the report should include the following information within the Event Description (section 5) of

the MedWatch 3500A form:

- Treatment regimen (dosing frequency, combination therapy)
- Protocol description (and number, if assigned)
- Description of event, severity, treatment, and outcome, if known
- Supportive laboratory results and diagnostics
- Investigator's assessment of the relationship of the adverse event to each investigational product and suspect medication

Additional information may be added to a previously submitted report by any of the following methods:

- Adding to the original MedWatch 3500A report and submitting it as follow-up
- Adding supplemental summary information and submitting it as follow-up with the original MedWatch 3500A form
- Summarizing new information and faxing it with a cover letter including patient identifiers (i.e. D.O.B. initial, patient number), protocol description and number, if assigned, suspect drug, brief adverse event description, and notation that additional or follow-up information is being submitted (The patient identifiers are important so that the new information is added to the correct initial report)

Occasionally Oncologie may contact the reporter for additional information, clarification, or current status of the patient for whom and adverse event was reported.

Assessing Causality:
Investigators are required to assess whether there is a reasonable possibility that bavituximab caused or contributed to an adverse event. The following general guidance may be used.
Yes: if the temporal relationship of the clinical event to bavituximab administration makes a causal relationship possible, and other drugs, therapeutic interventions or underlying conditions do not provide a sufficient explanation for the observed event.

No: if the temporal relationship of the clinical event to bavituximab administration makes a causal relationship unlikely, or other drugs, therapeutic interventions or underlying conditions provide a sufficient explanation for the observed event.

- **Safety Reporting Requirements for IND Holders**

In accordance with 21 CFR 212.32, investigator-investigators of studies conducted under an IND must comply with following safety reporting requirements:

a. Expedited IND Safety Reports:

7 Calendar-Day Telephone or Fax Report:
The Investigator-Investigator is required to notify the FDA of any fatal or life-threatening adverse event that is unexpected and assessed by the investigator to be possibly related to the use

of bavituximab. An unexpected adverse event is one that is not already described in the Investigator Brochure. Such reports are to be telephoned or faxed to the FDA and Oncologie within 7 calendar days of first learning of the event. Each telephone call or fax transmission (see fax number below) should be directed to the FDA new drug review division in the Center for Drug Evaluation and Research or in the product review division for the Center for Biologics Evaluation and Research, whichever is responsible for the review of the IND.

15 Calendar-Day Written Report:
The Investigator-Investigator is also required to notify the FDA and all participating investigators, in a written IND Safety Report, of any serious, unexpected AE that is considered possibly related to the use of bavituximab. An unexpected adverse event is one that is not already described in the Investigator Brochure.

Written IND Safety Reports should include an Analysis of Similar Events in accordance with regulation 21 CFR § 312.32. All safety reports previously filed with the IND concerning similar events should be analyzed. The new report should contain comments on the significance of the new event in light of the previous, similar reports.

Written IND safety reports with Analysis of Similar Events are to be submitted to the FDA, Oncologie Drug Safety, and all participating investigators within 15 calendar days of first learning of the event. The FDA prefers these reports on a MedWatch 3500A Form but alternative formats are acceptable (e.g. summary letter).

FDA fax number for IND Safety Reports:

1 (800) FDA - 0178

All written IND Safety Reports submitted to the FDA by the Investigator must also be emailed to: Oncologie Drug Safety

	North America
Hagop Youssoufian, MD	hagop@oncologie.international
Safety Reporting Email Fax	oncologiesafety@medassessment.com

For questions related to safety reporting, contact:

Oncologie Drug Safety (c/o MedAssessment)
Tel: 1-781-434-5010

b. IND Annual Reports
In accordance with the regulation 21 CFR § 312.32, the Investigator- Sponsor shall within 60

days of the anniversary date that the IND went into effect submit a brief report of the progress of the investigation. Please refer to Code of Federal Regulations, 21 CFR § 312.32 for a list of the elements required for the annual report. All IND annual reports submitted to the FDA by the Investigator should be copied to Oncologie. Copies of such reports should be provided to:

Susan MacIntyre, RN, BSN
Executive Director, Global Clinical Operations
Oncologie, Inc.
400 Totten Pond Rd
Waltham, MA
1-774-245-8299
susan@oncologie.international

7.3.3 DF/HCC Expedited Reporting Guidelines

Investigative sites within DF/HCC will report AEs directly to the DFCI Office for Human Research Studies (OHRS) per the DFCI IRB reporting policy.

7.4 Expedited Reporting to the Food and Drug Administration (FDA)

The Overall PI, as study sponsor, will be responsible for all communications with the FDA. The Overall PI will report to the FDA, regardless of the site of occurrence, any serious adverse event that meets the FDA's criteria for expedited reporting following the reporting requirements and timelines set by the FDA.

7.5 Expedited Reporting to Hospital Risk Management

Participating investigators will report to their local Risk Management office any participant safety reports or sentinel events that require reporting according to institutional policy.

7.6 Routine Adverse Event Reporting

All Adverse Events **must** be reported in routine study data submissions to the Overall PI on the toxicity case report forms. **AEs reported through expedited processes (e.g., reported to the IRB, FDA, etc.) must also be reported in routine study data submissions.**

8. PHARMACEUTICAL INFORMATION

A list of the adverse events and potential risks associated with the investigational or other agents administered in this study can be found in Section 7.1.

8.1 Bavituximab

8.1.1 Description

Bavituximab will be administered at 3 mg/kg body weight and diluted with normal saline to a volume no less than 100 mL. The total dose is only required to be recalculated if there is a ≥10% change in weight from Day 1 or per institutional policy. There will otherwise be no modification of dose level or schedule of bavituximab treatment.

8.1.2 Form

Bavituximab is supplied as 5 mL of a sterile solution in borosilicate type I glass vials and contains 120 mg bavituximab (24 mg/mL), 10 mM acetate at pH 5.0, and Water for Injection, United States Pharmacopeia.

8.1.3 Storage and Stability

Bavituximab is stored at 2°C to 8°C. Sites should monitor temperature conditions and report any temperature excursions. Once diluted, bavituximab should be stored at room temperature and infused within 8 hours. The infusion must be completed within 8 hours of dilution.

Bavituximab must be stored in a secure area and administered only to patients entered into the clinical study in accordance with the conditions specified in the protocol. Bavituximab will be managed by a pharmacist or designee at the site.

The pharmacist or designee must keep an accurate accounting of the number of investigational units received from Oncologie, dispensed to patients, and returned to the Oncologie or representative (or destroyed at the site) during and at the completion of the study. Standard operating procedures (SOPs) must be in place to control the use of the study drug. Bavituximab must be dispensed only by an appropriately qualified person and is to be used in accordance with the protocol in patients who are under the direct supervision of the investigator.

All used vials may be destroyed per institutional policy.

8.1.4 Compatibility

It is recommended that the investigational product is allowed to warm to room temperature for 30 to 60 minutes prior to dilution into normal saline Syringes and needles used for treatment preparation should be non-siliconized. The IV normal saline container admixed with IP may contain a small amount of intrinsic, translucent to white proteinaceous particles.

8.1.5 Handling

Qualified personnel, familiar with procedures that minimize undue exposure to themselves and the environment, should undertake the preparation, handling, and safe

disposal of the chemotherapeutic agent in a self-contained and protective environment.

8.1.6 **Availability**

Oncologie will supply drug and provide a request form for shipping. Drug will supplied free of charge.

8.1.7 **Preparation**

A pharmacist or designee at the study site will prepare bavituximab to be administered by study personnel to patients in accordance with the scheme. Infusion preparation and administration are to be performed as follows:

Using aseptic technique, withdraw the calculated dose volume of bavituximab from the vials using a sterile non-siliconized needle and syringe and inject contents of syringe into the normal saline container. Thoroughly mix the infusion container by gentle manual rotation. Avoid shaking or vigorous agitation during preparation and prior to administration.

The IV normal saline container admixed with IP may contain a small amount of intrinsic, translucent to white proteinaceous particles. Administer through a low protein binding 0.2-micrometer in-line filter (placed as proximal to the patient as practical).

8.1.8 **Administration**

DO NOT ADMINISTER TREATMENT AS AN IV PUSH OR BOLUS.

Pre-medication is no longer required prior to bavituximab administration as the steroids may interfere with drug efficacy and there have been limited reactions in patients so far. If pre-medication is warranted, one possible regimen is 250 mg hydrocortisone IV and 25-50 mg diphenhydramine IV administered ideally 30 minutes prior to infusion; regimen, dosages, and timing can be adjusted at the investigator's discretion. Patients receiving premedication for other therapy do not require additional corticosteroids on days when both are administered. Patients who tolerate bavituximab without reaction may not need pre-medication with subsequent doses and pre-medication use can be decided in conjunction with patient, nursing, treating physician.

Affix the infusion line and prime it with infusate before starting the infusion.
The first IV infusion must be administered over approximately 90 (±10) minutes. Patients should be watch for 180 minutes after the start of the first infusion. If the first IV infusion is tolerated, subsequent infusions may be administered over 60 minutes with no post-infusion observation as long as no hypersensitivity reactions occur.

Flush the main infusion line with normal saline after infusion.

NOTE: Bavituximab should only be administered in settings in which emergency

resuscitative equipment and personnel trained in the management of anaphylaxis are immediately available to treat systemic hypersensitivity reactions.

NOTE: These instructions must be consistent with protocol section 5: TREATMENT PLAN.

8.1.9 Ordering

A drug shipment request form will be supplied by Oncologie.

8.1.10 Accountability

The investigator, or a responsible party designated by the investigator, should maintain a careful record of the inventory and disposition of the agent using the NCI Drug Accountability Record Form (DARF) or another comparable drug accountability form. (See the NCI Investigator's Handbook for Procedures for Drug Accountability and Storage.)

8.1.11 Destruction and Return

All unused study treatments should be saved, accounted for, and processed after consulting with Oncologie.

At the time of study closure, unused, used and expired study drug will be destroyed at the site per Institutional SOPs if not already done so. For bavituximab, if site SOPs are not available, drug may be returned using the Agent Return Form.

8.2 *Other Agent #1: Temozolomide*

Temozolomide will be commercially obtained through a specialty pharmacy as it is a part of the treatment plan. Participants will be responsible for maintaining a pill diary for the 18-week treatment period.

9. BIOMARKER, CORRELATIVE, AND SPECIAL STUDIES

9.1 Biomarker Studies

As outlined in Section 2.5 MRI scans will be obtained at baseline, 4 weeks post-chemoradiation, and then every other month per standard of care. The MRIs at baseline, pre-study cycle 3 (i.e. prior to the first monthly cycle of temozolomide), and post-study cycle 4 (i.e. pre-third monthly cycle of temozolomide) will be performed at the Martinos Center for Biomedical Imaging. These scans will include anatomical MRI sequences as well as perfusion MR (dynamic susceptibility contrast MRI), permeability imaging (dynamic contrast enhanced MRI), MRSpectroscopy, and

resting state MRI. There will be 2 injections of gadolinium based contrast agent for a total 0.2 mmol/kg (less than the FDA limit of 0.3 mmol/kg). Patients will be required to have an eGFR>60 mL/min/1.73 m2 within 30 days prior to the MRI. Total scan time is approximately 50 minutes and data will be processed by Dr. Gerstner's group at the Martinos Center to extract information about tumor vascular structure and function, connectivity, as well as cell density and metabolism as outlined in Section 2.5 and per our standard analytical techniques (Batchelor 2013, Gerstner 2016). Quantitative markers to be evaluated as exploratory markers of response include cerebral blood flow/volume, relative tissue oxygenation saturation, apparent diffusion coefficient, intracellular volume fraction, ktrans, and metabolites (ex. Cr, NAA). We anticipate others arising as the field of physiological imaging for brain tumors is rapidly evolving with novel analysis approaches continually being proposed. These novel markers would be extracted from existing data.

9.2 Laboratory Correlative Studies

9.2.1.1 Blood for flow cytomety

Blood will be analyzed for circulating immune cells to help distinguish inflammation from active tumor by tracking T cell receptor (TCR) repertoire and its correlation with TCRs that expand in the tumor. In addition, myeloid cells will be analyzed to detect changes with therapy.

We will perform a flow cytometry analysis of myeloid cells, focusing on the number of (myeloid derived suppressor cells (MDSCs). We will use flow cytometry to analyze myeloid cells (HLA-DR+, CD3-, CD19-, CD56) cells, including CD14+ and CD16+ monocytes, DC subtypes (CLEC9A/CD141; CD1C+; CLEC9A-CD1C-), and MDSCs (CD33+CD11b+Lin-HLADR-, CD14+CD11b+Lin-HLADR- and CD15+CD11b+Lin-HLADR-) This will quantify the absolute number and frequencies of these cell types pre and post therapy. In addition, we will track TCR repertoire using standard next generation sequencing approaches (amplify TCRbeta from blood and analyze repertoire). A prior study in vaccinated glioblastoma patients demonstrated coordinate changes in TCR repertoire in tumor and blood in association with durable responses. (Hsu 2016)

9.2.1.2 Collection of Specimen(s)

Blood will be collected prior to start of treatment, at the end of chemoradiation (week 6 of chemoradiation), first week after radiation, and then monthly for the subsequent 2 months during bavituximab + monthly temozolomide treatment.

9.2.1.3 Handling of Specimens(s)

At appropriate time points, 50 cc of fresh whole blood will be collected in K2 EDTA 10 ml Blood Collection Tubes, delivered immediately after collection at room temperature (no ice) to Dr. Curry's laboratory (3rd Floor Simches Research Building , Room CPZN 3) for isolation and storage of peripheral blood mononuclear cells (PBMC's). Label subject ID, date and time of

collection. Site will be responsible for all of the supplies, inclusive of packaging and shipping materials and shipping charges. Contact Leland Richardson (LRICHARDSON7@mgh.harvard.edu) to arrange delivery (or phone 732-600-6514) Monday through Friday 8:00am - 4:00 pm.

9.2.1.4 Shipping of Specimen(s)

Dr. Curry's lab will arrange shipping to Dr. Hacohen's laboratory (Broad Institute, 7 Cambridge Center, Cambridge MA).

9.2.1.5 Site(s) Performing Correlative Study

Dr. Nir Hacohen's lab in conjunction with Dr. William Curry's lab will be responsible for analyzing the tissue and blood.

9.2.2 Tumor Tissue

For a subset of tumor samples (N=5 initially that will be identified once patients have consented to treatment and determined to have sufficient tissue), we will isolate myeloid cells (using a Lin- antibody cocktail to exclude all lymphocytes- CD56, CD2, CD3, CD19 and CD20) and use single cell RNA-seq (scRNA-seq) to characterize the types and activation status of these cells, as well as capture the paired TCR repertoire from T cells. If fresh tissue is not available, we will isolate RNA from fixed tissue and perform bulk sequencing of TCRs for the T cell repertoire, and mRNA for gene expression, from which we will infer myeloid cell populations and TCR repertoire. Fresh or fixed tissue will be obtained from pathology from the original surgery for analysis. Currently, pre-surgical patients are consented under DFHCC Protocol 10-417 to contribute fresh tissue as part of a tissue bank and we will petition this bank for fresh tissue. For patients requiring repeat resection while on study treatment, we will obtain fresh frozen tissue for analyze and comparison to the initial tumor tissue. Ideally we will obtain ~1cm^3 of tissue but if that amount of tissue is not available, Dr. Curry will review with Pathology what is available and determine if sufficient for the planned analyses.

9.2.2.1 Collection of Specimen(s)

Fresh frozen tissue or fixed if fresh is not available will be obtained at the time of diagnostic surgery or repeat surgery.

9.2.2.2 Handling of Specimens(s)

At the time of surgery, after confirmation that adequate specimen has been obtained for pathology review, fresh tumors will be collected and the presence of malignant cells will be confirmed in frozen sections on adjacent, representative pieces of tissue. Fresh tumor tissue will be delivered to Dr. Hacohen's laboratory minced with a scalpel and enzymatically dissociated using a gentle papain-based brain tumor dissociation kit (Miltenyi Biotec). From these cells, we

will isolate myeloid cells (using a Lin- antibody cocktail to exclude all lymphocytes- CD56, CD2, CD3, CD19 and CD20) and use single cell RNA-seq (scRNA-seq) to characterize the types and activation status of these cells, as well as capture the paired TCR repertoire from T cells.

9.2.2.3 Shipping of Specimen(s)

Tumor tissue will be delivered to Dr. Hacohen's laboratory at the Broad Institute. Please contact Karin Pelka 617-335-1190 (or backup is Moshe Sade-Feldman 857-500-0130) to arrange shipment.

9.2.2.4 Site(s) Performing Correlative Study

Dr. Nir Hacohen's lab in conjunction with Dr. William Curry's lab will be responsible for analyzing the tissue and blood.

10. STUDY CALENDAR

Baseline evaluations are to be conducted within 2 weeks prior to start of protocol therapy. Scans must be done ≤2 weeks prior to the start of therapy. In the event that the participant's condition is deteriorating, laboratory evaluations should be repeated within 48 hours prior to initiation of the next cycle of therapy.

Assessments must be performed prior to administration of any study agent. Study assessments and agents should be administered within ± 3 days of the protocol-specified date, unless otherwise noted.

Cycle 1: Radiation + temozolomide + bavituximab

	Pre-Study	Wk 1[g]	Wk 2	Wk 3	Wk 4	Wk 5	Wk 6	Off Treatment[e]
Bavituximab		X	X	X	X	X	X	
Temozolomide		X	X	X	X	X	X	
Radiation [J]		X	X	X	X	X	X	
Informed consent	X							
Demographics	X							
Medical history	X							
Concurrent meds	X	X		X			X	
Physical exam[l]	X	X		X			X	X
Vital signs[l]	X	X	X	X	X	X	X	X
Height	X							
Weight[l]	X	X		X			X	
Performance status[l]	X	X		X			X	X
Drug diary for temozolomide[h]		X	X	X	X	X	X	

CBC w/diff, plts	X	X	X	X	X	X	X	X
Serum chemistry[a]	X			X				X
PT/INR/aPTT	X							
ECG (as indicated)[i]	X							
Adverse event evaluation		X		X			X	X
Tumor measurements via MRI [b]	X							X
B-HCG [c]	X							
Tissue [d, k]	X							
Correlative Blood Draws [f]	X						X	

a- Serum chemistry panel should include albumin, alkaline phosphatase, total bilirubin, BUN, chloride, creatinine, glucose, potassium, total protein, AST/SGOT, ALT/SGPT, sodium.
b- MRI to be done at the Martinos Center for Biomedical Imaging
c – Serum Pregnancy Test (for women of childbearing potential)
d – Assessment to determine if there is sufficient tissue for correlative studies. Tissue may be sent as a batch later to minimize shipping requirements. Tissue is not required for registration.
e- This column included in case patient goes off study during cycle 1. This visit can occur within 2 weeks of the participant coming off study.
f – See section 9.2. Blood will be drawn prior to start of chemoradiation and during week 6 of chemoradiation.
g- First bavituximab infusion should happen after the 4th radiation treatment and before the 6th treatment.
h- See Appendix B for the required drug diary.
i – if ECG is clinically indicated, the QTc-B rather than QTc-F should be used to look for abnormalities
J – Radiation dose not have to be done at a DFHCC site but should be considered to allow for ease of dosing bavituximab during radiation
K – MGMT status will be collected but is not required for registration
l – if it is determined that a patient will not be coming in for bavituximab on a specific day (ex. due to treatment hold or scheduling conflicts), then the patient does not need to return to clinic solely for the physical exam, weight, KPS, and vital sign assessments as theses were intended to be done prior to the infusion as a safety check.

Cycle 2: Post-Radiation – bavituximab monotherapy
Labs should be drawn up to 3 days prior to bavituximab dosing.

	Wk 1	Wk 2	Wk 3	Wk 4	Off Treatment[e]
Bavituximab	X	X	X	X	
Concurrent meds		X		X	
Physical exam[g]		X		X	X
Vital signs[g]	X	X	X	X	X

Weight[g]		X		X	
Performance status[g]		X		X	X
CBC w/diff, plts	X				X
Serum chemistry[a]	X				X
Adverse event evaluation		X		X	X
Tumor measurements via MRI [b]				X	X
Correlative Blood draws [f]	X				

a- Serum chemistry panel should include albumin, alkaline phosphatase, total bilirubin, BUN, chloride, creatinine, glucose, potassium, total protein, AST/SGOT, ALT/SGPT, sodium.
b- MRI to be done at the Martinos Center for Biomedical Imaging. This should be done at any time during the 4-5 weeks after completion of radiation but within 1 week of starting post-RT temozolomide.
e- This column included in case patient goes off study during cycle 2. This visit can occur within 2 weeks of the participant coming off study.
f – See section 9.2
g – if it is determined that a patient will not be coming in for bavituximab on a specific day (ex. due to treatment hold or scheduling conflicts), then the patient does not need to return to clinic solely for the physical exam, weight, KPS, and vital sign assessments as theses were intended to be done prior to the infusion as a safety check.

Cycle 3: Monthly temozolomide + bavituximab therapy

	Wk 1	Wk 2	Wk 3	Wk 4	EOT/Off Study[e]	Follow-up[i]
Bavituximab	X	X	X	X		
Temozolomide[g]	X					
Drug diary for temozolomide[h]	X					
Concurrent meds		X				
Physical exam[k]		X		X	X	
Vital signs[k]	X	X	X	X	X	
Height						
Weight[k]		X			X	
Performance status[k]		X		X	X	
CBC w/diff, plts[j]			X	X	X	
Serum chemistry[a]	X				X	
EKG/coag panel (as indicated)						
Adverse event evaluation		X		X		
Tumor measurements via MRI[b]						X

Correlative Blood draws [f]	X					

a- Serum chemistry panel should include albumin, alkaline phosphatase, total bilirubin, BUN, chloride, creatinine, glucose, potassium, total protein, AST/SGOT, ALT/SGPT, sodium.
b- EOT MRI to be done at the Martinos Center for Biomedical Imaging if possible
e- EOT = end of treatment. For patients removed from the study for one of the criteria in Section 5.5 during cycle 3 or 4 or has completed treatment after cycle 4, this information should be collected within 28 days of the last bavituximab dose. Follow-up of AE resolution to </= grade 1 or baseline may be beyond 28 days.
f – See section 9.2
g- Temozolomide is administered for 5 consecutive days during week 1 of the cycle
h - See Appendix B for the required drug diary.
i – Participants will be followed for progression via MRI or clinic assessment per standard of care which is typically every 8 weeks as well as overall survival for up to 3 years. MRIs will be collected for central review. If MRI is not performed at Martinos, it should be sent to the study PI on a DVD or contact Dr. Gerstner for electronic transfer instructions.
j – CBC with diff should align with standard of care assessments on day 21± 3 days and day 28 ± 3 days of each cycle.
K - if it is determined that a patient will not be coming in for bavituximab on a specific day (ex. due to treatment hold or scheduling conflicts), then the patient does not need to return to clinic solely for the physical exam, weight, KPS, and vital sign assessments as theses were intended to be done prior to the infusion as a safety check.

Cycle 4: Monthly temozolomide + bavituximab therapy

	Wk 1	Wk 2	Wk 3	Wk 4	EOT/Off Study[e]	Follow-up[i]
Bavituximab	X	X	X	X		
Temozolomide[g]	X					
Drug diary for temozolomide[h]	X					
Concurrent meds		X				
Physical exam [k]		X		X	X	
Vital signs [k]	X	X	X	X	X	
Height						
Weight [k]		X			X	
Performance status [k]		X		X	X	
CBC w/diff, plts[j]			X	X	X	
Serum chemistry[a]	X				X	
EKG/coag panel (as indicated)						
Adverse event evaluation		X		X		
Tumor measurements via MRI [b]				X	X	X
Correlative Blood draws [f]	X					

a- Serum chemistry panel should include albumin, alkaline phosphatase, total bilirubin, BUN,

chloride, creatinine, glucose, potassium, total protein, AST/SGOT, ALT/SGPT, sodium.
b- MRI to be done at the Martinos Center for Biomedical Imaging. The MRI scan should be performed at any time during the 4th week of the cycle.
e- EOT = end of treatment. For patients removed from the study for one of the criteria in Section 5.5 during cycle 3 or 4 or has completed treatment after cycle 4, this information should be collected within 28 days of the last bavituximab dose. Follow-up of AE resolution to </= grade 1 or baseline may be beyond 28 days.
f – See section 9.2
g- Temozolomide is administered for 5 consecutive days during week 1 of the cycle
h - See Appendix B for the required drug diary.
i – Participants will be followed for progression via MRI or clinic assessment per standard of care which is typically every 8 weeks as well as overall survival for up to 3 years. MRIs will be collected for central review. If MRI is not performed at Martinos, it should be sent to the study PI on a DVD or contact Dr. Gerstner for electronic transfer instructions.
j – CBC with diff should align with standard of care assessments on day 21± 3 days and day 28 ± 3 days of each cycle.
k- if it is determined that a patient will not be coming in for bavituximab on a specific day (ex. due to treatment hold or scheduling conflicts), then the patient does not need to return to clinic solely for the physical exam, weight, KPS, and vital sign assessments as theses were intended to be done prior to the infusion as a safety check.

Please note, patients may continue bavituximab beyond 18 weeks at the discretion of the treating physician if the patient is deriving clinical benefit. If the patient also continues temozolomide, the dosing of concomitant temozolomide will be per institutional standard of care so not included in the Table for Cycle 5 and beyond but can be given in conjunction with bavituximab if previously tolerated.

Cycle 5 and beyond: Bavituximab therapy

	Wk 1	Wk 2	Wk 3	Wk 4	EOT/Off Study[e]	Follow-up[i]
Bavituximab[k]	X	X	X	X		
Concurrent meds		X				
Physical exam [l]		X		X	X	
Vital signs [l]	X	X	X	X	X	
Height						
Weight				X	X	
Performance status				X	X	
CBC w/diff, plts				X	X	
Serum chemistry[a]	X				X	
EKG/coag panel (as indicated)						
Adverse event evaluation		X		X		
Tumor measurements via MRI [b]				X[b]	X	X

Correlative Blood draws [f]	X					

a- Serum chemistry panel should include albumin, alkaline phosphatase, total bilirubin, BUN, chloride, creatinine, glucose, potassium, total protein, AST/SGOT, ALT/SGPT, sodium.
b- If possible, MRI to be done at the Martinos Center for Biomedical Imaging. The MRI scan should be performed at any time during the 4th week of every other cycle, starting with Cycle 6, to align with standard of care timepoints. If MRI during the study duration is not performed at Martinos, it should be sent to the study PI on a DVD or contact Dr. Gerstner for electronic transfer instructions.
e- EOT = end of treatment. For patients removed from the study for one of the criteria in Section 5.5 during cycle 5 or beyond, this information should be collected within 28 days of the last bavituximab dose. Follow-up of AE resolution to </= grade 1 or baseline may be beyond 28 days.
f – See section 9.2
i – Participants will be followed for progression via MRI or clinic assessment per standard of care which is typically every 8 weeks as well as overall survival for up to 3 years. MRIs will be collected for central review.
k – Weekly bavituximab infusion until progression or as outlined in Section 5.5.
l - if it is determined that a patient will not be coming in for bavituximab on a specific day (ex. due to treatment hold or scheduling conflicts), then the patient does not need to return to clinic solely for the physical exam, weight, KPS, and vital sign assessments as theses were intended to be done prior to the infusion as a safety check.

11. MEASUREMENT OF EFFECT

11.1 Antitumor Effect – Solid Tumors

For the purposes of this study, patients should be reevaluated for response every 8 weeks following completion of chemoradiation.

11.1.1 Definitions

Evaluable for Target Disease response. Only those participants who have measurable disease present at baseline, have received at least one cycle of therapy, and have had their disease re-evaluated will be considered evaluable for target disease response. These participants will have their response classified according to the definitions stated below. (Note: Participants who exhibit objective disease progression prior to the end of cycle 1 will also be considered evaluable.)

Evaluable Non-Target Disease Response. Participants who have lesions present at baseline that are evaluable but do not meet the definitions of measurable disease, have received at least one cycle of therapy, and have had their disease re-evaluated will be considered evaluable for non-target disease. The response assessment is based on the presence, absence, or unequivocal progression of the lesions.

11.1.2 Disease Parameters

Measurable disease. Measurable disease is defined as bidimensionally contrast enhancing lesions with clearly defined margins by CT or MRI scan, with two perpendicular diameters of at least 10 mm, visible on two or more axial slices that are preferably, at most, 5 mm apart with 0-mm skip. All tumor measurements must be recorded in millimeters (or decimal fractions of centimeters).

Non-measurable disease. All other lesions (or sites of disease), including small lesions (longest diameter <10 mm), are considered non-measurable disease. Bone lesions, leptomeningeal disease, ascites, pleural/pericardial effusions, lymphangitis cutis/pulmonitis, inflammatory breast disease, abdominal masses (not followed by CT or MRI), and cystic lesions are all considered non-measurable.

'Cystic lesions' thought to represent cystic metastases can be considered as measurable lesions, if they meet the definition of measurability described above. However, if non-cystic lesions are present in the same participant, these are preferred for selection as target lesions.

11.1.3 Methods for Evaluation of Disease

All measurements should be taken and recorded in metric notation using a ruler, calipers, or a digital measurement tool. All baseline evaluations should be performed as closely as possible to the beginning of treatment and never more than 4 weeks before the beginning of the treatment.

The same method of assessment and the same technique should be used to characterize each identified and reported lesion at baseline and during follow-up.

11.1.4 Response Criteria

Response and progression will be evaluated in this study using the Response Assessment in Neuro-Oncology Criteria (RANO) and incorporating the updated iRANO criteria proposed specifically for brain tumors treated with immunotherapy as it allows for early inflammation- related changes and potential for delayed responses.

11.1.4.1 Evaluation of Target Lesions

Complete Response (CR): Complete disappearance of all enhancing tumor determined by two consecutive MRI scans not less than 4 weeks apart and no corticosteroid usage with a stable or improved neurologic examination for a minimum of 4 weeks.

Partial Response (PR): Greater than or equal to 50% reduction in tumor size of all measurable lesions on a stable or decreasing dose of corticosteroids, with a stable or improving neurologic examination for a minimum of 4 weeks.

Progressive Disease (PD): Progressive neurologic abnormalities not explained by causes unrelated to tumor progression (example: anti-epileptic drug or corticosteroid toxicity, electrolyte abnormalities, hyperglycemia, etc.) or a greater than 25% increase in the volume of the tumor by MRI scan. If neurologic status deteriorates on a stable or increasing dose of corticosteroids, or if new lesions appear on serial MRI scans, this will also be considered PD.

iRANO recommends confirmation of disease progression on follow-up imaging 3 months after initial radiographic progression if there is no new or substantially worsened neurological deficits that are not due to comorbid events or concurrent medication, and it is 6 months or less from starting immunotherapy. This includes if there is appearance of new lesions that could represent a transient inflammatory response. If progression is subsequently confirmed, the progression date should be the date the new lesion or concern for progression occurred.

Stable Disease (SD): A patient whose clinical status and MRI measurements do not meet the criteria for CR, PR or PD.

Unknown (UN): Assessment of target lesions cannot be made due to insufficient or unevaluable data. In this case, a concise explanation must be given.

Note: If tumor response data is missing for target lesions, the overall assessment must be UN unless there is new disease that would result in an overall assessment of PD. However, if there is missing or unevaluable data for non-target lesions, but data is available for all target lesions, the overall response for that time point will be assigned based on the sum LD of all target lesions. Additionally, the assessment of CR cannot be made if there is missing or unevaluable data for non-target lesions. In this case, the overall assessment would be PR.

11.1.4.2 Evaluation of Non-Target Lesions

NA

11.1.4.3 Evaluation of New Lesions

The finding of a new lesion should be unequivocal (i.e. not due to difference in scanning technique, imaging modality, or findings thought to represent something other than tumor (for example, some ‘new’ bone lesions may be simply healing or flare of pre-existing lesions). However, a lesion identified on a follow-up scan in an anatomical location that was not scanned at baseline is considered new and will indicate PD. If a new lesion is equivocal (because of small size etc.), follow-up evaluation will clarify if it truly represents new disease and if PD is confirmed,

progression should be declared using the date of the initial scan on which the lesion was discovered.

11.1.4.4 Evaluation of Best Overall Response

The best overall response is the best response recorded from the start of the treatment until disease progression/recurrence (taking as reference for progressive disease the smallest measurements recorded since the treatment started).

11.1.5 Duration of Response

Duration of overall response: The duration of overall response is measured from the time measurement criteria are met for CR or PR (whichever is first recorded) until the first date that recurrent or progressive disease is objectively documented (taking as reference for progressive disease the smallest measurements recorded since the treatment started, or death due to any cause. Participants without events reported are censored at the last disease evaluation).

Duration of overall complete response: The duration of overall CR is measured from the time measurement criteria are first met for CR until the first date that progressive disease is objectively documented, or death due to any cause. Participants without events reported are censored at the last disease evaluation.

Duration of stable disease: Stable disease is measured from the start of the treatment until the criteria for progression are met, taking as reference the smallest measurements recorded since the treatment started, including the baseline measurements.

11.1.6 Progression-Free Survival

Overall Survival: Overall Survival (OS) is defined as the time from start of treatment to death due to any cause, or censored at date last known alive.

Progression-Free Survival: Progression-Free Survival (PFS) is defined as the time from start of treatment to the earlier of progression or death due to any cause. Participants alive without disease progression are censored at date of last disease evaluation.

Time to Progression: Time to Progression (TTP) is defined as the time from start of treatment to progression, or censored at date of last disease evaluation for those without progression reported.

11.1.7 Response Review

Response will be determined by the treating physician but all MRIs scans will be collected for later, central review of response by Dr. Gerstner and her team members.

12. DATA REPORTING / REGULATORY REQUIREMENTS

Adverse event lists, guidelines, and instructions for AE reporting can be found in Section 7.0 (Adverse Events: List and Reporting Requirements).

12.1 Data Reporting

12.1.1 Method

The Office of Data Quality (ODQ) will collect, manage, and perform quality checks on the data for this study.

12.1.2 Responsibility for Data Submission

Investigative sites within DF/HCC or DF/PCC are responsible for submitting data and/or data forms to the Office of Data Quality in accordance with DF/HCC SOPs.

12.2 Data Safety Monitoring

The DF/HCC Data and Safety Monitoring Committee (DSMC) will review and monitor toxicity and accrual data from this study. The committee is composed of clinical specialists with experience in oncology and who have no direct relationship with the study. Information that raises any questions about participant safety will be addressed with the Overall PI and study team.

The DSMC will review each protocol up to four times a year or more often if required to review toxicity and accrual data. Information to be provided to the committee may include: up-to-date participant accrual; current dose level information; DLT information; all grade 2 or higher unexpected adverse events that have been reported; summary of all deaths occurring with 30 days of intervention for Phase I or II protocols; for gene therapy protocols, summary of all deaths while being treated and during active follow-up; any response information; audit results, and a summary provided by the study team. Other information (e.g. scans, laboratory values) will be provided upon request.

Although an increase in toxicity is not anticipated with combination radiation, temozolomide, and bavituximab based on the mechanisms of action of these therapies and prior experience combining bavituximab with other cytotoxic chemotherapies, we will closely monitor patients for any unanticipated adverse events. Specifically, if 5 or more patients out of the first 20 treated and followed over a duration of 4 weeks are observed with CTCAE grade 4 or higher toxicity that is unexpected (i.e. not expected due to radiation, temozolomide, or bavituximab), then the treatment will be halted to determine if any changes need to be made to the design of the trial. This rule provides a one-sided 95% upper limit of 45.5%. Toxicities expected and related to temozolomide – specifically thrombocytopenia, leukopenia, lymphopenia, neutropenia, nausea or vomiting – will not be included in this early stopping rule since these are expected from temozolomide in rates as high as 46% of patients (Stupp 2005). Alopecia from radiation will also

not be included and neither will toxicities related to the underlying disease itself (ex. seizures). For any toxicity that may arise, the PI will review all unexpected CTCAE grade ≥3 AEs to determine if any would warrant halting the trial. As stated above the DFHCC DSMB will also be monitoring this trial as an independent review.

13. STATISTICAL CONSIDERATIONS

13.1 Study Design/Endpoints

The primary endpoint will be the proportion of patients alive at 12 months (OS12). OS12 was selected as the primary endpoint as this avoids the confound of measuring progression in immunotherapy trials where pseudoprogression can complicate the analysis. OS12 will be defined from the time of study entry to death from any cause. Patients will be enrolled as one cohort (i.e. 1 stage design).

Recent studies in newly diagnosed GBM testing novel agents have suggested that OS12 is ~60% (Gilbert 2014, Lee 2015, Gilbert 2013). This number serves as a framework for our sample size calculations. The primary outcome will be evaluated by the Kaplan-Meier estimate as well a simple proportion. If the true OS12 increased by 20%, there would definitely be interest in further investigation of bavituximab. This study will test the null hypothesis (H0) with power and significance level of 0.89 and 0.09 respectively, that OS12 is ≤ 60% versus the alternative hypothesis (Ha) that OS12 is ≥ 80%. The total evaluable sample size is 33 with a total of 36 potential patients to be enrolled if there are inevaluable patients. Inevaluable patients are defined as patients who drop out prior to receiving bavituximab. Data for subjects who are lost to follow up prior to documented death will be counted as deaths at the last assessment date when the subject is known to be alive. Given our patient population, we anticipate the number of patients lost to follow-up to be very small and we will able to establish vital status at 12 months (the primary endpoint) even if patients drop out prior to 12 months. Vital status can be determined by searching death records as well.

We will test the relationship between other prognostic factors (specifically MGMT methylation status, age) and survival by using a Cox proportional hazards model. Another prognostic factor, KPS, was not included because all the patients have to have a good functional status to enroll.

13.2 Sample Size, Accrual Rate and Study Duration

A total of 36 evaluable participants will be accrued within 2 years, anticipating 3 patients enrolled per month. An additional 12 months of follow-up will be required on the last participant accrued to observe the participant's OS12. Patients will be followed for an additional 3 years for progression and survival data for secondary endpoint analysis.

Accrual targets below reflect the demographics of our area as well as the slight preponderance of GBM in males.

Accrual Targets	
Ethnic Category	**Sex/Gender**

	Females		Males		Total
Hispanic or Latino	2	+	3	=	5
Not Hispanic or Latino	14	+	17	=	31
Ethnic Category: Total of all subjects	16	+	20	=	36
Racial Category					
American Indian or Alaskan Native	0	+	0	=	0
Asian	2	+	3	=	5
Black or African American	2	+	2	=	4
Native Hawaiian or other Pacific Islander	0	+	0	=	0
White	11	+	15	=	27
Racial Category: Total of all subjects	16	+	20	=	36

13.3 Analysis of Primary Endpoints

See above – Section 13.1

13.4 Analysis of Secondary Endpoints

The radiographic response proportion (RR), progression-free survival (PFS), overall survival (OS), and toxicity proportion will each be described with 95% confidence limits. Radiographic response is defined as a complete response or partial response, as determined by the RANO criteria with incorporation of iRANO criteria (Okada 2015). The radiographic response rate is the proportion of subjects who have a radiographic response. Data for subjects without disease progression or death at the time of analysis or who are lost to follow up prior to the documented disease progression will be censored at the last tumor assessment date when the subject is known to be progression free. PFS is defined as the time of study entry to time of disease progression. Overall survival is defined as the time of study entry to death from any cause. Data for subjects who are lost to follow up will be censored at the date when the subject is last known to be alive. Kaplan-Meier methods will be used to estimate PFS/OS.

As data permit, exploratory analyses of blood biomarkers and MRI parameters will be summarized by descriptive statistics, including mean, median and standard deviation. Differences in these biomarkers between responders and non-responders will be compared with parametric or nonparametric techniques as permitted by the data. For correlative analyses, Cox proportional hazards model will be used to explore the relationship between these biomarkers measured at baseline and PFS and OS. Change in biomarkers during treatment will also be explored and their ability to serve as pharmacodynamics markers of response will be assessed.

13.5 Reporting and Exclusions

13.5.1 Evaluation of Toxicity

All participants will be monitored for 90 days from completion of radiation in order to assess for any AEs related to radiation and bavituximab regardless of duration of bavituximab.

Otherwise, all participants will be evaluable for toxicity from the time of their first treatment until 30 days after the last dose of bavituximab.

13.5.2 Evaluation of the Primary Efficacy Endpoint

Intent to treat analysis will be used for the primary endpoint.

14. PUBLICATION PLAN

The results should be made public within 24 months of reaching the end of the study. The end of the study is the time point at which the last data items are to be reported, or after the outcome data are sufficiently mature for analysis, as defined in the section on Sample Size, Accrual Rate and Study Duration. If a report is planned to be published in a peer-reviewed journal, then that initial release may be an abstract that meets the requirements of the International Committee of Medical Journal Editors. A full report of the outcomes should be made public no later than three (3) years after the end of the study.

REFERENCES

Batchelor TT, Gerstner ER, Emblem KE, Duda DG, Kalpathy-Cramer J, Snuderl M, Ancukiewicz M, Polaskova P, Pinho MC, Jennings D, Plotkin SR, Chi AS, Eichler AF, Dietrich J, Hochberg FH, Lu-Emerson C, Iafrate AJ, Ivy SP, Rosen BR, Loeffler JS, Wen PY, Sorensen AG, Jain RK (2013) Improved tumor oxygenation and survival in glioblastoma patients who show increased blood perfusion after cediranib and chemoradiation. Proc Natl Acad Sci U S A 110: 19059-19064.

Batchelor TT, Sorensen AG, di Tomaso E, Zhang WT, Duda DG, Cohen KS, Kozak KR, Cahill DP, Chen PJ, Zhu M, Ancukiewicz M, Mrugala MM, Plotkin S, Drappatz J, Louis DN, Ivy P, Scadden DT, Benner T, Loeffler JS, Wen PY, Jain RK (2007) AZD2171, a pan-VEGF receptor tyrosine kinase inhibitor, normalizes tumor vasculature and alleviates edema in glioblastoma patients. Cancer Cell 11: 83-95

Beck A W, Luster TA, Miller AF, et al. Combination of a monoclonal anti-phosphatidylserine antibody with gemcitabine strongly inhibits the growth and metastasis of orthotopic pancreatic tumors in mice. Int J Cancer 2006; 118(10):2639-42.

Birge RB, Boeltz S, Kumar S, et al. Phosphatidylserine is a global immunosuppressive signal in efferocytosis, infectious disease, and cancer. Cell Death Differ. 2016;23(6):962-78.

Blanco VM, Chu Z, Vallabhapurapu SD, Sulaiman MK, Kendler A, Rixe O, Warnick RE, Franco RS, Qi X (2014) Phosphatidylserine-selective targeting and anticancer effects of SapC-DOPS nanovesicles on brain tumors. Oncotarget. 2014 Aug 30;5(16):7105-18.

Blanco VM, Curry R, Qi X (2015) SapC-DOPS nanovesicles: a novel targeted agent for the imaging and treatment of glioblastoma. Oncoscience 2: 102-110

Burrows FJ, Thorpe PE. Vascular targeting--a new approach to the therapy of solid tumors. Pharmacol Ther. 1994;64(1):155-74.

CBTRUS (2008) Statistical report: primary brain tumors in the United States, 2000-2004. Central Brain Tumor Registry of the United States.

Chalasani, P, Marron M, Roe D, Clarke K, Iannone M, Livingston RB, et al. A phase I clinical trial of bavituximab and paclitaxel in patients with HER2-negative metastatic breast cancer. Cancer Med. 2015 Mar 31.

Curry WT, Jr., Gorrepati R, Piesche M, Sasada T, Agarwalla P, Jones PS, Gerstner ER, Golby AJ, Batchelor TT, Wen PY, Mihm MC, Dranoff G (2016) Vaccination with Irradiated Autologous Tumor Cells Mixed with Irradiated GM-K562 Cells Stimulates Antitumor Immunity and T Lymphocyte Activation in Patients with Recurrent Malignant Glioma. Clin Cancer Res 22: 2885-2896 doi:10.1158/1078-0432.CCR-15-2163

Digernes I, Saxhaug C, Geier O, Reitan E, Waldeland E, Hole K, Jacobsen K, Helland A, Emblem K. Immunotherapy revised; Ipilimumab potentiates the vascular response to stereotactic radiosurgery in patients with brain metastases. AACR 2016: abstract 1471.

Denekamp J. Angiogenesis, neovascular proliferation and vascular pathophysiology as targets for cancer therapy. Br J Radiol. 1993;66(783):181-96.

DeRose P, Thorpe PE, Gerber DE. Development of bavituximab, a vascular targeting agent with immune-modulating properties, for lung cancer treatment. Immunotherapy. 2011;3(8):933-44.

Emblem KE, Farrar CT, Gerstner ER, Batchelor TT, Borra RJ, Rosen BR, Sorensen AG, Jain RK (2014) Vessel caliber--a potential MRI biomarker of tumour response in clinical trials. Nature reviews Clinical oncology 11: 566-584

Gerber DE, Spigel DR, Giorgadze D, Shtivelband M, Ponomarova OV, Shan JS, Menander KB, Belani CP. Docetaxel Combined With Bavituximab in Previously Treated, Advanced Nonsquamous Non-Small-Cell Lung Cancer. Clin Lung Cancer. 2016 May;17(3):169-76.

Gerstner E, Zhang Z, Fink J, Muzi M, Hanna L, Greco E, Prah M, Schmainda K, Mintz A, Kostakoglu L, Eikman EA, Ellingson BM, Ratai EM, Sorensen AG, Barboriak D, Mankoff DA. ACRIN 6684: Assessment of tumor hypoxia in newly diagnosed GBM using 18F-FMISO PET and MRI. Clin Cancer Res. 2016 Oct 15;22(20):5079-5086.

Gerstner ER, Ye X, Duda DG, Levine MA, Mikkelsen T, Kaley TJ, Olson JJ, Nabors BL, Ahluwalia MS, Wen PY, Jain RK, Batchelor TT, Grossman S (2015) A phase I study of cediranib in combination with cilengitide in patients with recurrent glioblastoma. Neuro Oncol 17: 1386-1392

Gilbert MR, Sulman EP, Mehta MP (2014) Bevacizumab for newly diagnosed glioblastoma. N Engl J Med 370: 2048-2049

Gilbert MR, Wang M, Aldape KD, Stupp R, Hegi ME, Jaeckle KA, Armstrong TS, Wefel JS, Won M, Blumenthal DT, Mahajan A, Schultz CJ, Erridge S, Baumert B, Hopkins KI, Tzuk-Shina T, Brown PD, Chakravarti A, Curran WJ, Jr., Mehta MP (2013) Dose-dense temozolomide for newly diagnosed glioblastoma: a randomized phase III clinical trial. J Clin Oncol 31: 4085-4091.

He J, Yin Y, Luster TA, Watkins L, Thorpe PE (2009) Antiphosphatidylserine antibody combined with irradiation damages tumor blood vessels and induces tumor immunity in a rat model of glioblastoma. Clin Cancer Res. Nov 15;15(22):6871-80.

Huang X, Bennet M, Thorpe PE. A monoclonal antibody that binds anionic phospholipids on tumor blood vessels enhances the antitumor effect of docetaxel on human breast tumors in mice. Cancer Res 2005; 65:4408-4416.

Hsu MS, Sedighim S, Wang T, Antonios JP, Everson RG, Tucker AM, Du L, Emerson R, Yusko

E, Sanders C, Robins HS, Yong WH, Davidson TB, Li G, Liau LM, Prins RM (2016) TCR Sequencing Can Identify and Track Glioma-Infiltrating T Cells after DC Vaccination. Cancer Immunol Res 4: 412-418.

Jain M, Raizada N, Kuttan R, et al. Phase II study of bavituximab plus paclitaxel and carboplatin in locally advanced or metastatic breast cancer; interim results. J Clin Oncol 2010; 28(15s) Abstract 1062.

Lee EQ, Kaley TJ, Duda DG, Schiff D, Lassman AB, Wong ET, Mikkelsen T, Purow BW, Muzikansky A, Ancukiewicz M, Huse JT, Ramkissoon S, Drappatz J, Norden AD, Beroukhim R, Weiss SE, Alexander BM, McCluskey CS, Gerard M, Smith KH, Jain RK, Batchelor TT, Ligon KL, Wen PY (2015) A Multicenter, Phase II, Randomized, Noncomparative Clinical Trial of Radiation and Temozolomide with or without Vandetanib in Newly Diagnosed Glioblastoma Patients. Clin Cancer Res 21: 3610-3618

Li W, Graeber MB (2012) The molecular profile of microglia under the influence of glioma. Neuro Oncol 14: 958-978

Nida Hussain HD, Subrahmanya Vallabhapurapu, Zhengtao Chu, Victor Blanco and Xiaoyang Qi (2015) ATPS-35 RADIATION TREATMENT INCREASES PHOSPHATIDYLSERINE EXTERNALIZATION ON GLIOBLASTOMA CELLS – INDICATES POTENTIAL TARGET FOR THERAPY. Neuro Oncol 17 (suppl 5): v25-v26

Ran S, He J, Huang X, Soares M, et al. Anti-tumor effects of a monoclonal antibody directed against anionic phospholipids on the surface of tumor blood vessels in mice. Clin Cancer Res 2005; 11:1551-1162.

Okada H, Weller M, Huang R, Finocchiaro G, Gilbert MR, Wick W, Ellingson BM, Hashimoto N, Pollack IF, Brandes AA, Franceschi E, Herold-Mende C, Nayak L, Panigrahy A, Pope WB, Prins R, Sampson JH, Wen PY, Reardon DA (2015) Immunotherapy response assessment in neuro-oncology: a report of the RANO working group. The Lancet Oncology 16: e534-542

Rooney MS, Shukla SA, Wu CJ, Getz G, Hacohen N (2015) Molecular and genetic properties of tumors associated with local immune cytolytic activity. Cell 160: 48-61.

Stupp R, Mason WP, van den Bent MJ et al. Radiotherapy plus Concomitant and Adjuvant Temozolomide for Glioblastoma. N Engl J Med 2005;352:987-96.

Sorensen AG, Emblem KE, Polaskova P, Jennings D, Kim H, Ancukiewicz M, Wang M, Wen PY, Ivy P, Batchelor TT, Jain RK (2012) Increased survival of glioblastoma patients who respond to antiangiogenic therapy with elevated blood perfusion. Cancer Res 72: 402-407

Tabagari D, Nemsadze G, Janjalia M, et al. Phase II study of bavituximab plus docetaxel in locally advanced or metastatic breast cancer. J Clin Oncol 2010; 28(15s) Abstract 1042.

Winter PM, Pearce J, Chu Z, McPherson CM, Takigiku R, Lee JH, Qi X. Imaging of brain

tumors with paramagnetic vesicles targeted to phosphatidylserine. J Magn Reson Imaging. 2015 Apr;41(4):1079-87.

Wojton J, Chu Z, Mathsyaraja H, Meisen WH, Denton N, Kwon CH, Chow LM, Palascak M, Franco R, Bourdeau T, Thornton S, Ostrowski MC, Kaur B, Qi X (2013) Systemic delivery of SapC-DOPS has antiangiogenic and antitumor effects against glioblastoma. Mol Ther. 2013 Aug;21(8):1517-25.

Wojton J, Meisen WH, Jacob NK, Thorne AH, Hardcastle J, Denton N, Chu Z, Dmitrieva N, Marsh R, Van Meir EG, Kwon CH, Chakravarti A, Qi X, Kaur B (2014) SapC-DOPS-induced lysosomal cell death synergizes with TMZ in glioblastoma. Oncotarget. 2014 Oct 30;5(20):9703-9.

Yin Y, Huang X, Lynn KD, Thorpe PE (2013) Phosphatidylserine-targeting antibody induces M1 macrophage polarization and promotes myeloid-derived suppressor cell differentiation. Cancer Immunol Res 1: 256-268

Yin Y, Huang X, Barbero G, et al. Phosphatidylserine-targeting antibody reactivates tumor immunity and destroys tumor vasculature in mice. American Association of Cancer Research Meeting 2013; Poster #1244.

Yopp, AC, Singal, AG, Yull, EA, Udit, NV, Kallinteris, NL, Shan, J, Beg, S, Mansour, JC, Zhu, H. A phase II study of bavituximab and sorafenib in advanced hepatocellular carcinoma (HCC). J Clin Oncol 2015; Abstract 4109.

Zhang L, Habib AA, Zhao D. Phosphatidylserine-targeted liposome for enhanced glioma-selective imaging. Oncotarget. 2016 Jun 21;7(25):38693-38706.

APPENDIX A PERFORMANCE STATUS CRITERIA

ECOG Performance Status Scale		**Karnofsky Performance Scale**	
Grade	Descriptions	Percent	Description
0	Normal activity. Fully active, able to carry on all pre-disease performance without restriction.	100	Normal, no complaints, no evidence of disease.
		90	Able to carry on normal activity; minor signs or symptoms of disease.
1	Symptoms, but ambulatory. Restricted in physically strenuous activity, but ambulatory and able to carry out work of a light or sedentary nature (*e.g.*, light housework, office work).	80	Normal activity with effort; some signs or symptoms of disease.
		70	Cares for self, unable to carry on normal activity or to do active work.
2	In bed <50% of the time. Ambulatory and capable of all self-care, but unable to carry out any work activities. Up and about more than 50% of waking hours.	60	Requires occasional assistance, but is able to care for most of his/her needs.
		50	Requires considerable assistance and frequent medical care.
3	In bed >50% of the time. Capable of only limited self-care, confined to bed or chair more than 50% of waking hours.	40	Disabled, requires special care and assistance.
		30	Severely disabled, hospitalization indicated. Death not imminent.
4	100% bedridden. Completely disabled. Cannot carry on any self-care. Totally confined to bed or chair.	20	Very sick, hospitalization indicated. Death not imminent.
		10	Moribund, fatal processes progressing rapidly.
5	Dead.	0	Dead.

APPENDIX B DRUG DIARY

Study Participant Self-Administration Drug Diary

DFCI Study Number: 17-037

Participant Name: _____

Your Doctor_____ Phone_____

Your Nurse_____ Phone_____

Dosing Instructions for Temozolomide: Your dose of temozolomide is _____ mg made up of _____ - _____ mg capsules. ☐You will take temozolomide once daily for up to 42 days in cycle 1 and once daily for the first 5 days of adjuvant cycles.

- You will take your dose of temozolomide orally 1-3 hours before each session of radiotherapy during the week and on the weekends temozolomide is to be taken in the morning for cycle 1.
- During adjuvant cycles it does not matter when you take your dose of Temozolomide as long as you take it at the same time for the first 5 days of the cycle.
- If you vomit your dose of temozolomide do not take another dose until your next scheduled dose.
- If you miss your dose of Temozolomide it may be made up within 8 hours of the missed dose otherwise take your next dose as regularly scheduled.
- The capsules are to be swallowed whole, in rapid succession, without chewing, crushing, or dissolving them.
- You should take your dose of temozolomide on an empty stomach with a minimum of 2 hours after eating and no food consumption for at least 1 hour after your dose of temozolomide.
- This diary should be brought to your week 1-6 appointments during cycle 1 and to your week 1 appointments during adjuvant cycles.
- In the event of an emergency please contact Massachusetts General Hospital at 617-724-8770 or Dana-Farber Cancer Institute at (617) 632-3455 and ask for your doctor to be paged.
- Temozolomide should be stored at room temperature.

DOSING LOG: Cycle 1

Dosing Log Instructions

- Make sure to indicate the date, time, amount taken and any comments immediately following each dose.
- Once complete, provide this signed or initialed and dated dosing log to your study doctor or nurse.

Day	Date	Time of Dose	Number of Temozolomide Capsules	Comments *If dose was vomited, missed or skipped, indicate reason below.*
1		□ am / □ pm		
2		□ am / □ pm		
3		□ am / □ pm		
4		□ am / □ pm		
5		□ am / □ pm		
6		□ am / □ pm		
7		□ am / □ pm		
8		□ am / □ pm		
9		□ am / □ pm		
10		□ am / □ pm		
11		□ am / □ pm		
12		□ am / □ pm		
13		□ am / □ pm		
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39		□ am / □ pm		
40		□ am / □ pm		
41		□ am / □ pm		
42		□ am / □ pm		

Participant Signature or Initials **Date**

DOSING LOG: Cycle _____

Dosing Log Instructions

- Make sure to indicate the date, time, amount taken and any comments immediately following each dose.
- This log covers an entire 31 day month but only mark the days you take your Temozolomide.
- Once complete, provide this signed or initialed and dated dosing log to your study doctor or nurse.

Day	Date	Time of Dose	Number of Temozolomide Capsules	Comments *If dose was vomited, missed or skipped, indicate reason below.*
1		□ am / □ pm		
2		□ am / □ pm		
3		□ am / □ pm		
4		□ am / □ pm		
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29		□ am / □ pm		
30		□ am / □ pm		
31		□ am / □ pm		

Participant Signature or Initials **Date**

APPENDIX C INFORMATION ON POSSIBLE DRUG INTERACTIONS

Information on Possible Interactions with Other Agents for Patients and Their Caregivers and Non-Study Healthcare Team

The participant _____ is enrolled on a clinical trial using the experimental agent Bavituximab. This form is addressed to the participant, but includes important information for others who care for this participant.

It is very important to tell your study doctors about all of your medicine before you start this study. It is also very important to tell them if you stop taking any regular medicine, or if you start taking a new medicine while you take part in this study. When you talk about your medicine with your study doctor, include medicine you buy without a prescription at the drug store (over-the-counter remedy), or herbal supplements such as St. John's wort.

Many health care prescribers can write prescriptions. You must also tell your other prescribers (doctors, physicians' assistants or nurse practitioners) that you are taking part in a clinical trial. **Bring this paper with you and keep the attached information card in your wallet**. These are the things that you and they need to know:

Other medicines can be a problem with your study drugs.

- You should check with your doctor or pharmacist whenever you need to use an over-the-counter medicine or herbal supplement.
- Your regular prescriber should check a medical reference or call your study doctor before prescribing any new medicine for you. Your study doctor's name is

_____

and he or she can be contacted at

_____