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IRB Proposal for CRC Rotation

PHASE 1/2 Investigation of HSPPC-96 Vaccine in Patients with non-Glioblastoma Solid CNS Tumors

1. Study Purpose and Rationale:

The estimated US incidence rate for primary brain and nervous system tumors is approximately 19.34 per 100,000 persons, with a mortality rate of 4.6 per 100,000 person-years. Depending on the specific histopathologic grading of the tumor, 5-year survival rates range from less than 5 percent to over 90 percent. [1]

Current standard of care for treatment of most brain malignancies is maximal surgical resection followed by chemotherapy and/or radiation therapy depending on the specific pathology of the tumor. Location of tumor and eloquence of surrounding brain parenchyma largely influence the resectability of a given tumor, as one has to balance the goal of maximal resection with that of minimal postoperative functional deficit. Following resection, the specific chemotherapeutic agents of choice depend on the histopathologic grade and type of tumor; however, all agents come with a their own level of toxicity, both systemic and CNS-specific.

Often gross total surgical resection is not achievable given the location of the tumor. Even when gross total resection is said to be achieved, many tumors recur, and five-year survival rates for certain types of brain cancer are still dismal. Clearly there is room for improvement in terms of treatment. To that end, our study has the following aims:

Primary Endpoints:
- To evaluate the safety profile of HSPPC-96, an autologous tumor-derived heat shock protein peptide-complex therapeutic vaccine, administered to patients with non-glioblastoma solid CNS tumors, including any adverse effects.
- To evaluate progression-free survival (PFS) in patients treated with HSPPC-96, from date of surgical resection.

Secondary Endpoints:
- To evaluate median overall survival in patients treated with HSPPC-96 vaccine.
- To evaluate the immunologic response to vaccine treatment.

2. Study Design and Statistical Procedures:

This is a Phase 1 and 2 single-arm open label study designed to evaluate feasibility, safety and efficacy of tumor-derived heat-shock protein peptide complex (HSPPC-96) vaccine in the treatment of patients with non-glioblastoma solid CNS tumors, following initial surgical resection.

During the initial phase of this trail, at least 10 patients will be enrolled for the purposes of evaluating initial safety of HSPCC-96 administration. A safety evaluation and immune testing will be completed once at least 10 patients have received at least 6 doses of vaccine. Treatment with vaccine will continue during the safety evaluation and immune testing to confirm safety and assess the dosing schedule, without interruption to study enrollment or treatment procedures.

In order to evaluate efficacy, we will measure the proportion of patients who remain disease free at three years post surgical resection and compare this to historical norms. Initial efficacy evaluations will focus on patients with anaplastic meningiomas, as this is the largest group of patients we expect to enroll. Based on a study done by Yang et al. in 2008, the 3-year recurrence-free survival rate for anaplastic meningioma patients is 50%.[1] We can reasonably expect to see a 50% increase in this number to 75% of patients treated with the vaccine being recurrence-free at 3 years post-resection. This effect size is based on the Phase I/II trials that have been carried out using HSPPC-96 vaccine treatment for patients with glioblastoma multiforme. Historical median survival for patients with glioblastoma is 26 months. In the clinical trial, 93% of patients were still alive at 26 months—an effect size of 43%. Using an arguably modest effect size of 25%, and a fixed comparison group, we will need to enroll 33 patients to show this level of efficacy. This calculation is done using a chi-square test, with alpha = 0.05, and a power of 80%, as follows:

n (# needed in each group) = 8(p1q1+p2q1)/(effect^2) + 2/(effect) +2
n = 8[(.5)(.5) + (.75)(.25)]/(.25^2) + 2/(.25) + 2
n = 66 patients
However, because we are comparing to a fixed proportion of .5 for our control group (as it is based on historical norms), we will only need half as many subjects in our treatment group, giving: n = 66/2
n = 33 patients

For all patients enrolled in the study, time to recurrence, as well as overall survival will be measured. For each event (event = tumor recurrence or death), a z-score can be calculated comparing the patient's time to event to historical norms. Using z-scores, we will be able to evaluate efficacy across different tumor types. Using an effect size of half a standard deviation prolonged survival, alpha = 0.05, and power = 80%, one again gets n = 33 patients needed to enroll, based on an unpaired t-test as follows:

n (# needed in each group) = 1 + 16(SD/effect)^2
n = 1 + 16[(SD)/(SD/2)]^2
n = 1 + 16(2)^2
n = 65
Again, this number can be divided by two, as we are comparing to historical norms, which eliminates the uncertainty in one of the groups, giving n = 65/2 = 32.5
n = 33 patients

This n represents 33 patients with events (primary event: recurrence), rather than 33 patients followed to a certain time point. However, it allows us to include patients with more rare tumor types in our analysis. Final statistical analysis will be conducted using Kaplan-Meier survival curves and a Log-Rank analysis.

3. Study Procedures:

Patients will be treated with standard of care maximal surgical resection, followed by confirmation of eligibility for vaccine therapy 1-3 weeks postoperatively. Initial eligibility assessment will consist of clinical and radiographic confirmation of disease stability. Vaccine administration will then be initiated at 2-5 weeks postop, and patients will receive 4 weekly injections of HSPPC-96 followed by a 5th vaccine injection administered 2 weeks following vaccine administration #4. Subsequent vaccine injections will be administered monthly, and will continue until depletion of vaccine or progression of disease.

- Safety monitoring and investigational product interruption: All patients on study will be monitored for safety. The safety parameters include all clinical laboratory testing for abnormalities, physical & neurological examination findings, and reporting of adverse events reported to the investigator by patients. AEs will be documented from the date of the first treatment with HSPPC-96 until 30 days following the protocol defined administration period. All toxicities encountered during the study will be evaluated according to the NCI CTCAE, version 4.0, and recorded prior to each course of therapy. All serious adverse events (SAEs) should be reported within 24 hours to Agenus, Inc. The FDA will be notified about any SAE with suspected relationship to HSPPC-96 as required in 21 CFR 312.32 (c).

All patients will be monitored for toxicities. No treatment modifications are necessary for grade 1 toxicities. Patients who experience grade 2 toxicities felt to be secondary to the vaccine, but not severe enough to be removed from treatment, may continue treatment without modification. If the toxicity increases to grade 3 or higher and is felt to be related to the vaccine, the PI will consult with the Agenus medical monitor to determine if the patient should discontinue vaccine treatments.

- Tumor evaluation: Following the baseline visit (pre-vaccination), tumor assessment evaluations will be performed every 4 weeks by clinical examination and every 8 weeks by radiographic imaging (scheduled from the first tumor evaluation visit) to assess disease status until disease recurrence or the patient comes off-study.

Disease status and tumor response will be used primarily to evaluate progression and will be assessed by one or more of the following which includes, but is not limited to: clinical evaluation (neurology examination, physical examination, Karnofsky score), radiographic evaluation, and surgical biopsy.
- Immune Blood Draws: Immune blood draws will consist of (14) 10-mL vials of blood drawn during each of the following visits: baseline immune blood draws will be completed preoperatively, intraoperatively, and within 48 hours postoperatively. Additionally, immune blood draws will be completed prior to the first and the 5th vaccine administrations. Subsequent immune blood draws consisting of (10) 10-mL vials of blood will be completed at the time of tumor evaluations #2 (week 9), TE #3 (week 13), TE #8 (week 37) and TE #12 (week 53). An optional final immunologic assessment may be completed a minimum of 4-weeks after the last vaccine administration if the patient has not completed all protocol scheduled immunological testing through TE #12. All immunologic blood draws will be conducted prior to vaccine administration.

Immunomonitoring will be completed via enzyme-linked immunospot (ELISPOT) assay at Agenus. All patients who have completed at least 4 vaccinations and immunomonitoring through the blood draw prior to vaccine administration #5, will be evaluable for immune analysis.

See Appendix A for Schedule of Events.

4. Study Drugs or Devices:

HSPPC-96: HSPPC-96 is an autologous, tumor-derived heat shock protein (glycoprotein 96 or gp96)-peptide complex vaccine that is individually prepared from specimens of the patient's own tumor. Patients will undergo standard surgical resection of intracranial tumor. Tissue will be adequately stored for immediate overnight shipment to Agenus. Tumor designated for vaccine production will be evaluated and processed upon arrival at Agenus.

HSPPC-96 is currently under clinical investigation for the treatment of a variety of cancer types, including glioblastoma multiforme. 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 via specific receptors, including CD91. Once internalized by APCs, the HSP acts as a chaperone to shuttle the tumor-associated antigen peptides to both the endogenous and exogenous immune pathways, so that they are transferred to major histocompatibility complex (MHC) class I and II molecules in intracellular compartments and subsequently expressed at the cell surface. This results in T cell recognition and stimulation. HSPPCs are unique in their ability to elicit an antigen-specific, cytotoxic T-cell response, as well as a CD4+ T cell, natural killer (NK) cell, and innate immune response, including cytokine and chemokine release by macrophage and dendritic cells. 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.

HSPPC-96 for clinical use is individually prepared from specimens of the patient's own tumor, using multistep chromatography involving affinity and non-affinity matrices. It is formulated in a 9% sucrose-potassium phosphate for intradermal (ID) injection. Each product batch is screened prior to release for administration to the patient through a series of quality control tests, including identity, purity, strength, potency, pH, appearance, sterility and endotoxin levels.

Preclinical toxicology studies investigating HSPPC-96 were performed prior to the clinical trials using HSPPC-96 in the treatment of glioblastoma multiforme. Given that patients are treated with multiple doses of HSPPC-96, single-dose toxicity studies were considered to be not reflective of the product, and so, repeat dose toxicology studies were performed to reflect the clinical situation instead. A total of 5 toxicology studies were performed in mice. HSPPC-96 was administered subcutaneously in all studies. In some studies, mice were administered HSPPC-96 only, whereas in others, HSPPC-96 was administered either before or after live tumor cell challenge. The number of HSPPC-96 injections ranged from 2 to 4 (at 1-week intervals) and the dose ranged from 0-100 mcg. The total cumulative

exposure to HSPPC-96 for all studies was 200 mcg. No adverse effects on metabolism (as assessed by measuring body weight), clinical chemistry, hemoglobin, hematocrit, erythrocyte, leukocyte and platelet levels were detected among the groups. Minimal to mild subacute inflammation of the subcutaneous tissue was observed in a limited number of animals. This was not attributed to the test article but rather to the injection itself.
In one of the studies in which HSPPC-96 was administered to tumor-bearing animals, lymphoid hyperplasia was observed in all peripheral lymphoid organs, as reflected by an increase in absolute lymphocyte numbers in the peripheral blood. Other changes consisted of marked vacuolation of hepatocytes of all 5 treated mice. The livers of control animals were not available for examination. Of the 5 treated mice, focal degenerative renal tubular change was observed in 1 mouse and focal scars (fibrosis) in 1 mouse. These changes were likely attributable to resolution of metastatic lesions.

Clinical trials to date have supported the use of HSPPC vaccines in the treatment of human tumors. A Phase I/II trial evaluating HSPPC-96 vaccine in patients with recurrent high-grade glioma has been completed. Immune assessment demonstrated a significant Adaptive and Innate Immune response peripherally, and at the site of tumor resection in situ when biopsies were performed in all patients. Additionally, there were no grade 3 or 4 adverse events directly referable to the vaccine.

Among the 30 evaluable patients, who completed 4 or more doses of the vaccine, median survival was 47.6 weeks, which compares favorably with the historical median control time of 26-weeks. In fact, study patients had a 93% survival rate at 26-weeks.

We confirm that the use and storage of the investigational drugs in this study will comply with the Columbia University Medical Center Policy relating to the use and control of investigational drugs for outpatients, at New York-Presbyterian Hospital and the Investigational Drugs: Use and Control (New York-Presbyterian Hospital P168). In addition, the drug will be dispensed by the Research Pharmacy.

Radiation
: The radiological procedures required by this research protocol will be conducted in accordance with standard practice, both from the perspective of the actual procedure and the frequency of the procedure. The schedule of scans is SOC and as such does not necessitate review and approval by the JSRC.

5. Study Questionnaires:

N/A

6. Study Subjects:

Study subjects will be those with newly diagnosed non-glioblastoma solid CNS tumors, eligible for surgical resection.

Pre-surgery tissue acquisition eligibility:

Inclusion Criteria:
- At least 18 years old
- Life expectancy of greater than 12 weeks
- Able to read and understand the informed consent document; must sign the informed consent
- Must have suspected diagnosis of non-Glioblastoma solid CNS tumor with a surgical intent to resect at least 90% of disease. MRI/CT must be done within 30 days of surgery
- Must be eligible for post-surgical treatment with standard of care

Exclusion Criteria:
- Current diagnosis of Human Immunodeficiency Virus (HIV testing is not required per protocol)
- Any prior diagnosis of any other cancer or other concurrent malignancy, with the exception of adequately treated non-metastatic in situ carcinoma of the uterine cervix or non-metastatic non-melanoma skin cancer provided that in complete remission and off all therapy for that disease for a minimum of 5 years

- Any systemic autoimmune disease (e.g., Hashimoto thyroiditis) and/or any history of primary or secondary immunodeficiency
- Any prior therapy for CNS malignancy
- Planned use or current use of other investigational therapy for the treatment of CNS tumor

Post-radiation therapy/Pre-vaccine eligibility:
Inclusion criteria:
- Agree to use contraception or abstain from sexual activity from the time of consent through 1 month after the end of study drug
- Negative serum pregnancy test for female patients of childbearing potential
- Patients with histologically proven, non-glioblastoma CNS tumors
- Patient must have received standard of care treatment for given malignancy
- Must have undergone at least a 90% resection (determined by the PI) measured by postoperative magnetic resonance imaging (MRI) scan, T1-weighted contrast scan, or CT scan if clinically indicated, performed within 72 hours after surgery
- All radiotherapy must be completed at least 2 weeks prior and no more than 5 weeks prior to the first planned vaccine administration
- Availability of at least 4 doses of vaccine (at least 4 vials for clinical administration produced from the tumor provided)
- Karnofsky functional status rating greater than or equal to 70
- Adequate bone marrow function (ANC greater than or equal to 1,500/ mm3; platelet count greater than or equal to 100,000/mm3), adequate liver function (serum glutamic oxaloacetic transaminase/ aspartate aminotransferase [AST] and alkaline phosphatase <2.5 times institutional upper limit of normal [IULN] and bilirubin (total) <1.5 mgIULN), and adequate renal function (BUN and creatinine <1.5 times IULN)

Exclusion criteria:
- Inability to comply with study-related procedures
- Prior diagnosis of any other cancer or other concurrent malignancy, with the exception of adequately treated non-metastatic in situ carcinoma of the uterine cervix or non-metastatic non-melanoma skin cancer provided that patient is in complete remission and off all therapy for that disease for a minimum of 5 years
- Current or active use of chemotherapy

- Contrast MRI findings (or CT scan if MRI is clinically contraindicated) consistent with progression of disease
- Patients with active uncontrolled infection
- Evidence of bleeding diathesis
- Unstable or severe concurrent medical conditions
- Female patients who are pregnant or breastfeeding

7. Recruitment:

Patients will be recruited from the Oncology services at Columbia University Medical Center. All patients will be seen by an oncology attending. Ample time will be provided to answer any questions and the consent form will be given to the patient to review at home.

Please note that the medical records to be reviewed are from the PIs clinic; as such, the PI will be their treating physician. HIPAA D is put into place to allow the study team to review patient charts prior to their visit to the clinic to identify if they are eligible for a certain trial. As such the PI will be the individual who will be discussing the study with potential subjects. No letters or information sheets will be utilized. The informed consent form will be provided as a document to provide information regarding the trial.

8. Confidentiality of Study Data:

The patients will be followed and their privacy maintained according to HIPAA guidelines and GCP recommendations. The research file that links their name to the code number will be kept in a locked file cabinet on the 9th floor of the

Herbert Irving Pavilion and only the investigator and study staff will have access to the file. The study data collected, specimens and questionnaire responses will be assigned a code number, and separated from the patient name or any other information that could identify them. Data will be sent through a secure electronic website. The sponsor will have access to identifiers.

If the results of this research project are published or presented at a scientific or medical meeting, the patient will not be identified. Otherwise, all results will be kept confidential and will not be divulged (except as required by law) without permission.
Except when required by law, study information shared with persons and organizations outside of Columbia University Medical Center will not identify the patient by name, social security number, address, telephone number, or any other direct personal identifier.

9. Potential Conflict of Interest:

The investigators have no conflicts of interest to declare.

10. Location of the Study:

All office/clinic visits will take place in neurosurgical clinical care areas in the Neurological Institute and Milstein Hospital. Initial surgical procedures will take place in the neurosurgical operating rooms on the 4th floor of the Milstein Hospital Building. Vaccine manufacturing will be outsourced to Agenus Inc.

11. Potential Risks:

There are potential complications associated with a CNS tumor as well as the surgical treatment for this indication that are not necessarily associated with HSPPC-96 and include: infection, increased intracranial pressure, aseptic meningitis, cerebral edema, hemorrhage as well as focal neurological deficits related to the anatomical location of tumor.

See the Investigator's Brochure for a complete listing of HSPPC-96 risks.

12. Potential Benefits:

It is unknown at this time if there are any benefits, however, information gained will help in the treatment of future cancer patients.

13. Alternatives:

The alternatives are standard of care therapy for treatment of this disease or not to participate in this research.
Other choices may include:
- Getting treatment or care for your cancer without being in a study
- Taking part in another study
- Getting no treatment
- Getting comfort care, also called palliative care. This type of care helps reduce pain, tiredness, appetite problems and other problems caused by the cancer. It does not treat the cancer directly, but instead tries to improve how you feel. Comfort care tries to keep you as active and comfortable as possible.

14. Compensation to Subjects:

No compensation will be offered to subjects.

15. Costs to Subjects:

No costs will be incurred by subjects. Subjects will not be billed for charges related to vaccine administration and monitoring.

References:

1. Yang, S.Y., et al., *Atypical and anaplastic meningiomas: prognostic implications of clinicopathological features.* J Neurol Neurosurg Psychiatry, 2008. **79**(5): p. 574-80.

2. Central Brain Tumor Registry of the United States. 2011 CBTRUS Statistical Report: Primary Brain and Central Nervous System Tumors Diagnosed in the United States in 2004-2007., 2011.

Appendix A: Schedule of Events

Tests / Procedures	Pre-surgical eval	Surgical visit + 72hrs post-op	Pre-vaccine eval	Week 1	Week 2	Week 3	Week 4		Week 6	Week 9	Week 13		Week 37	Week 53
Pre-trial screenings/ consent	**X**	**X**	**X**											
Vaccine administration[1]				**X (#1)**	**X (#2)**	**X (#3)**	**X (#4)**		**X (#5)**	**X (#6)**	**X**		**X**	**X**
Injection site rxn assessments				**X**	**X**	**X**	**X**		**X**	**X**	**X**		**X**	**X**
Vital signs			**X**	**X**	**X**	**X**	**X**		**X**	**X**	**X**		**X**	**X**
Physical Exam			**X**				**X**		**X**	**X**	**X**		**X**	**X**
Neurological evaluation			**X**				**X**		**X**	**X**	**X**		**X**	**X**
Karnofsky score			**X**				**X**		**X**	**X**	**X**		**X**	**X**
Brain CT or MRI	**X**	**X**	**X**	Every 8 weeks following baseline evaluation (prior to vaccine administration)										
Hematology[2]			**X**	**X**			**X**		**X**		Every 8 weeks			
Serum Chemistry[3]			**X**	**X**			**X**		**X**		Every 8 weeks			
Adverse event reporting[4]				**X**	**X**	**X**	**X**		**X**	**X**	**X**		**X**	**X**
Immune blood draws	**X**			**X**					**X**	**X**	**X**		**X**	**X**

[1]Four weekly injections followed by a 5th injection administered 2 weeks following vaccine administration #4. Subsequent vaccine injections will be administered monthly until vaccine depletion or another off-treatment criterion is met

[2]RBC, platelets, hematocrit, hemoglobin, WBC, plus WBC differential with absolute counts for neutrophils, eosinophils, basophils, lymphocytes, monocytes

[3]Albumin, alkaline phosphatiase, AST, ALT, bilirubin (total), lugcose, BUN, creatinine, potassium and sodium

[4]Adverse Events will be captured from the date of the first dose of vaccine until 30 days following the protocol defined administration period