**Abbreviated Title:** GBM Pembro HSPPC

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**Title:** A randomized, double blind phase II trial of Surgery, Radiation Therapy plus Temozolomide and Pembrolizumab with and without HSPPC-96 in newly diagnosed Glioblastoma (GBM)

**Coordinating Center:** BTTC Coordinating Center, Center for Cancer Research, NCI

**Responsible DSMB:** NCI Data Safety Monitoring Board

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| Drug Name: | HSPPC-96 | Pembrolizumab | Temozolomide |
|---|---|---|---|
| IND Number: | 17203 | 17203 | 17203 |
| Sponsor: | National Cancer Institute, Center for Cancer Research | National Cancer Institute, Center for Cancer Research | National Cancer Institute, Center for Cancer Research |
| Manufacturer: | Agenus | Merck | generic |
| Supplier: | Agenus | Merck | CC Pharmacy |

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## BTTC PARTICIPATING INVESTIGATORS/SITES (ENROLLING):

The BTTC Participating Investigators/Sites listed in the NIH IRB system will enroll study subjects as described in the protocol. Each site will obtain IRB Approval at their site.

## A DISCLAIMER STATEMENT FOR BTTC PROTOCOLS

This is a research protocol of the Brain Tumor Trials Collaborative describing an experimental treatment procedure. It is a privileged document and is not intended to be circulated or used for other purposes. The Brain Tumor Trials Collaborative assumes no responsibility for its use outside of the constraints of this research protocol or by investigators other than those approved by the Consortium.

## PRÉCIS

### Background:

- Malignant gliomas are unfortunately, in most cases, a uniformly fatal tumor. Despite aggressive surgery, radiation treatment (RT) and chemotherapy at initial diagnosis these tumors almost always recur.
- Many clinical trials in glioblastoma (GBM) have evaluated the addition of agent(s) to standard therapy which consists of concurrent radiation with temozolomide chemotherapy after maximal surgical resection in patients with newly diagnosed disease and salvage chemotherapy with either rechallenge with temozolomide or an alternative alkylating agent such as CCNU or cisplatin. To date, none of the combination strategies have demonstrated clinical benefit. Furthermore, in subjects with an unmethylated MGMT ($O^6$-methylguanine-DNA methyltransferase) promoter temozolomide has only modest benefit and salvage therapies have not demonstrated a significant impact in this subject group underscoring the need for more research.
- Immunotherapy offers the promise of improving outcomes for patients with GBM by evoking specific immune responses that may produce a more sustained and less toxic effect than conventional therapy. Heat-shock proteins (HSPs), which function as intracellular chaperones, can be used to deliver a variety of tumor antigens to antigen presenting cells for immune stimulation.
- Heat Shock Protein-Peptide Complex-96 (HSPPC-96) consists of the heat shock protein glycoprotein-96 (HSP gp-96) and a wide array of chaperoned proteins, including autologous antigenic peptides (aka “vaccine”). Heat shock proteins (HSP) are molecules that respond to cellular stress and counteract abnormal protein folding. They are known to modulate immune responses, especially the HSP gp-96. In a stressful environment, such as a tumor, HSPs are upregulated and highly expressed on tumor cells. This protects the tumor and leads to resistance to therapy. HSP expression is associated with cellular proliferation, apoptosis evasion, tissue invasion, metastasis, and angiogenesis.
- Pembrolizumab is a potent and highly selective humanized monoclonal antibody (mAb) of the IgG4/kappa isotype designed to directly block the interaction between PD-1 and its ligands, PD-L1 and PD-L2. Additionally, pembrolizumab is thought to also have activity in the peripherally circulating T-effector cells by reversing lymphocyte exhaustion. It is currently FDA approved for use in patients with unresectable or metastatic melanoma and disease progression following ipilimumab and, if BRAF V600 mutation positive, a BRAF inhibitor and NSCLC with elevated PD-L1 in the tumor. recurrent or metastatic HNSCC with disease progression on or after platinum-containing chemotherapy. It is also FDA approved for use with advanced (metastatic) non-small cell lung cancer (NSCLC) whose disease has progressed after other treatments and with tumors that express a protein called PD-L1 and for the treatment of patients with recurrent or metastatic head and neck squamous cell carcinoma (HNSCC) with disease progression on or after platinum-containing chemotherapy
- This study will be the first to evaluate this combination of vaccine (HSPPC-96) and PD-1 inhibition (pembrolizumab) in newly diagnosed GBM patients whose tumors are MGMT promoter unmethylated and are isocytrate dehydrogenase (IDH) wildtype; and

will provide important data on immune-modulatory effect of this combination. This may be of particular value in patients with high peripheral PD-L1 expression, but also the value of PD-1 added to standard GBM therapy. As vaccine needs to be generated from the patient's tumor, patients will need to be identified prior to surgery.

**Eligibility:**

- MRI findings consistent with a suspected GBM or histologically confirmed newly diagnosed GBM that has not been treated and would benefit from further surgical resection.
- Tumor must be supratentorial.
- GBM diagnosis must be made by surgical excision (stereotactic biopsy will not be allowed unless there is plan for second surgery to remove ≥ 80 % of the tumor) and patients tumors must not have MGMT promoter methylation and must be IDH wildtype.
- No prior treatment with radiation or chemotherapy for their GBM.
- Age ≥ 18 years on day of signing informed consent

**Objective:**

- The primary endpoint is to determine whether the one-year overall survival (OS) rate is improved in newly diagnosed GBM patients whose tumors have an unmethylated MGMT promoter and are IDH wildtype treated with RT + TMZ + Pembrolizumab followed by TMZ + Pembrolizumab + HSPPC-96 vaccine or Placebo vaccine x 6 cycles (1 cycle is 9 weeks).

**Design:**

- This will be a randomized, double blind phase II trial of surgery, RT + TMZ + Pembrolizumab followed by TMZ + Pembrolizumab +/- HSPPC-96 in newly diagnosed GBM patients whose tumors have an unmethylated MGMT promoter and are IDH wildtype.
- Subjects will be assigned to intervention based on ability to generate vaccine as follows:
    - If ≥ 80 % of tumor removed and ≥7 g of tumor is resected but HSPPC-96 cannot be generated, subjects will be treated on the ancillary arm of RT+TMZ +Pembrolizumab followed by TMZ+ Pembrolizumab.
    - If ≥ 80% of contrasting enhanced tumor removed (based on T1 Post contrast MRI using cross sectional measurement), ≥7 g of tumor is resected and sufficient HSPPC-96 is generated, subjects will be included in the main cohort and will be randomized on a 1:1 basis to receive:
        - RT+TMZ +Pembrolizumab followed by TMZ+Pembrolizumab + Placebo

          OR
        - RT+TMZ +Pembrolizumab followed by TMZ+Pembrolizumab+HSPPC-96
- Subjects whose tumor does not meet the criteria (unmethylated MGMT promoter and IDH wildtype by pathology) and for whom < 80 % of tumor is removed or < 7g of tumor is resected are not eligible for further intervention. Approximately 8 potentially eligible

patients are screened per month, and it is anticipated that at least 1-2 per month will be accrued per site.

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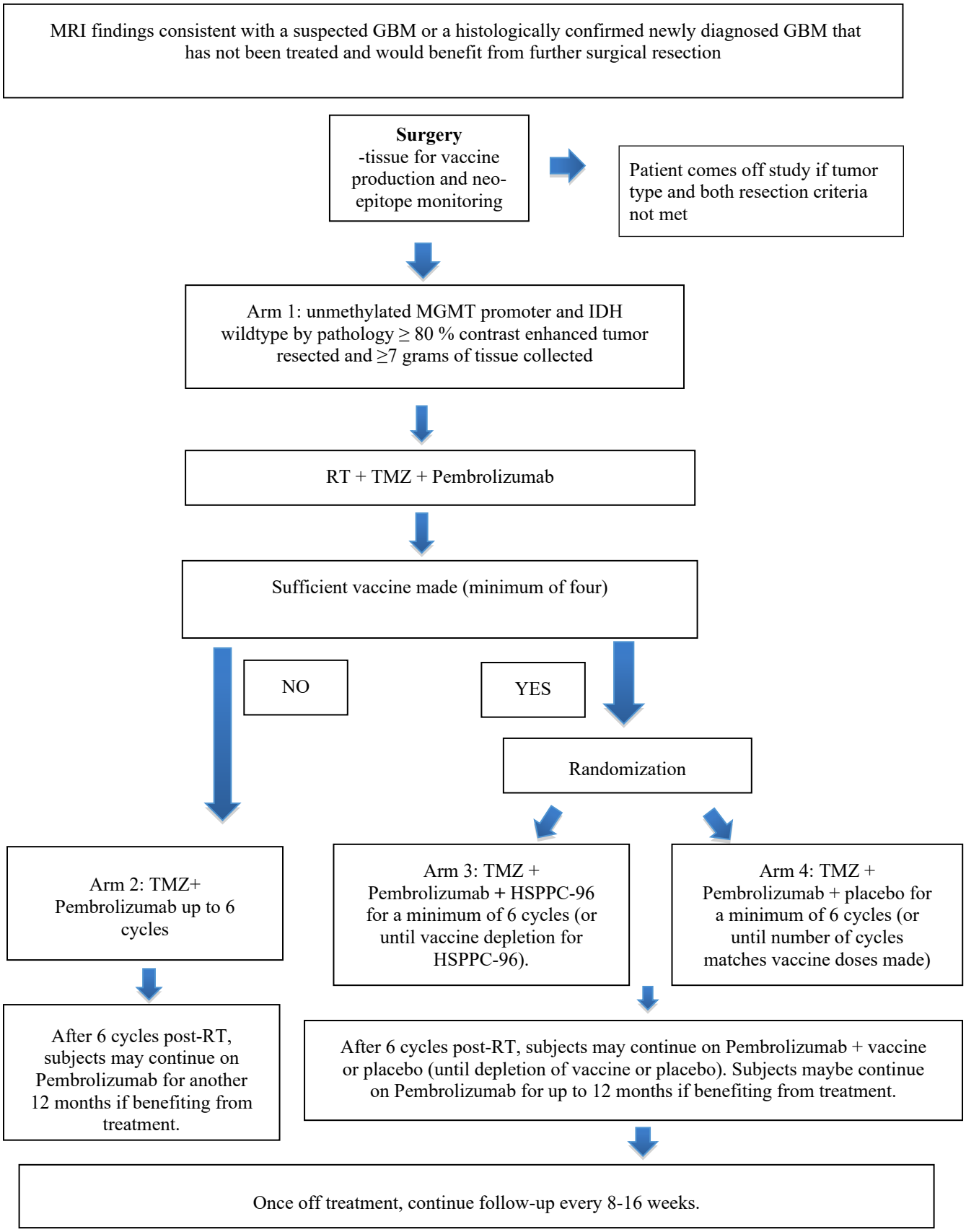
## SCHEMA


MRI findings consistent with a suspected GBM or a histologically confirmed newly diagnosed GBM that has not been treated and would benefit from further surgical resection
Surgery
-tissue for vaccine production and neo-epitope monitoring
Patient comes off study if tumor type and both resection criteria not met
Arm 1: unmethylated MGMT promoter and IDH wildtype by pathology ≥ 80 % contrast enhanced tumor resected and ≥7 grams of tissue collected
RT + TMZ + Pembrolizumab
Sufficient vaccine made (minimum of four)
NO
YES
Randomization
Arm 2: TMZ+ Pembrolizumab up to 6 cycles
Arm 3: TMZ + Pembrolizumab + HSPPC-96 for a minimum of 6 cycles (or until vaccine depletion for HSPPC-96).
Arm 4: TMZ + Pembrolizumab + placebo for a minimum of 6 cycles (or until number of cycles matches vaccine doses made)
After 6 cycles post-RT, subjects may continue on Pembrolizumab for another 12 months if benefiting from treatment.
After 6 cycles post-RT, subjects may continue on Pembrolizumab + vaccine or placebo (until depletion of vaccine or placebo). Subjects maybe continue on Pembrolizumab for up to 12 months if benefiting from treatment.
Once off treatment, continue follow-up every 8-16 weeks.

| |
|---|
| CR/PR/SD: continue treatment until PD or 2 years. |

CR: complete response; PD: Progressive Disease; GBM: glioblastoma multiforme; PR: partial response; RT: radiation therapy; SD: stable disease; TMZ: temozolomide.

## STATEMENT OF COMPLIANCE

The trial will be carried out in accordance with International Council for Harmonisation Good Clinical Practice (ICH GCP) and the following:

- United States (US) Code of Federal Regulations (CFR) applicable to clinical studies (45 CFR Part 46, 21 CFR Part 50, 21 CFR Part 56, 21 CFR Part 312, and/or 21 CFR Part 812)

National Institutes of Health (NIH)-funded investigators and clinical trial site staff who are responsible for the conduct, management, or oversight of NIH-funded clinical trials have completed Human Subjects Protection and ICH GCP Training.

The protocol, informed consent form(s), recruitment materials, and all participant materials will be submitted to the Institutional Review Board (IRB) for review and approval. Approval of both the protocol and the consent form must be obtained before any participant is enrolled. Any amendment to the protocol will require review and approval by the IRB before the changes are implemented to the study. In addition, all changes to the consent form will be IRB-approved; an IRB determination will be made regarding whether a new consent needs to be obtained from participants who provided consent, using a previously approved consent form.

# 1. INTRODUCTION

## 1.1 STUDY OBJECTIVES

### 1.1.1 Primary Objective:

- To determine whether the one-year overall survival rate (OS) is improved in newly diagnosed GBM patients whose tumors have an unmethylated MGMT promoter and are IDH wildtype treated with RT + Temozolomide (TMZ) + Pembrolizumab followed by Pembrolizumab + TMZ and either HSPPC-96 x 6 cycles (1 cycle is 9 weeks) or placebo vaccine.

*Nota Bene*: Those patients whose tumor does not meet the criteria (unmethylated MGMT promoter and IDH wildtype by pathology) and with < 80% of contrast enhanced tumor removed or < 7g of tumor resected are ineligible.

### 1.1.2 Secondary Objectives:

- To determine the following
- Progression Free Survival-6 **(PFS-6)** (percentage of patients alive and progression-free at 6 months post registration).
- Overall Survival **(OS).**
- Overall Survival (**OS) OS-6** and **24** (percentage of patients alive at 6 and 24 months respectively).
- Response will be determined by **Table 2** below using RANO as main criteria. If a lesion is 25% larger but there remains a question as to disease progression or pseudoprogression due to treatment, the patient can remain on treatment until this can be defined, if true progression occurred earlier, the earlier date will be used.
- Safety of Pembrolizumab +/- HSPPC-96 in GBM.
- To evaluate the occurrence of symptoms and correlate to disease progression and tolerance to treatment using the MD Anderson Symptom Inventory-Brain Tumor Module (MDASI-BT). This will include:
  - To longitudinally evaluate patient reported outcome measures using self-reported symptom severity and interference with daily activities using the MDASI-BT questionnaire.
  - To measure symptom burden over the course of therapy to evaluate differences between patients individual symptom severity, overall mean symptom severity, and difference in scores on the interference items between responders and non-responders.
  - To describe the variability of symptom severity longitudinally over the treatment course and follow-up period.

### 1.1.3 Exploratory Objectives:

- To determine if there is a correlation with clinical outcome of PDL1 expression and T-Cell infiltrate in pathology from first, and if applicable, second surgical specimens with outcome.

- To assess tumor tissue for neo-antigens.
- To collect peripheral monocytes for PD-L1 levels to confirm that low level expression leads to better outcome and if higher levels that are treated with Pembrolizumab might improve outcome.
- Assess changes in peripheral T-Cell activation and tryptophan metabolites.
- Evaluate response using iRANO criteria.
- Agenus will isolate additional heat shock protein (HSP)-peptide complexes from "waste streams" generated during the manufacture of HSPPC-96 (Prophage) from GBM tumor tissue. From these HSP-peptide complexes, Agenus may generate a novel "multi-chaperone" vaccine that Agenus will evaluate in preclinical studies.

## 1.2 BACKGROUND AND RATIONALE

### 1.2.1 Disease Background

Malignant gliomas, particularly GBM are nearly always fatal. Despite aggressive surgery, radiation treatment (RT) and chemotherapy at initial diagnosis these tumors almost always recur. There have been many studies in patients with newly diagnosed GBM, either altering the dosing schedule of the temozolomide (RTOG 0525) [1] or adding anti-angiogenic agents to the standard treatment regimen. None have proven to prolong survival and the latter treatments targeting neovascularization were particularly disappointing given that angiogenesis is a cardinal feature of GBM. [2] [3]

Many clinical trials in GBM have evaluated the addition of agent(s) to standard therapy which consists of concurrent radiation with temozolomide chemotherapy after maximal surgical resection in patients with newly diagnosed disease and salvage chemotherapy with either rechallenge with temozolomide or an alternative alkylating agent such as CCNU or cisplatin. To date, none of the combination strategies have demonstrated clinical benefit. Furthermore, in subjects with an unmethylated MGMT ($O^6$-methylguanine-DNA methyltransferase) promoter temozolomide has only modest benefit and salvage therapies have not demonstrated a significant impact in this subject group underscoring the need for more research.

Novel approaches being evaluated in GBM treatment include immunologic therapies, such as therapeutic vaccines that are either derived from the subject's own tumor (HSPPC-96) or have been designed to target specific epitopes on tumor cells (CDX-110, ICT-107). Preliminary results were promising from Phase 2 trials of CDX-110 (rindopeptimet), ICT 107, HSPPC-96, and others. [4-6]. However, a randomized Phase 3 trial evaluating the CDX-110 did not demonstrate efficacy [7].

There are several agents currently under investigation that target the local immune response in solid tumors, particularly in melanoma, such as ipilimumab (blockade of cytotoxic T lymphocyte–associated protein 4 [CTLA-4]) and programmed cell death protein 1 (PD-1; PD-L1 blockade). Such agents are either FDA approved or being evaluated in various solid tumors with the most activity seen in [8].

There are many immunologic approaches to treatment of GBM. Gliomas can upregulate B7-H1 expression in circulating monocytes and tumor-infiltrative macrophages through modulation of

autocrine/paracrine interleukin 10 (IL-10) signaling, resulting in an immunosuppressive phenotype.[9] Glioma-associated cancer-initiating cells have been shown to express the costimulatory inhibitory molecule B7-H1, [10] which is a key factor in mediating immune resistance in glioma[11] and can induce T-cell apoptosis. [12].

Direct cell-to-cell contact experiments have shown that the glioma-associated cancer-initiating cells induced T-cell apoptosis in the setting of B7-H1 expression. [10] found that cancer-initiating cells markedly inhibited T-cell proliferation and activation, induced regulatory T cells, and triggered T-cell apoptosis that was mediated by B7-H1 and soluble galectin-3. Loss of tumor suppressor phosphotase and tensin homolog (PTEN) function increases B7-H1 expression and immunoresistance in glioma. [11] It also been shown that glioma-associated cancer-initiating cells induce immunosuppression, thereby allowing tumor cells to evade immunosurveillance.

Yao et al. [13] found that B7-H1 was correlated with the malignancy grade of human astrocytic tumors, but preferentially in the non-dividing tumor cells. It was also upregulated at the growing edge of the tumors and had a negative correlation with tumor-infiltrating CD8 T cells. Tumor-infiltrating lymphocytes and PD-L1 expression are seen in the majority of GBM samples. [14, 15] Data show that lymphopenia induced by temozolomide (TMZ) can actually potentiate cancer immunotherapy, which may be due in part to cell death, which releases cellular contents that increase the immunoreactivity of the tumor environment. [16]

### 1.2.2 Intervention Background & Overview

#### 1.2.2.1 Programmed Cell Death Protein 1

The importance of intact immune surveillance in controlling outgrowth of neoplastic transformation has been known for decades.[17] Accumulating evidence shows a correlation between tumor-infiltrating lymphocytes (TILs) in cancer tissue and favorable prognosis in various malignancies. [12] [18] [19, 20] [21]. In particular, the presence of CD8+ T-cells and the ratio of CD8+ effector T-cells / FoxP3+ regulatory T-cells seems to correlate with improved prognosis and long-term survival in many solid tumors.

The PD-1 receptor-ligand interaction is a major pathway hijacked by tumors to suppress immune control. The normal function of PD-1, expressed on the cell surface of activated T-cells under healthy conditions, is to down-modulate unwanted or excessive immune responses, including autoimmune reactions. PD-1 (encoded by the gene Pdcd1) is an Ig superfamily member related to CD28 and CTLA-4 which has been shown to negatively regulate antigen receptor signaling upon engagement of its ligands (PD-L1 and/or PD-L2). [22, 23] The structure of murine PD-1 has been resolved. [24] PD-1 and family members are type I transmembrane glycoproteins containing an Ig Variable-type (V-type) domain responsible for ligand binding and a cytoplasmic tail which is responsible for the binding of signaling molecules. The cytoplasmic tail of PD-1 contains 2 tyrosine-based signaling motifs, an immunoreceptor tyrosine-based inhibition motif (ITIM) and an immunoreceptor tyrosine-based switch motif (ITSM). Following T-cell stimulation, PD-1 recruits the tyrosine phosphatases SHP-1 and SHP-2 to the ITSM motif within its cytoplasmic tail, leading to the dephosphorylation of effector molecules such as CD3ζ, PKCθ and ZAP70 which are involved in the CD3 T-cell signaling cascade [23, 25-27]. The

mechanism by which PD-1 down modulates T-cell responses is similar to, but distinct from that of CTLA-4 as both molecules regulate an overlapping set of signaling proteins[25] [26]. PD-1 was shown to be expressed on activated lymphocytes including peripheral CD4+ and CD8+ T-cells, B-cells, T regs and Natural Killer cells. [28] [29] Expression has also been shown during thymic development on CD4-CD8- (double negative) T-cells as well as subsets of macrophages and dendritic cells. [30] The ligands for PD-1 (PD-L1 and PD-L2) are constitutively expressed or can be induced in a variety of cell types, including non-hematopoietic tissues as well as in various tumors. [30] [31] [32, 33] Both ligands are type I transmembrane receptors containing both IgV- and IgC-like domains in the extracellular region and contain short cytoplasmic regions with no known signaling motifs. Binding of either PD-1 ligand to PD-1 inhibits T-cell activation triggered through the T-cell receptor. PD-L1 is expressed at low levels on various non- hematopoietic tissues, most notably on vascular endothelium, whereas PD-L2 protein is only detectably expressed on antigen-presenting cells found in lymphoid tissue or chronic inflammatory environments. PD-L2 is thought to control immune T-cell activation in lymphoid organs, whereas PD-L1 serves to dampen unwarranted T-cell function in peripheral tissues. [34] Although healthy organs express little (if any) PD-L1, a variety of cancers were demonstrated to express abundant levels of this T-cell inhibitor. PD-1 has been suggested to regulate tumor-specific T-cell expansion in subjects with melanoma. [35] This suggests that the PD- 1/PD-L1 pathway plays a critical role in tumor immune evasion and should be considered as an attractive target for therapeutic intervention.

Pembrolizumab (previously known as SCH 900475) is a potent and highly selective humanized monoclonal antibody (mAb) of the IgG4/kappa isotype designed to directly block the interaction between PD-1 and its ligands, PD-L1 and PD-L2. It is currently FDA approved for use in patients with unresectable or metastatic melanoma and disease progression following ipilimumab and, if BRAF V600 mutation positive, a BRAF inhibitor. It is also FDA approved for advanced (metastatic) non-small cell lung cancer (NSCLC) whose disease has progressed after other treatments and with tumors that express a protein called PD-L1 and for the treatment of patients with recurrent or metastatic head and neck squamous cell carcinoma (HNSCC) with disease progression on or after platinum-containing chemotherapy.

### 1.2.3 Rationale for the Pembrolizumab dose

Selection of 200 mg as the appropriate dose for a switch to fixed dosing is based on simulation results indicating that 200 mg will provide exposures that are reasonably consistent with those obtained with 2 mg/kg dose and importantly will maintain individual patient exposures within the exposure range established in melanoma as associated with maximal clinical response. A population PK model, which characterized the influence of bodyweight and other patient covariates on exposure, has been developed using available data from 476 subjects from PN001. The distribution of exposures from the 200 mg fixed dose are predicted to considerably overlap those obtained with the 2 mg/kg dose, with some tendency for individual values to range slightly higher with the 200 mg fixed dose. The slight increase in PK variability predicted for the fixed dose relative to weight-based dosing is not expected to be clinically important given that the range of individual exposures is well contained within the range of exposures shown in the melanoma studies of 2 and 10 mg/kg to provide similar efficacy and safety. The population PK evaluation revealed that there was no significant impact of tumor burden on exposure. In addition, exposure was similar between the NSCLC

and melanoma indications. Therefore, there are no anticipated changes in exposure between different tumor types and indication settings.

Available pharmacokinetic results in subjects with melanoma, NSCLC, and other solid tumor types support a lack of meaningful difference in pharmacokinetic exposures obtained at a given dose among tumor types. Population PK analysis has been performed and has confirmed the expectation that intrinsic factors do not affect exposure to Pembrolizumab to a clinically meaningful extent. Taken together, these data support the use of lower doses (with similar exposure to 2 mg/kg Q3W) in all solid tumor indications. 2 mg/kg Q3W is being evaluated in NSCLC in PN001, Cohort F30 and PN010, and 200 mg Q3W is being evaluated in head and neck cancer in PN012, which are expected to provide additional data supporting the dose selection.

This trial will use Pembrolizumab 200 mg every three weeks to fit with treatment schedules currently used.

### 1.2.4 Heat Shock Protein-Peptide Complex-96 (HSPPC-96)

Immunotherapy offers the promise of improving outcomes for patients with GBM by evoking specific immune responses that may produce a more sustained and less toxic effect than conventional therapy. Heat-shock proteins (HSPs), which function as intracellular chaperones, can be used to deliver a variety of tumor antigens to antigen presenting cells for immune stimulation.

Heat Shock Protein-Peptide Complex-96 (HSPPC-96) consists of the heat shock protein glycoprotein-96 (HSP gp-96) and a wide array of chaperoned proteins, including autologous antigenic peptides. Heat shock proteins (HSP) are molecules that respond to cellular stress and counteract abnormal protein folding. They are known to modulate immune responses, especially the HSP gp-96. In a stressful environment, such as a tumor, HSPs are upregulated and highly expressed on tumor cells. This protects the tumor and leads to resistance to therapy. HSP expression is associated with cellular proliferation, apoptosis evasion, tissue invasion, metastasis, and angiogenesis.

HSPPC-96 immunization works mechanistically by interacting with antigen presenting cells (APCs) via specific receptors, including CD91. [36, 37] highly specific nature of the interaction between HSPPC-96 and APCs is a significant advantage over other cancer vaccine approaches; and has been shown to facilitate robust CD4+ and CD8+ T-cell immune responses. There are multiple mechanisms by which the vaccine works. The first is the following: The autologous antigens in the vaccine are bound to the chaperone proteins (HSP) with noncovalent bonds. Macrophages and other transfer the antigens from HSP to antigen presenting cells (e.g., DCs) bind and take up the HSP-antigen complexes, via receptor- mediated endocytosis (with receptors such as CD91). The antigens are then expressed via MHC class I and II on antigen presenting cells (APCs), interacting with naïve T cells, which are activated into specific CD8 T and CD4 T cells. Another mechanism of the vaccine is an innate immunity pathway cells.

HSPPC-96 Ag is internalized into the APC, which activates the APC. This leads to the following:

- Activated APCs express co-stimulatory molecules (CD80, CD86, and CD40) and express MHC class II, which interacts with T cells and produces tumor specific CD4 cells.
- Activated APCs cause cytokine release (IL-1b, IL-12, IL-6, TNF-alph, GMCSF, chemokines, and nitric oxide). IL-12 stimulates NK cells, which enhance the antitumor activity of T cells.
- The interaction between HSP-antigen and APCs causes dendritic cell maturation, which stimulates NK cells.

A final mechanism of the vaccine is that exposure to Gp96 leads to translocation of NFkB into the APC nucleus, which then regulates cancer cell growth, progression, and apoptosis.

The advantage of the HSPPC-96 vaccine over others is the following:

- Multifactorial mechanism: as described above, the vaccine works by multiple pathways to exert its effect, rather than 1 pathway.
- The vaccine is individualized and does not require identification of immunogenic antigens required by other vaccines.
- Multiple antigens are used, which decreases the chance of immune evasion that occurs with a single antigen vaccine.

Clinical grade HSPPC-96 can be easily purified from solid tumor and has been safely tested in hundreds of patients with newly diagnosed and recurrent melanoma and renal cell cancer. The toxicity encountered in renal cell carcinoma in prior trials is the following: injection site erythema (50%), injection site induration (48%), back pain (12%), headache (12%), fatigue (10%), nasopharyngitis (9%), incision site complication (9%), arthralgia (9%), hypertension (9%), urinary tract infection (8%), nausea (7%), diarrhea (6%), asthenia (6%), influenza (6%), constipation (5%), pyrexia (5%), and dizziness (5%). In melanoma, toxicity seen was pyrexia (8%), fatigue (6%), and nausea (5%). Between the trials there were 3 serious adverse events, 2 were thyroid dysfunction, and 1 was cellulitis. [38] [39] [11]

The rate of successful HSPPC-96 vaccine manufacture is anticipated to be 80% or greater based on prior clinical trials. A prior phase 2 study of new diagnosed glioblastoma, vaccine was successfully manufactured in 84% (63/77) of attempted cases. The majority of manufacture failures were caused by insufficient number of vials available, below the required 4, for clinical use (9 cases).

A single center single arm Phase I clinical trial and a multicenter single arm Phase II clinical trial for recurrent GBM patients treated with HSPPC-96 have been completed. Collectively these studies have shown that: 1) HSPPC-96 vaccine made from recurrent GBM tumor is safe and well tolerated, 2) HSPPC-96 vaccine extends survival in recurrent GBM patients over historical controls and 3) HSPPC-96 is immunogenic in recurrent GBM patients, inducing both an immunologic response locally and peripherally and via innate and adaptive immune mechanisms [39] and confers clinical efficacy in patients with newly diagnosed and recurrent GBM. Importantly, in patients with newly diagnosed GBM, HSPPC-96 vaccine in combination with standard of care temozolomide extended response. Specifically, in the phase I trial, testing was performed pre- and post-vaccine on all patients. Eleven of 12 patients were shown to have a significant immune response, which was measured by testing peripheral blood lymphocytes (PBLs). PBLs were tested for phenotype, reactivity to vaccine, IFN gamma production, and

proliferation. Vaccine was well tolerated with no serious adverse events and the only toxicity noted was injection site erythema or induration. The median progression survival in responders was 47 weeks, compared to 16 weeks in the non-responder. In the phase II trial, 33 patients were treated. Again, there were no serious adverse events attributed to vaccine. Toxicities reported were injection site reaction in 12 patients, and fatigue in 9 patients. Eleven patients were still alive at time of analysis, with median survival of 333 days for the evaluable population. Also, all patients studied showed immune response, based on CD8 IFN gamma production [9].

In a separate single arm phase II trial of newly diagnosed GBM patients who were treated with RT + TMZ + HSPPC-96, median PFS and overall survival were two to three times longer than expected based on historical controls (median PFS was 18 v 6-9 mos and median OS was 24 v 14-16 mos) of 24 months. Importantly, patients with low PDL1 expression on peripheral monocytes had longer survival than those with high PD-L1 expression, suggesting that an optimal group for HSPPC-96 use with a checkpoint inhibitor would be in the high PD-L1 expressing patients.

Importantly, in data presented at ASCO 2015, in patients with less elevated PD-L1 expression on peripheral monocytes, treatment with HSPPC-96 was associated with an unexpected and impressively longer survival than those with high more elevated PD-L1 on circulating monocytes (mPFS of 27 v 6-9 mos and mOS 47 v 14-16; [38] ).

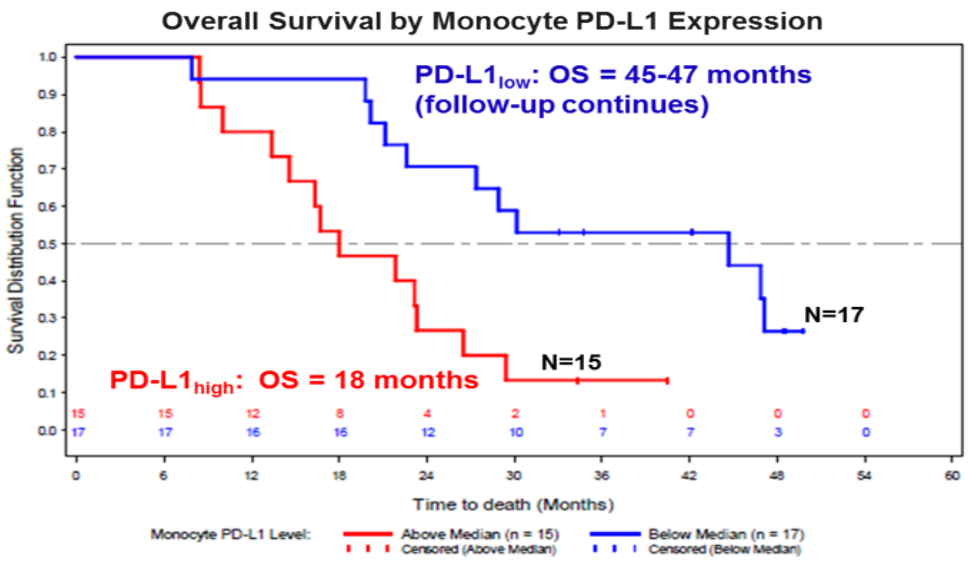



**Figure 1: Overall Survival by Monocyte PD-L1**

### 1.2.5 Rationale for the Current Study

The preclinical data about the immunologic aspects of gliomas and the data using vaccines, suggest that there is a strong rationale to evaluate these agents in GBM. While the blood brain

barrier (BBB) is often an issue for drug penetration, this would be less of an issue for checkpoint inhibitors as the effect is on trafficking T-Cells which can migrate peripherally to the brain. [15] Nonetheless, in the setting of contrast enhancement, the BBB is leaky, allowing drug penetration.

Taken together, these data support the rationale that the combination of an autologous cancer vaccine and an anti-PD-1 inhibitor may synergistically result in an increase in tumor specific immune response with downregulation of tumor immunosuppression resulting in important clinical responses. There is pre-clinical and some clinical precedence for using an immune educating strategy with a release of tumoral immunosuppression. Several studies have shown a benefit of the combination over check point modulators alone or vaccine alone in several types of solid tumors. [19, 40]

This study will be the first to evaluate this combination of vaccine (HSPPC-96) and PD-1 inhibition (Pembrolizumab) in newly diagnosed GBM with unmethylated MGMT promoter and will provide important data on immune-modulatory effect of this combination. This may be of particular value in patients with high peripheral PD-L1 expression, but also there also be value adding PD-1 to standard GBM therapy. We will be assessing OS-12 and other PFS and OS rates.

A phase I study adding Pembrolizumab (200 mg) to RT+TMZ using standard starting doses was found to be safe and without a DLT.[41] The DLT window was from the start of RT until 4 weeks post Radiation. There were no grade 3 or 4 toxicities seen. Related grade 1 and 2 toxicities were anemia, lymphopenia, neutropenia, thrombocytopenia, fatigue, rash, and GI upset/poor PO intake. Pseudo-progression and inflammatory responses were not seen in in a phase I of RT + TMZ + Pembrolizumab. We do not believe there will be any added toxicity of adding HSPPC-96 to RT + TMZ + Pembrolizumab; while an increased inflammatory reaction might occur, these patients will all have achieved ≥ 80% resection.

Tumor tissue will be analyzed to assess for MGMT, PDL-1 levels and T-cell infiltration as well as collection of peripheral markers of T-Cell activation and tryptophan metabolites. Patients who progress and will benefit from further surgery will have their tumor tissue collected and analyzed.

### 1.2.6 Rationale for the patient-reported outcomes:

This study seeks to establish effective therapies. We hypothesize that using a combination of pembrolizumab, temozolomide and HSPPC-96 will result in improved survival. However, given the intensive nature of this regimen, it will be important to determine whether any determined survival benefit is associated with improvements in symptoms or whether a worsening of symptoms offsets the increase in survival.

Precedence for measuring “non-therapeutic” endpoints exists in oncology research. For example, Gemcitabine was approved by the FDA partially as a consequence of the decrease in pain reported in pancreatic patients who were treated, not on the basis of survival improvement which was modest at best [42]. There have been efforts in neuro-oncology to evaluate secondary endpoints using validated instruments as an additional indicator of benefit.

The M.D. Anderson Symptom Inventory-Brain Tumor Module (MDASI-BT) allows the self-reporting of symptom severity and interference with daily activities. The MDASI-BT has demonstrated reliability and validity in the adult primary brain tumor patient population. [43] This tool represents a modification of the widely used and validated M.D. Anderson Symptom Inventory, with particular attention to symptoms common in patients with brain tumors. The availability of validated instruments provides an opportunity to prospectively assess the impact of treatment, both positive and negative, on patients. The evaluation of symptom burden in this study will assist in finding the best possible treatment with the least toxicity.

# 2 ELIGIBILITY ASSESSMENT AND ENROLLMENT

## 2.1 ELIGIBILITY CRITERIA

The target population for this study is patients with newly diagnosed GBM with an un-methylated MGMT promoter and IDH wild type. **NOTE**: Enrollment to this protocol is closed. See Study Implementation (Section **3.1**) regarding study termination and procedures for unblinding participants .

Eligibility will be evaluated by the study team according to the following criteria:

Eligibility waivers are not permitted.

Subjects must meet all of the inclusion and none of the exclusion criteria to be registered to the study. Study treatment may not begin until a subject is registered. Please refer to Section **2.2** for complete instructions regarding registration procedures.

### 2.1.1 Inclusion Criteria

**Pre-Surgery (Step 1) Inclusion:**

2.1.1.1 MRI findings consistent with a suspected GBM or a histologically confirmed newly diagnosed GBM that has not been treated and would benefit from further surgical resection. As vaccine needs to be generated from the patient's tumor, patients will need to be identified prior to definitive surgery.

2.1.1.2 Preliminary assessment by the neurosurgeons that >80% of the tumor can be resected with an expectation that >7gm of tissue would be resected

2.1.1.3 Age ≥ 18 years on day of signing informed consent.

2.1.1.4 Karnofsky performance status ≥ 70%.

2.1.1.5 Tumor must be supratentorial only.

2.1.1.6 Stereotactic biopsy will not be allowed unless there is plans for second surgery to remove ≥ 80 % of the tumor.

2.1.1.7 No prior treatment with radiation or chemotherapy for their GBM.

2.1.1.8 No prior treatment with carmustine wafers.

**Post-Surgery (Step 2) Inclusion:**

2.1.1.9 Pathology must be a GBM, MGMT promoter region determined to be unmethylated and IDH wild type, ≥80 % resection of contrast enhanced tumor on post operative MRI and ≥ 7 grams of tumor resected are required otherwise patient is ineligible .

2.1.1.10 Treatment must be initiated ≥ 14 days and < 6 weeks from surgery.

2.1.1.11 Craniotomy site must be adequately healed and free of drainage or cellulitis, and the underlying cranioplasty must appear intact at the time of radiation. Radiation must start within 6 weeks of surgery.

2.1.1.12 Dexamethasone dose should be ≤ 4 mg/day or steroid equivalent prior to starting treatment. If higher doses are needed, consult with Study Chair.

2.1.1.13 Female subjects of childbearing potential should have a negative urine or serum pregnancy within 7 days prior to receiving the first dose of study medication. If the urine test is positive or cannot be confirmed as negative, a negative serum pregnancy test will be required.

2.1.1.14 Patients must have adequate organ and bone marrow function within 14 days prior to step 2 registration, as defined below:

2.1.1.15 Absolute neutrophil count (ANC) > $1.5 \times 10^9$/L; platelet count > $100 \times 10^9$/L; and hemoglobin (Hb) >9.0 g/dL within 7 days prior to step 2 registration. Note: The use of transfusion or other intervention to achieve Hb ≥ 9.0 g/dL is acceptable.

2.1.1.16 Total bilirubin < 1.5 × ULN (except in patients diagnosed with Gilbert's disease)

2.1.1.17 AST (SGOT), ALT (SGPT), and alkaline phosphatase (ALP) < 2.5 × ULN

2.1.1.18 Serum creatinine < 1.5 × ULN

2.1.1.19 International normalized ratio (INR), prothrombin time (PT), or activated partial thromboplastin time (APTT) as follows: In the absence of therapeutic intent to anticoagulate the patient: INR < 1.5 or PT < 1.5×ULN or aPTT < 1.5×ULN. In the presence of therapeutic intent to anticoagulate the patient: INR or PT and aPTT within therapeutic limits (according to the medical standard in the institution) and the patient has been on a stable dose of anticoagulants for at least 2 weeks before registration.

2.1.1.20 Females of child-bearing potential (FOCBP) and males must agree to use two adequate contraception methods (give examples, e.g. hormonal or barrier method of birth control; abstinence) prior to study entry, for the duration of study participation, and for 120 days following completion of therapy. Should a female patient become pregnant or suspect she is pregnant while participating in this study, she should inform her treating physician immediately. Male patients who father a child should notify the treating physician.

NOTE: A FOCBP is *any woman* (regardless of sexual orientation, having undergone a tubal ligation, or remaining celibate by choice) who meets the following criteria:

- *Has not* undergone a hysterectomy or bilateral oophorectomy
- *Has had* menses at any time in the preceding 12 consecutive months (and therefore has not been naturally postmenopausal for > 12 months)

2.1.1.21 Patients must have the ability to understand and the willingness to sign a written informed consent prior to registration on study.

2.1.1.22 Diagnosis must be made by surgical excision.

2.1.1.23 Patients should not be on antibiotics for any infection but post operative antibiotics are allowed if used prophylactically but should be completed prior to starting RT.

### 2.1.2 Exclusion Criteria

**Pre-Surgery (Step 1) Exclusion:**

2.1.2.1 Known history of immunodeficiency (HIV). This medical entity can be exacerbated by PD-1 blockade.

2.1.2.2 History of another malignancy in the previous 3 years, with a disease-free interval of < 3 years. Exceptions include basal cell carcinoma of the skin, squamous cell carcinoma of the skin, or in situ cervical cancer that has undergone potentially curative therapy. Patients who have undergone a bone marrow or stem-cell transplant for any malignancy are excluded.

2.1.2.3 Has an active autoimmune disease requiring systemic treatment within the past 3 months or a documented history of clinically severe autoimmune disease, or a syndrome that requires chronic systemic steroids or immunosuppressive agents except as noted above in **2.1.2.13** and **2.1.2.14** Subjects with vitiligo or resolved childhood asthma/atopy would be an exception to this rule. Subjects that require intermittent use of bronchodilators or local steroid injections will not be excluded from the study. Subjects with hypothyroidism stable on hormone replacement or Sjorgen's syndrome will not be excluded from the study.

2.1.2.4 Has a history of interstitial lung disease, non-infectious pneumonitis or pneumonitis.

2.1.2.5 Has a history or current evidence of any condition, therapy, or laboratory abnormality that might confound the results of the trial, interfere with the subject's participation for the full duration of the trial, or is not in the best interest of the subject to participate, in the opinion of the treating investigator. Examples include:

- Hypertension (defined as 160/95) that is not controlled on medication
- Ongoing or active infection requiring systemic treatment
- Symptomatic congestive heart failure
- Unstable angina pectoris
- Cardiac arrhythmia
- Psychiatric illness/social situations or substance abuse disorders that would limit compliance with study requirements
- Any other illness or condition that the treating investigator feels would interfere with study compliance or would compromise the patient's safety or study endpoints.

2.1.2.6 Is pregnant or breastfeeding or expecting to conceive or father children within the projected duration of the trial, starting with the pre-screening or screening visit through 120 days after the last dose of trial treatment.

2.1.2.7 The effects of pembrolizumab and HSPPC-96 on the developing human fetus are unknown. For this reason and because checkpoint inhibitors and immunotherapeutic vaccines as well as other therapeutic agents used in this trial are known to be teratogenic, 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.

2.1.2.8 Has received prior therapy with an anti-PD-1, anti-PD-L1, anti-PD-L2, anti-CD137, or anti- Cytotoxic T-lymphocyte-associated antigen-4 (CTLA-4) antibody (including ipilimumab or any other antibody or drug specifically targeting T-cell co-stimulation or checkpoint pathways).

2.1.2.9 On treatment for Hepatitis B or Hepatitis C or history of TB.

2.1.2.10 Patients who have a history of allergic reactions attributed to compounds of similar chemical or biologic composition to Pembrolizumab are not eligible. Known hypersensitivity to any excipients of Pembrolizumab.

**Post-Surgery (Step 2) Exclusion:**

2.1.2.11 Patients are ineligible if the tumor is not a GBM, MGMT promoter region determined to be unmethylated and IDH wild type, or if < 80 % resection of contrast enhanced tumor on post-operative MRI or < 7 grams of tumor is resected.

2.1.2.12 Patients who are receiving any other investigational agents.

2.1.2.13 Known history of immunodeficiency (HIV). This medical entity can be exacerbated by PD-1 blockade.

2.1.2.14 Any form of immunosuppressive therapy within 7 days prior to the first dose of trial treatment excluding steroids. Attempts should be made to have patient on lowest possible dose of steroids. These medical entities can be exacerbated by PD-1 blockade.

2.1.2.15 History of another malignancy in the previous 3 years, with a disease-free interval of < 3 years. Exceptions include basal cell carcinoma of the skin, squamous cell carcinoma of the skin, or in situ cervical cancer that has undergone potentially curative therapy. Patients who have undergone a bone marrow or stem-cell transplant for any malignancy are excluded.

2.1.2.16 Has an active autoimmune disease requiring systemic treatment within the past 3 months or a documented history of clinically severe autoimmune disease, or a syndrome that requires chronic systemic steroids or immunosuppressive agents except as noted above in **2.1.2.13** and **2.1.2.14** Subjects with vitiligo or resolved childhood asthma/atopy would be an exception to this rule. Subjects that require intermittent use of bronchodilators or local steroid injections will not be excluded from the study.

Subjects with hypothyroidism stable on hormone replacement or Sjorgen's syndrome will not be excluded from the study.

2.1.2.17 Has a history of interstitial lung disease, non-infectious pneumonitis or pneumonitis.

2.1.2.18 Has an active infection requiring systemic antibiotics within 10 days of surgery.

2.1.2.19 Has a history or current evidence of any condition, therapy, or laboratory abnormality that might confound the results of the trial, interfere with the subject's participation for the full duration of the trial, or is not in the best interest of the subject to participate, in the opinion of the treating investigator. Examples include:

- Hypertension (defined as 160/95) that is not controlled on medication
- Ongoing or active infection requiring systemic treatment
- Symptomatic congestive heart failure
- Unstable angina pectoris
- Cardiac arrhythmia
- Psychiatric illness/social situations or substance abuse disorders that would limit compliance with study requirements
- Any other illness or condition that the treating investigator feels would interfere with study compliance or would compromise the patient's safety or study endpoints.

2.1.2.20 Is pregnant or breastfeeding or expecting to conceive or father children within the projected duration of the trial, starting with the pre-screening or screening visit through 120 days after the last dose of trial treatment.

2.1.2.21 The effects of pembrolizumab and HSPPC-96 on the developing human fetus are unknown. For this reason and because checkpoint inhibitors and immunotherapeutic vaccines as well as other therapeutic agents used in this trial are known to be teratogenic, 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.

2.1.2.22 Has received prior therapy with an anti-PD-1, anti-PD-L1, anti-PD-L2, anti-CD137, or anti- Cytotoxic T-lymphocyte-associated antigen-4 (CTLA-4) antibody (including ipilimumab or any other antibody or drug specifically targeting T-cell co-stimulation or checkpoint pathways).

2.1.2.23 On treatment for Hepatitis B or Hepatitis C or history of TB.

2.1.2.24 Has received a live vaccine within 30 days prior to the first dose of trial treatment (see Section **4.2**)

2.1.2.25 Patients who have a history of allergic reactions attributed to compounds of similar chemical or biologic composition to Pembrolizumab are not eligible. Known hypersensitivity to any excipients of Pembrolizumab.

### 2.1.3 Recruitment Strategies

This protocol may be abstracted into a plain language announcement posted on NIH websites and on NIH social media platforms. Patients will be referred by BTTC site PIs. An NIH/IRB approved patient flyer is used to recruit patients.

## 2.2 SCREENING EVALUATION

### 2.2.1 Screening activities performed prior to obtaining informed consent

Minimal risk activities that may be performed before the subject has signed a consent include the following:

- Email, written, in person or telephone communications with prospective subjects
- Review of existing medical records to include H&P, laboratory studies, etc.
- Review of existing MRI, x-ray, or CT images
- Review of existing photographs or videos
- Review of existing pathology specimens/reports from a specimen obtained for diagnostic purposes

A waiver of consent for these activities has been requested in section **12.6.2**.

### 2.2.2 Screening activities performed after a consent for screening has been signed

The following activities will be performed only after the subject has signed the consent for this study for screening. Assessments performed at outside facilities or on another NIH protocol within the timeframes below may also be used to determine eligibility once a patient has signed the consent.

The screening evaluation elements must be completed within (+/-) 14 days of signing consent unless otherwise noted:

Outside lab/procedure results for standard of care studies or another protocol that are performed within this timeframe may be used to determine eligibility.

- Medical History
- Prior and Concomitant medication review
- Karnofsky performance status
- Physical and Neurological Exam
- Vital signs and Weight
- Tumor Imaging (by MRI unless contraindicated) showing tumor consistent with a GBM.
- Tumor Pathology (before Step 3)
- Pregnancy Test
- PT/INR or aPTT
- CBC with differential
- Na, K, CL, Bicarbonate, BUN/Cr, AST, ALT, ALP, T. Bili, Calcium, Albumin
- Urinalysis

## 2.3 PARTICIPANT REGISTRATION AND STATUS UPDATE PROCEDURES

Patients who are candidates for the study will first be evaluated for eligibility by the local investigator. All patients must be registered both locally and centrally with BTTC prior to surgery (see Section **3.3.1**, Surgery).

BTTC patients will be registered with the BTTC Coordinating Center. All subjects must be registered through the BTTC NCI Central Registration Office (CRO). The CRO is open from 8:30 am to 5:00 pm EST Monday through Friday, excluding federal holidays.

If any emails described in sections **2.3.2** through **2.3.6** contain protected health information (PHI), they must be encrypted.

### 2.3.1 Informed Consent/Authorization

Prior to protocol enrollment and initiation of treatment, subjects must sign and date an Institutional Review Board (IRB) approved consent form.

Registrations must be completed after the patient has signed the informed consent and has been determined to be eligible by the local investigator.

At the time of registration, the following information will be requested by the BTTC Coordinating Center:

- A copy of a completed and signed, protocol specific, Registration Form (eligibility checklist)
- One copy of the signed and dated Informed Consent/Authorization.
- Copies of all source documents to support protocol specific eligibility. Please refer to the BTTC Operations Manual for specific source documents required.

### 2.3.2 Registration at the NCI

Registration and status updates (e.g., when a participant is taken off protocol therapy and when a participant is taken off-study) will take place per CCR SOP ADCR-2, CCR Participant Registration & Status Updates found here.

Registration will be a three part process as patients are screened on this protocol. Authorized staff must register an eligible candidate with NCI Central Registration Office (CRO) within 24 hours of signing consent. **See Section 2.3.5 for Step 3 of Registration Form (Randomization at NCI).**

### 2.3.3 Registration at Participating BTTC Sites

Registration will be a three part process as patients are screened on this protocol. A protocol registration form and cover memo will be supplied by the Coordinating Center, NCI CCR and updates will be provided as needed. Registration will be completed within 2 business days of receipt of documents required for registration.

**Step 1:** All patients must be registered both locally and centrally with BTTC prior to surgery. To initially register a subject, after the participant has signed the consent, site will complete and send Step 1 of the Registration Form, source documents, and consent form, indicating that the patient is being registered for screening and surgery, to the BTTC Coordinating Center via secure fax (240-541-4432) or send to the BTTC CC research nurse via Secure Email and File Transfer [SEFT] (preferred). Upon confirmation of consent and available slot, the BTTC CC

research nurse will send the Registration Form to NCI Central Registration Office ncicentralregistration-l@mail.nih.gov via encrypted email. Within 1 business day, NCI Central Registration Office will send the Verification of Registration with the assigned subject ID number to the BTTC CC research nurse via encrypted email. The BTTC CC research nurse will email the Verification of Registration to the site and Agenus (redacted with subject ID number and patient initials). The subject ID number is unique to the patient and must be written on all data and correspondence for the patient and used to enter data into the NCI-designated eCRF database.

**Step 2:** Study treatment may not begin until a subject is registered. After completion of screening and surgery, site will complete Step 2 of the Registration Form indicating that the patient has met all inclusion and no exclusion criteria. The Registration Form and source documents will be sent to the BTTC CC via secure fax (240-541-4432) or send to the BTTC CC research nurse via Secure Email and File Transfer [SEFT] (preferred). Upon confirmation of eligibility, the BTTC CC research nurse will sign the Registration Form (eligibility checklist) and send it via encrypted email to the CRO at NCI Central Registration Office ncicentralregistration-l@mail.nih.gov. The CRO will send the Verification of Registration to the BTTC CC research nurse via encrypted email. The BTTC CC research nurse will email the Verification of Registration to the site and Agenus (redacted with study ID number and patient initials).

When a participant has a status change (e.g., subject screened on the study, does not meet eligibility criteria and is removed from the study, participant is taken off protocol therapy or off study, etc.), the Participant Status Update Form will be supplied by the BTTC Coordinating Center research nurse coordinator. Send the completed form to the BTTC Research Nursevia secure fax: 240-541-4432, or via Secure Email and File Transfer (SEFT)..

**See Section 2.3.6 for Step 3 of Registration Form (Randomization for Participating Sites).**

### 2.3.4 Initiation of Therapy (Radiation Therapy, Temozolomide and Pembrolizumab)

Treatment may not be initiated until the participating institution receives a faxed or emailed copy of the patient's Registration Verification Letter from the Central Registration Office.

Treatment must be initiated $\geq$ 14 days and < 6 weeks from surgery.

The BTTC Coordinating Center must be notified in writing of any exceptions to this policy.

### 2.3.5 Registration Step 3: Randomization for NCI

Following vaccine manufacture and completion of quality testing, Agenus will send the Vaccine Production Notification form to NCI. This Form indicates whether manufacture of vaccine was successful. The site will send Step 3 of the Registration form and Vaccine Production Notification Form to BTTC CC research nurse via secure fax: 240-541-4432, hand-delivery, or encrypted email. BTTC CC will confirm verification and send via encrypted email to CRO and via Secure Email and File Transfer [SEFT] to Agenus. CRO will send the Verification of Registration with the assigned sequence number to the site and the BTTC CC research nurse via encrypted email. The BTTC CC research nurse will send the Verification of Registration with the assigned sequence number (vaccine assignment) to Agenus (redacted with subject ID number

and patient initials). Agenus will send the blinded vaccine or placebo directly to the site. In parallel, CRO will also notify the NCI Pharmacy to release blinded vaccine or placebo.

### 2.3.6 Registration Step 3: Randomization for Participating Sites

Following vaccine manufacture and completion of quality testing, Agenus will send the Vaccine Production Notification form to the site. This Form indicates whether manufacture of vaccine was successful. The site will send Step 3 of the Registration form and Vaccine Production Notification Form to BTTC CC research nurse via secure fax: 240-541-4432 or via Secure Email and File Transfer [SEFT] (preferred). BTTC CC will confirm verification and send via encrypted email to CRO and via Secure Email and File Transfer [SEFT] to Agenus. CRO will send the Verification of Registration with the assigned sequence number to the BTTC CC research nurse. The BTTC CC research nurse will send the Verification of Registration with the assigned sequence number (vaccine assignment) to the site and Agenus (redacted with subject ID number and patient initials). Agenus will send the blinded vaccine or placebo directly to the site.

### 2.3.7 Study Status Updates for Participating Sites

For participating BTTC Sites: The Participant Status Updates Form will be supplied by the BTTC Coordinating Center, NCI, CCR. Send the completed Participant Status Updates Form via secure fax: 240-541-4432, or via Secure Email and File Transfer (SEFT).

### 2.3.8 Screen Failures

Screen failures are defined as participants who consent to participate in the clinical trial but are not subsequently assigned to the study intervention or entered in the study. A minimal set of screen failure information is required to ensure transparent reporting of screen failure participants, to meet the Consolidated Standards of Reporting Trials (CONSORT) publishing requirements and to respond to queries from regulatory authorities. Minimal information includes demography, screen failure details, eligibility criteria, and any serious adverse event (SAE).

## 2.4 TREATMENT ASSIGNMENT AND RANDOMIZATION /STRATIFICATION PROCEDURES

### Cohorts

| Number | Name | Description |
|---|---|---|
| 1 | Patients with Glioblastoma eligible to receive radiation therapy | GBM patients with MRI findings consistent with a suspected GBM or a histologically confirmed newly diagnosed GBM that has not been treated, have met all inclusion and no exclusion criteria, and have completed surgery (i.e., Step 2 of registration) |
| 2 | Patients with Glioblastoma with sufficient vaccine created | GBM patients with MRI findings consistent with a suspected GBM or a histologically confirmed newly diagnosed GBM that has not been treated, whose tumors have an unmethylated MGMT promoter and are IDH wildtype with sufficient vaccine created (i.e., Step 3 of registration) |
| 3 | Patients with Glioblastoma without sufficient vaccine created | GBM patients with MRI findings consistent with a suspected GBM or a histologically confirmed newly diagnosed GBM that has not been treated, whose tumors have an unmethylated MGMT promoter and are IDH wildtype without sufficient vaccine created (i.e., Step 3 of registration) |

### Arms

| Number | Name | Description |
|---|---|---|
| 1 | Radiation Therapy (RT) | Standard radiation therapy |
| 2 | RT+TMZ + Pembrolizumab | Standard treatment with experimental treatment (pembro) added |
| 3 | RT+TMZ+Pembrolizumab+HSPPC-96 Vaccine | Standard treatment with experimental treatment (pembro+ vaccine) added |
| 4 | RT+TMZ+Pembrolizumab+Placebo Vaccine | Standard treatment with experimental treatment and placebo added |

### Randomization and Arm Assignment

All study patients satisfying the eligibility criteria and who have completed surgery will be assigned to Cohort 1 and Arm 1, Radiation Therapy. All study patients that satisfy the eligibility criteria post-radiation therapy, will be assigned to Cohort 2 or 3, based on ability or inability to make sufficient vaccine. Study patients in Cohort 2 will then be randomized 1:1 in a blinded fashion between arms 3 and 4. Patients in cohort 3 will be assigned directly to the ancillary arm, Arm 2.

**Stratifications**

| Name | Distinct Options | Notes |
|---|---|---|
| None | | |

# 3 STUDY IMPLEMENTATION

## 3.1 EARLY TERMMINATION

On May 20, 2021 the NCI/CCR Data Safety and Monitoring Board met to review the progress of this protocol, including the results of an unscheduled interim analysis.

Prior to the meeting, the board agreed to a request by the Lead NCI PI to look at outcome data by arm and agreed on principles for interpretation of outcome and accrual data.

Based on the presented data, the board recommended the study be stopped because of poor accrual. The board deferred to the principal investigator on how to enact an orderly termination of the study including continued treatment and unblinding of patients.

Because this is a multi-institutional trial, the IRBs of all institutions participating in this study must be informed by the Lead NCI PI about this DSMB's decision.

Sections throughout the protocol have been revised to reflect that the study is closed to new enrollment and to describe the early termination procedures. The revisions include decisions for how long participants currently enrolled will remain on study after the unblinding.

Below are guidelines for unblinding and follow up per Study Arm:

- **Ancillary Arm 2**: For participants assigned to RT+TMZ + Pembrolizumab, they will remain on treatment and will be followed for overall survival per Study Calendar.
- **Arm 3**: For participants randomized to the RT+TMZ+Pembrolizumab+HSPPC-96 Vaccine, they will be unblinded but may remain on treatment to receive TMZ, Pembro and/or HSPPC-96 vaccine and will be followed for overall survival per Study Calendar.
- **Arm 4**: For participants randomized to RT+TMZ+Pembrolizumab+Placebo Vaccine, they will be unblinded but may remain on treatment to receive TMZ and/or Pembro and will be followed for overall survival per Study Calendar. Placebo vaccine will be discontinued as permitted per Section **3.10**.

Participants who decide to discontinue treatment will continue to be followed for overall survival.

## 3.2 STUDY DESIGN

This will be a randomized, double blind phase II trial of surgery, RT + TMZ + Pembrolizumab followed by TMZ + Pembrolizumab with HSPPC-96 vaccine or placebo vaccine in newly diagnosed GBM patients whose tumor has an unmethylated MGMT promoter and is IDH wildtype.

Subjects will be randomized without stratification on a 1:1 basis, using variable block sizes. The first block will be of size n=6 in order to facilitate the evaluation of toxicity and safety in the first 3 patients on each arm.

The study will utilize the Recommended Phase 2 Dose (RP2D) for RT + TMZ + Pembrolizumab which was TMZ 75 mg/m$^2$, RT approximately 60Gy and Pembrolizumab 200 mg every 3 weeks. HSPPC-96 is not expected to result in additional toxicity although the first 3 subjects treated in each arm will be observed for toxicity before further enrollment.

The primary endpoint is:

- To determine whether the one-year overall survival rate (OS) is improved in newly diagnosed GBM patients whose tumor is MGMT promoter unmethylated and is IDH wildtype treated with RT + TMZ + Pembrolizumab followed by Pembrolizumab + TMZ + HSPPC-96 vaccine (or placebo vaccine) x 6 cycles (1 cycle is 9 weeks).

*Nota Bene:* Those patients whose tumor does not meet the criteria (unmethylated MGMT promoter and IDH wildtype by pathology) and with < 80% of contrast enhanced tumor or < 7grams of tumor resected are ineligible.

All eligible patients will be treated with standard RT to approximately 60 Gy (180 cGy (1.8 Gy) x 33 or 200 cGy (2.0 Gy) x 30). Initial dose of TMZ during radiation will be 75 mg/m$^2$/day 7 days per week and Pembrolizumab 200 mg every 3 weeks.

### 3.2.1 Blinding

This study will utilize a placebo control for the vaccine in order to fulfill the double blind design of the study. This protocol design minimizes the risk of bias and enhances the impartial bias analysis of the study vaccine versus placebo effect. However, it is important to note that in case this information is required for safety purposes (affects care or medically needed), it will be provided in an expedited manner.

In this double blind study, neither the study personnel (including pharmacists) nor the subject will have knowledge of the vaccine assignment being administered. Data analysis personnel and the Central Registration Office will have access to vaccine assignment.

Blinded vaccine (HSPPC-96 or placebo) will be prepared by Agenus and shipped directly to sites. HSPPC-96 or placebo will be dispensed in identical appearing single-use vial as a clear, colorless solution. Each vial of vaccine or placebo is labeled with the subject/patient number, subject/patient initials, and subject/patient date of birth (DOB).

Unblinding may occur at the discretion of the principal investigator and only where knowledge of treatment affects patient care. Site would need to submit the document found in Appendix **16.11** to request unblinding. Unblinding will only be done upon approval by the lead PI as documented in Appendix **16.12**.

**NOTE**: Based on the DSMB's May 2021 decision for early termination, unblinding treatment assignments for participants receiving HSPPC-96 vaccine or placebo will be provided to the Lead NCI PI by the NCI Central Registration Office (CRO) in consultation with and confirmed by Agenus, the vaccine manufacturer. The Lead NCI PI or designee at the BTTC Coordinating

Center will be responsible for notifying site PIs (at the participating BTTC site) of the unblinded treatment assignments for participants receiving HSPPC-96 vaccine or placebo. Participants receiving pembrolizumab, temozolomide and/or HSPPC-96 vaccine will continue to receive those study treatments per Study Calendar (See Section **3.11**). Participants who are on placebo will stop receiving placebo but may continue to receive pembrolizumab and/or temozolomide per Section **3.10**. Participants who decide to discontinue treatment will continue to be followed for overall survival, and the procedures for removal from protocol therapy will be followed (see Section **3.14**). Off-Study Procedures will be followed per protocol for patients who decide to withdraw from the study (see Section **3.14**).

#### 3.2.1.1 Rationale for Double Blinding

This study has been designed to be double blinded despite having an objectively ascertained primary endpoint for the following reasons: (1) This limits study defection due to randomization disappointment (so subjects randomized to placebo will not quit the study), (2) there will be no difference in the interpretation of biomarkers or lab tests due to knowing the intervention being delivered. (3) Adverse event reporting will not be impacted with the knowledge of the treatment arm. (4) Subject/patient responses in the MDASI will not be impacted with the knowledge of the treatment arm.

## 3.3 SURGERY & TISSUE COLLECTION FOR VACCINE ADMINISTRATION

### 3.3.1 Surgery

Patients with presumed GBM based on MRI or if they require a more definitive surgery may be consented for the trial. At the time of surgery, tissue will be collected for vaccine production (≥ 7 grams is required) and processed per Appendix **16.2** Tumor must be confirmed to be MGMT promoter unmethylated and IDH wildtype prior to starting treatment.

GBM patients whose tumor is MGMT promoter methylated and is not IDH wildtype are not eligible for treatment on this protocol and will be removed from the protocol.

Those patients with < 80% of contrast enhanced tumor resected or < 7 grams of tissue will be no longer eligible for treatment with SOC (RT + TMZ) + Pembrolizumab. Tumor will be measured per RANO criteria. The determination of ≥ 80% resection will be based on the pre-operative and post operative cross sectional measurements, the residual tumor, if any, should be < 20% or original cross sectional measurements.

Post tumor resection, all patients must have a post-operative MRI done no more than 72 hours after surgery. This MRI can be used as baseline prior to treatment.

### 3.3.2 Tissue Collection for Vaccine Production at NCI

Tumor sample will be collected at time of surgery by the Laboratory of Pathology NCI. Tissue sections will be obtained and will be labeled with the Study Subject ID number. The relationship between the Study Subject ID number and the patient clinical information will be stored in a secure database that is maintained and regularly backed up by the Laboratory of Pathology NCI. The Laboratory of Pathology NCI will submit tumor tissue for vaccine manufacture according to Appendix **16.2**.

Tumor tissue that does not qualify either because of incorrect diagnosis, MGMT status, or presence of IDH mutation will remain at or be returned to the site of origin.

Tissue will also be collected at NCI for Correlatives. Refer to **5.1.2**, **5.1.3**, and **5.1.4** for tissue collection for Correlatives.

Tumor tissue that does not qualify either because of incorrect diagnosis, MGMT status, or presence of IDH mutation will remain at or be returned to the site of origin.

### 3.3.3 Tissue Collection for Vaccine Production at Participating BTTC Sites

Tumor sample will be collected at time of surgery at the participating sites. Tissue sections will be obtained and will be labeled with the Study Subject ID number. The participating BTTC site will submit tumor tissue for vaccine manufacture according to Appendix **16.2**.

Tumor tissue that does not qualify either because of incorrect diagnosis, MGMT status, or presence of IDH mutation will remain at or be returned to the site of origin.

Tissue will also be collected at the participating BTTC sites for Correlatives. Refer to **5.1.2** and **5.1.3** for tissue collection for Correlatives.

## 3.4 Radiation Therapy (RT)

All eligible patients will be treated with standard RT to approximately 60 Gy (180 cGy (1.8 Gy) x 33 fractions or 200 cGy (2.0 Gy) x 30 fractions).

## 3.5 Drug Administration

### 3.5.1 (RT+TMZ+Pembro)

TMZ will be administered orally on day 1 of radiation therapy (RT) 1-2 hours before each session of radiotherapy or 1 night before RT (within 24 hours before Day 1); and it should be administered the same time throughout the course of RT. TMZ should continue throughout RT at the dose of 75 mg/m2/day. During weekends without radiotherapy (Saturday and Sunday), the drug will be taken at the same time as on radiotherapy days.

Pembrolizumab at 200 mg will be administered on day 1 as a 30 minute IV infusion (+/- 24 hours day of RT) every 3 weeks during RT Phase on days 1, 22 and 43 (+/- 24 hours for each timepoint). Sites should make every effort to target infusion timing to be as close to 30 minutes as possible. However, given the variability of infusion pumps from site to site, a window of -10 minutes and +10 minutes is permitted (i.e., infusion time is 30 minutes: -10 min/+10 min).

Observation for one hour after the infusion of the first dose of Pembrolizumab is recommended.

### 3.5.2 HSPPC-96 Vaccine or Placebo

HSPPC-96 vaccine will be produced prior to the end of RT. If $\geq$ 4 doses can be made, the patient will then be randomized 1:1 to the treatment with TMZ + Pembrolizumab with HSPPC-96 vaccine or placebo.

## 3.6 POST RT TREATMENT REGIMEN

### 3.6.1 For patients not randomized (ancillary arm 2)

Four weeks post end of RT, an MRI will be performed and then patients will start TMZ (days 1-5 and 29-33) + Pembrolizumab (days 1, 22 and 43 of each cycle +/- 24 hours for each timepoint). 1 cycle = 9 weeks. Please note that if days 1-5 of TMZ is well tolerated, the dose of TMZ will be increased for days 29-33 as described below.

The starting TMZ dose will be 150 mg/m2/day for cycle 1, days 1-5, with a single dose escalation to 200 mg/m2/day for all subsequent treatment if no treatment-related adverse events greater than Grade 2 are noted.

After completion of 6 cycles, patients may continue on Pembrolizumab for an additional 12 months if felt to be of benefit by the treating physician.

**NOTE**: For early terminationArm 2 ancillary participants that were not randomized and were assigned to Arm 2 will continue on treatment andfollowed for overall survival per Study Calendar (See Section **3.11**).

### 3.6.2 For patients randomized to vaccine or placebo

One week post end of RT, patients will receive a dose of HSPPC-96 vaccine or placebo vaccine given as approximately 0.4mL intradermal weekly x 4 (see Section **14.3.8** details on intradermal administration).

Within 7 days of the last dose of HSPPC-96 vaccine or placebo, an MRI will be performed and then patients will start TMZ (days 1-5 and 29-33) + Pembrolizumab (days 1, 22 and 43 of each cycle +/- 24 hours for each timepoint). 1 cycle = 9 weeks.

The starting TMZ dose will be 150 mg/$m^2$/day for cycle 1, days 1-5, with a single dose escalation to 200 mg/$m^2$/day for all subsequent treatment if no treatment-related adverse events greater than Grade 2 are noted.

HSPPC-96 vaccine or placebo will then be given 21 days (+/- 24 hours) after the day 5 and day 33 doses of TMZ. TMZ will be given for 6 cycles. HSPPC-96 vaccine or placebo vaccine will be given for 6 cycles or until supply runs out. Patients who receive placebo will be matched for number of vaccine injections that were generated by their tumor tissue. After the completion of 6 cycles (54 weeks), they may continue pembrolizumab for 12 more months alone or in conjunction with HSPPC-96 or placebo vaccine if any is available, if felt to be of benefit by the treating physician.

**NOTE**: Early Termination procedures require that participants randomized to HSPPC-96 or placebo be unblinded. Participants randomized to HSPPC-96 vaccine may continue on treatment to receive TMZ, Pembro and/or HSPPC-96 vaccine. Patients randomized to placebo vaccine may continue on treatment to receive TMZ and Pembro. Placebo vaccine will be discontinued. Participants randomized to HSPPC-96 vaccine or placebo will be followed for overall survival per Study Calendar (see Section **3.11**).

### 3.7 Baseline Assessments (Post-Surgery And Pre-Treatment RT+TMZ+Pembro)

Prior to RT, once patient has recovered from surgery, all patients will undergo the following baseline evaluations or procedures:

- Karnofsky performance status
- Physical and Neurological Exam
- Vital signs and Weight
- Pregnancy Test
- PT/INR or aPTT
- CBC with differential
- Na, K, CL, Bicarbonate, BUN/Cr, AST, ALT, ALP, T. Bili, Calcium, Albumin
- Urinalysis
- T3, T4, and TSH
- MDASI-BT*
- Correlative Studies Blood Collection (LabConnect Kit B refer to Appendix **16.3**)
- Baseline tumor imaging (by MRI unless contraindicated) to be performed within 72 hours after surgery.

*The MDASI-BT (Appendix **16.9**) will be administered at time of baseline MRI or up to 14 days prior to treatment initiation.

Baseline assessments can be performed up to 7 days before RT+TMZ+Pembro treatment starts. If treatment is delayed (e.g. complications of wound healing, infection), repeat all baseline assessments (except for the MDASI-BT and Correlative research blood samples which do not need to be repeated).

### 3.8 Assessments While On Trial During And Post RT

- Patients will have a physical (including vital signs and weight), KPS, and neurological exam every 3 weeks (- 3 days to + 7 days) during RT and then every 4 weeks (+/- 7 days) after RT.
- Patients on ancillary arm do not require vaccine associated physical exam.
- Adverse events will be assessed during Physical Exams and on treatment days.
- For subjects not randomized: An MRI will be done pre surgery, post surgery (within 72 hours), 4 weeks post RT (+/- 1 week), and then every 9 weeks +/- 1 week.
- For subjects who are randomized: An MRI will be done pre surgery, post surgery (within 72 hours), within 7 days of last dose of vaccine or placebo induction, and then every 9 weeks +/- 1 week.
- Pregnancy test will be done at the start of each cycle.
- A CBC with differential will be done weekly +/- 3 days during RT and then weekly +/- 3 days after RT for the first year and then every 2 weeks for the year after if the patient remains on Pembrolizumab.

- A Comprehensive Chemistry Panel with LFTs and TFTs, T3, T4, TSH will be done on day 21 of RT +\- 3 days, at the completion of RT and then day 1 (+/- 1 day) and day 29 (-7 days) of each cycle.
- Blood for correlative labs will be collected for different immune monitoring assessments that may include, but are not limited to: whole exome sequencing (WES), immune phenotyping of immune cell subsets, functional characterization of immune cell subsets, measurement of immune mediators, peripheral lymphocyte, monocyte and tryptophan and the assessment of PD-L1 expression and circulating tumor at the following time points: For subjects who are randomized: Pre-RT treatment (RT+TMZ+Pembro), Post-RT treatment (RT+TMZ+Pembro)/Pre-vaccine (or placebo) induction, Post Vaccine (or placebo) induction/Pre-cycle 1 maintenance, Every Day 1 on Cycle 2, C4, C6, C8 (if continuing on Pembrolizumab), then at discretion of Study Chair and BTTC Coordinating Center, every other cycle, at time of progression or at end of treatment and for subjects not randomized: Pre-RT treatment (RT+TMZ+Pembro), Post-RT treatment (RT+TMZ+Pembro)/Pre-cycle 1 maintenance, Every Day 1 on Cycle 2, C4, C6, C8 (if continuing on Pembrolizumab); then at discretion of Study Chair and BTTC Coordinating Center, every other cycle, at time of progression, or at end of treatment (See Study Calendar **3.11**).
- Archival tissue for PDL1 expression and mutation analysis will be collected after surgery irrespective of whether the patient is randomized or not.
- If a patient has tumor progression while on trial and undergoes surgery, patients will have their tumor evaluated for PDL1 expression and repeat mutational analysis.

### 3.8.1 Questionnaires

The MDASI-BT will be administered prior to the initiation of radiation, temozolomide and pembrolizumab (baseline) prior to initiation of adjuvant therapy and then at the time of each evaluation that also includes imaging. Of note, every attempt should be made to administer the MDASI-BT prior to informing the patients about the results of the imaging study. The MDASI-BT will be completed only by the patient, unless changes in vision or weakness make this difficult. If this occurs, then the caregiver or research assistant may read the questions to the patient or assist with marking the severity number or score as described by the patient. A caregiver may complete the questionnaires as a patient proxy if the patient's deficits preclude self-report; however, this must be done at every assessment from baseline to end of treatment. Full instruments are provided in the Appendix **16.9**. In addition, information regarding demographics and treatment history will be collected as part of the larger study and used in this analysis.

The MDASI-BT consists of symptoms rated on an 11-point scale (0 to 10) to indicate the presence and severity of the symptom, with 0 being "not present" and 10 being "as bad as you can imagine." Each symptom is rated at its worst in the last 24 hours. Symptoms included on the instrument include those commonly associated with cancer therapies, those associated with increased intracranial pressure, and those related to focal deficits. The questionnaire also includes ratings of how much symptoms interfered with different aspects of a patient's life in the last 24 hours. These interference items include: general activity, mood, work (includes both work outside the home and housework), relations with other people, walking, and enjoyment of life.

The interference items are also measured on 0 - 10 scales. The average time to complete these instruments is 5 minutes. The MDASI-BT has been translated into multiple languages, but the English language version will be used for this study. [43]

### 3.9 TELEHEALTH/REMOTE VISITS

In the extenuating event that a subject cannot travel to be seen in person at the study site, or any other crisis causing travel and staffing restriction, the provider assessment may occur via telehealth visits. Telehealth visits (synonyms: telemedicine, videoconferencing, virtual visits, video phone call or remote visits) will be allowed for patients on-study unless these visits are essential to maintaining patient safety while on-study, refer to study calendar section **3.11** for more details. Decisions regarding the use of telehealth will be based on the patient's physician's decision regarding the risk of an in-person clinic visit compared with the potential benefit of a comprehensive in-person evaluation. The medical record or progress note for the telehealth visit should state that the risks were greater than the benefit.

Telehealth visits for clinical assessments will be conducted in compliance with local guidelines and FDA regulations. For example, to ensure privacy, the patients will confirm their identities before the interaction or exchange of private information begins; guidance on this issue can be found in the National Institute of Standards and Technology (NIST) Digital Identity Guidelines, Special Publication 800-63A —*Enrollment and Identity Proofing Requirements When Developing an Identity Verification Plan*. In addition, documentation of telehealth visits must comply with local requirements (e.g., date and time of the real-time video interaction, location of the subject, individuals present, assessments performed, etc.).

General interviews may be conducted adequately via telehealth visits. For example, to obtain a patient's medical history, check-in with a participant regarding their current health status, or to administer a questionnaire to a participant related to their level of functioning or activities of daily living.

Clinical assessments via telehealth visits for patients on study will not compromise either the primary or secondary objectives and will likely have minimal impact on the ability to complete this study as long as administration of treatment (e.g., radiation, temozolomide, pembrolizumab and vaccine/placebo) is not compromised. Given that the study requires only standard, clinical laboratory assessments and imaging per routine radiologic parameters/procedures, any possible interlaboratory or facility variability is not expected to be a concern. If any question about possible progression, a patient may be asked to have repeat imaging and/or additional assessments to confirm response.

Further, a patient may be referred to another local provider or asked to come to the NIH for an in-person assessment, if clinically indicated, and at the discretion of the investigator. In the case of any visits with participants' local providers, records will be obtained for the research records.

**The following clinical assessments generally obtained during a physical examination (i.e., H&P) can be conducted via telehealth visits through discussion and remote/video observation**:

- Karnofsky Performance Status.

- Neurological exam to assess cognition remotely will use the American Academy of Neurology Telemedicine guidance document for evaluation of memory, attention, concentration, language, eye movements, facial weakness, strength, walking and coordination.
- Physical exam to observe rashes, thrust, surgical scar, and swelling will use the American Academy of Dermatology Teledermatology guidance document for physical evaluation during telehealth vistis.

**The following clinical assessments are not feasible via telehealth visits**:

- Hearing and vision
- Listening to your heart and lungs
- Assessing weakness
- Checking reflexes
- Checking for abnormalities in sensation
- Feeling lymph nodes, areas of swelling

## 3.10 DOSE MODIFICATIONS

If Pembrolizumab and/or HSPPC-96 are stopped or discontinued, the patient can continue treatment on TMZ alone. If TMZ is stopped or discontinued, Pembrolizumab or HSPPC-96/placebo can continue. If HSPPC-96/Placebo is stopped or discontinued, TMZ and Pembrolizumab can continue.

### 3.10.1 Pembrolizumab Dose Modifications

Adverse events (AE) (both non-serious and serious) associated with pembrolizumab exposure, including coadministration with additional compounds, may represent an immunologic etiology. These immune-related adverse events (irAEs) may occur shortly after the first dose or several months after the last dose of treatment and may affect more than one body system simultaneously. Pembrolizumab must be withheld for drug-related toxicities and severe or life-threatening AEs as per **Table 1: Dose Modification and Toxicity Management Guidelines for Immune-related AEs Associated with Pembrolizumab**.

See Section **3.10.3** for supportive care guidelines, including use of corticosteroids.

3.10.1.1 Attribution of Toxicity:

When study interventions are administered in combination, attribution of an adverse event to a single component is likely to be difficult. Therefore, while the investigator may attribute a toxicity event [to the combination, to Temozolomide alone or to pembrolizumab alone, for adverse events listed in [**Table 1**], both interventions must be held according to the criteria in [Table 1 Dose Modification and Toxicity Management Guidelines for Immune-Related Adverse Events Associated with Pembrolizumab].

3.10.1.2 Holding Study Interventions:

When study interventions are administered in combination, if the AE is considered immune-related, both interventions should be held according to recommended dose modifications.

3.10.1.3 Restarting Study Interventions:

Participants may not have any dose modifications (no change in dose or schedule) of pembrolizumab in this study, as described in [**Table 1**].

- If the toxicity does not resolve or the criteria for resuming treatment are not met, the participant must be discontinued from all study interventions.

If the toxicities do resolve and conditions are aligned with what is defined in [**Table 1**], the combination of temozolomine and pembrolizumab may be restarted at the discretion of the investigator.[In these cases where the toxicity is attributed to [the combination or to temozolomine alone, re-initiation of pembrolizumab as a monotherapy may be considered at the principal investigator's discretion .

**Table 1: Dose Modification and Toxicity Management Guidelines for Immune-related AEs Associated with Pembrolizumab**

General instructions:

1. Severe and life-threatening irAEs should be treated with IV corticosteroids followed by oral steroids. Other immunosuppressive treatment should begin if the irAEs are not controlled by corticosteroids.
2. Study intervention must be permanently discontinued if the irAE does not resolve or the corticosteroid dose is not ≤10 mg/day within 12 weeks of the last study intervention treatment.
3. The corticosteroid taper should begin when the irAE is ≤ Grade 1 and continue at least 4 weeks.
4. If study intervention has been withheld, study intervention may resume after the irAE decreased to ≤ Grade 1 after corticosteroid taper.

| irAEs | Toxicity Grade (CTCAE v5.0) | Action With Pembrolizumab | Corticosteroid and/or Other Therapies | Monitoring and Follow-up |
|---|---|---|---|---|
| Pneumonitis | Grade 2 | Withhold | • Administer corticosteroids (initial dose of 1 to 2 mg/kg prednisone or equivalent) followed by taper<br>• Add prophylactic antibiotics for opportunistic infections | • Monitor participants for signs and symptoms of pneumonitis<br>• Evaluate participants with suspected pneumonitis with radiographic imaging and initiate corticosteroid treatment |
| | Recurrent Grade 2, Grade 3 or 4 | Permanently discontinue | | |
| Diarrhea/Colitis | Grade 2 or 3 | Withhold | • Administer corticosteroids (initial dose of 1 to 2 mg/kg prednisone or equivalent) followed by taper | • Monitor participants for signs and symptoms of enterocolitis (ie, diarrhea, abdominal pain, blood or mucus in stool with or without fever) |

| irAEs | Toxicity Grade (CTCAE v5.0) | Action With Pembrolizumab | Corticosteroid and/or Other Therapies | Monitoring and Follow-up |
|---|---|---|---|---|
| | Recurrent Grade 3 or Grade 4 | Permanently discontinue | | and of bowel perforation (ie, peritoneal signs and ileus)<br>• Participants with ≥Grade 2 diarrhea suspecting colitis should consider GI consultation and performing endoscopy to rule out colitis<br>• Participants with diarrhea/colitis should be advised to drink liberal quantities of clear fluids. If sufficient oral fluid intake is not feasible, fluid and electrolytes should be substituted via IV infusion |
| AST or ALT Elevation or Increased Bilirubin | Grade 2 [a] | Withhold | • Administer corticosteroids (initial dose of 0.5 to 1 mg/kg prednisone or equivalent) followed by taper | • Monitor with liver function tests (consider weekly or more frequently until liver enzyme value returned to baseline or is stable) |
| | Grade 3 [b] or 4 [c] | Permanently discontinue | • Administer corticosteroids (initial dose of 1 to 2 mg/kg prednisone or equivalent) followed by taper | |
| T1DM or Hyperglycemia | New onset T1DM or Grade 3 or 4 hyperglycemia associated with | Withhold [d] | • Initiate insulin replacement therapy for participants with T1DM | • Monitor participants for hyperglycemia or other signs and symptoms of diabetes |

| irAEs | Toxicity Grade (CTCAE v5.0) | Action With Pembrolizumab | Corticosteroid and/or Other Therapies | Monitoring and Follow-up |
|---|---|---|---|---|
| | evidence of β-cell failure | | • Administer antihyperglycemic in participants with hyperglycemia | |
| Hypophysitis | Grade 2 | Withhold | • Administer corticosteroids and initiate hormonal replacements as clinically indicated | • Monitor for signs and symptoms of hypophysitis (including hypopituitarism and adrenal insufficiency) |
| | Grade 3 or 4 | Withhold or permanently discontinue [d] | | |
| Hyperthyroidism | Grade 2 | Continue | • Treat with nonselective beta-blockers (eg, propranolol) or thionamides as appropriate | • Monitor for signs and symptoms of thyroid disorders |
| | Grade 3 or 4 | Withhold or permanently discontinue [d] | | |
| Hypothyroidism | Grade 2, 3 or 4 | Continue | • Initiate thyroid replacement hormones (eg, levothyroxine or liothyronine) per standard of care | • Monitor for signs and symptoms of thyroid disorders |
| Nephritis: grading according to increased creatinine or acute kidney injury | Grade 2 | Withhold | • Administer corticosteroids (prednisone 1 to 2 mg/kg or equivalent) followed by taper | • Monitor changes of renal function |
| | Grade 3 or 4 | Permanently discontinue | | |
| | Grade 2 | Withhold | | |

| irAEs | Toxicity Grade (CTCAE v5.0) | Action With Pembrolizumab | Corticosteroid and/or Other Therapies | Monitoring and Follow-up |
|---|---|---|---|---|
| Neurological Toxicities | Grade 3 or 4 | Permanently discontinue | • Based on severity of AE administer corticosteroids | • Ensure adequate evaluation to confirm etiology and/or exclude other causes |
| Myocarditis | Grade 1 | Withhold | • Based on severity of AE administer corticosteroids | • Ensure adequate evaluation to confirm etiology and/or exclude other causes |
| | Grade 2, 3 or 4 | Permanently discontinue | | |
| Exfoliative Dermatologic Conditions | Suspected SJS, TEN, or DRESS | Withhold | • Based on severity of AE administer corticosteroids | • Ensure adequate evaluation to confirm etiology or exclude other causes |
| | Confirmed SJS, TEN, or DRESS | Permanently discontinue | | |
| All Other irAEs | Persistent Grade 2 | Withhold | • Based on severity of AE administer corticosteroids | • Ensure adequate evaluation to confirm etiology or exclude other causes |
| | Grade 3 | Withhold or discontinue based on the event [e] | | |
| | Recurrent Grade 3 or Grade 4 | Permanently discontinue | | |

AE(s)=adverse event(s); ALT= alanine aminotransferase; AST=aspartate aminotransferase; CTCAE=Common Terminology Criteria for Adverse Events; DRESS=Drug Rash with Eosinophilia and Systemic Symptom; GI=gastrointestinal; IO=immuno-oncology; ir=immune related; IV=intravenous; SJS=Stevens-Johnson Syndrome; T1DM=type 1 diabetes mellitus; TEN=Toxic Epidermal Necrolysis; ULN=upper limit of normal.

**Note: Non-irAE will be managed as appropriate, following clinical practice recommendations.**

| irAEs | Toxicity Grade (CTCAE v5.0) | Action With Pembrolizumab | Corticosteroid and/or Other Therapies | Monitoring and Follow-up |
|---|---|---|---|---|
| [a] AST/ALT: >3.0 to5.0 x ULN if baseline normal; >3.0 to 5.0 x baseline, if baseline abnormal; bilirubin:>1.5 to 3.0 x ULN if baseline normal; >1.5 to 3.0 x baseline if baseline abnormal<br>[b] AST/ALT: >5.0 to 20.0 x ULN, if baseline normal; >5.0 to 20.0 x baseline, if baseline abnormal; bilirubin:>3.0 to 10.0 x ULN if baseline normal; >3.0 to 10.0 x baseline if baseline abnormal<br>[c] AST/ALT: >20.0 x ULN, if baseline normal; >20.0 x baseline, if baseline abnormal; bilirubin: >10.0 x ULN if baseline normal; >10.0 x baseline if baseline abnormal<br>[d] The decision to withhold or permanently discontinue pembrolizumab is at the discretion of the investigator or treating physician. If control achieved or ≤ Grade 2, pembrolizumab may be resumed.<br>[e] Events that require discontinuation include, but are not limited to: encephalitis and other clinically important irAEs (eg, vasculitis and sclerosing cholangitis). | | | | |

### 3.10.2 Guidelines for Drug-Related Adverse Events due to Pembrolizumab

Dosing interruptions are permitted in the case of medical / surgical events or logistical reasons not related to study therapy (e.g., elective surgery, unrelated medical events, patient vacation, and/or holidays). Subjects should be placed back on study therapy within 3 weeks of the scheduled interruption, unless otherwise discussed with the Study Chair. The reason for interruption should be documented in the patient's study record.

### 3.10.3 Pembrolizumab Supportive Care

Participants should receive appropriate supportive care measures as deemed necessary by the treating investigator. Suggested supportive care measures for the management of adverse events with potential immunologic etiology are outlined below. Where appropriate, these guidelines include the use of oral or intravenous treatment with corticosteroids as well as additional anti-inflammatory agents if symptoms do not improve with administration of corticosteroids. Note that several courses of steroid tapering may be necessary as symptoms may worsen when the steroid dose is decreased. For each disorder, attempts should be made to rule out other causes such as metastatic disease or bacterial or viral infection, which might require additional supportive care. The treatment guidelines are intended to be applied when the investigator determines the events to be related to pembrolizumab.

Note: If after the evaluation the event is determined not to be related, the investigator does not need to follow the treatment guidance (as outlined below). Refer to Section **3.10** for dose modification.

It may be necessary to perform conditional procedures such as bronchoscopy, endoscopy, or skin photography as part of evaluation of the event.

- **Pneumonitis**:
    - For **Grade 2 events**, treat with systemic corticosteroids. When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks.
    - For **Grade 3-4 events**, immediately treat with intravenous steroids. Administer additional anti-inflammatory measures, as needed.
    - Add prophylactic antibiotics for opportunistic infections in the case of prolonged steroid administration.
- **Diarrhea/Colitis**:

Subjects should be carefully monitored for signs and symptoms of enterocolitis (such as diarrhea, abdominal pain, blood or mucus in stool, with or without fever) and of bowel perforation (such as peritoneal signs and ileus).

- All subjects who experience diarrhea/colitis should be advised to drink liberal quantities of clear fluids. If sufficient oral fluid intake is not feasible, fluid and electrolytes should be substituted via IV infusion. For Grade 2 or higher diarrhea, consider GI consultation and endoscopy to confirm or rule out colitis.
- For **Grade 2 diarrhea/colitis,** administer oral corticosteroids.

- For **Grade 3 or 4 diarrhea/colitis,** treat with intravenous steroids followed by high dose oral steroids.
- When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks.

- **Type 1 diabetes mellitus (if new onset, including diabetic ketoacidosis [DKA]) or ≥ Grade 3 Hyperglycemia, if associated with ketosis (ketonuria) or metabolic acidosis (DKA)**
  - For **T1DM** or **Grade 3-4** Hyperglycemia
    - Insulin replacement therapy is recommended for Type I diabetes mellitus and for Grade 3-4 hyperglycemia associated with metabolic acidosis or ketonuria.
    - Evaluate patients with serum glucose and a metabolic panel, urine ketones, glycosylated hemoglobin, and C-peptide.
- **Hypophysitis**:
  - For **Grade 2** events, treat with corticosteroids. When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks. Replacement of appropriate hormones may be required as the steroid dose is tapered.
  - For **Grade 3-4** events, treat with an initial dose of IV corticosteroids followed by oral corticosteroids. When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks. Replacement of appropriate hormones may be required as the steroid dose is tapered.
- **Hyperthyroidism or Hypothyroidism**:

Thyroid disorders can occur at any time during treatment. Monitor patients for changes in thyroid function (at the start of treatment, periodically during treatment, and as indicated based on clinical evaluation) and for clinical signs and symptoms of thyroid disorders.

- **Grade 2** hyperthyroidism events (and **Grade 2-4** hypothyroidism):
  - In hyperthyroidism, non-selective beta-blockers (e.g. propranolol) are suggested as initial therapy.
  - In hypothyroidism, thyroid hormone replacement therapy, with levothyroxine or liothyronine, is indicated per standard of care.
- **Grade 3-4** hyperthyroidism
  - Treat with an initial dose of IV corticosteroid followed by oral corticosteroids. When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks. Replacement of appropriate hormones may be required as the steroid dose is tapered.

- **Hepatic**:
    - For **Grade 2** events, monitor liver function tests more frequently until returned to baseline values (consider weekly).
        - Treat with IV or oral corticosteroids
    - For **Grade 3-4** events, treat with intravenous corticosteroids for 24 to 48 hours.
    - When symptoms improve to Grade 1 or less, a steroid taper should be started and continued over no less than 4 weeks.
- **Renal Failure or Nephritis**:
    - For **Grade 2** events, treat with corticosteroids.
    - For **Grade 3-4** events, treat with systemic corticosteroids.
    - When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks.
- **Management of Infusion Reactions**: Signs and symptoms usually develop during or shortly after drug infusion and generally resolve completely within 24 hours of completion of infusion.

Refer to **Infusion Reaction Treatment Guidelines** Appendix **16.8** for subjects who experience an infusion reaction associated with administration of pembrolizumab.

### 3.10.4 Temozolomide Dose Modifications

**NOTE**: TMZ dose is modified ONLY if toxicity is specifically attributed to TMZ.

TMZ can be delayed at the beginning of a cycle (e.g., C2D1) and then restarted when the reason for a delay has resolved. If TMZ is held (e.g., for toxicity) at mid-cycle (e.g., C2D29) it can be delayed up to 2 weeks. When TMZ is restarted, for example, 1 week later (e.g., D36) the cycle date entered in the CRF will automatically be labelled as D36, not D29; please enter a comment in the CRF that TMZ was held for X amount of days.

If during a TMZ dosing period, there is a drop in ANC to < 1.5 or platelets < 75 000 a dose reduction should occur in the subsequent cycle. See tables.

| **RT Phase** | | |
|---|---|---|
| **Dose level** | **Temozolomide dose** | **Dose reduce to** |
| 0 (starting dose) | 75 mg/m$^2$ | 60 mg/m$^2$ |
| -1b | 60 mg/m$^2$ | 50 mg/m$^2$ |
| -2b | 50 mg/m$^2$ | 40 mg/m$^2$ |

| Post RT phase | | |
|---|---|---|
| **Dose level** | **Temozolomide dose** | **Dose reduce to** |
| 0<br>(starting dose for cycle 2 and beyond) | 200 mg/m$^2$ | 150 mg/m$^2$ |
| -1<br>(starting dose for cycle 1) | 150 mg/m$^2$ | 125 mg/m$^2$ |
| -2 | 125 mg/m$^2$ | 100 mg/m$^2$ |

### 3.10.5 HSPPC-96/Placebo Dose Modifications

For grade 1 or 2 reactions considered related to HSPPC-96/Placebo, manage symptoms according to institutional procedures. Continue on study.

For grade 3 or 4 allergic reactions considered related to HSPPC-96/Placebo, discontinue HSPPC-96/Placebo.

### 3.10.6 Vaccine Injection Site Reactions

For grade 1 or 2 injection site reactions, continue on study; make sure to rotate injection sites as instructed in Administration Section **14.3.8.** Manage symptoms according to institutional procedures.
For grade 3 or 4 injection site reactions, discontinue HSPPC-96/Placebo.

**For other grade 1 or 2 non-hematologic toxicity,** continue on protocol once toxicity is resolved to < grade 1.
**For other grade 3 or 4 non-hematologic toxicity considered at least possibly related to HSPPC-96,** discontinue HSPPC-96/Placebo.

## 3.11 STUDY CALENDAR

| **Time Period** | **Screening[7]** | | **Treatment** | | | | **Follow Up** | |
|---|---|---|---|---|---|---|---|---|
| | **Pre-Surgery Screening** | **Surgery** | **RT + TMZ + Pembrolizumab[16]** | **HSPPC-96/Vaccine Induction or Break[14] (4 weeks)** | **TMZ+Pembrolizumab +/- HSPPC-96/Vaccine (6 cycles)** | **End of Treatment or Discontinuation** | **30 days after last dose** | **Long-term Follow-up[8]** |
| **Scheduling Window (Days)** | **+/-14 of signing consent** | | **- 7 (before treatment)** | | **- 7 (before treatment)** | **+/- 3** | | **Every 8-16 weeks** |
| Informed Consent | X | | | | | | | |
| Medical history | X | | | | | | | |
| Prior & concomitant medications review | X | | | | | | | |
| Assess and document first post-protocol treatment | | | | | | | | X |
| Assess and document Adverse Events[12] | | | X[12] | X[12] | X[12] | X | X | |
| Physical, KPS & neurological exam[13] | X | | X[13] | X[13] | X[13] | X | X | |
| Vital Signs and Weight[1] | X | | X | X | X | X | X | |
| Pregnancy test[2] | X | | X[2] | X[2] | X[2] | | | |
| PT/INR or aPTT | X | | X | | | | | |
| CBC with diff | X | | X[4] | X[4] | X[4] | X | X | |
| Comprehensive Chemistry panel[9] | X | | X[5] | X[5] | X[5] | X | | |
| Urinalysis | X | | X | | | | | |
| T3, T4, and TSH | | | X[5] | X[5] | X[5] | X | X | |
| Tumor Imaging[3] | X | X[3] | | X[3] | X[3] | X | X | |
| Archival or Collection Tumor | | X | | | | | | |
| MDASI-BT[11] | | | X | | X | X | X | |
| Correlative Studies Blood Collection | | | X[6] | X[6] | X[6] | X[6] | | |

| Time Period | Screening[7] | | Treatment | | | | Follow Up | |
|---|---|---|---|---|---|---|---|---|
| | Pre-Surgery Screening | Surgery | RT + TMZ + Pembrolizumab[16] | HSPPC-96/Vaccine Induction or Break[14] (4 weeks) | TMZ+Pembrolizumab +/- HSPPC-96/Vaccine (6 cycles) | End of Treatment or Discontinuation | 30 days after last dose | Long-term Follow-up[8] |
| **Scheduling Window (Days)** | **+/-14 of signing consent** | | **- 7 (before treatment)** | | **- 7 (before treatment)** | **+/- 3** | | **Every 8-16 weeks** |
| Drug Administration | | | X[15] | X[10] | X[10] | | | |
| Assess and document Survival Status | | | | | | | | X |

[1] Includes vital signs (pulse, respirations, blood pressure) and height (baseline only) and weight.
[2] Serum or urine test for females of child-bearing potential prior to the start of RT+TMZ+Pembro and at the start of every cycle.
[3] By MRI (unless contraindicated): For subjects not randomized: An MRI will be done pre surgery, post surgery (within 72 hours), 4 weeks post RT, and then every 9 weeks +/- 1 week. For subjects who are randomized: An MRI will be done pre surgery, post surgery (within 72 hours), within +/- 10 days of last dose of vaccine or placebo induction, and then every 9 weeks +/- 1 week. This schedule of MRIs is consistent with standard of care.
[4] A CBC with differential will be done weekly during RT and then weekly after RT for the first year and then every 2 weeks for the year after if the patient remains on Pembrolizumab**.**
[5] Chemistry should be done at beginning of RT**,** Day 21 of RT +/- 3 days, at completion of RT, and on Days 1(+/- 1 day) and Day 29 (- 7 days) of each cycle.
[6] Randomized - 8x8mL CPT blue and black top tubes (LabConnect Kit B) will be used to collect correlative blood samples at the following time points for patients randomized to study vaccine or placebo: 1) Pre-RT treatment (RT+TMZ+Pembro) 2) Post-RT treatment (RT+TMZ+Pembro)/Pre-vaccine (or placebo) induction 3) Post Vaccine (or placebo) induction/Pre-cycle 1 maintenance 4) Every Day 1 on Cycle 2, C4, C6, C8 (if continuing on Pembrolizumab); then, at discretion of Study Chair and BTTC Coordinating Center, every other cycle; 5) At time of progression or at End of Treatment.
Ancillary - 8x8mL CPT blue and black top tubes (LabConnect Kit B) will be used to collect correlative blood samples at the following time points for patients not randomized (ancillary cohort): 1) Pre-RT treatment (RT+TMZ+Pembro) 2) /Pre-cycle 1 maintenance 3) Every Day 1 on Cycle 2, C4, C6, C8 (if continuing on Pembrolizumab); then, at discretion of Study Chair and BTTC Coordinating Center, every other cycle 4) At time of progression, or at End of Treatment.
[7] Screening labs will be completed within (+/-) 14 days of signing consent.
[8] Patients will be followed for overall survival. Patients withdrawn from treatment will follow the same schedule.
[9] Comprehensive Serum Chemistry panel=Na, K, CL, Bicarbonate, BUN/Cr, AST, ALT, ALP T. Bili, Calcium, Albumin.

**[10]** HSPPC-96 Vaccine/Placebo given weekly post RT x 4 weeks, then 21 days (+/- 24 hours) after the day 5 and day 33 doses of TMZ of each cycle. TMZ doses Cycle 1, days 1-5 at 150 mg/m2. If tolerated, the dose will be escalated to 200 mg/m2 for all remaining doses (This would begin with Cycle 1 days 29-33.). Pembrolizumab dosed on day 1, 22, and 43 (+/- 24 hours for each timepoint) of RT then day 1, 22, 43 (+/- 24 hours for each timepoint) of each 9 week cycle.
**[11]** MDASI-BT assessments will take place along with imaging studies. MDASI-BT will need to be given prior to informing the patients about the results of the imaging study, within +/- 1 week of the MRI assessment date.
**[12]** Assess patients for adverse events during Physical Exam and treatment days.
**[13]** Physical, KPS and neurological exam every 3 weeks (- 3 days to + 7 days) during RT and then every 4 weeks (+/- 7 days) after RT. If needed, telehealth assessments will be allowed for the physical, KSP & neurological exams. For additional details on allowed telehealth assessments, please see Section **3.9**.
**[14]** For those not randomized (ancillary arm), no drug administration during the 4 weeks prior to Pembro and TMZ, therefore no laboratory tests or scheduled assessments are required.
**[15]** TMZ may be given night before RT (within 24 hours before day 1).
**[16]** Baseline assessments will be done prior to the start of RT+TMZ+Pembro (see **3.7**).

## 3.12 SURGICAL GUIDELINES

Surgical guidelines will be according to Standard of Care at each institution.

## 3.13 COST AND COMPENSATION

### 3.13.1 Costs

Subjects costs will be based on local guidelines as described in the site specific consent.

### 3.13.2 Compensation

Participants will not be compensated on this study.

### 3.13.3 Reimbursement

Reimbursement is based on site policy.

At the NCI site, the NCI will cover the costs of some expenses associated with protocol participation. Some of these costs may be paid directly by the NIH and some may be reimbursed to the participant/guardian as appropriate. The amount and form of these payments are determined by the NCI Travel and Lodging Reimbursement Policy.

## 3.14 CRITERIA FOR REMOVAL FROM PROTOCOL THERAPY AND OFF STUDY CRITERIA

Patients can be taken off the protocol therapy and/or study as a whole at any time at their own request, or they may be withdrawn at the discretion of the investigator for safety, behavioral or administrative reasons.

Prior to documenting removal from study, effort must be made to have all subjects complete a safety visit approximately 30 days following the last dose of study therapy. The following evaluation will occur:

- KPS
- Toxicity Assessment
- Physical & Neurological Exam
- Vital Signs and Weight
- CBC with Diff, Comprehensive Chemistry Panel, T3, T4, and TSH
- Tumor Imaging
- MDASI-BT
- Correlative Blood Collection

### 3.14.1 Criteria for Removal from Protocol Therapy

The reason(s) for discontinuation must be clearly documented on the appropriate eCRF and may include:

- Patient voluntarily withdraws from treatment (follow-up permitted)
- Patient demonstrates disease progression after beginning treatment
- Patient experiences unacceptable toxicity
- Treating physician determines that continuation with treatment on the study would not be in the patient’s best interest

- Patient develops a second malignancy that requires treatment which would interfere with this study
- Patient becomes pregnant
- Patient completes treatment as outlined in section **3.3**
- Screen failure

### 3.14.2 Off Study Criteria

The reason(s) for discontinuation must be clearly documented on the appropriate eCRF and may include:

- Screen failure
- Tumor is identified as a methylated MGMT promoter
- Tumor is not IDH wildtype by pathology
- Investigator decision to end study
- Patient withdraws consent (no follow-up permitted)
- Patient is unable to comply with protocol requirements
- Patient becomes lost to follow-up (LTF)
- Death

### 3.14.3 Lost to Follow-up

A participant will be considered lost to follow-up if he or she fails to return for 3 scheduled visits and is unable to be contacted by the study site staff.

The following actions must be taken if a participant fails to return to the clinic for a required study visit:

- The site will attempt to contact the participant and reschedule the missed visit within 24 hours, and reschedule within 7 days and counsel the participant on the importance of maintaining the assigned visit schedule and ascertain if the participant wishes to and/or should continue in the study.
- Before a participant is deemed lost to follow-up, the investigator or designee will make every effort to regain contact with the participant (where possible, 3 telephone calls and, if necessary, a certified letter to the participant's last known mailing address or local equivalent methods). These contact attempts should be documented in the participant's medical record or study file.
- Should the participant continue to be unreachable, he or she will be considered to have withdrawn from the study with a primary reason of lost to follow-up.

## 4 CONCOMITANT MEDICATIONS/VACCINATIONS (ALLOWED & PROHIBITED)

Medications or vaccinations specifically prohibited in the exclusion criteria are not allowed during the ongoing trial. If there is a clinical indication for one of these or other medications or vaccinations specifically prohibited during the trial, discontinuation from trial therapy or vaccination may be required. The investigator should discuss any questions regarding this with

the coordinating center. The final decision on any supportive therapy or vaccination rests with the investigator and/or the subject's primary physician.

## 4.1 ACCEPTABLE CONCOMITANT MEDICATIONS

All treatments that the investigator considers necessary for a subject's welfare may be administered at the discretion of the investigator in keeping with the community standards of medical care. All concomitant medication will be recorded on the case report form (CRF) including all prescription, over-the-counter (OTC), herbal supplements, and IV medications and fluids. If changes occur during the trial period, documentation of drug dosage, frequency, route, and date may also be included on the CRF.

All concomitant medications received within 28 days before the first dose of trial treatment and 30 days after the last dose of trial treatment should be recorded. Concomitant medications administered after 30 days after the last dose of trial treatment should be recorded for SAEs and ECIs as defined in Section **8.1**.

## 4.2 PROHIBITED CONCOMITANT MEDICATIONS

Subjects are prohibited from receiving the following therapies during the Screening and Treatment Phase (including retreatment for post-complete response relapse) of this trial:

- Antineoplastic systemic chemotherapy or biological therapy
- Immunotherapy not specified in this protocol
- Chemotherapy not specified in this protocol
- Investigational agents other than pembrolizumab and HSPPC-96
- Live vaccines within 30 days prior to the first dose of trial treatment and while participating in the trial. Examples of live vaccines include, but are not limited to, the following: measles, mumps, rubella, varicella/zoster, yellow fever, rabies, BCG, and typhoid vaccine.
- Systemic glucocorticoids for any purpose other than to modulate symptoms from an event of clinical interest of suspected immunologic etiology. The use of physiologic doses of corticosteroids may be approved after consultation with the Sponsor. The exception is for cerebral edema, but dose should be $\leq$ 4 mg once treatment begins.
- Bevacizumab can be used to control edema, but not as a biologic treatment intended for the patient's disease under study.

Subjects who, in the assessment by the investigator, require the use of any of the aforementioned treatments for clinical management should be removed from the trial. Subjects may receive other medications that the investigator deems to be medically necessary.

The Exclusion Criteria describes other medications which are prohibited in this trial.

There are no prohibited therapies during the Post-Treatment Follow-up Phase.

## 4.3 DIET/ACTIVITY/OTHER CONSIDERATIONS

### 4.3.1 Diet

Subjects should maintain a normal diet unless modifications are required to manage an AE such as diarrhea, nausea or vomiting.

### 4.3.2 Contraception

Pembrolizumab may have adverse effects on a fetus in utero. Furthermore, it is not known if pembrolizumab has transient adverse effects on the composition of sperm.

For this trial, male subjects will be considered to be of non-reproductive potential if they have azoospermia (whether due to having had a vasectomy or due to an underlying medical condition).

Female subjects will be considered of non-reproductive potential if they are either:

(1) postmenopausal (defined as at least 12 months with no menses without an alternative medical cause; in women < 45 years of age a high follicle stimulating hormone (FSH) level in the postmenopausal range may be used to confirm a post-menopausal state in women not using hormonal contraception or hormonal replacement therapy. In the absence of 12 months of amenorrhea, a single FSH measurement is insufficient.);

OR

(2) have had a hysterectomy and/or bilateral oophorectomy, bilateral salpingectomy or bilateral tubal ligation/occlusion, at least 6 weeks prior to screening;

OR

(3) has a congenital or acquired condition that prevents childbearing.

Female and male subjects of reproductive potential must agree to avoid becoming pregnant or impregnating a partner, respectively, while receiving study drug and for 120 days after the last dose of study drug by complying with one of the following:

(1) practice abstinence† from heterosexual activity;

OR

(2) use (or have their partner use) acceptable contraception during heterosexual activity.

Acceptable methods of contraception are‡:

Single method (one of the following is acceptable):

- intrauterine device (IUD)
- vasectomy of a female subject's male partner
- contraceptive rod implanted into the skin

Combination method (requires use of two of the following):

- diaphragm with spermicide (cannot be used in conjunction with cervical cap/spermicide)
- cervical cap with spermicide (nulliparous women only)
- contraceptive sponge (nulliparous women only)

- male condom or female condom (cannot be used together)
- hormonal contraceptive: oral contraceptive pill (estrogen/progestin pill or progestin-only pill), contraceptive skin patch, vaginal contraceptive ring, or subcutaneous contraceptive injection

†Abstinence (relative to heterosexual activity) can be used as the sole method of contraception if it is consistently employed as the subject's preferred and usual lifestyle and if considered acceptable by local regulatory agencies and ERCs/IRBs. Periodic abstinence (e.g., calendar, ovulation, sympto-thermal, post-ovulation methods, etc.) and withdrawal are not acceptable methods of contraception.

‡If a contraceptive method listed above is restricted by local regulations/guidelines, then it does not qualify as an acceptable method of contraception for subjects participating at sites in this country/region.

Subjects should be informed that taking the study medication may involve unknown risks to the fetus (unborn baby) if pregnancy were to occur during the study. In order to participate in the study subjects of childbearing potential must adhere to the contraception requirement (described above) from the day of study medication initiation (or 14 days prior to the initiation of study medication for oral contraception) throughout the study period up to 120 days after the last dose of trial therapy. If there is any question that a subject of childbearing potential will not reliably comply with the requirements for contraception, that subject should not be entered into the study.

# 5 BIOSPECIMEN COLLECTION

## 5.1 CORRELATIVE STUDIES FOR RESEARCH/PHARMACOKINETIC STUDIES

The purpose of the correlative studies is to determine if there is a correlation with clinical outcome of PD1 or PDL1 expression and T-Cell infiltrate in pathology from first, and if applicable, second surgical specimens with outcome.

- To assess tumor tissue for neo-antigens (initial tumor pathology).
- To collect peripheral monocytes for PD-L1 levels to confirm that low level expression leads to better outcome and if higher levels that are treated with Pembrolizumab might improve outcome. To perform other immune monitoring assessments including, but are not limited to: whole exome sequencing (WES), immune phenotyping of immune cell subsets, functional characterization of immune cell subsets, measurement of immune mediators and the assessment of circulating tumor DNA (ctDNA).
- To correlate MGMT and IDH status with outcome (local pathology report).

At each timepoint noted in the study calendar, correlative blood will be collected in 8 mononuclear cell preparation tubes (LabConnect Kit B, 8mL CPT™ blue and black top tubes) and processed for the isolation of both PBMCs and plasma. As detailed in the LabConnect lab manual, Correlative blood will be collected at NCI and participating BTTC sites and sent to:

NCI Frederick Lab
Leidos Biomedical Research

Attn: Theresa Burks
1050 Boyles Street
Bldg. 469/Room 121
Frederick, MD 21702
Phone 301-846-5125, or 301-846-6865
Email: burkst@mail.nih.gov, cc: jon.inglefield@nih.gov

### 5.1.1 Collection of correlative blood time points:

**NOTE:** The schedule of correlative blood sample collection should be held/delayed when treatment dosing is held. See section 5 Biospecimen Collection, and at the end of Table 1, and/or the calendar.

**For participants randomized to study vaccine or placebo:**

1) Pre-RT treatment (RT+TMZ+Pembro)
2) Post-RT treatment (RT+TMZ+Pembro)/Pre-vaccine (or placebo) induction
3) Post Vaccine (or placebo) induction/Pre-cycle 1 maintenance
4) Every Day 1 on Cycle 2, C4, C6, C8 (if continuing on Pembrolizumab); then, at discretion of Study Chair and BTTC Coordinating Center, every other cycle
5) At time of progression or at end of treatment

**For participants not randomized (ancillary cohort 3)**:

1) Pre-RT treatment (RT+TMZ+Pembro)
2) Pre-cycle 1 maintenance
3) Every Day 1 on Cycle 2, C4, C6, C8 (if continuing on Pembrolizumab); then, at discretion of Study Chair and BTTC Coordinating Center, every other cycle
4) At time of progression or at end of treatment

LabConnect (Fee for Service Provider) will supply kits and track shipments for correlative blood collection. (See LabConnect lab manual.)

### 5.1.2 PD-L1 expression on tumor cells

A collection of sectioned slides from tumor samples for the purpose of immunohistochemical (IHC) analysis will be sent for PD-L1 analysis at the end of the study. Sectioned slides from formalin-fixed, paraffin-embedded (FFPE) samples will be stained for the biomarker(s) of interest, and then evaluated by a pathologist for review of biomarker staining expression levels.

This sample will be taken at time of surgery. At the end of the study, the BTTC CC Research Nurse will contact the participating site to request the slides.

### 5.1.3 Tissue will be interrogated by Next Generation Sequencing for tumor antigens

Generation of the HSPPC-96 vaccine requires ≥ 7 grams of tissue. **All available** tumor aside from what will be used for routine pathologic studies and diagnosis should be sent for HSPPC-96 vaccine preparation. At the end of the study, up to 18 FFPE unstained slides (formalin fixed

paraffin embedded) may be collected. If subsequent tumor biopsies are obtained, including at time of progression, additional tissue specimens may be collected following the same method described above. **These tissue collections for next generation sequencing are at the investigator's discretion.**

The **FFPE Tumor Tissue Slide** specimens are maintained at room temperature and shipped ambient by the site upon request by BTTC Coordinating Center to:

Attn: BTTC Coordinating Center Research Nurse Coordinator
NCI / Neuro-Oncology Branch / Brain Tumor Trials Collaborative
National Institutes of Health
Building 82 , Room 215
Bethesda, MD 20892
Phone: 240-858-7083
Email: ukeme.ikiddeh-barnes@nih.gov

### 5.1.4 Tissue Collection at NCI

Tumor samples will be collected at time of surgery by the Laboratory of Pathology NCI. Tissue sections will be obtained and will be labeled with the Study Subject ID number. The relationship between the Study Subject ID number and the patient clinical information will be stored in a secure database that is maintained and regularly backed up by the Laboratory of Pathology NCI.

The Laboratory of Pathology at NCI will process the tissue samples per Appendices **16.2**, **16.3**, **16.4**, **16.5**.

### 5.1.5 Sample Storage, Tracking and Disposition

All specimens obtained in the protocol are used as defined in the protocol. Any specimens that are remaining at the completion of the protocol will be stored in the conditions described below. The study will remain open so long as sample or data analysis continues. Samples from consenting subjects will be stored until they are no longer of scientific value or if a subject withdraws consent for their continued use, at which time they will be destroyed.

If the patient withdraws consent the participant's data will be excluded from future distributions, but data that have already been distributed for approved research use will not be able to be retrieved.

The PI will record any loss or unanticipated destruction of samples as a deviation. Reporting will be per the requirements of section **7.2**.

## 5.2 SAMPLES FOR GENETIC/GENOMIC ANALYSIS

### 5.2.1 Scope of the analysis

Patients' tissue specimens will be analyzed for exploratory tumor-derived biomarkers. Assays performed may include, but are not limited to: a) immunohistochemistry (IHC) of FFPE samples to determine the frequency of tumor infiltrating lymphocytes (TILs) such as CD8 T cells and CD4 regulatory T cells, as well as the expression of predictive biomarkers such as PD-L1; b) whole exome sequencing (WES) and transcriptome analysis (RNAseq) of flash frozen/OCT

samples to evaluate mutational burden and expression of neo-epitopes; and c) functional and phenotypic characterization of vitally frozen immune cell subsets.

### 5.2.2 Certificate of Confidentiality

As part of study efforts to provide confidentiality of subject information, this study has obtained a Certificate of Confidentiality, which helps to protect personally identifiable research information. The Certificate of Confidentiality allows investigators on this trial to refuse to disclose identifying information related to the research participants, should such disclosure have adverse consequences for subjects or damage their financial standing, employability, insurability or reputation. The informed consent includes the appropriate coverage and restrictions of the Certificate of Confidentiality.

### 5.2.3 Incidental Findings

Subjects will be contacted if a clinically actionable gene variant is discovered. Clinically actionable findings for the purpose of this study are defined as disorders appearing in the American College of Medical Genetics and Genomics recommendations for the return of incidental findings that is current at the time of primary analysis. (A list of current guidelines is maintained on the CCR intranet: https://ccrod.cancer.gov/confluence/display/CCRCRO/Incidental+Findings+Lists). Subjects will be contacted at this time with a request to provide a blood sample to be sent to a CLIA certified laboratory. If the research findings are verified in the CLIA certified lab, the subject will be offered the opportunity to come to NIH (at our expense) to have genetic education and counseling to explain this result. If the subject does not want to come to NIH, a referral to a local genetic healthcare provider will be provided (at their expense). Subjects enrolled at participating BTTC sites will be referred to a local genetics health care provider.

This is the only time during the course of the study that incidental findings will be returned. No interrogations regarding clinically actionable findings will be made after the primary analysis.

# 6 DATA COLLECTION AND EVALUATION

## 6.1 DATA COLLECTION

The PI will be responsible for overseeing entry of data into a 21 CFR Part 11-compliant data capture system provided by the NCI CCR and ensuring data accuracy, consistency and timeliness. The MDASI-BT Survey responses will be entered by study subjects directly into Scribe/Labmatrix. The principal investigator, associate investigators/research nurses and/or a contracted data manager will assist with the data management efforts. All data will have identifiers so that research data can be attributed to an individual human subject participant.

All adverse events, including clinically significant abnormal findings on laboratory evaluations, regardless of severity, will be followed until return to baseline or stabilization of event. Such events will be recorded at each examination on the Adverse Event case report forms/worksheets. The investigator will make every attempt to follow all subjects with non-serious adverse events for outcome.

For the time period beginning at treatment initiation through 90 days following cessation of treatment, or 30 days following cessation of treatment if the subject initiates new anticancer therapy, whichever is earlier, any serious adverse event, or follow up to a serious adverse event, including death due to any cause, whether or not related to the study treatments, must be reported.

### 6.1.1 Exceptions to Data Collection

Adverse events that occur after the consent form is signed but before treatment (RT, Pembrolizumab, TMZ) must be collected by the investigator only if they cause the subject to be excluded from the trial. Sites (NCI or participating BTTC sites) will be responsible for entering data collected at their site.

Changes resulting from normal growth and development that do not vary significantly in frequency or severity from expected levels are not to be considered adverse events. Examples of this may include, but are not limited to, onset of menses or menopause occurring at a physiologically appropriate time.

An abnormal laboratory value will be considered an AE if the laboratory abnormality is characterized by any of the following:

- Results in discontinuation from the study
- Is associated with clinical signs or symptoms
- Requires treatment or any other therapeutic intervention
- Is associated with death or another serious adverse event, including hospitalization.
- Is judged by the Investigator to be of significant clinical impact
- If any abnormal laboratory result is considered clinically significant, the investigator will provide details about the action taken with respect to the test drug and about the patient's outcome.

**End of study procedures:** Data will be stored according to HHS, FDA regulations and NIH Intramural Records Retention Schedule as applicable.

**Loss or destruction of data:** Should we become aware that a major breach in our plan to protect subject confidentiality and trial data has occurred, this will be reported expeditiously per requirements in section **7.2.1**.

### 6.1.2 Source Documentation Timeframes

The following information will be entered into a 21 CFR Part 11-compliant data capture system provided by the NCI CCR and Scribe/Labmatrix within the indicated timeframes. In addition, the source documents should be provided to the BTTC Coordinating Center's Research Nurse at secure fax: 240-541-4432, encrypted email, or Secure Email and File Transfer within the indicated timeframe. The MDASI-BT responses will be entered by study subjects directly into Scribe/LabMatrix.

| Data Set / Source Documents | Schedule for Submission |
|---|---|
| Regulatory Documents<br>(as described in the BTTC Operations Manual) | Prior to Step 1 of registration |

| Data Set / Source Documents | Schedule for Submission |
|---|---|
| Registration Form (eligibility checklist) | Within 24 hours of signing Informed Consent (Step 1 registration)<br>Following surgery and confirmation of diagnosis (Step 2 registration)<br>Prior to end of RT in order to randomize subject (Step 3 registration) |
| Copy of signed & dated Informed Consent w/ HIPAA Authorization | Within 24 hours of signing Informed Consent (Step 1 registration) |
| Pathology Report (from the most recent pre-registration diagnostic biopsy or surgery) | At time of Step 2 of registration |
| Baseline MDASI – BT Questionnaire | Within 14 days (+ 3 working days) after enrollment |
| Screening/Baseline Source Documents | At time of registration (step 1 or 2, as applicable) |
| Other Source Documents | With Participant Status Updates Form |

### 6.1.3 Database Entry Timeframes

The following data should be entered into a 21 CFR Part 11-compliant data capture system provided by the NCI CCR and Scribe/Labmatrix within the specified timeframes. Source documentation will be kept at the participating site.

Sites (NCI or participating BTTC sites) will be responsible for entering data collected at their site.

| Data Set | Schedule for Data Entry |
|---|---|
| Screening data | Within 14 days of Step 1 Registration |
| Baseline data | No later than 14 days after Day 1 of radiation (RT cycle) |
| Treatment (Course/Cycle) Data<br>(To include treatment, response, AE, and clinical evaluation information) | Course initiation case report form (CRF) no later than 7 days after the cycle begins<br>All other course/cycle CRFs must be entered no later than 21 days after the cycle is complete |
| Off Treatment Data | Within 21 days after the:<br>• last date of any modality of protocol treatment; or,<br>• study discontinuation visit. |
| Follow-up (Survival) Data<br>(To include post-protocol treatment and long-term follow up survival data)<br><br>Note: Long-term follow up is required every 8-16 weeks for two years until patient is off study. | No later than 21 days after follow up visit or survival call or post-treatment medication begins. |
| Non-Treatment Data<br>(May include Quality of Life questionnaires (MDASI-BT), Specimen Tracking information, Pathology Specimen Submission, etc.) | Within 10 days after each scheduled assessment, event, or activity |
| Off Study Data | No later than 21 days after the date the patient is removed from the study. |

### 6.1.4 Confidentiality

All documents, investigative reports, or information relating to the patient are strictly confidential. Any source documents submitted to the BTTC Coordinating Center with protected health information or personally identifiable information (e.g. patient's name) must be sent via Secure Email and File Transfer (SEFT) or secure fax. For NIH only, documents can also be sent via hand-delivery or encrypted email.

### 6.1.5 Safety Data

All patients receiving study agents will be evaluated for safety. The safety parameters include all laboratory tests and hematological abnormalities, CNS observations, physical examination findings, and spontaneous reports of adverse events reported to the investigator by patients. Life-threatening toxicities that are unexpected and assessed to be possibly related to the study agent/s should be reported immediately.

Information about all adverse events, whether volunteered by the subject, discovered by investigator questioning, or detected through physical examination, laboratory test or other means, will be collected and recorded on the Adverse Event Case Report Form and followed as appropriate.

Medical conditions/diseases present before starting study treatment are only considered adverse events if they worsen after starting study treatment (any procedures specified in the protocol). Adverse events occurring before starting study treatment but after signing the informed consent form are recorded on the Baseline Evaluations Case Report Form.

### 6.1.6 Data Monitoring

All submitted data will be monitored by the BTTC Clinical Research Nurse specifically assigned to this protocol. Requests for correction of data deficiencies will be sent via emailed to the Institutional Coordinator. Any major deficiencies will be corrected by telephone and email communication. All data will be monitored for completeness. Key parameters such as drug dosages including attenuations and escalations, toxicity documentation and tumor measurements will be analyzed. All data deficiencies will be corrected within two weeks.

## 6.2 DATA SHARING PLANS

### 6.2.1 Human Data Sharing Plan

**What data will be shared?**

I will share human data generated in this research for future research as follows:

- Coded, linked data in an NIH-funded or approved public repository.
- Coded, linked data in another public repository.
- Coded, linked data in BTRIS (automatic for activities in the Clinical Center)
- Coded, linked or identified data with approved outside collaborators under appropriate agreements.

**How and where will the data be shared?**

Data will be shared through:

- An NIH-funded or approved public repository: clinicaltrials.gov.
- BTRIS (automatic for activities in the Clinical Center)
- Approved outside collaborators under appropriate individual agreements.
- Publication and/or public presentations.

**When will the data be shared?**

- Before publication.
- At the time of publication or shortly thereafter.

### 6.2.2 Genomic Data Sharing Plan

Unlinked genomic data will be deposited in public genomic databases such as dbGaP in compliance with the NIH Genomic Data Sharing Policy.

## 6.3 RESPONSE CRITERIA

### 6.3.1 Definitions

6.3.1.1 Evaluable for toxicity: All patients will be evaluable for toxicity from the time of their first treatment with Pembrolizumab and/or HSPPC-96.

6.3.1.2 Evaluable for objective response: Only those patients 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 response. These patients will have their response classified according to the definitions stated below. (Note: Patients who exhibit objective disease progression prior to the end of cycle 1 will also be considered evaluable.)

6.3.1.3 Evaluable Non-Target Disease Response: Patients 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.

### 6.3.2 Disease Parameters

6.3.2.1 Measurable disease: Based on RANO criteria [44]

### 6.3.3 RANO Criteria for Response

**Table 2: RANO Criteria for Response**

| RANO Criteria for Response [44] | | | | |
|---|---|---|---|---|
| | CR | PR | SD | PD |
| T1 gadolinium enhancing disease | None | ≥ 50% ↓ | < 50% ↓ but < 25% ↑ | ≥ 25% |
| T2/FLAIR | Stable or ↓ | Stable or ↓ | Stable or ↓ | ↑[1] |
| New lesion | None | None | None | Present[1] |
| Corticosteroids | None | Stable or ↓ | Stable or ↓ | n/a[†] |
| Clinical status | Stable or ↑ | Stable or ↑ | Stable or ↑ | ↓[1] |
| Requirement for response | All | All | All | Any[1] |

| RANO Criteria for Response [44] | | | | |
|---|---|---|---|---|
| | CR | PR | SD | PD |

[1] Progression occurs when this criterion is present

†Increase in corticosteroids alone will not be taken into account in determining progression in the absence of persistent clinical deterioration.

↓ = decreased, ↑ = increased,

CR = complete response, PR = partial response, SD = stable disease, PD = progressive disease. If PD is unclear based on white matter changes, patients may be kept on trial.

FLAIR, fluid-attenuated inversion recovery; NA, not applicable.

### 6.3.4 iRANO (Exploratory)[45]

**Table 3: iRANO**

Table 11

| | RANO Criteria | | |
|---|---|---|---|
| | **Malignant Glioma[16]** | **Low-Grade Glioma[17]** | **Brain Metastases[18]** |
| **Complete Response** | -Disappearance of all enhancing disease for ≥ 4 weeks *AND*<br>- No new lesions *AND*<br>- Stable/improved T2/FLAIR *AND*<br>- No more than physiologic steroids *AND*<br>- Stable/improved clinically | -Disappearance of all enhancing and T2/FLAIR disease for ≥ 4 weeks *AND*<br>- No new lesions *AND*<br>- No more than physiologic steroids *AND*<br>- Stable/improved clinically | -Disappearance of all enhancing target and non-target lesions for ≥ 4 weeks *AND*<br>- No new lesions *AND*<br>- No steroids *AND*<br>- Stable/improved clinically |
| **Partial Response** | - ≥ 50% ↓ sum of biperpendicular diameters of enhancing disease for ≥ 4 weeks *AND*<br>- No new lesions *AND*<br>- Stable/improved T2/FLAIR *AND*<br>- Stable/improved steroids *AND*<br>- Stable/improved clinically | - ≥ 50% ↓ sum of biperpendicular diameters of T2/FLAIR disease for ≥ 4 weeks *AND*<br>- No new lesions *AND*<br>- Stable/improved steroids *AND*<br>- Stable/improved clinically | - ≥ 30% ↓ sum of longest diameters of target lesions for ≥ 4 weeks *AND*<br>- No new lesions *AND*<br>- Stable/improved steroids *AND*<br>- Stable/improved clinically |
| **Minor Response** | - Non-applicable | - 25-49% ↓ sum of biperpendicular diameters of T2/FLAIR disease for ≥ 4 weeks *AND*<br>- No new lesions *AND*<br>- Stable/improved clinically | Not applicable |
| **Stable Disease** | - Does not qualify for CR, PR, PD *AND*<br>- No new lesions *AND*<br>- Stable/improved T2/FLAIR *AND*<br>- Stable/improved steroids *AND*<br>- Stable/improved clinically | - Does not qualify for CR, PR, PD *AND*<br>- No new lesions *AND*<br>- Stable/improved T2/FLAIR *AND*<br>- Stable/improved steroids *AND*<br>- Stable/improved clinically | - Does not qualify for CR, PR, PD |
| **Progressive Disease** | - ≥ 25% ↑ sum of biperpendicular diameters of enhancing disease *OR*<br>- New lesions *OR*<br>- Significant worsened T2/FLAIR *OR*<br>- Significant clinical decline | - ≥ 25% ↑ sum of biperpendicular diameters of T2/FLAIR disease *OR*<br>- New lesions *OR*<br>- Significant clinical decline | - ≥ 20% ↑ sum of longest diameters of target lesions *OR*<br>-Unequivocal progression of enhancing non-target lesions *OR*<br>- New lesions *OR*<br>- Significant clinical decline |
| **iRANO** | Confirmation of progression on follow-up imaging 3 months after initial radiographic progression if:<br>1. No new or significantly worsened neurologic deficits not due to co-morbid event or concurrent medication *AND*<br>2. ≤ 6 months from initiation of immunotherapy<br><br>If follow-up imaging confirms progression, the date of actual progression should be back-dated to the date of initial radiographic progression | | |

### 6.3.5 Evaluation of Best Overall Response

6.3.5.1 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).

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.

6.3.5.2 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.

### 6.3.6 Dose Limiting Toxicity

Any of the following adverse events not due to disease progression or any underlying disease will be documented as a DLT during the first 4 weeks of treatment with Pembrolizumab, TMZ and HSPPC-96/Placebo (attribution should be assigned to drug most suspected) during the first post RT cycle, after completion of induction:

- Grade 4 neutropenia for more than 7 days
- Grade 3 or 4 febrile neutropenia during the first 4 weeks
- Grade 3 or 4 thrombocytopenia for more than 7 days
- Grade 4 anemia for more than 7 days
- Grade 3 or 4 non-hematologic adverse events.
- Exceptions include:
- Grade 3 nausea, vomiting or diarrhea will only be considered a DLT if not adequately managed with maximum supportive care within 48 hours.
- More than 14 days of treatment delay due to the failure to recover from attributable toxicity.

Subjects experiencing irAEs due to Pembrolizumab and are successfully treated for toxicity, and able to be re-dosed will not be considered to have experienced DLTs. The frequency, grade, and nature of irAE will be recorded.

### 6.3.7 Progression-Free Survival

Progression free survival will be defined from the time of registration to the time of confirmed progression. In cases where this might be unclear, patients may continue on trial.

### 6.3.8 Overall survival

Defined as the time from registration to the time of death

### 6.3.9 Response Review

Response review will be done locally and by Study Chair if response is questionable.

### 6.4 TOXICITY CRITERIA

The following adverse event management guidelines are intended to ensure the safety of each patient while on the study. The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 5.0 will be utilized for AE reporting. An investigator who is a qualified physician will evaluate all adverse events according to (CTCAE), version 5.0. Any adverse event which changes CTCAE grade over the course of a given episode will have each change of grade recorded on the adverse event case report forms/worksheets.

All adverse events regardless of CTCAE grade must also be evaluated for seriousness.

All appropriate treatment areas should have access to a copy of the CTCAE version 5.0. A copy of the CTCAE version 5.0 can be downloaded from the CTEP web site (http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm ).

## 7 NIH REPORTING REQUIREMENTS/DATA AND SAFETY MONITORING PLAN

### 7.1 DEFINITIONS

Please refer to definitions provided in Policy 801: Reporting Research Events found at: https://irbo.nih.gov/confluence/pages/viewpage.action?pageId=36241835#Policies&Guidance-800Series-ComplianceandResearchEventReportingRequirements.

### 7.2 OHSRP OFFICE OF COMPLIANCE AND TRAINING / IRB REPORTING

#### 7.2.1 Expedited Reporting

Please refer to the reporting requirements in Policy 801: Reporting Research Events and Policy 802 Non-Compliance Human Subjects Research found at: https://irbo.nih.gov/confluence/pages/viewpage.action?pageId=36241835#Policies&Guidance-800Series-ComplianceandResearchEventReportingRequirements. **Note**: Only IND Safety Reports that meet the definition of an unanticipated problem will need to be reported per these policies.

#### 7.2.2 IRB Requirements for NCI PI Reporting at Continuing Review

Please refer to the reporting requirements in Policy 801: Reporting Research Events found at: https://irbo.nih.gov/confluence/pages/viewpage.action?pageId=36241835#Policies&Guidance-800Series-ComplianceandResearchEventReportingRequirements.

### 7.3 NCI CLINICAL DIRECTOR REPORTING

**NIH site only**:

Problems expeditiously reviewed by the OHSRP in the NIH eIRB system will also be reported to the NCI Clinical Director/designee; therefore, a separate submission for these reports is not necessary.

In addition to those reports, all deaths that occur within 30 days after receiving a research intervention should be reported via email to the Clinical Director unless they are due to progressive disease.

To report these deaths, please send an email describing the circumstances of the death to NCICCRQA@mail.nih.gov within one business day of learning of the death.

## 7.4 NCI Guidance For Reporting Expedited Adverse Events For Multi-Center Trials

The site PI (at the participating BTTC site) must immediately report to the BTTC Coordinating center PI any serious adverse event, whether or not considered drug related, including those listed in the protocol or investigator brochure and must include an assessment of whether there is a reasonable possibility that the drug caused the event within 24 hours of PI awareness of the event.

CCR Reportable Event Form (REF) will be used to submit Serious Adverse Events to both the CCR Office of Sponsor and Regulatory Oversight at OSROSafety@mail.nih.gov and the BTTC Coordinating Center. https://ccrod.cancer.gov/confluence/display/CCRCRO/Templates

Participating BTTC sites must also report events to the Reviewing IRB as per its policy.

For IND studies, the site PI will also directly submit reports to the CCR as IND sponsor per section **8.3**. Please also notify the coordinating center PI and study coordinator of your submission at the time you make it.

The BTTC Coordinating Center will maintain documentation of all Serious Adverse Events from each institution. The BTTC Coordinating Center will notify all investigators of any serious and unexpected adverse experiences that are possibly related to the study agent/s. The investigators are to file a copy with their protocol file and send a copy to their IRB according to their local IRB's policies and procedures.

- For the time period beginning when the consent form is signed until treatment initiation, any serious adverse event, or follow up to a serious adverse event, including death due to any cause that occurs to any subject must be reported within 24 hours to the Sponsor (NCI) if it causes the subject to be excluded from the trial, or is the result of a protocol-specified intervention, including but not limited to washout or discontinuation of usual therapy, diet, placebo treatment or a procedure.
- For the time period beginning at treatment initiation through 90 days following cessation of treatment, or 30 days following cessation of treatment if the subject initiates new anticancer therapy, whichever is earlier, any serious adverse event, or follow up to a serious adverse event, including death due to any cause, whether or not related to the study treatments, must be reported within 24 hours to the Sponsor (NCI).
- Additionally, any serious adverse event, considered by an investigator who is a qualified physician to be related to study treatment that is brought to the attention of the investigator at any time following consent through the end of the specified safety follow-up period specified in the paragraph above, or at any time outside of the time period specified in the previous paragraph also must be reported immediately to the Sponsor.

All subjects with serious adverse events must be followed up for outcome.

### 7.4.1 Reporting Deviations and Unanticipated Problems for Multi-Center Trials

Deviation or Unanticipated Problem Reports and any accompanying documentation (to include the local IRB acknowledgement of the event when applicable) are to be submitted by the participating BTTC site to the BTTC Coordinating Center using the CCR Reportable Event (REF) Form in Appendix **16.10** by email nci_bttc@mail.nih.gov or via fax at: 240-541-4432.

Deviations or Unanticipated problems must be submitted to the BTTC Coordinating Center within 5 days after becoming aware of the event. When Deviations or Unanticipated Problems are reported to BTTC, but, the local IRB does not require a report, the report that is submitted to the BTTC Coordinating Center must be accompanied by a formal memo explaining the local policy and the rationale for not reporting the event to the local IRB.

Neither the FDA nor the ICH GCP guidelines define the term "protocol deviation." The definition is often left to the Lead Institution IRB. Accordingly, since NCI, Center for Cancer Research is the Coordinating Center and the Lead PI must adhere to those policies set by the NIH Intramural IRB, the definitions for unanticipated problem and protocol deviation as described by the NIH Intramural IRB will be applied for reporting purposes for all institutions participating in this study. Definitions are listed in Section **7.1**.

Protocol Deviations or Unanticipated problems occurring at a participating institution will be submitted to that institution's own IRB in accordance with local policies and procedures. However, the participating institution must submit a report to the BTTC Coordinating Center even in instances where the local IRB does not require a report.

**NCI Center for Cancer Research BTTC Coordinating Center**: Upon receipt of the deviation/unanticipated reportable event report from the participating institution, the BTTC Coordinating Center will submit the report to the Study Chair for review. Subsequently, the participating institution's IRB deviation/unanticipated reportable event report will be submitted to the NIH Intramural IRB if it meets the criteria under Policy 801: Reporting Research Events found at:
https://irbo.nih.gov/confluence/pages/viewpage.action?pageId=36241835#Policies&Guidance-800Series-ComplianceandResearchEventReportingRequirements.

## 7.5 NIH Required Data and Safety Monitoring Plan

The clinical research team will have a teleconference every other week when patients are being actively treated on the trial to discuss each patient.

All data will be collected in a timely manner and reviewed by the Principal investigator or a lead associate investigator. Events meeting requirements for expedited reporting as described in section **7.2.1** will be submitted within the appropriate timelines.

The Principal Investigator will review adverse event and response data on each patient to ensure safety and data accuracy. The Principal Investigator will personally conduct or supervise the investigation and provide appropriate delegation of responsibilities to other members of the research staff.

### 7.5.1 Data Safety Monitoring Board (DSMB)

As this is a prospective, double-blinded, randomized controlled trial, the NCI/CCR DSMB will be used to monitor the trial. This monitoring will be done for futility, as well as for safety. Since the meetings of the NCI DSMB are annual, futility monitoring may be performed by the DSMB at a special meeting requested by the study team. These ad-hoc DSMB meetings may convene by phone in the event of rapid accrual.

The interim data analyses needed for the DSMB futility monitoring will be performed by the BTTC biostatistician.

Subjects will be randomized without stratification on a 1:1 basis, using variable block sizes. The first block will be of size n=6 in order to facilitate the evaluation of toxicity and safety in the first 3 patients on each arm.

The futility monitoring will be performed as follows: at the first DSMB meeting after which 22 patients have been enrolled into each of the experimental arms, given an experimental arm,

> *(i) if 16 or more patients survive at 8 months, additional 23 patients will be enrolled into that arm;*
>
> *(ii) if 7 or more patients fail to survive at 8 months, the experimental arm will be terminated early.*

If neither of these two conditions are satisfied, in other words, some patients are followed less than 8 months and their survival at 8 month is unknown, the posterior probability $Pr(OS8 < 0.75 \mid data)$ will be evaluated using the method of Cai, Liu and Yuan (2014) [48]. If $Pr(OS8 < 0.75 \mid data) > 0.6$, the experimental arm will be terminated early; otherwise additional 23 patients will be enrolled into that arm. Note that this posterior stopping rule is consistent with rules (i) and (ii). Interim outcome results will not be revealed to the investigators of the trial; results will be presented to the investigators prior to final accrual to the trial only if the DSMB recommends early termination of the trial. The DSMB will review the rules for early termination for toxicity and provide feedback to the BTTC Biostatistician, the Study Chair, and lead PI who will confirm the condition for early termination. Once confirmed, the Biostatistician will then communicate to CRO (Central Registration Office) to break the blind and provide the listing to the BTTC Biostatistician and Study Chair.

The evaluation for safety will be performed independently for each of the two experimental arms as follows:

> *For each of the experimental arms, stop the enrollment if [# of patients with DLT]/[# of patients evaluated] ≥ 2/3, 4/6, 5/10 or 8/20; otherwise continue the enrollment.*

In the case that one arm is discontinued due to futility or safety, enrollment would shift to the remaining arm as the primary objective of this trial is not to compare two arms.

### 7.5.2 Patient-Reported Outcomes

Received MDASI-BT forms will be checked versus the timing schedule and considered as valid if they fall within 14 days of the scheduled assessment window. Compliance rates will be calculated as the number of received valid forms over the number of expected forms. Differences between groups in compliance will be tested by use of Fisher's exact test at every time point.

We will use descriptive statistics to describe how patients rate symptom severity and interference with function at each time point. Error bar graphs for each of the symptoms will be constructed at each time point. The proportion of patients rating their symptoms to be 5 or greater (on a 0-10 scale) will also be reported. We will construct individual patient profiles for each of the selected symptoms to describe the individual patients' patterns of change over time. We will calculate the mean symptom severity and mean symptom interference at the time of clinical evaluation. Estimates of differences in the mean symptom severity and mean symptom interference between responders and non-responders will be estimated in the intent to treat population. All patients with at least one valid questionnaire will be included in the analyses. Questionnaires completed at study registration will be considered baseline. All questionnaire data received after randomization will be used in the primary analyses.

Differences of at least 2 points will be classified as the minimum clinically meaningful change in the symptom severity and symptom interference measures. For example, an increase of 2 points or more would mean a moderate improvement, whereas a decrease of 2 points or more would be interpreted as moderate worsening. For individual symptoms, a rise in a symptom score means deterioration, whereas a reduced score means improvement of the specific symptom.

### 7.6 EXPLORATORY OBJECTIVES & ENDPOINTS

PDL1 expression and T-Cell infiltrate in tumor will be correlated using Spearman correlation coefficients. Peripheral T-Cell activation and tryptophan metabolites will be summarized by reporting descriptive statistics such as median and inter-quartile range for all time points.

## 8 SPONSOR PROTOCOL/SAFETY REPORTING

### 8.1 DEFINITIONS

#### 8.1.1 Adverse Event

Any untoward medical occurrence in a patient or clinical investigation subject administered a pharmaceutical product and which does not necessarily have a causal relationship with this treatment. An adverse event (AE) can therefore be unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease, temporally associated with the use of a medicinal (investigational) product, whether or not related to the medicinal (investigational) product (ICH E6 (R2)).

#### 8.1.2 Serious Adverse Event (SAE)

An adverse event or suspected adverse reaction is considered serious if in the view of the investigator or the sponsor, it results in any of the following:

- Death,
- A life-threatening adverse event (see section **8.1.3**)
- Inpatient hospitalization or prolongation of existing hospitalization
    - A hospitalization/admission that is pre-planned (i.e., elective or scheduled surgery arranged prior to the start of the study), a planned hospitalization for pre-existing condition, or a procedure required by the protocol, without a serious deterioration in health, is not considered a serious adverse event.

- A hospitalization/admission that is solely driven by non-medical reasons (e.g., hospitalization for patient or subject convenience) is not considered a serious adverse event.
- Emergency room visits or stays in observation units that do not result in admission to the hospital would not be considered a serious adverse event. The reason for seeking medical care should be evaluated for meeting one of the other serious criteria.

- Persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions
- A congenital anomaly/birth defect.
- Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered a serious adverse drug experience when, based upon appropriate medical judgment, they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition.

**Note**: In addition to the above criteria, adverse events meeting either of the below criteria, although not serious per ICH definition, are reportable to the Merck in the same timeframe as SAEs to meet certain local requirements. Therefore, these events are considered serious by Merck for collection purposes.

- Is a new cancer (that is not a condition of the study);
- Is associated with an overdose.

Refer to Section **8.1.1** "Adverse Events" for additional details regarding each of the above criteria.

8.1.2.1 Protocol-Specific Exceptions to Serious Adverse Event Reporting

Efficacy endpoints as outlined in this section will not be reported to Merck as described in Immediate Reporting of Adverse Events to the Sponsor (NCI), unless there is evidence suggesting a causal relationship between the drug and the event. Any such event will be submitted to the Merck within 24 hours.

Specifically, the suspected/actual events covered in this exception include any event that is disease progression of the cancer under study.

Hospitalization related to convenience (e.g. transportation issues, etc.) will not be considered a SAE.

### 8.1.3 Life-threatening

An adverse event or suspected adverse reaction is considered "life-threatening" if, in the view of either the investigator or sponsor, its occurrence places the patient or subject at immediate risk of death. It does not include an adverse event or suspected adverse reaction that, had it occurred in a more severe form, might have caused death. (21CFR312.32)

### 8.1.4 Severity

The severity of each Adverse Event will be assessed utilizing the CTCAE version 5.

### 8.1.5 Relationship to Study Product

All AEs will have their relationship to study product assessed using the terms: related or not related.

- Related – There is a reasonable possibility that the study product caused the adverse event. Reasonable possibility means that there is evidence to suggest a causal relationship between the study product and the adverse event.
- Not Related – There is not a reasonable possibility that the administration of the study product caused the event.

### 8.1.6 Events Of Clinical Interest (ECI) For This Trial Include:

- an overdose of Merck product, as defined in Section **8.1.7** Definition of an Overdose for This Protocol and Reporting of Overdose to the Sponsor, that is not associated with clinical symptoms or abnormal laboratory results.
- an elevated AST or ALT lab value that is greater than or equal to 3X the upper limit of normal and an elevated total bilirubin lab value that is greater than or equal to 2X the upper limit of normal and, at the same time, an alkaline phosphatase lab value that is less than 2X the upper limit of normal, as determined by way of protocol-specified laboratory testing or unscheduled laboratory testing.

*Note: These criteria are based upon available regulatory guidance documents. The purpose of the criteria is to specify a threshold of abnormal hepatic tests that may require an additional evaluation for an underlying etiology.

### 8.1.7 Definition of an Overdose for This Protocol and Reporting of Overdose to the Sponsor

For purposes of this trial, an overdose of pembrolizumab will be defined as any dose of 1,000 mg or greater (≥5 times the indicated dose). No specific information is available on the treatment of overdose of pembrolizumab. Appropriate supportive treatment should be provided if clinically indicated. In the event of overdose, the subject should be observed closely for signs of toxicity. Appropriate supportive treatment should be provided if clinically indicated.

If an adverse event(s) is associated with ("results from") the overdose of a Merck product, the adverse event(s) is reported as a serious adverse event, even if no other seriousness criteria are met.

If a dose of Merck's product meeting the protocol definition of overdose is taken without any associated clinical symptoms or abnormal laboratory results, the overdose is reported as a non-serious Event of Clinical Interest (ECI), using the terminology "accidental or intentional overdose without adverse effect."

All reports of overdose with and without an adverse event must be reported within 24 hours to the Sponsor.

## 8.2 ASSESSMENT OF SAFETY EVENTS

AE information collected will include event description, date of onset, assessment of severity and relationship to study product and alternate etiology (if not related to study product), date of resolution of the event, seriousness and outcome. The assessment of severity and relationship to the study product will be done only by those with the training and authority to make a diagnosis and listed on the Form FDA 1572 as the site principal investigator or sub-investigator. AEs occurring during the collection and reporting period will be documented appropriately regardless of relationship. AEs will be followed through resolution.

SAEs will be:

- Assessed for severity and relationship to study product and alternate etiology (if not related to study product) by a licensed study physician listed on the Form FDA 1572 as the site principal investigator or sub-investigator.
- Recorded on the appropriate SAE report form, the medical record and captured in the clinical database.
- Followed through resolution by a licensed study physician listed on the Form FDA 1572 as the site principal investigator or sub-investigator.

For timeframe of recording adverse events, please refer to section **6.1**. All serious adverse events recorded from the time of first investigational product administration must be reported to the sponsor with the exception of any listed in section **8.4**.

## 8.3 REPORTING OF SERIOUS ADVERSE EVENTS

Any AE that meets protocol-defined serious criteria or meets the definition of Adverse Event of Special Interest that require expedited reporting must be submitted immediately (within 24 hours of awareness) to OSRO Safety using the CCR SAE report form. Any exceptions to the expedited reporting requirements are found in section **8.4**.

All SAE reporting must include the elements described in section **8.2**.

SAE reports will be submitted to the NCI Center for Cancer Research (CCR) at: OSROSafety@mail.nih.gov and to the BTTC PI and study coordinator. CCR SAE report form and instructions can be found at: https://ccrod.cancer.gov/confluence/display/CCRCRO/Forms+and+Instructions

Following the assessment of the SAE by OSRO, other supporting documentation of the event may be requested by the OSRO Safety and should be provided as soon as possible.

## 8.4 SAFETY REPORTING CRITERIA TO THE PHARMACEUTICAL COLLABORATORS

### 8.4.1 To be Sent by OSRO Safety

OSRO Safety will only provide the following: SAEs, SUSARs, overdoses and pregnancies. OSRO Safety will not send routine AEs or periodic reports.

### 8.4.2 To be Sent by BTTC study team

#### 8.4.2.1 Adverse Events

An investigator who is a qualified physician, will evaluate all adverse events.

#### 8.4.2.2 Duration

Record the start and stop dates of the adverse event. If less than 1 day, indicate the appropriate length of time and units.

#### 8.4.2.3 Action taken

Did the adverse event cause Merck product to be discontinued?

#### 8.4.2.4 Relationship to Merck Product

**Did Merck product cause the adverse event?** The determination of the likelihood that Merck product caused the adverse event will be provided by an investigator who is a qualified physician. The investigator's signed/dated initials on the source document or worksheet that supports the causality noted on the AE form, ensures that a medically qualified assessment of causality was done. This initialed document must be retained for the required regulatory time frame. The criteria below are intended as reference guidelines to assist the investigator in assessing the likelihood of a relationship between the test drug and the adverse event based upon the available information.

The following components are to be used to assess the relationship between Merck product and the AE; the greater the correlation with the components and their respective elements (in number and/or intensity), the more likely Merck product caused the adverse event (AE):

#### 8.4.2.5 Exposure

Is there evidence that the subject was actually exposed to Merck product such as: reliable history, acceptable compliance assessment (pill count, diary, etc.), expected pharmacologic effect, or measurement of drug/metabolite in bodily specimen?

#### 8.4.2.6 Time Course

Did the AE follow in a reasonable temporal sequence from administration of Merck product?

Is the time of onset of the AE compatible with a drug-induced effect (applies to trials with investigational medicinal product)?

#### 8.4.2.7 Likely Cause

Is the AE not reasonably explained by another etiology such as underlying disease, other drug(s)/vaccine(s), or other host or environmental factors

#### 8.4.2.8 Dechallenge

Was Merck product discontinued or dose/exposure/frequency reduced?

If yes, did the AE resolve or improve?

If yes, this is a positive dechallenge. If no, this is a negative dechallenge.

(Note: This criterion is not applicable if: (1) the AE resulted in death or permanent disability; (2) the AE resolved/improved despite continuation of the Sponsor's product; or (3) the trial is a single-dose drug trial); or (4) Sponsor's product(s) is/are only used one time.)

#### 8.4.2.9 Rechallenge

Was the subject re-exposed to Merck product in this study?

If yes, did the AE recur or worsen?

If yes, this is a positive rechallenge. If no, this is a negative rechallenge.

(**Note**: This criterion is not applicable if: (1) the initial AE resulted in death or permanent disability, or (2) the trial is a single-dose drug trial); or (3) Sponsor's product(s) is/are used only one time).

**NOTE**: IF A RECHALLENGE IS PLANNED FOR AN ADVERSE EVENT WHICH WAS SERIOUS AND WHICH MAY HAVE BEEN CAUSED BY MERCK PRODUCT, OR IF REEXPOSURE TO MERCK PRODUCT POSES ADDITIONAL POTENTIAL SIGNIFICANT RISK TO THE SUBJECT, THEN THE RECHALLENGE MUST BE APPROVED IN ADVANCE BY THE SPONSOR AS PER DOSE MODIFICATION GUIDELINES IN THE PROTOCOL.

#### 8.4.2.10 Consistency with Trial Treatment Profile

Is the clinical/pathological presentation of the AE consistent with previous knowledge regarding Merck product or drug class pharmacology or toxicology?

The assessment of relationship will be reported on the case report forms /worksheets by an investigator who is a qualified physician according to his/her best clinical judgment, including consideration of the above elements.

Record one of the following: Use the following scale of criteria as guidance (not all criteria must be present to be indicative of Merck product relationship).

1. **Yes, there is a reasonable possibility of Merck product relationship**. There is evidence of exposure to Merck product. The temporal sequence of the AE onset relative to the administration of Merck product is reasonable. The AE is more likely explained by Merck product than by another cause.
2. **No, there is not a reasonable possibility of Merck product relationship**. Subject did not receive the Merck product OR temporal sequence of the AE onset relative to administration of Merck product is not reasonable OR the AE is more likely explained by another cause than the Merck product. (Also entered for a subject with overdose without an associated AE.)

### 8.4.3 Events of Clinical Interest

These selected non-serious and serious adverse events are also known as Events of Clinical Interest (ECI) and must be reported within 24 hours to Merck. For reporting of ECI refer to section **8.3**.

For the time period beginning when the consent form is signed until treatment initiation, any ECI, or follow up to an ECI, that occurs to any subject must be reported within 24 hours to the

Sponsor if it causes the subject to be excluded from the trial, or is the result of a protocol-specified intervention, including but not limited to washout or discontinuation of usual therapy, diet, placebo treatment or a procedure.

For the time period beginning at treatment initiation through 90 days following cessation of treatment, or 30 days following cessation of treatment if the subject initiates new anticancer therapy, whichever is earlier, any ECI, or follow up to an ECI, whether or not related to study treatment, must be reported within 24 hours to the Sponsor (NCI).

Merck Global Safety facsimile number: +1-215-993-1220 (Sponsor [NCI] will contact Merck)

### 8.4.4 Reporting Serious Adverse Events to Merck and Agenus

Upon receipt, Sponsor (NCI) will forward SAE reports and any other relevant safety information to the Merck Global Safety facsimile number: +1-215-993-1220 and to Agenus Pharmacovigilance via email: adverse.events@agenusbio.com or fax: 1-781-674-4261.

A copy of all 15 Day Reports and Annual Progress Reports will be submitted to Merck & Co., Inc. (Attn: Worldwide Product Safety; FAX 215 993-1220) and to Agenus Pharmacovigilance (email: adverse.events@agenusbio.com or fax: 1-781-674-4261) at the time of submission to FDA.

For the time period beginning at treatment initiation through 90 days following cessation of treatment, or 30 days following cessation of treatment if the subject initiates new anticancer therapy, whichever is earlier, any ECI, or follow up to an ECI, whether or not related to study treatment, must be reported within 24 hours to the Sponsor (NCI).

Sites will submit all reportable events to the Sponsor (NCI), and NCI will submit to Agenus and Merck within 2 working days.

The DSMB Sponsor will monitor unblinded aggregated efficacy endpoint events and safety data to ensure the safety of the subjects in the trial. Any suspected endpoint which upon review is not progression of the cancer under study will be forwarded to Merck Global Safety and Agenus Pharmacovigilance as a SAE within 2 working days of determination that the event is not progression of the cancer under study.

For reporting pregnancy: The designated Sponsor representative will work with the investigator to ensure that all relevant information is provided to Merck and Agenus within 1 to 5 calendar days for SAEs and within 30 days for all other pregnancies.

The same timelines apply when outcome information is available.

## 8.5 Reporting Pregnancy

All required pregnancy reports/follow-up to OSRO will be submitted to: OSROSafety@mail.nih.gov and to the CCR PI and study coordinator. Forms and instructions can be found here: https://ccrod.cancer.gov/confluence/display/CCRCRO/Forms+and+Instructions.

### 8.5.1 Maternal exposure

If a participant becomes pregnant during the course of the study, the study treatment should be discontinued immediately, and the pregnancy reported to the Sponsor no later than 24 hours of

when the Investigator becomes aware of it. The Investigator should notify the Sponsor no later than 24 hours of when the outcome of the Pregnancy becomes known,

Pregnancy itself is not regarded as an SAE. However, congenital abnormalities or birth defects and spontaneous miscarriages that meet serious criteria (section **8.3**) should be reported as SAEs.

The outcome of all pregnancies should be followed up and documented.

### 8.5.2 Paternal exposure

Male participants should refrain from fathering a child or donating sperm during the study and for 120 days after the last dose of pembrolizumab.

Pregnancy of the participant's partner is not considered to be an AE. The outcome of all pregnancies occurring from the date of the first dose until 120 days after the last dose should, if possible, be followed up and documented. Pregnant partners may be offered the opportunity to participate in an institutional pregnancy registry protocol (e.g., the NIH IRP pregnancy registry study) to provide data about the outcome of the pregnancy for safety reporting purposes.

## 8.6 REGULATORY REPORTING FOR STUDIES CONDUCTED UNDER CCR SPONSORED IND

Following notification from the investigator, CCR, the IND sponsor, will report any suspected adverse reaction that is both serious and unexpected in expediated manner to the FDA in accordance to 21 CFR 312. 32. CCR will report an AE as a suspected adverse reaction only if there is evidence to suggest a causal relationship between the study product and the adverse event. CCR will notify FDA and all participating investigators (i.e., all investigators to whom the sponsor is providing drug under its INDs or under any investigator's IND) in an IND safety report of potential serious risks from clinical trials or any other source, as soon as possible, in accordance to 21 CFR Part 312.32.

All serious events will be reported to the FDA at least annually in a summary format.

# 9 CLINICAL MONITORING

Clinical site monitoring is conducted to ensure:

- that the rights of the participants are protected;
- that the study is implemented per the approved protocol, Good Clinical Practice and standard operating procedures; and,
- the quality and integrity of study data and data collection methods are maintained.

Monitoring for this study will be performed by NCI CCR Office of Sponsor and Regulatory Oversight (OSRO) Sponsor and Regulatory Oversight Support (SROS) Services contractor. Clinical site monitoring activities will be based on OSRO standards, FDA Guidance E6(R2) Good Clinical Practice: Integrated Addendum to ICH E6(R1) March 2018, and applicable regulatory requirements.

Details of clinical site monitoring will be documented in a Clinical Monitoring Plan (CMP) developed by OSRO. CMPs will be protocol-specific, risk-based and tailored to address human subject protections and integrity of the study data. OSRO will determine the intensity and frequency of monitoring based on several factors, including study type, phase, risk, complexity,

expected enrollment rate, and any unique attributes of the study and the site. The Sponsor will conduct a periodic review of the CMP to confirm the plan's continued appropriateness. A change to the protocol, significant or pervasive non-compliance with GCP, or the protocol may trigger CMP updates.

OSRO SROS Monitoring visits and related activities will be conducted throughout the life cycle of each protocol. The first activity is before the study starts to conduct a Site Assessment Visit (SAV) (as warranted), followed by a Site Initiation Visit (SIV), Interim Monitoring Visit(s) (IMVs), and a study Close-Out Visit (COV).

Some monitoring activities may be performed remotely, while others will occur at the study site(s). Monitoring visit reports will describe visit activities, observations, and associated action items or follow-up required for resolution of any issues, discrepancies, or deviations. Monitoring reports will be distributed to the study PI, NCI CCR QA, CCR Protocol Support Office, coordinating center (if applicable), and the Sponsor regulatory file.

The site Monitor will inform the study team of any deviations observed during monitoring visits. If unresolved, the Monitor will request that the site Staff enter the deviations in the CCR Protocol Deviation Tracking System (PDTS) for deviation reporting to the Sponsor and as applicable per institutional and IRB guidance.

## 10 STATISTICAL CONSIDERATIONS

This is a phase II blinded, randomized trial in newly diagnosed GBM. Evaluable patients must complete 1 cycle of treatment post RT. A Bayesian two-stage design will be used to monitor the efficacy of two experimental arms independently based on the OS at 8 months (OS8) using the following Bayesian rule (Thall and Simon, 1994) [46]:

*Stop the experimental arm for futility if Pr (OS8 < 0.75 | data ) > 0.6.*

That is, if the observed data indicate that there is greater than 60% chance that the OS8 is smaller than 0.75, we stop the experimental arm. We choose 0.75 as the threshold because that is the estimate of OS8 for the standard of care (RT+TMZ). In order to speed up the trial, we use the OS8, rather than the OS at 12 months (i.e., the primary endpoint), for the purpose of interim monitoring. Assuming vague Beta prior Beta (0.25, 0.25) for the OS8, the two-stage Bayesian design can be described as follows:

*Enroll 22 patients for each of the experimental arms, if 16 or more patients survive at 8 months, we continue to enroll the remaining 23 patients; otherwise stop that experimental arm.*

At the interim analysis, if some patients are followed less than 8 months, we will calculate Pr (OS8 < 0.75 | data) based on the methodology of Cai, Liu and Yuan (2014) by imputing the OS8 for these patients. The following table shows the operating characteristics of the Bayesian phase II design:

| | True OS8 | | | |
|---|---|---|---|---|
| | 0.6 | 0.7 | 0.8 | 0.9 |

| | | | | |
|---|---|---|---|---|
| Stopping probability | 0.845 | 0.507 | 0.135 | 0.004 |
| Average sample size | 25.6 | 33.3 | 41.9 | 44.9 |

Similar Bayesian method will also be used to monitor the rate of DLT. We monitor two experimental arms independently-patients that are randomized. Specifically, for each of the arms, we will stop enrolling patients and inspect the safety data if the Pr(DLT > 25% | data) > 0.9. That is, we will stop enrolling patients if the data indicate that there is more than 90% chance that the true DLT rate is higher than 25%. This decision rule gives the following stopping rule, assuming a Beta(0.1, 0.4) prior distribution for DLT rate,

*Stop enrolling pts if [# of pts with DLT] / [# of pts evaluated] ≥ 2/3, 4/6, 5/10 or 8/20.*

Below are the operating characteristics of the toxicity monitoring rule.

| | True toxicity rate | | | | |
|---|---|---|---|---|---|
| | 0.1 | 0.2 | 0.3 | 0.4 | 0.5 |
| Early stop probability | 0.028 | 0.136 | 0.372 | 0.692 | 0.906 |
| Average sample size | 27.3 | 25.0 | 20.6 | 14.9 | 9.8 |

In the case that one arm is discontinued due to futility or safety, enrollment would shift to the remaining arm as the primary objective of this trial is not to compare two arms.

### 10.1 Sample Size and Accrual

Based on previous experience of newly diagnosed (ND) unmethylated GBM patients treated with Pembrolizumab, the estimated one-year OS for the RT+TMZ+Pembrolizumab arm is 70%, and the estimated one-year OS for the RT+TMZ+Pembrolizumab +HSPPC-96 arm is 90%. Given one-sided significance level of 0.10, the sample size of 45 patients per arm provides 80% power to detect the difference between two experimental arms.

Treatment accrual for the randomization arms will be 90 patients (45 patients per arm). Considering vaccine production failures for patients when HSPPC-96 is not produced, accrual in the ancillary cohort is up to 18 patients. Therefore, the randomized treatment accrual ceiling will be 108 patients. Assuming a screen failure rate of 65%, the total accrual ceiling will be 310 patients.

Approximately 8 potentially eligible patients are seen per month, and it is anticipated that at least 1-2 per month will be accrued per site. NOTE: DSMB recommended in May 2021 to terminate study early due to slow accrual. Study is closed to enrolment. Patients will be unblinded but will be kept on treatment until completion of TMZ, Pembro, and/or HSPPC-96 and followed for overall survival. Placebo will be discontinued.

### 10.2 DATA ANALYSES PLANS

Progression-free and overall survival will be determined using a Kaplan-Meier survival curve. Median PFS and OS will be determined from this curve. A one-sided lower 90% confidence limit for median PFS and OS will be determined by the method of Brookmeyer and Crowley [47]. The Chi-square test will be used to compare the one-year OS between two experimental arms. In addition, the weighted log rank test will also be used to compare the OS and PFS between two experimental arms.

### 10.3 SECONDARY OBJECTIVES & ENDPOINTS

- PFS-6 (percentage of subjects alive and progression-free at 6 months post-registration): determined from the value of the Kaplan-Meier curve at 6 months.
- Response rate: determined as described by RANO **Table 2**. Rates and 95% confidence intervals will be determined using exact binomial probability distributions.
- Overall survival: Kaplan-Meier curves will be used to analyze overall survival.
- OS-6, 12 and 24 (overall survival at 6, 12, and 24 months post-registration, respectively): determined from the level of the Kaplan-Meier curves at these time points.
- Safety of Pembrolizumab in GBM: Adverse events will be described descriptively by giving frequencies of each event by type, severity, frequency, timing, and attribution.
- To evaluate the occurrence of symptoms and correlate to disease progression and tolerance to treatment using the MD Anderson Symptom Inventory-Brain Tumor Module (MDASI-BT).

## 11 COLLABORATIVE AGREEMENTS

### 11.1 AGREEMENT TYPE

There is a BTTC consortia agreement in place between all of the participating institutions listed in the NIH IRB system. In addition, there are CRADA agreements in place with Merck (03128), and Agenus (03127) who are supporting this study.

Transfers that are associated with correlative studies conducted under an approved protocol: Investigators in the NIH intramural program may participate in multi-site clinical trials (either as a site or as the coordinating center) under which human materials are transferred from the intramural program to another site for correlative studies that are part of the approved protocol. In such a situation, the protocol clearly documents the tests conducted under the correlative studies, and each institution participating in the clinical study is bound by the terms of their Protocol and their obligations under the statutes and regulations. In addition, intramural protocols are cleared by the IC Clinical Director. In such a situation, use of an HM-MTA is not necessary for these transfers.

### 11.2 MULTI-INSTITUTIONAL GUIDELINES

#### 11.2.1 Participating Institution Site Initiation

Before an institution may begin participating in a BTTC protocol, they must complete the following steps:

- Submit all required regulatory documents to the BTTC Coordinating Center
- Participate in a site initiation visit, webcast, or conference call
- Receive training regarding study specific CRF's and/or databases

After these requirements have been fulfilled, the participating institution will receive by fax, e-mail, or hard copy, a Site Activation Memo. Once the Site Activation Memo has been received, the participating institution may begin to register patients to the protocol.

### 11.2.2 IRB Approvals

The lead PI will provide the NIH Intramural IRB with a copy of the participating institution's initial approval and approved yearly continuing review. Registration will be halted at any participating institution in which a current continuing approval is not on file at the NIH Intramural IRB.

## 12 HUMAN SUBJECTS PROTECTIONS

### 12.1 RATIONALE FOR SUBJECT SELECTION

This study was designed to include women and minorities, but was not designed to measure differences of intervention effects. Males and females will be recruited with no preference to gender. No exclusion to this study will be based on race. Minorities will actively be recruited to participate. High grade gliomas occur in patients of all races and although there is a slight predominance in men, this is a disease that is also common in women. The molecular targets of pembrolizumab and HSPPC-96 within the tumor are not known to be different among patients based on gender or race; hence this study will be open to all adults.

### 12.2 PARTICIPATION OF CHILDREN

Individuals under the age of 18 will not be eligible to participate in this study because they are unlikely to have glioblastoma, and because of unknown toxicities of the study agents in the pediatric patient. Furthermore, the targets of pembrolizumab and HSPPC-96 are not as prevalent in pediatric malignant gliomas and therefore, the efficacy of this regimen will be initially determined in the adult population before consideration of its use in pediatrics.

### 12.3 PARTICIPATION OF SUBJECTS UNABLE TO GIVE CONSENT (NCI)

Adults unable to give consent are excluded from enrolling in the protocol. However, re-consent may be necessary and there is a possibility, though unlikely, that subjects could become decisionally impaired. In circumstances where the subject becomes decisionally impaired due to disease progression, the subject will be taken off-treatment per protocol, but may continue in long term follow-up. In long term follow-up, the risks associated with study related interventions are no more than minimal.

However, because in other circumstances, there is a prospect of direct benefit from research participation, all subjects ≥ age 18 will be offered the opportunity to fill in their wishes for research and care, and assign a substitute decision maker on the "NIH Advance Directive for Health Care and Medical Research Participation" form so that another person can make decisions about their medical care in the event that they become incapacitated or cognitively

impaired during the course of the study. Note: The PI or AI will contact the NIH Ability to Consent Assessment Team (ACAT) for evaluation to assess ongoing capacity of the subjects and to identify an LAR, as needed. Please see section **12.6.1** for consent procedure.

## 12.4 PARTICIPATION OF ADULTS UNABLE TO GIVE CONSENT (BTTC)

Each institution will follow their own procedure for re-consenting subjects unable to re-consent and will notify the Coordinating Center at time of continuing review of these occurrences.

## 12.5 EVALUATION OF BENEFITS AND RISKS/DISCOMFORTS

### 12.5.1 Risks

#### 12.5.1.1 Study Agents

The primary risk to patients participating in this research study is from the toxicity of pembrolizumab or HSPPC-96 or all drugs as described in section **14** and the consent document. Pembrolizumab and HSPPC-96 are investigational agents in the treatment of gliomas. The protocol provides for detailed and careful monitoring of all patients to assess for toxicity. Toxicity data from the current dose level will be collected and reviewed to ensure that there were no severe toxicities that would preclude further patient enrollment. Patients will be treated with therapeutic intent and response to the therapy will be closely monitored.

#### 12.5.1.2 Imaging risks

The MRIs to be done in this study will involve contrast. The risks associated with MRIs and contrast are discussed in the consent form.

Although MR imaging with gadolinium has been used as part of the standard care for patients with high grade gliomas, long term toxicities of gadolinium as a consequence of gadolinium accumulation in the body has been reported. All investigators will ascertain the MR imaging history of all prospective study participants to determine the extent of prior exposure to gadolinium contrast. Radiology will be consulted if there is a history of repeated gadolinium exposure.

#### 12.5.1.3 Blood Collection

The risk of blood draws in this study is that it may cause pain, bleeding, and/or bruising. The risks associated with blood collection are discussed in the consent form. Participants may faint and/or develop an infection with redness and irritation of the vein at the site where blood is drawn. Frequent blood collection may cause anemia (low red blood cell count), which may create a need for blood transfusions.

#### 12.5.1.4 Questionnaires

The risk of questionnaires in this study is that they may contain questions sensitive in nature. Participants may refuse to answer any question that makes them feel uncomfortable. The primary use of the information collected is to assess the severity of symptoms related to study treatment. Submission of this information is voluntary.

#### 12.5.1.5 Non-Physical Risks of Genetic Research

##### 12.5.1.5.1 Risk Of Receiving Unwanted Information

Anxiety and stress may arise as a result of the anticipation that unwanted information regarding disease related DNA sequencing or disease tendencies, or misattributed paternity. Patients will be clearly informed that the data related to DNA sequencing and genetic analysis is coded, investigational and will not be shared with patients, family members or health care providers.

#### 12.5.1.5.2 Risk Related To Possibility That Information May Be Released

This includes the risk that data related to genotype, DNA sequencing or risk for disease tendency or trait can be released to members of the public, insurers, employers, or law enforcement agencies. Although there are no plans to release results to the patients, family members or health care providers, this risk will be included in the informed consent document.

### 12.5.2 Benefits

The potential benefit to a patient on this study is a reduction in the bulk of their tumor and improvement in cancer lesions, which may or may not have favorable impact on symptoms and/or survival.

## 12.6 CONSENT PROCESS AND DOCUMENTATION

The informed consent document will be provided as a physical or electronic document to the participant or consent designee as applicable, for review prior to consenting. A designated study investigator will carefully explain the procedures and tests involved in this study, and the associated risks, discomforts, and benefits. In order to minimize potential coercion, as much time as is needed to review the document will be given, including an opportunity to discuss it with friends, family members and/or other advisors, and to ask questions of any designated study investigator. A signed informed consent document will be obtained prior to entry onto the study.

The initial consent process as well as re-consent, when required, may take place in person or remotely (e.g., via telephone or other site approved remote platforms used in compliance with policy, including HRPP Policy 303) per discretion of the designated study investigator and with the agreement of the participant/consent designee(s). Whether in person or remote, the privacy of the subject will be maintained. Consenting investigators (and participant/consent designee, when in person) will be located in a private area (e.g., clinic consult room). When consent is conducted remotely, the participant/consent designee will be informed of the private nature of the discussion and will be encouraged to relocate to a more private setting if needed.

**Note**: When required, witness signature will be obtained similarly as described for the investigator and participant as described below.

Consent will be documented with required signatures on the physical document (which includes the printout of an electronic document sent to participant. Signatures on electronic documents are described below. Note: FDA only regulates electronic signatures (i.e., an electronic timestamp is generated at the time of signature) in FDA regulated research.

Manual (non-electronic) signature on electronic document:

When a manual signature on an electronic document is used for the documentation of consent at the NIH Clinical Center, this study will use the Adobe platform (which is not 21 CFR Part 11 compliant) to obtain the required signatures.

During the consent process, participants and investigators will view individual copies of the approved consent document on screens at their respective locations.

Both the investigator and the subject will sign the document using a finger, stylus or mouse.

Electronic signature on electronic document:

When permitted by the NIH Clinical Center, an electronic signature may be obtained using the iMedConsent platform to obtain the required signatures.

During the consent process, participants and investigators will view individual copies of the approved consent document on screens at their respective locations.

The identity of the participant will be determined by a prompt which will require the provision of information from a form of government-issued identification prior to obtaining the signature. If participant does not have such identification available, security questions will be used to confirm identity.

Both the investigator and the subject will sign the document electronically per system prompts.

**For BTTC Participating multicenter sites**:

Each institution will follow their own local procedure for electronic signature requirements at participating site.

### 12.6.1 Consent Process for Adults Who Lack Capacity to Consent to Research Participation

For NIH participants addressed in section **12.3**, an LAR will be identified consistent with Policy 403 and informed consent obtained from the LAR, as described in Section **12.6**.

### 12.6.2 Request for Waiver of Consent for Screening Activities

Prior to the subject signing the consent for this study pre-screening activities listed in section **2.2.1** may be performed.

We request a waiver of consent for these activities as they involve only minimal risk to the subjects. A waiver will not adversely affect the rights and welfare of the subjects given that the activities are only intended to determine suitability for screening for participation in research protocols. These activities could not practicably be carried out without the wavier as central recruiting services, utilized in the NIH Clinical Center, perform pre-screening activities for multiple studies and obtaining consent for each one is beyond their resources. The subjects will be provided with additional pertinent information after participation as they will be informed whether or not they are eligible to sign a consent for additional screening.

### 12.6.3 Reconsent via phone at BTTC Sites (outside of NCI)

Each institution will follow their own procedure for reconsenting subjects.

# 13 REGULATORY AND OPERATIONAL CONSIDERATIONS

## 13.1 STUDY DISCONTINUATION AND CLOSURE

This study may be temporarily suspended or prematurely terminated if there is sufficient reasonable cause. Written notification, documenting the reason for study suspension or termination, will be provided by the suspending or terminating party to study participants, site investigators, funding agency, the Investigational New Drug (IND) or Investigational Device Exemption (IDE) sponsor and regulatory authorities. If the study is prematurely terminated or suspended, the Principal Investigator (PI) will promptly inform study participants, the Institutional Review Board (IRB), and sponsor and will provide the reason(s) for the termination or suspension. Study participants will be contacted, as applicable, and be informed of changes to study visit schedule.

Circumstances that may warrant termination or suspension include, but are not limited to:

- Determination of unexpected, significant, or unacceptable risk to participants
- Demonstration of efficacy that would warrant stopping
- Insufficient compliance to protocol requirements
- Data that are not sufficiently complete and/or evaluable
- Determination that the primary endpoint has been met
- Determination of futility

Study may resume once concerns about safety, protocol compliance, and data quality are addressed, and satisfy the sponsor, IRB and as applicable, Food and Drug Administration (FDA).

## 13.2 QUALITY ASSURANCE AND QUALITY CONTROL

Each clinical site will perform internal quality management of study conduct, data and biological specimen collection, documentation and completion. An individualized quality management plan will be developed to describe a site's quality management.

Quality control (QC) procedures will be implemented beginning with the data entry system and data QC checks that will be run on the database will be generated. Any missing data or data anomalies will be communicated to the site(s) for clarification/resolution.

Following written Standard Operating Procedures (SOPs), the monitors will verify that the clinical trial is conducted and data are generated and biological specimens are collected, documented (recorded), and reported in compliance with the protocol, International Council for Harmonisation Good Clinical Practice (ICH GCP), and applicable regulatory requirements (e.g., Good Laboratory Practices (GLP), Good Manufacturing Practices (GMP)).

The investigational site will provide direct access to all trial related sites, source data/documents, and reports for the purpose of monitoring and auditing by the sponsor, and inspection by local and regulatory authorities.

## 13.3 CONFLICT OF INTEREST POLICY

The independence of this study from any actual or perceived influence, such as by the pharmaceutical industry, is critical. Therefore, any actual conflict of interest of persons who have a role in the design, conduct, analysis, publication, or any aspect of this trial will be disclosed and managed. Furthermore, persons who have a perceived conflict of interest will be

required to have such conflicts managed in a way that is appropriate to their participation in the design and conduct of this trial. The study leadership in conjunction with the National Cancer Institute has established policies and procedures for all study group members to disclose all conflicts of interest and will establish a mechanism for the management of all reported dualities of interest.

Participating sites will adhere to local conflict of interest policy guidelines.

### 13.4 CONFIDENTIALITY AND PRIVACY

Participant confidentiality and privacy is strictly held in trust by the participating investigators, their staff, and the sponsor(s). This confidentiality is extended to cover testing of biological samples and genetic tests in addition to the clinical information relating to participants. Therefore, the study protocol, documentation, data, and all other information generated will be held in strict confidence. No information concerning the study or the data will be released to any unauthorized third party without prior written approval of the sponsor.

All research activities will be conducted in as private a setting as possible.

The study monitor, other authorized representatives of the sponsor, representatives of the Institutional Review Board (IRB), and/or regulatory agencies may inspect all documents and records required to be maintained by the investigator, including but not limited to, medical records (office, clinic, or hospital) and pharmacy records for the participants in this study. The clinical study site will permit access to such records.

The study participant's contact information will be securely stored at each clinical site for internal use during the study. At the end of the study, all records will continue to be kept in a secure location for as long a period as dictated by the reviewing IRB, Institutional policies, or sponsor requirements.

Study participant research data, which is for purposes of statistical analysis and scientific reporting, will be transmitted to and stored at the BTTC Coordinating Center. This will not include the participant's contact or identifying information. Rather, individual participants and their research data will be identified by a unique study identification number. The study data entry and study management systems used by clinical sites and by BTTC Coordinating Center research staff will be secured and password protected. At the end of the study, all study databases will be archived at the BTTC Coordinating Center.

To further protect the privacy of study participants, a Certificate of Confidentiality has been issued by the National Institutes of Health (NIH). This certificate protects identifiable research information from forced disclosure. It allows the investigator and others who have access to research records to refuse to disclose identifying information on research participation in any civil, criminal, administrative, legislative, or other proceeding, whether at the federal, state, or local level. By protecting researchers and institutions from being compelled to disclose information that would identify research participants, Certificates of Confidentiality help achieve the research objectives and promote participation in studies by helping assure confidentiality and privacy to participants.

# 14 PHARMACEUTICAL INFORMATION

## 14.1 PEMBROLIZUMAB/KEYTRUDA ®

### 14.1.1 Chemical Name

Humanized X PD-1_mAb (H409A11) IgG4

### 14.1.2 CAS Name and Number

Anti-(human protein PDCD1 (programmed cell death 1)) immunoglobulin G4 (human-Mus musculus monoclonal heavy chain) disulfide with human-Mus musculus monoclonal light chain, dimer / 1374853-91-4

### 14.1.3 Molecular Formula: $C_{6534}H_{10004}N_{1716}O_{2036}S_{46}$ (peptide)

### 14.1.4 Molecular Weight: 149 kDa.

### 14.1.5 Appearance

Sterile, preservative-free, white to off-white lyophilized powder in single-use vials. Each vial is reconstituted and diluted for intravenous infusion. Each 2 mL of reconstituted solution contains 50 mg of pembrolizumab and is formulated in L-histidine (3.1 mg), polysorbate-80 (0.4 mg), sucrose (140 mg). May contain hydrochloric acid/sodium hydroxide to adjust pH to 5.5

### 14.1.6 Storage and Stability

KEYTRUDA is supplied in a carton containing one 50 mg single-use vial (NDC 0006-3029-02). Store vials under refrigeration at 2°C to 8°C (36°F to 46°F).

### 14.1.7 Supplier

Pharmacy will receive Merck Marketed Drug (MMD) (i.e., commercial supply) of pembrolizumab for use after 30 April 2020. This will replace the previous arrangement in which Merck was providing Pembrolizumab Lot R022867, which expires on 30 Apr 2020.

### 14.1.8 Mechanism of Action

Pembrolizumab (previously known as SCH 900475) is a potent and highly selective humanized monoclonal antibody (mAb) of the IgG4/kappa isotype designed to directly block the interaction between PD-1 and its ligands, PD-L1 and PD-L2. This blockade enhances functional activity of the target lymphocytes to facilitate tumor regression and ultimately immune rejection.

### 14.1.9 Administration for this study

Intravenous

### 14.1.10 Preparation

| Product Name & Potency | Dosage Form |
|---|---|
| Pembrolizumab 50 mg | Lyophilized Powder for Injection |
| Pembrolizumab 100 mg/ 4mL | Solution for Injection |

Drug will be supplied either as powder for injection, 50 mg single-use vial, or a liquid, DP 100 mg/vial (25 mg/mL), both in Type I glass vials intended for single use only.

Reconstitution of Pembrolizumab Lyophilized Powder for Injection:

Add 2.3 mL of Sterile Water for Injection, USP by injecting the water along the walls of the vial and not directly on the lyophilized powder (resulting concentration 25 mg/mL).

Slowly swirl the vial. Allow up to 5 minutes for the bubbles to clear. Do not shake the vial.

Pembrolizumab Solution for Infusion 100 mg/vial is a liquid DP (manufactured using the fully formulated DS), and has the identical formulation as that of the reconstituted lyophilized vial.

### 14.1.11 Preparation for Intravenous Infusion

The liquid drug product is a clear to opalescent solution which may contain proteinaceous and extraneous particulates. Do not shake or freezer the vial(s).

The liquid product is intended for IV administration.

The liquid DP can be further diluted with 0.9% Sodium Chloride (normal saline) in IV containers made of polyvinyl chloride (PVC) or non-PVC material. The final concentration of the diluted solution should be between 1 mg/ml to 10 mg/ml. Do not dilute or combine product as an infusion with other medicinal products. Do not co-administer other drugs through the same infusion line. Administer infusion solution intravenously over 30 minutes through an intravenous line containing a sterile, non-pyrogenic, low- protein binding 0.2 micron to 5 micron in-line or add-on filter. The infusion solution should be immediately administered. If not used immediately, vials and/or IV bags may be stored at 2-8 °C for up to a cumulative time of 20 hours. If refrigerated, the vials and/or IV bags should be allowed to equilibrate to room temperature prior to subsequent use. Pembrolizumab solutions may be stored at room temperature for a cumulative time of up to 6 hours. This includes room temperature storage of reconstituted or liquid DP solution in vials, room temperature storage of infusion solution in the IV bag and the duration of infusion.

### 14.1.12 Incompatibilities

No formal pharmacokinetic drug interaction studies have been conducted with Pembrolizumab. Live vaccines should be prohibited within 30 days prior to the first dose of trial treatment and while participating in the trial.

### 14.1.13 Side Effects

14.1.13.1 Non-Clinical Toxicology Summary of Results

In the 1-month and 6-month toxicology study in cynomolgus monkeys, Pembrolizumab administered once a week and once every other week respectively, intravenously up to 200 mg/kg resulted in no adverse treatment related effects. The exposure multiple based on a predicted AUC 0-tau of 4464 μg.day/mL at the maximum anticipated human clinical dose of 10 mg/kg Q2W or Q3W is 15-fold at 200 mg/kg, the NOAEL for the 6-month monkey study. Additionally, in the tissue cross-reactivity study of Pembrolizumab with human and monkey tissues demonstrated the expected on-target staining of the membranes of mononuclear leukocytes in both species. Off-target cross-reactivity staining was also noted in both species but was limited to cytoplasm of various cell types/tissues and the stroma (extracellular connective tissue matrix), and was considered related to the experimental method artifacts, i.e. tissue processing for IHC, that are well recognized limitations of tissue cross-reactivity studies and, thus not considered toxicologically relevant.

No reproductive or developmental toxicity studies are planned with Pembrolizumab. Therefore, inclusion of women of childbearing potential in clinical trials should be in accordance with the study protocol and applicable regulatory guidance (e.g., ICH M3(R2): Nonclinical Safety Studies for the Conduct of Human Clinical Trials and Marketing Authorization for Pharmaceuticals).

14.1.13.2 Clinical Summary of Results

As of 18-Oct-2013, 1,000 patients have been treated with Pembrolizumab at several dose schedules, including 10 mg/kg every 2 weeks. Pembrolizumab has been generally well tolerated, as expected based on preclinical findings and other anti-PD-1 monoclonal antibodies. As of 18-Oct-2013 no serious infusions reactions have been reported in PN001, however, since the potential exists in anti-PD-1 monoclonal antibodies, investigators should be vigilant to this possibility. Less than 1% of patients thus far assayed had confirmed positive ADA samples and among these, no or no clear impact on exposure has been observed.

There is no contraindication to further clinical investigation with Pembrolizumab. Pharmacokinetics were as expected, based on Pembrolizumab being an IgG mAb and based on preclinical data, which support dosing once every 2 or 3 weeks. Pembrolizumab monotherapy induces an ORR of 25%/27% in patients with ipilimumab exposed melanoma by central independent RECIST and oncology review/investigator assessed irRC, respectively. Pembrolizumab monotherapy induces an ORR of 39%/43% in patients with ipilimumab-naive melanoma by central independent RECIST and oncology review/investigator assessed irRC, respectively. These responses are remarkably durable. The preliminary 1-year survival rate for patients, many of whom have had multiple therapies, including ipilimumab, who receive Pembrolizumab is 81%. Pembrolizumab monotherapy induces an ORR of 21%/24% in patients with previously-treated NSCLC by central independent RECIST/investigator assessed irRC, respectively, with these responses also remarkably durable. Preliminary data suggest higher levels of PD-L1 expression in tumors of NSCLC are associated with increased activity (ORR 67% by investigator assessed irRC/57% by central independent RECIST); additional data are required to define the optimal PD-L1 cut point. The most commonly reported treatment emergent AEs experienced are fatigue (43.8%), nausea (26.7%), cough (25.3%), pruritus (24.6%), diarrhoea (22.3%) and rash (21.5%). Immune-related adverse events were reported in 21.4% of melanoma patients; most of these events (15.8%) were considered drug-related by the

investigator. The most commonly reported, immune-related adverse events across the dose-schedules are rash (3.2%), pruritus (2.9%), vitiligo (2.9%), hypothyroidism (2.7%), arthralgia (2.2%), diarrhoea (2.2%), and pneumonitis (1.9%). Review of the overall benefit: risk ratio of Pembrolizumab favors enrollment of eligible patients into clinical trials of Pembrolizumab. The preliminary data suggest that a dose of Pembrolizumab at 2 mg/kg Q3W is appropriate for patients with melanoma.

#### 14.1.13.3 Important Safety Considerations

Based on results from the nonclinical studies, there are currently no specific safety considerations. Pembrolizumab is a humanized monoclonal Ab. Thus far, no serious infusion reactions have been reported however, patients should be closely monitored for potential AEs during antibody infusion and potential AEs throughout the study. In the event that a subject experiences an allergic reaction to Pembrolizumab, treatment (i.e., vasopressors, H2-blockers, antihistamines, H1-blockers, steroids) should be administered, as appropriate, and prophylaxis should be considered. Surveillance for the appearance of ADAs is included in all protocols.

Pembrolizumab has the same mechanism of action as other anti-PD-1 monoclonal antibodies. Preclinical studies have suggested similar potency, and PK modeling has suggested similar human PK in the class. Accordingly, the AEs observed with other anti-PD-1 antibodies may serve as in indicator for the AEs to expect for Pembrolizumab in cancer patients. Furthermore, AEs from other immunotherapies for cancer were considered in the Events of Clinical Interest (Section ) and Immune-Related Guidance.

#### 14.1.13.4 Immune-Related Adverse Events (I-R AE)

An I-R-AE is defined as a clinically significant AE of any organ that is associated with study drug exposure, is of unknown etiology, and is consistent with an immune-related mechanism. Immune-related adverse events were reported in 21.4% of melanoma patients; most of these events (15.8%) were considered drug- related by the investigator.

The most commonly reported, immune-related adverse events across the dose-schedules are rash (3.2%), pruritus (2.9%), vitiligo (2.9%), hypothyroidism (2.7%), arthralgia (2.2%), diarrhoea (2.2%), and pneumonitis (1.9%). The organ most frequently affected by irAEs with Pembrolizumab is the skin. Less frequently affected tissues include thyroid gland, colon, lung, kidney, and liver. Based on the mechanism of action of Pembrolizumab and similar immunomodulatory agents, the sponsor is keenly interested in potential events of colitis, hepatitis, hyper/hypothyroidism, nephritis, pneumonitis. Therefore, the sponsor encourages appropriate academic investigation of signs and symptoms suggestive of any of the above events. Consultation with the appropriate medical specialist should be considered when investigating a possible irAE. These events can occur after the first dose to several months after the last dose of treatment. Mild irAEs are usually treated symptomatically and do not require dosing delays or discontinuation. Higher grade and persistent lower grade irAEs typically necessitate withholding or discontinuing treatment and administration of systemic steroids or other immunosuppressive agents (such as tumor necrosis factor blockers), when systemic steroids are not effective. Early recognition of irAEs and initiation of treatment are critical to reduce the risk of complications, since the majority of irAEs are reversible with the use of steroids and other immune suppressants.

14.1.13.4.1 Other immune-mediated adverse reactions

The following additional clinically significant, immune-mediated adverse reactions

were reported in less than 1% of patients treated with KEYTRUDA (pembrolizumab) in KEYNOTE-001, KEYNOTE-002, KEYNOTE-006, and KEYNOTE-010:
uveitis, myositis, Guillain-
Barré syndrome, pancreatitis, encephalitis, sarcoidosis, myasthenic syndrome/myasthenia gravis (including exacerbation) and), myelitis., and vasculitis.

The following were reported in other clinical studies with KEYTRUDA or in postmarketing use: myocarditis and sclerosing cholangitis.

Cases of these immune-mediated adverse reactions, some of which were severe, have been reported in clinical trials or in postmarketing use.

14.1.13.5 Nursing Implications

Standard treatment protocols per hospital guidelines will be used for this Pembrolizumab. Dose will be administered prior to RT.

## 14.2 TEMOZOLOMIDE

Temozolomide during Concomitant Radiation Therapy (RT)

TMZ will be administered orally on day 1 of radiation therapy (RT) 1-2 hours before each session of radiotherapy or night before RT; and it should be administered the same time throughout the course of RT. TMZ should continue throughout RT at the dose of 75 mg/m2/day. During weekends without radiotherapy (Saturday and Sunday), the drug will be taken at the same time as radiotherapy days.

### 14.2.1 Chemical Name

Temozolomide is 3,4-dihydro-3methyl-4-oxoimidazo[5,1-d]-as-tetrazine-8-carboxamide

### 14.2.2 Molecular Formula: $C_6H_6N_6O_2$

### 14.2.3 Molecular Weight: 194.15

### 14.2.4 Appearance

white to light tan/light pink powder

### 14.2.5 Supply: Commercially Available.

### 14.2.6 Pill Diary

Temozolomide will be properly accounted for, handled, and disposed in accordance with existing federal regulations and principles of Good Clinical Practice. All oral study drugs will be recorded in the patient diaries found in Appendix **16.14**. This will be used as a memory aide for subjects. A clinical research team maintains the primary source record.

### 14.2.7 Storage and Stability

The capsules are packaged in amber glass bottles and should be stored at 25°C. Temperature excursions between 15 and 30°C are permissible. Refer to the commercially labeled bottles for expiration dating.

Temozolomide is rapidly and completely absorbed after oral administration; peak plasma concentrations occur in 1 hour. Food reduces the rate and extent of temozolomide absorption. Mean peak plasma concentration and AUC decreased by 32% and 9%, respectively, and Tmax increased 2-fold (from 1.1 to 2.25 hours) when temozolomide was administered after a modified high-fat breakfast.

Temozolomide is rapidly eliminated with a mean elimination half-life of 1.8 hours and exhibits linear kinetics over the therapeutic dosing range. Temozolomide has a mean apparent volume of distribution of 0.4 L/kg (%CV=13%). It is weakly bound to human plasma proteins; the mean percent bound of drug- related total radioactivity is 15%.

### 14.2.8 Metabolism and Elimination

Temozolomide is spontaneously hydrolyzed at physiologic pH to the active species, 3-methyl-(triazen-1-yl)imidazole-4-carboxamide (MTIC) and to temozolomide acid metabolite. MTIC is further hydrolyzed to 5-amino-imidazole-4-carboxamide (AIC), which is known to be an intermediate in purine and nucleic acid biosynthesis and to methylhydrazine, which is believed to be the active alkylating species.

Cytochrome P450 enzymes play only a minor role in the metabolism of temozolomide and MTIC.

### 14.2.9 Mechanism of Action

An oral cytotoxic alkylating agent is the lead agent in a new class of compounds known as imidazotetrazines. Cytotoxic agents are designed to prevent the replication of cells that divide rapidly, including those in tumors.

### 14.2.10 Special Populations

14.2.10.1 Creatinine Clearance:

Caution should be exercised when temozolomide is administered to patients with severe renal impairment. Temozolomide has not been studied in patients on dialysis.

14.2.10.2 Hepatically Impaired Patients:

In a pharmacokinetic study, the pharmacokinetics of temozolomide in patients with mild to moderate hepatic impairment (Child's-Pugh Class I-II) were similar to those observed in patients with normal hepatic function. Caution should be exercised when temozolomide is administered to patients with severe hepatic impairment.

14.2.10.3 Gender:

Population pharmacokinetic analysis indicates that women have an approximately 5% lower clearance (adjusted for body surface area) for temozolomide than men. Women have higher incidences of Grade 4 neutropenia and thrombocytopenia in the first cycle of therapy than men.

14.2.10.4 Age:

Population pharmacokinetic analysis indicates that age (range 19-78 years) has no influence on the pharmacokinetics of temozolomide.

### 14.2.11 Drug-Drug Interactions

In a multiple dose study, administration of temozolomide with ranitidine did not change the Cmax or AUC values for temozolomide or MTIC. Population analysis indicates that administration of valproic acid decreases the clearance of temozolomide by about 5%. The clinical implication of this effect is not known. Population analysis failed to demonstrate any influence of co-administered dexamethasone, prochlorperazine, phenytoin, carbamazepine, ondansetron, H2-receptor antagonists, or phenobarbital on the clearance of orally administered temozolomide.

### 14.2.12 Adverse Events:

- Hematologic: Thrombocytopenia, leukopenia, myelodysplastic syndrome
- Gastrointestinal: Nausea, vomiting, anorexia
- Hepatic: Elevated liver enzymes (reversible)
- Skin: Rash
- Neurologic: Convulsions, weakness on one side of the body, abnormal coordination, paralysis
- Other: Constipation, diarrhea, stomatitis, fatigue, decreased performance status, headache

### 14.2.13 Special Handling and Precautions

Capsules should not be opened or damaged. Rigorous precautions should be taken to avoid capsule contents having contact with skin or mucous membranes. Capsule contents may be irritating to skin and eyes. Mutagenic and prolonged exposure may cause serious health effects (outside of prescribed dosage in this trial).

Temozolomide is potentially mutagenic and should be handled with appropriate precautions by both staff and patients. Capsules should not be opened. If capsules are accidentally opened or damaged, rigorous precautions should be taken with the capsule contents to avoid inhalation or contact with the skin or mucous membranes. Procedures for proper handling and disposal of anticancer drugs should be considered.

### 14.2.14 Contraindications

Temozolomide is contraindicated in patients who have a history of a hypersensitivity reaction to any of its components or to DTIC.

### 14.2.15 Pregnancy Category D

Temozolomide may cause fetal harm when administered to a pregnant woman. There are no adequate and well-controlled studies in pregnant women. If this drug is used during pregnancy, or if the patient becomes pregnant while taking this drug, the patient should be apprised of the potential hazard to the fetus. **Women of childbearing potential should be advised to avoid becoming pregnant during therapy with temozolomide.**

### 14.2.16 Fertility

Treatment of a man with temozolomide may increase the risk of birth defects if he causes a woman to become pregnant while he is taking temozolomide. Men treated with temozolomide may have difficulty causing a woman to become pregnant after their treatment is completed. Men receiving temozolomide should be directed to use effective contraception while they are being treated. There is insufficient data to know what the risk of subsequent problems with fertility will be. Similarly, women who are treated with temozolomide may have difficulty becoming pregnant in the future and may at be at increased risk of having children with birth defects. There is insufficient evidence to determine what the risk of these complications will be.

## 14.3 HEAT SHOCK PROTEIN-PEPTIDE COMPLEX-96 VACCINE (HSPPC-96)

HSPPC-96 is an autologous tumor-derived vaccine under clinical investigation for the treatment of a variety of cancer types. It is composed of the 96-kDa heat shock protein gp96 in complex with autologous tumor-derived peptides. The gp96 in HSPPC-96 is a highly conserved, abundant, nonpolymorphic stress protein found in every cell type of the body. The gp96 isolated from normal or tumor tissues is found in complex with a diverse repertoire of peptides, which are specific to the tissue of origin. Published studies in mouse tumor models have shown that HSPPC-96 confers protective immunity only to the tumor from which it is derived and not to antigenically distinct tumors. The specific immunogenicity of the HSP preparations can be attributed to the unique repertoire of antigenic peptides that exists in different cancers.

When injected into the host, HSPPC-96 interacts with antigen presenting cells (APCs) such as macrophages, dendritic cells or Langerhans cells, which take up HSPPCs via HSP receptors, including CD91. Once internalized by APCs, the peptides chaperoned by the HSPs are transferred to major histocompatibility complex (MHC) class I and II molecules in intracellular compartments and subsequently expressed at the cell surface. The cells then migrate to lymph nodes, where the complexes are processed so that the peptides chaperoned by the HSPs are re-presented to naive T cells. The T cells recognize the peptides and, as a result, are stimulated by them. Vaccination with HSPPCs thus elicits both a CD8+ and CD4+ T-cell response targeting potentially all relevant tumor antigens. The interaction of HSPs with receptors on APCs also leads to activation of various components of innate immunity, including cytokine and chemokine release by macrophage and dendritic cells, maturation of dendritic cells and activation of natural killer cells, as shown in both murine and human systems. Tumor immunity is largely mediated by T cells and the ability of HSPPC-96 to stimulate both T-cell arms to recognize a large variety of tumor antigens, coupled with the ability to activate innate immune responses, uniquely positions the product among other cancer vaccine strategies.

### 14.3.1 Molecular Formula

The molecular formula of HSPPC-96 is unknown since the antigenic peptide repertoire isolated from tumor cells is patient specific.

### 14.3.2 Peptide Component

The peptide component of HSPPC-96 is not characterized on a patient specific basis due to the limited quantity of product available and the technical challenges in performing this type of peptide analysis.

### 14.3.3 Appearance

HSPPC-96 is supplied in a single-use vial as a clear, colorless solution. Each vial contains 25 µg of HSPPC-96 in a solution of 9% sucrose-potassium phosphate for intradermal (ID) injection. The total volume of each vial of HSPPC-96 is 0.47 mL. The total volume that should be administered is approximately 0.4 mL.

### 14.3.4 Availability

Blinded vaccine (HSPPC-96 or placebo) will be prepared by Agenus Inc. and shipped directly to sites within approximately 4-6 weeks of Agenus' receipt of tumor tissue for randomized patients. The number of vaccine treatments will be based on the amount of vaccine generated. Although HSPPC-96 vaccine will be prepared for patients on the placebo arm, it will not be shipped to sites. Vaccine produced for patients randomized to placebo will be used for quality evaluation and will not be available for clinical use.

### 14.3.5 HSPPC-96 Vaccine Label

Each vial of vaccine vial is labeled with the subject/patient study number, subject/patient initials, and subject/patient date of birth (DOB).

### 14.3.6 Storage and Stability

Following successful production of HSPPC-96 vaccine at Agenus Inc. and randomization, vials of blinded vaccine are shipped overnight on dry ice and must be stored at -80 °C ± 20 °C until administration to the patient.

NOTE: Once HSPPC-96 is drawn up into the syringe, the vaccine is stable for up to 2 hours at ambient temperature if immediate administration is not possible. Please refer to the Investigator's Brochure for further details.

Any unused or expired vaccine remaining after a patient has completed vaccine treatment should be returned to Agenus or destroyed at the site upon approval from Agenus. Destruction records are to be forwarded to Agenus. The shipping kit used to send vaccine from Agenus to the site may be stored until the patient has completed vaccine therapy and utilized for return of any unused or expired vaccine. See Appendix **16.6** for further detail.

### 14.3.7 Preparation

Withdraw the vial from the freezer and roll gently between two fingers until it is completely thawed (5-10 minutes). Withdraw approximately 0.4 mL, using a low hold up TB or insulin syringe.

### 14.3.8 Administration

The route of administration is intradermal injection. Inject into 1 site or into 2 adjacent sites (0.2 mL each) a few centimeters apart. Appropriate sites for vaccination include the anterior deltoid regions or subclavicular region bilaterally.

Do not administer HSPPC-96/placebo to any side where lymph nodes have been removed or damaged. Rotate the injection sites so that injections are not repeated at the same site at 2 consecutive administrations, and utilize all potential sites for the patient before repeating

injections at a previously used injection site. Syringes with slip-tip detachable needles or luer hubs with greater than 0.1 mL dead space should not be utilized.

### 14.3.9 Toxicities

Local injection-site reactions can occur and are usually of minimal extent and mild severity. If a local injection-site reaction requires medical attention, the investigator can provide local therapy as needed. Occasional systemic toxicities have been reported that have been mild in severity. The most frequently reported events include injection- site reactions of erythema and induration, fatigue, nausea, pyrexia, headache, back pain, arthralgia, constipation, and asthenia.

## 14.4 PLACEBO

### 14.4.1 Supply

Placebo will be prepared by Agenus Inc. and shipped directly to sites within approximately 4-6 weeks of Agenus' receipt of tumor tissue for randomized patients. The number of placebo treatments will be based on the amount of vaccine generated.

### 14.4.2 Product Description

Each dose of placebo is supplied in a single-use vial as a clear, colorless solution. Each vial contains a solution of 9% sucrose-potassium phosphate for intradermal (ID) injection. The total volume in each vial of placebo is 0.47 mL. The total volume that should be administered is approximately 0.4 mL.

### 14.4.3 Placebo Label

Each vial of vaccine vial is labeled with the subject/patient study number, subject/patient initials, and subject/patient date of birth (DOB).

### 14.4.4 Storage

Vials of blinded vaccine are shipped overnight on dry ice and must be stored at–-80 °C ± 20 °C until administration to the patient.

**NOTE**: Once the placebo is drawn up into the syringe, it is stable for up to 2 hours at ambient temperature if immediate administration is not possible.

### 14.4.5 Administration

Withdraw the vial from the freezer and roll gently between two fingers until it is completely thawed (5-10 minutes). Withdraw approximately 0.4 mL, using a low hold up TB or insulin syringe.

### 14.4.6 Dose

The route of administration is intradermal injection. Inject into 1 site or into 2 adjacent sites (0.2 mL each) a few centimeters apart. Appropriate sites for vaccination include the anterior deltoid regions or subclavicular region bilaterally.

Do not administer placebo to areas distal to lymph node basins that have been resected or in areas just distal to a surgical scar. Rotate the injection sites so that injections are not repeated at the same site at 2 consecutive administrations, and utilize all potential sites for the patient before repeating injections at a previously used injection site. Syringes with slip-tip detachable needles or luer hubs with greater than 0.1 mL dead space should not be utilized.

## 14.5 ORDERING STUDY AGENT

### 14.5.1 Heat Shock Protein-Peptide Complex-96

Following vaccine manufacture and completion of quality testing, Agenus will send the site the Vaccine Production Notification form indicating whether manufacture of vaccine was successful. Upon randomization, Agenus will then send the appropriate blinded vaccine to the site.

Agenus Contact Information for Tissue Procurement and Kit Ordering:
supply&logistics@agenusbio.com or fax: 781-674-4220.

See Appendix **16.3** and Appendix **16.4** for more details.

### 14.5.2 Pembrolizumab

Pembrolizumab may be requested by the Principal Investigator (or their authorized designees) at each participating institution. All regulatory document requirements, as described in the BTTC Operations Manual, must be current and up to date in the BTTC Coordinating Center. A Study Drug Order Form and Pharmacy Initiation Worksheet Form will need to be completed; these forms can be obtained from the BTTC Coordinating Center. The participating institution must have received a Site Activation Memo from the BTTC Coordinating Center prior to requesting study agents.

Signed and Dated drug requests should be emailed to:

Merck
Attn: Eleonora Veglia, PhD
Email: nciisdrugorder@merck.com
Cc: eleonora.veglia@msd.com
Phone: +44 (0) 7824524450

When a number of investigators are participating on a clinical study at the same institution, one investigator should be considered or designated the principal or lead investigator under whom all investigational agents for that protocol should be ordered.

### 14.5.3 Temozolomide

Temozolomide is commercially available.

### 14.5.4 Agent Storage and Accountability:

The investigator is responsible for the proper and secure physical storage and record keeping of investigational agents received for BTTC protocols. Specifically, the investigator must:

- Maintain a careful record of the receipt, use and final disposition of all investigational agents received, using the NCI Investigational Agent Accountability Record Form (DARF), **http://ctep.cancer.gov/forms/index.html**
- Store the agent in a secure location, accessible to only authorized personnel, preferably in the pharmacy.
- Maintain appropriate storage of the investigational agent to ensure the stability and integrity of the agent.
- Return or destroy any unused investigational agents at the completion of the study or upon notification that an agent is being withdrawn.

The intent of the agent accountability procedures described in this section is to assist the investigator in making certain that agents received from BTTC are used only for patients entered onto an approved protocol. The record keeping described in this section is required under FDA regulation. Investigators are responsible for the use of investigational agents shipped in their name. Even if a pharmacist or chemotherapy nurse has the actual task of handling these agents upon receipt, the investigator is the responsible individual and has agreed to accept this responsibility by signing the FDA 1572: https://www.fda.gov/science-research/clinical-trials-and-human-subject-protection/clinical-trial-forms

### 14.5.5 BTTC Procedures for Agent Accountability and Storage

- Each investigational agent should be stored separately by protocol. If an agent is used for more than one protocol, there should be separate physical storage for each protocol. Remember that agents are provided and accounted for on a protocol-by-protocol basis.
- Each agent should be accounted for separately by protocol. If an agent is used for more than one protocol, there should be a separate Investigational Agent Accountability Record Form (DARF) for each protocol, http://ctep.cancer.gov/forms/index.html. There should be a separate DARF for each agent in a multi-agent protocol.
- Separate accountability forms should be maintained for each different strength or dosage form of a particular agent (e.g., an agent with a 1-mg vial and a 5-mg vial would require a different DARF for the 1-mg vial than for the 5-mg vial).
- The DARF has been designed for use at each location where agents are stored, e.g., main pharmacy, satellite pharmacy, physician's office, or other dispensing areas.
- The DARF is also designed to accommodate both dispensing records and other agent transaction documentation (e.g., receipt of agent, returns, broken vials, etc.). A copy of the DARF may be found at http://ctep.cancer.gov/forms/index.html.
- Unauthorized inter-institutional transfer of BTTC investigational agents from one participating institution to another is not permitted. For some protocols the lead institution may enter into contractual agreements to forward agents to participating institutions (Refer to the BTTC Operations Manual).
- BTTC Supplied agents must not be repackaged and forwarded to patients on a routine basis. Refer to the BTTC Operations Manual for BTTC polices on forwarding BTTC supplied agents under certain limited circumstances.

### 14.5.6 Verification of Compliance

Investigators are reminded that compliance with procedures to ensure proper agent usage will be reviewed during site visits conducted under the monitoring program. Specifically, site visitors will check that the agent accountability system is being maintained, and will spot-check the agent accountability records by comparing them with the patients' medical records to verify that the agents were administered to a patient entered in the recorded protocol.

## 14.6 RETURNING OR DESTROYING UNUSED AND/OR DEFECTIVE AGENT:

### 14.6.1 Pembrolizumab Return Procedure

All unused and returned pembrolizumab may be destroyed on site. The site must follow their local drug destruction policy and procedure. A copy of the drug destruction record must be submitted to BTTC to send to Merck..

**Contact**: Eleonora Veglia, PhD
Scientific Leadership & Research Manager (EU SLRM)
Merck Investigator Initiated Studies Program/ Scientific Engagements
MSD, MRL Innovation Hub
Two Pancras Square, Kings Cross
London N1C 4AG
Tel: +44 (0) 7824524450
Email: eleonora.veglia@msd.com

The investigator is responsible for keeping accurate records of the clinical supplies received from Merck or designee, the amount dispensed to and returned by the subjects and the amount remaining at the conclusion of the trial.

Upon completion or termination of the study, all used and/or partially used investigational product will be destroyed at the site per institutional policy. It is the Investigator's responsibility to arrange for disposal of all empty containers, provided that procedures for proper disposal have been established according to applicable federal, state, local and institutional guidelines and procedures, and provided that appropriate records of disposal are kept.

### 14.6.2 HSPPC-96 or Placebo Return Procedure

If a subject is randomized to receive placebo or if there is any remaining vaccine after the subject has finished treatment, it will be coded and linked and used by Agenus or other approved investigators for development and research purposes. Refer to section **16.6** for HSPPC-96 or Placebo return instructions.

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# 16 APPENDICES

## 16.1 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. |

## 16.2 TUMOR TISSUE SUBMISSION FOR VACCINE MANUFACTURE

See Appendix **16.3** for detailed instructions for handling, preparation, and submission of tumor tissue for HSPPC-96 vaccine manufacture.

The Tumor Procurement Form (TPF) must be included with the tissue sent to Agenus Inc.

**Kits for shipping tissue will be supplied by Agenus and should be ordered at time of IRB approval because they will be needed at time of surgery:**

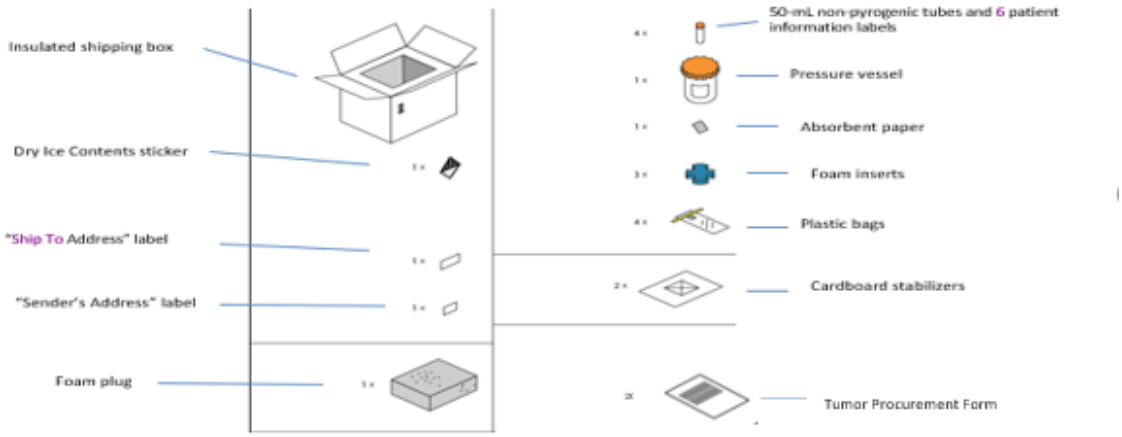
Supply and Logistics
Agenus Inc.
Primary Contact: Tel: +1 (781) 674-4400 x4486 (office)
Email: supply&logistics@agenusbio.com

The dimensions of the kits are 17" x 17" x 15". Please order one or two kits at a time, based on your anticipated use, and please allow 3-4 days for shipping. Upon receipt of a kit with patient tumor at Agenus, a new kit will automatically be replenished for the site.

## 16.3 VACCINE TISSUE PROCUREMENT AND SHIPPING INSTRUCTIONS

**Tissue procurement kits provided by Agenus Inc. include:**

- Tumor Procurement Kits provided by Agenus will include the following materials:




  - 50-mL nonpyrogenic tubes and patient information labels
  - Dry Ice Contents Sticker
  - Absorbent paper
  - Plastic bags
  - Orange-capped pressure vessels (with foam insert)
  - Cardboard pieces (to support base of pressure vessel and top of dry ice)
  - Tumor Procurement Form (TPF)
  - Shipping box and all shipping/address labels **Sites must provide the dry ice (8kg).**
  -

**The site should contact Agenus at supply&logistics@agenusbio.com to request courier documents (pre-paid shipping label for tumor shipment from site to Agenus) 1 day in advance of the desired shipment and the shipping label will be emailed to the site.**

The shipment of tissue to Agenus for vaccine creation should be delayed until confirmation of histologic diagnosis, MGMT status and IDH mutational status that would make the tumor eligible for vaccine production. Tumor specimens for vaccine production must be frozen and stored at -80°C ± 20°C. The tumor specimen must not arrive on the weekend or a holiday unless this has been agreed to in advance by Agenus.

If the tumor specimen will not be shipped immediately, it should be placed into an -80° (+/- 20°) C freezer until ready to pack and ship. Do this within a maximum of 30 minutes of removal.

For tumor specimen stored at sites prior to shipping to Agenus (i.e. overnight or for weekends/holidays), the freezer does not need to be sterile, but it is recommended that it be monitored for temperature (24/7) with alarms or other mechanism for identifying when temperature goes out of specification. The freezer or its location should have controlled access

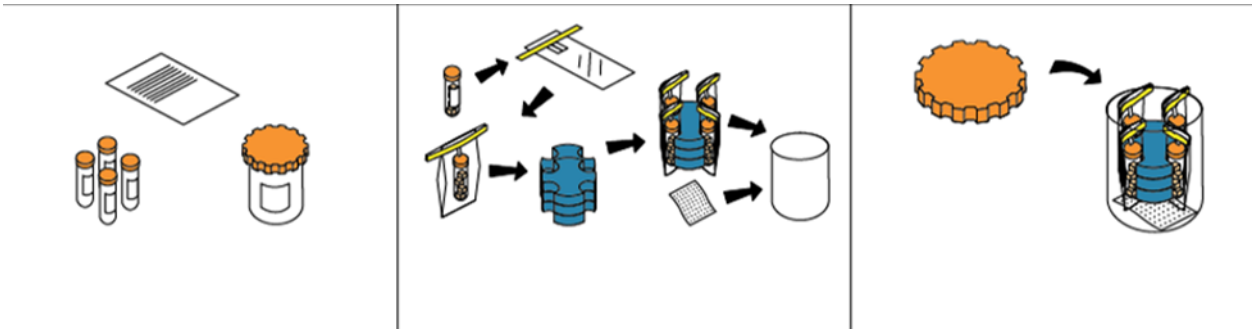
(locked with specified group members having access). The freezer should also be on a preventative maintenance program and ideally be hooked up to a back-up power system.

In the OR, wrap or place the tumor specimen in a sterile specimen cup, cloth or towel, and place in a basin, on ice (not dry ice). Transport the specimen immediately to the pathology/frozen section suite.

**Share the following steps with the surgical pathologist, along with the tissue procurement kit:**

a. Use basic sterile technique. Care should be taken to avoid any possible cross-contamination with tissues or fluids from other patients.
b. As small a section of tumor as possible should be kept for routine pathologic studies and diagnosis. Tumor for routine pathologic diagnosis should be processed according to institutional standards.
c. The pathologist should assess viability of the tumor.
d. When there is necrosis and/or cystic degeneration in the tumor specimen, the necrotic tissue and/or cystic component should be removed and should not be sent to Agenus.
e. **All available** tumor aside from what will be used for routine pathologic studies and diagnosis should be sent for HSPPC-96 vaccine preparation. A minimum of ≥ 7 grams of fresh, non-necrotic tumor should be sent (if <7 grams, may not produce sufficient vaccine).

**Tumor procurement and shipping procedure**



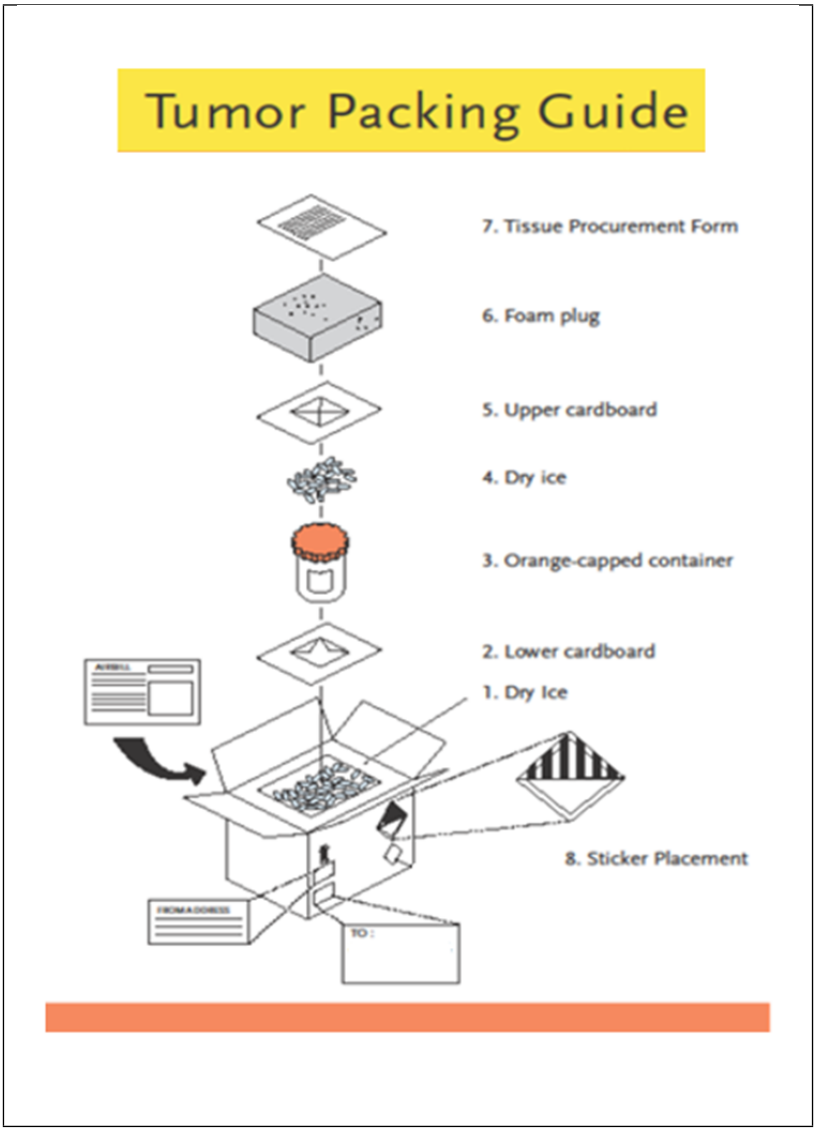
a. The pathologist should section the tissue into 1-2 $cm^2$ sections. The sections should then be placed carefully, maintaining sterility, into the 50-mL nonpyrogenic tubes. Each tube should be filled no more than ½-⅔ full to allow for expansion of tumor during freezing.
b. Each tube must be properly labeled (patient/subject identifiers are a match on all tubes).
c. Each tube should be placed in a plastic pouch, and the pouch sealed closed.
d. Wrap the tubes in bubble wrap or place in the foam insert in the upright position.
e. Absorbent paper should be placed in the bottom of the orange capped pressure vessel. The sealed tubes in the foam insert are then placed into the pressure vessel

and the vessels lid is screwed shut. Dry ice should NOT be placed inside the pressure vessel.

f. Place some dry ice in the bottom of the insulated shipper box.

g. The pressure vessel should be placed above the dry ice, with the base seated in the cardboard support, and more dry ice packed surrounding the container.

h. Dry ice should be added to about 10 cm from the top of the box, a second cardboard insert placed above the ice, and the foam plug placed into the box. If an insufficient amount of dry ice is placed in the box, and the tissue is not frozen on arrival, it cannot be processed and the patient must be withdrawn from the study.




i. The **Tumor Procurement Form** (top sections) must be completed with all required information ensuring patient/subject identifiers are a match on the form and tube labels.

The form should be scanned and e-mailed to: supply&logistics@agenusbio.com so

that the shipment is expected and can be traced by airbill number if it does not arrive. A copy of the form should also be kept for the records of the study center.

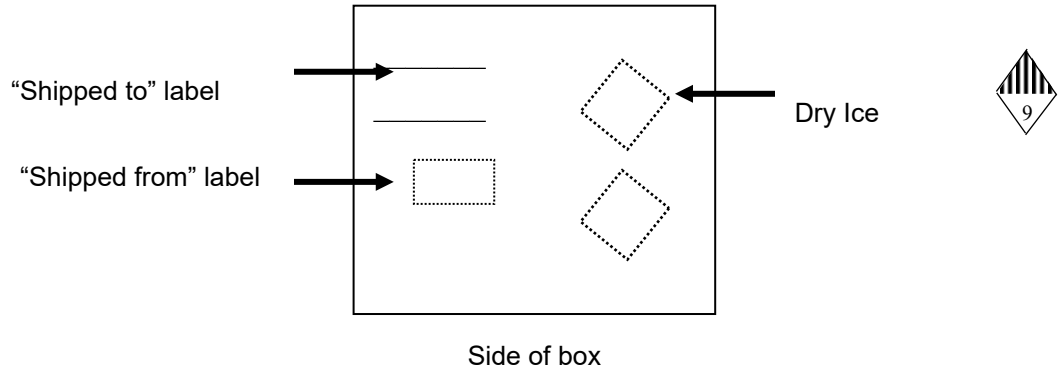
j. The original **Tumor Procurement Form** should be placed in the clear documentation holder on the inside carboard flap of the box. Close the cardboard flaps and securely taped shut.

k. A copy of the TPF should be kept for site records.

l. The following labels should be over the appropriate areas:

1. Dry Ice (#9) With Weight
2. "Shipping To" label
3. "Shipped From" label

m. Record the amount (in kilograms) of dry ice placed in the package on the Dry Ice label.




**Attach the courier air bill to the box. The shipment should be sent to:**

Supply and Logistics
Agenus Inc.
3 Forbes Road
Lexington, MA 02421
United States
Tel: +1 (781) 674-4486

**Below is a list of some common issues that have been encountered with completing the Tumor Procurement Form (TPF).**

Please take great care in filing out this form correctly, as any such issues may lead to delays in vaccine production and potential rejection of the material if the correct identity cannot be confirmed.

- Study subject ID number does not match – tube labels vs. procurement form
- Patient/subject date of birth does not match – tube labels vs. procurement form
- Patient/subject initials do not match – tube labels vs. procurement form (i.e., A-C ≠ C-A)

- Study subject ID number, date of birth or patient initials are illegible on procurement form or tube labels
- Tube labels used are not the same as those issued by Agenus
- Tubes are not labeled at all, or information is recorded directly onto body of tube and is then smeared upon receipt
- Pen or marker used to record information on tube label is not indelible ink, and information is then smeared upon receipt
- Tubes for more than 1 patient/subject are placed into one pressure vessel
- Initials and date of correction are not recorded when cross-outs are made on tube labels or Tumor Procurement Form

## 16.4 TUMOR PROCUREMENT FORM





**TUMOR PROCUREMENT FORM (TPF) FOR PROTOCOL 17C0034**

Principal Investigator: _____ Site Name: _____

**PATIENT AND TUMOR INFORMATION (ALL FIELDS MUST BE COMPLETED BY INSTITUTION)**

**Study Subject ID Number:** ☐☐☐-☐☐☐☐

Patient Initials: ☐ - ☐ (First - Last)

Patient Date of Birth: ☐☐/☐☐☐/☐☐☐☐ (Day / Month / Year)
Example: 30JAN 2017

Check here to confirm above information (mistakes in identity will cause delay in tumor processing) ☐

Date of Excision: ☐☐/☐☐☐/☐☐☐☐
Example: 30JAN2017

Number of Tubes: ☐

Time of Excision of tumor: ☐☐:☐☐ (24-hour clock)

Time tumor placed at -80°C or on dry ice: ☐☐:☐☐ (24-hour clock)

Tumor Sample Weight: ☐☐.☐ Grams

Has all visible necrotic tissue been removed to leave viable tissue? ☐ Yes ☐ No
*i.e., pinkish/yellowish (may be darker) homogenous and firm in consistency*

COMMENT: _____ Recorded by: _____ (Signature)

Date of Shipping: ☐☐/☐☐☐/☐☐☐☐
Example: 30JAN 2017

Courier: _____

Air waybill Number: _____

Email or Fax # to send confirmation of tumor receipt: _____

PACKED FOR SHIPPING BY:
_____ _____
Print Name Date

Shipping Instructions Followed: ☐ Shipping container filled with at least 8 kg of dry ice: ☐

**Email this form to supply&logisitics@agenusbio.com or Fax this form to +1-781-674-4220 as soon as completed. Maintain a copy and enclose the original form with tumor shipment.**

**TO BE COMPLETED BY AGENUS**

PLACE LABEL HERE

RECEIVED BY:
_____ _____
Print Name Date

Did tumor and documentation meet acceptance criteria? Yes ☐ No ☐ If no, why: _____

Deviation Number (if applicable): _____

Agenus Document IS-31748, Ver. 01

## 16.5 LabConnect Manual for Whole Blood PBMC Processing and Shipping

This manual is a separate document.

## 16.6 HSPPC-96 Vaccine or Placebo Return Instructions

1. Any unused or expired vaccine remaining after a subject has completed vaccine treatment, should be returned to Agenus Inc. according to the following instructions.
2. Please contact the Agenus Team at BTTC17C0034@agenusbio.com in advance (i.e., minimum of 1 day) to inform Agenus when you plan to return unused or expired vaccine. Also copy nci_bttc@mail.nih.gov on this return request communication.
3. Agenus will provide a pre-paid courier air-way bill via email for this shipment to your site.
4. Use the qualified HSPPC-96 shipping kit that was received with the original shipment of vaccine from Agenus. The shipping kit should have the following materials, which came as part of the vaccine shipment from Agenus:

    - Outer cardboard shipping box.
    - Insulated liner and foam plug.
    - Pressure vessel container (with foam inserts).
    - Cardboard pieces (2: 1 for upright positioning of base of pressure vessel and 1 for placement on top of pressure vessel surrounded with dry ice).
    - Absorbent paper (on bottom of pressure vessel)

5. Place vials to be returned inside of the pressure vessel, inserted upright in the holes of the foam inserts. Screw the lid shut on the pressure vessel. Multiple subjects' /patients' vaccines may be returned in 1 one pressure vessel.
6. Place a layer of dry ice on the bottom of the insulated liner that is within the outer cardboard shipping box. The pressure vessel container should then be placed above this layer of dry ice, with the base of the pressure vessel inserted into the cardboard support. Additional dry ice is then packed around the closed pressure vessel. The shipping container must be filled with at least 8 kg of dry ice (i.e., level with the top of the pressure vessel). A second cardboard insert should be placed above the dry ice, and the foam plug placed into the box. The box is then sealed with packing tape.
7. The following labels should be placed on the exterior of the outer cardboard shipping box:

    - Dry ice (#9) with weight (8 kg).
    - Shipping address/content label (where the package is being sent).
    - Shipped from label (Where the package is shipping from)

8. Contact the courier to arrange for a pick-up, if a standard pick- up time is not established for your site or if this was not done in advance.
9. Attach the courier air bill to the top of the box. The shipment should be sent overnight to:

Supply and Logistics
Agenus Inc.
3 Forbes Road
Lexington, MA 02421
United States
Tel: +1 (781) 674-4486
supply&logistics@agenusbio.com

16.7 **HSPPC-96 Intradermal Injection Instructions**

# HSPPC-96 Intradermal Injection Instructions

*****FOR CLINICAL STAFF ONLY*****

**IRB APPROVAL IS REQUIRED IF IT IS INTENDED FOR PATIENT USE.**

agenus

# HSPPC-96 Intradermal Injection Instructions

The route of administration is intradermal injection. Using a 25 g needle or equivalent, inject into 1 site or into 2 adjacent sites (0.2 mL each) a few centimeters apart. Appropriate sites for vaccination include the anterior deltoid regions, subclavicular region bilaterally, and medial inguinal regions of the upper thighs (please see the figure below).

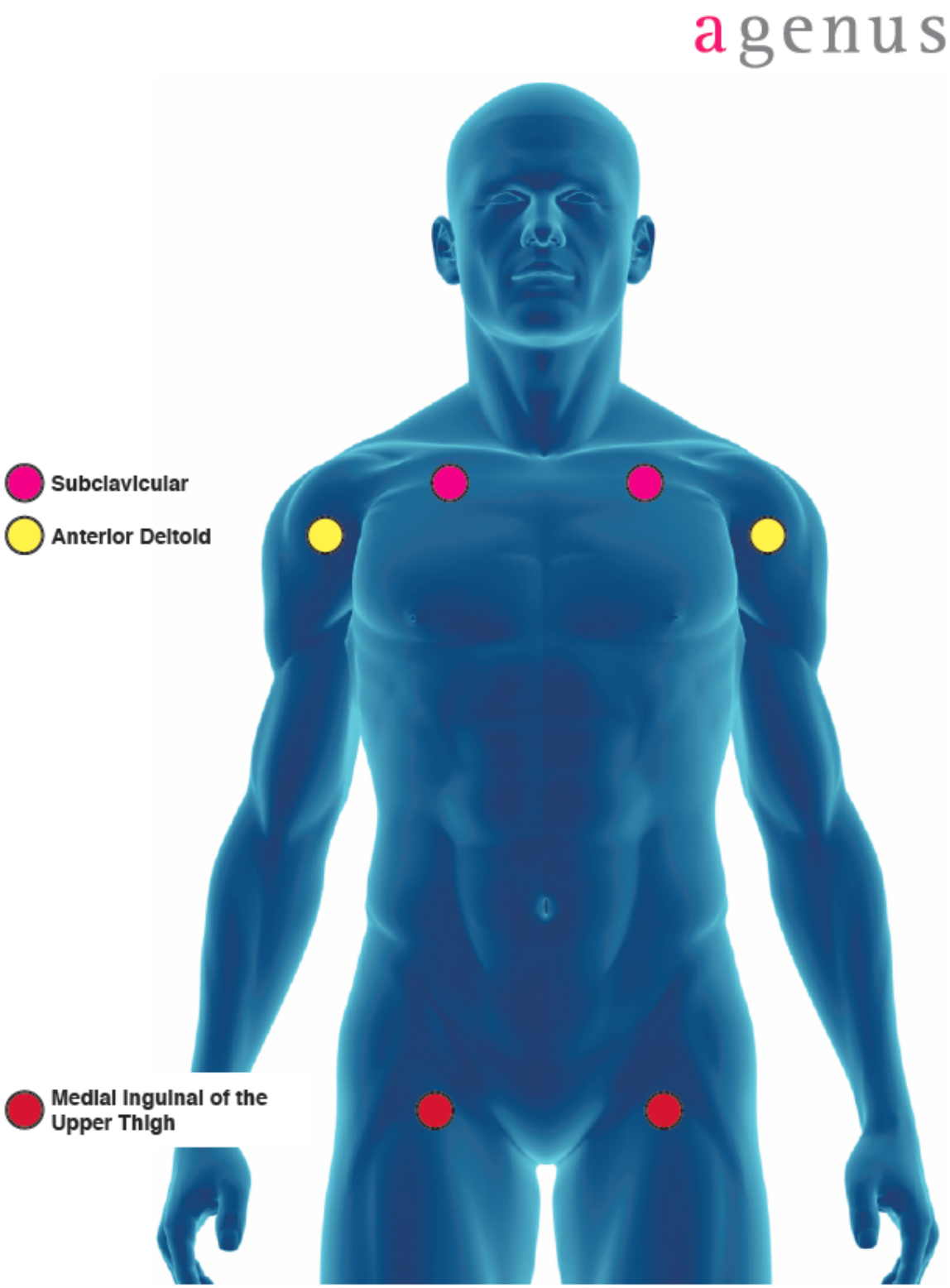
Intradermal injections are administered into the outer layers of the skin. The needle should be inserted at a

10-15 degree angle so that it just punctures the skin's surface. Insert the needle about 0.3 cm below the epidermis and stop when the needle's tiup is under the skin. You should feel some resistance as you make the injection and a wheal should form as you complete the injection. If the needle tip moves freely, you have inserted the needle too deeply. At this point, withdraw needle slightly and check again for resistance. The appearance of a wheal indicates that the vaccine has entered the area between the intradermal tissues. If a wheal does not appear, withdraw the needle and repeat the procedure in another site a few centimeters apart.

Do not administer HSPPC-96 to areas distal to lymph node basins that have been resected or in areas just distal to a surgical scar.

Rotate the injection sites so injections are not repeated at the same site forat 2 consecutive administrations and utilize all potential sites for the patient before repeating injections at a previously used injection site.

Syringes with slip-tip detachable needles or luer hubs with greater than 0.1 mL dead space should not be utilized.


agenus
Subclavicular
Anterior Deltoid
Medial Inguinal of the Upper Thigh

## 16.8 INFUSION REACTION TREATMENT GUIDELINES

| NCI CTCAE Grade | Treatment | Premedication at subsequent dosing |
|---|---|---|
| Grade 1<br>Mild reaction; infusion interruption not indicated; intervention not indicated | Increase monitoring of vital signs as medically indicated until the subject is deemed medically stable in the opinion of the investigator. | None |
| Grade 2<br>Requires infusion interruption but responds promptly to symptomatic treatment (e.g., antihistamines, NSAIDS, narcotics, IV fluids); prophylactic medications indicated for < =24 hrs | **Stop Infusion and monitor symptoms.**<br>Additional appropriate medical therapy may include but is not limited to:<br>IV fluids<br>Antihistamines<br>NSAIDS<br>Acetaminophen<br>Narcotics<br>Increase monitoring of vital signs as medically indicated until the subject is deemed medically stable in the opinion of the investigator.<br>If symptoms resolve within one hour of stopping drug infusion, the infusion may be restarted at 50% of the original infusion rate (e.g., from 100 mL/hr to 50 mL/hr). Otherwise dosing will be held until symptoms resolve and the subject should be premedicated for the next scheduled dose.<br>**Subjects who develop Grade 2 toxicity despite adequate premedication should be permanently discontinued from further trial treatment administration.** | Subject may be premedicated 1.5h (± 30 minutes) prior to infusion of pembrolizumab with:<br>Diphenhydramine 50 mg po (or equivalent dose of antihistamine).<br>Acetaminophen 500-1000 mg po (or equivalent dose of antipyretic). |

| NCI CTCAE Grade | Treatment | Premedication at subsequent dosing |
|---|---|---|
| <u>Grades 3 or 4</u><br>Grade 3:<br>Prolonged (i.e., not rapidly responsive to symptomatic medication and/or brief interruption of infusion); recurrence of symptoms following initial improvement; hospitalization indicated for other clinical sequelae (e.g., renal impairment, pulmonary infiltrates)<br>Grade 4:<br>Life-threatening; pressor or ventilatory support indicated | **Stop Infusion.**<br>Additional appropriate medical therapy may include but is not limited to:<br>IV fluids<br>Antihistamines<br>NSAIDS<br>Acetaminophen<br>Narcotics<br>Oxygen<br>Pressors<br>Corticosteroids<br>Epinephrine<br>Increase monitoring of vital signs as medically indicated until the subject is deemed medically stable in the opinion of the investigator.<br>Hospitalization may be indicated.<br>**Subject is permanently discontinued from further trial treatment administration.** | No subsequent dosing |
| Appropriate resuscitation equipment should be available in the room and a physician readily available during the period of drug administration. | | |
| | | |

## 16.9 MD ANDERSON SYMPTOM INVENTORY (MDASI-BT)

This questionnaire is a separate document.

## 16.10 CCR REPORTABLE EVENT FORM (REF)

| | |
|---|---|
| **NCI Protocol #:** | |
| **Protocol Title:** | |
| **Report version:** *(select one)*<br>☐ Initial Report<br>☐ Follow-up | |
| **Site Principal Investigator:** | |
| **Date site PI was notified of the problem:** | **Date of problem:** |
| **If delay in reporting to the coordinating center, please explain:** | |
| **Location of problem:** *(e.g., patient's home, doctor's office )* | |
| **Description of Subject**<br>Does this problem apply to a subject?<br>☐ yes<br>☐ not applicable (more than one subject is involved)<br><br>**If yes, enter details below:**<br>Subject ID: (*do not use medical record number)*<br>Sex: ☐Male ☐ Female<br>Age: | |

| |
|---|
| Diagnosis: |
| **Name the problem:** *(select all that apply)*<br>☐ Specimen collection issue<br>☐ Informed consent issue<br>☐ Ineligible for enrollment<br>☐ Breach of PII<br>☐ Other, briefly state the nature of the problem:<br><br>**Detailed Description of the problem:** (*Include any relevant treatment, outcomes or pertinent history*): |
| **What are you reporting?**<br>☐ **unanticipated problem**<br>☐ **death**<br>☐ **non-compliance (other than a protocol deviation)**<br>☐ **protocol deviation**<br>☐ **new information that might affect the willingness of subjects to enroll or continue participation in this study** |
| **If interventional or expanded access study, please answer the following questions about your site:** |

| | |
|---|---|
| How many participants are still receiving the study intervention?<br><br>How many participants completed study interventions but remain in follow up?<br><br>How many participants are enrolled but not yet receiving study interventions? | |
| **Have similar problems occurred on this protocol at your site?**<br>**☐ Yes ☐ No** | |
| **Describe what steps you have already taken or will be taking as a result of this problem:** | |
| **INVESTIGATOR'S SIGNATURE:** | **DATE:** |

## 16.11 UNBLINDING REQUEST MEMO

MEMORANDUM

DATE:

TO: Lead PI

FROM: _____MD

Phone Number:_____

Email address:_____

Site:_____

RE: Patient Unblinding Request for A randomized, double blind phase II trial of Surgery, Radiation Therapy plus Temozolomide and Pembrolizumab with and without HSPPC-96 in newly diagnosed Glioblastoma (GBM)

I am requesting the following patient to be unblinded:

Subject ID # _____

I am requesting this patient to be unblinded because:

_____
_____
_____
_____

Thank you,

_____ _____

Signature of requesting physician Date

I have reviewed this request and agree that the circumstances warrant patient unblinding

_____ _____

Signature of approving MD Date

I have reviewed this request but do not agree that the circumstances warrant patient unblinding. The reason I am denying this request is:

_____
_____
_____
_____

_____ _____

Signature of disapproving MD Date

**16.12 UNBLINDING NOTIFICATION MEMO**

MEMORANDUM

DATE: _____

TO: _____MD

Site:_____

FROM: Lead PI, MD

RE: Patient Unblinding Notification for protocol titled - A randomized, double blind phase II trial of Surgery, Radiation Therapy plus Temozolomide and Pembrolizumab with and without HSPPC-96 in newly diagnosed Glioblastoma (GBM)

With concurrence from the Data Safety Monitoring Board, the Lead PI MD has requested unblinding for the following patient:

Subject ID #_____

Date of unblinding: _____

The patient's treatment assignment is: _____HSPPC-96 _____Placebo

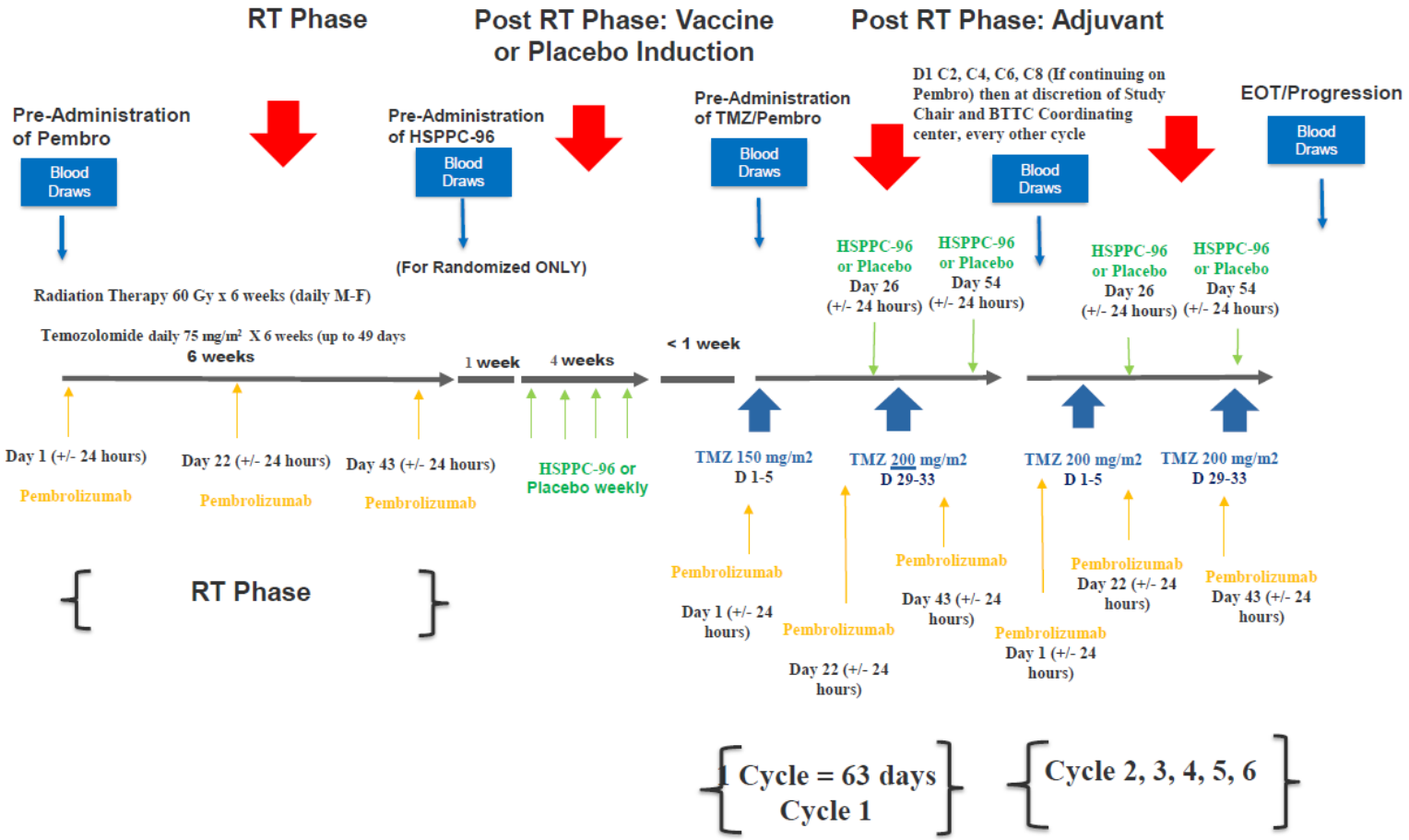
Be sure to place this memo in the patient's clinic or hospital chart following your institution's guidelines.

Thank you,

_____ _____

SIGNATURE DATE

## 16.13 STUDY PHASES AND CORRELATIVE BLOOD DRAWS



## 16.14 PILL DIARY

**Pill Diary Instructions**: Use this pill diary as an aid to record when you took your daily chemotherapy. Place a check mark or your initials after each daily dose. If you miss a dose, add a reason in the space provided. Bring this diary and all study drug bottle(s) back to your next clinic.

| | | | |
|---|---|---|---|
| **Patient Name:** | | | **Study ID #:** |
| **Study Phase/Cycle:** | | | **Today's date:** |
| **Week** | **Day** | **Date** | **Temozolomide (TMZ) Dose**<br>**Instructions: Take TMZ _____mg by mouth before bed daily** |
| 1 | Pre | | X_____ |
| | 1 | | X_____ |
| | 2 | | X_____ |
| | 3 | | X_____ |
| | 4 | | X_____ |
| | 5 | | X_____ |
| | 6 | | X_____ |
| 2 | 7 | | X_____ |
| | 8 | | X_____ |
| | 9 | | X_____ |
| | 10 | | X_____ |
| | 11 | | X_____ |
| | 12 | | X_____ |
| | 13 | | X_____ |
| 3 | 14 | | X_____ |
| | 15 | | X_____ |
| | 16 | | X_____ |
| | 17 | | X_____ |
| | 18 | | X_____ |
| | 19 | | X_____ |
| | 20 | | X_____ |
| 4 | 21 | | X_____ |
| | 22 | | X_____ |
| | 23 | | X_____ |
| | 24 | | X_____ |
| | 25 | | X_____ |
| | 26 | | X_____ |
| | 27 | | X_____ |
| 5 | 28 | | X_____ |
| | 29 | | X_____ |
| | 30 | | X_____ |
| | 31 | | X_____ |
| | 32 | | X_____ |

| | | | |
|---|---|---|---|
| | 33 | | X_____ |
| | 34 | | X_____ |
| 6 | 35 | | X_____ |
| | 36 | | X_____ |
| | 37 | | X_____ |
| | 38 | | X_____ |
| | 39 | | X_____ |
| | 40 | | X_____ |
| | 41 | | X_____ |
| | 42 | | X_____ |

## 16.15 INVESTIGATOR AGREEMENT

I have received and reviewed the Investigator Brochure for Pembrolizumab and HSPPC-96.

I have read this protocol and agree that the study is ethical.

I agree to conduct the study as outlined and in accordance with all applicable regulations and guidelines.

I agree to maintain the confidentiality of all information received or developed in connection with this protocol.

Protocol Title: A randomized, double blind phase II trial of Surgery, Radiation Therapy plus Temozolomide and Pembrolizumab with and without HSPPC-96 in newly diagnosed Glioblastoma (GBM)

Protocol Version Date: 06/25/2025

_____ _____

Signature of Principal Investigator Date

_____

Name of Principal Investigator (printed or typed)