

**TITLE: A Phase 2 Study of GVAX Colon Vaccine (with Cyclophosphamide) and Pembrolizumab in Patients with Mismatch Repair-Proficient (MMR-p) Advanced Colorectal Cancer**

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**JHU Supplied Agent:** GVAX Colon Vaccine (Allogeneic SW 837 and SW 620 cell lines admixed with GM-CSF Producing Bystander K562 cell line)

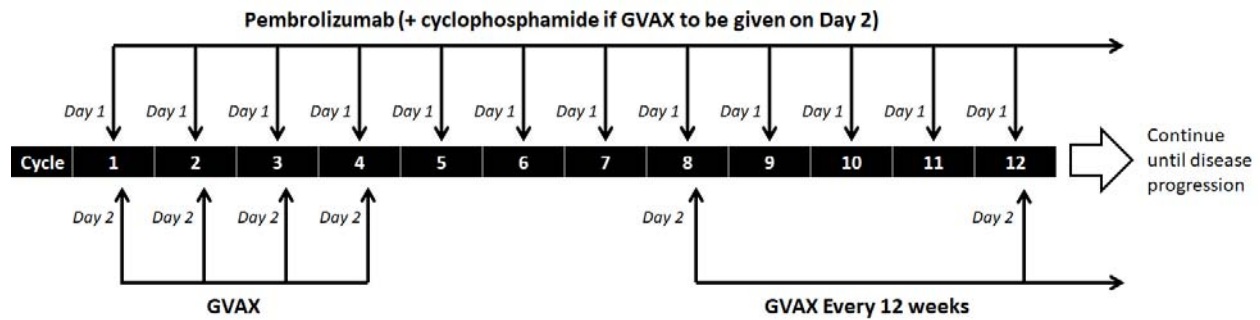
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## TRIAL DESIGN



- This is an open-label phase 2 trial evaluating the GVAX Colon Vaccine with Cyclophosphamide and Pembrolizumab in Patients with Mismatch Repair-Proficient (MMR-p) Advanced Colorectal Cancer
- Each treatment cycle = 21 days
- Patients will receive pembrolizumab on day 1 of each cycle
- Patients will also receive cyclophosphamide on day 1, and GVAX on day 2, for each of the first four cycles, and then every fourth cycle thereafter
- For all patients, imaging scans will be performed at baseline and at least every 12 weeks thereafter, irrespective of the treatment schedule.

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## **1. OBJECTIVES**

### **1.1 Primary Objectives**

- 1.1.1 To determine the objective response rate (ORR) of GVAX colon vaccine in combination with pembrolizumab in patients with mismatch repair-proficient (MMR-p), advanced colorectal cancer who have progressed after at least two lines of prior therapy.

### **1.2 Secondary Objectives**

- 1.2.1 To assess the safety and tolerability of the combination GVAX/pembrolizumab in subjects with MMR-p, advanced colorectal cancer
- 1.2.2 To assess the progression free survival (PFS) of subjects with MMR-p, advanced colorectal cancer who receive GVAX in combination with pembrolizumab using immune related response criteria.
- 1.2.3 To assess the duration of response among subjects who demonstrate an objective response to treatment with GVAX in combination with pembrolizumab.
- 1.2.4 To determine the overall survival (OS) of subjects with MMR-p, advanced colorectal cancer who receive GVAX in combination with pembrolizumab.

### **1.3 Exploratory Objectives**

- 1.3.1 To evaluate the effects of combination therapy with pembrolizumab and Cy/GVAX on multiple immune pathways including suppressive pathways (including PD-L1, PD-L2, LAG3, BTLA, TIM3, IDO1, CTLA-4, and Tregs), activation pathways (including CD137, OX40, CD40, CD40L), cytokines/chemokines or their receptors (including CCL12, CXCR4, CCL2, CCL5, CCR2, CCR5), and tumor infiltrating lymphocytes.
- 1.3.2 To assess tumor burden dynamics through measurements of circulating tumor biomarkers including carcinoembryonic antigen (CEA) as well as other exploratory circulating biomarkers in serial collections of sera and plasma at baseline and throughout treatment.
- 1.3.3 To assess the baseline characteristic of the subjects enrolled and to correlate these molecular and clinicopathologic criteria with treatment response and toxicity.
- 1.3.4 To assess changes in peripheral blood lymphocytes (PBL) to explore the association of lymphocyte activation markers with clinical responses.

### **1.4 Primary Endpoint**

- 1.4.1 ORR, defined as the proportion of patients achieving a complete response (CR) or partial response (PR) based on the Response Evaluation Criteria in Solid Tumors (RECIST 1.1) at any time during the study, of GVAX in combination with pembrolizumab in patients with MMR-p, advanced colorectal cancer.

## **1.5 Secondary Endpoints**

- 1.5.1 Adverse events as graded by NCI CTCAE active version
- 1.5.2 Progression free survival
- 1.5.3 Duration of response
- 1.5.4 Overall Survival

## **1.6 Exploratory endpoints**

- 1.6.1 Intratumoral changes in pre- and post-treatment core biopsy specimens will be studied using analyses such as immunohistochemistry (IHC) and transcriptional analysis (Merck's nanostring panels)
- Immune suppressive pathways including PD-L1, PD-L2, LAG3, BTLA, TIM3, IDO1, CTLA-4, and Tregs
  - Immune activation pathways including CD137, OX40, CD40, CD40L
  - Cytokines/chemokines or their receptors including CCL12, CXCR4, CCL2, CCL5, CCR2, CCR5.
  - T cell receptor (TCR) repertoire (and compared to PBL)
- 1.6.2 Biomarker marker changes (including standard protein biomarkers such as CEA) and correlation to evaluate for prognostic or predictive factors
- 1.6.3 Clinicopathologic characteristics (including but not limited to age, sex, histology, grade, tumor mutations) and biomarkers of response (OS, PFS, and best overall response) or toxicity.
- 1.6.4 Measure pre- and post-treatment changes in PBLs including effector, helper, and regulatory T cells through cell phenotyping analysis and gene expression profiling in association with clinical responses. We will also measure specific T cell responses as a parameter of immune response to treatment with immunotherapy.

## **1.7 Study Design**

This is an open-label, single-arm phase II study of Cy/GVAX in combination with pembrolizumab in subjects with MMR-p, advanced colorectal cancer who have progressed on at least two lines of prior therapies. Subjects with microsatellite repair deficiency (MMR-d; MSI) are not eligible for this study.

Therapy is administered on days 1 and 2 of a 21 day cycle. On day 1, participants will receive their first dose of cyclophosphamide (Cy) 200 mg/m<sup>2</sup> and pembrolizumab 200

mg, both administered intravenously (IV). On day 2, participants will receive GVAX, administered as 8-9 intradermal injections for a final dose of  $5 \times 10^8$  colon cancer cells +  $5 \times 10^7$  GM-CSF secreting cells. Participants may continue therapy every 21 days until there is disease progression. After completing four cycles of therapy, patients will continue pembrolizumab with every cycle of therapy and cyclophosphamide and GVAX will be administered every 4<sup>th</sup> cycles of therapy.

Due to the expectation that some patients may experience delayed clinical responses to therapy, patients with disease progression by radiographic imaging or laboratory parameters during a 12 week evaluation period but without rapid clinical deterioration or significant change in performance status that requires additional immediate therapy may continue to receive treatment on study. In this setting, a repeat scans will be obtained at weeks 6 and 12, and every 12 weeks thereafter from time of initial radiographic progression. This extra scan at 6 weeks after initial radiographic progression will allow earlier recognition of true progression and will ensure that the treatment is stopped adequately if not working. Subjects who meet the above criteria and continue to receive treatment on study despite disease progression by radiographic imaging must discontinue treatment upon documentation of disease progression on the second scan (week 6). The date of progression will be backdated to the time of first RECIST criteria progression. Tumor assessments will be made using RECIST 1.1 and immune-related response criteria (irRC). All patients will be followed by this protocol for at least 30 days after their last dose of study drug or until initiation of a new anti-cancer treatment, whichever occurs first.

NOTE: For patients who progress and remain on study, at the time of the next restaging, stable disease with clinical stability as determined by their treating physician/team can remain on study. Specifically, these patients do not have to have regression to <20% growth from baseline. If they do not have 20% increase from the last restaging scan, they may remain on study.

Patients who are discontinued from the study due to an unacceptable drug-related AE will be followed until the resolution of the AE to Grade 0-1 or stabilization or until initiation of a new therapy for their cancer, whichever occurs first. Patients who discontinue from treatment will be contacted every three months to monitor overall survival. Information about other cancer therapies after discontinuation from the study treatment will be collected.

At the end of the study, subjects will be required to enroll in a long-term follow-up protocol, an FDA requirement for genetically modified products, in which they will be followed for at least five years (or until death) after receiving the last dose of any investigational agent through this protocol for disease progression, survival, and potential long term toxicity of gene therapy. The subjects will be asked to enroll in the study entitled, “Long-Term Follow-Up of Patients who Received the Allogeneic Colon Cancer Cell Vaccine Administered with a GM-CSF Producing Bystander Cell Line.”

## **2. BACKGROUND**



## 2.1 Disease Type

Despite the existence of excellent screening and preventative strategies, colorectal carcinoma remains a major public health problem in industrialized countries and a rising health problem in developing countries. About 102,480 new cases of colon cancer and 40,340 cases of rectal cancer are expected to occur in the United States in 2013, with about 50,830 deaths, accounting for approximately 9% of cancer deaths. Colorectal carcinoma is the third leading cause of death from cancer in both males and females, comprising 9% of all cancers diagnosed in men and women. It also is the third most common malignancy in both men (after prostate and lung cancers) and women (after breast and lung cancers) [1]. Surgical resection of the primary colorectal lesions combined with adjuvant chemotherapy and radiation, when indicated, remain the mainstay of therapy. Unfortunately, approximately 30% of these patients will be diagnosed with metastatic disease at initial presentation, and an additional 25-30% of patients will subsequently develop metastatic disease. Among these patients, approximately 20% will have disease confined to the liver, which is the dominant site of metastatic involvement [2]. Despite recent advances in earlier detection and improvements in chemotherapy, the median survival for all patients with metastatic colorectal carcinoma ranges from 11-20 months with five-year survival < 5% [3-8].

## 2.2 Vaccine Therapy (GVAX)

Immunotherapy is a potentially effective therapeutic approach to the treatment of colorectal carcinoma for several reasons. First, immune-mediated tumor killing acts by mechanisms distinct from standard chemotherapy or radiation therapy and may represent a non-cross resistant treatment modality. Second, the immune system is capable of recognizing a diverse array of potential antigens while orchestrating selective and specific cytotoxic responses. This may be particularly important in the killing of a heterogeneous tumor population while avoiding normal tissue toxicity. Third, preclinical animal models using a vaccine approach for immunotherapy have been able to eliminate small burdens of established tumors, a situation that corresponds to a state of minimal residual disease commonly found after resection of all gross disease [9-14]. Fourth, completed Phase I and II trials evaluating irradiated, GM-CSF secreting allogeneic pancreatic cancer tumor vaccines have demonstrated both clinical and immunologic response [15-17].

The use of whole-cell vaccines is promising because it delivers a range of peptide antigens without the need for specific knowledge of the relevant target antigens. Preclinical studies show that among tumor cells genetically modified to express various cytokines, GM-CSF is the cytokine most effective in inducing anti-tumor immunity [18]. GM-CSF is an important growth and differentiation factor for dendritic cells, which are potent antigen-presenting cells. The use of allogeneic tumor cells for vaccine development over autologous tumor cells is attractive for several reasons. Autologous tumor cells are not always available and the production of an autologous vaccine is technically difficult, costly, and inefficient. Supporting the use of allogeneic tumor cells is the characterization of tumor-associated antigens in melanoma, which revealed that regardless of human leukocyte antigen (HLA) type, 50% of tumors share common antigens [19, 20]. In addition, both preclinical and human data support that the antigen-

presenting cells important in GM-CSF based vaccination are host-derived suggesting that the vaccine cells and the host do not have to be HLA compatible [21, 22].

### **Phase I Study of an Allogeneic GM-CSF-Secreting Tumor Vaccine in Patients with Resected Pancreatic Cancer**

This study was the first clinical trial to test the hypothesis that GVAX can prime a systemic immune response in patients with resected pancreatic adenocarcinoma [15]. Fourteen patients with stage II or III disease received an initial vaccination 8 weeks following resection. This was a dose escalation study in which 3 patients each received  $1 \times 10^7$ ,  $5 \times 10^7$ , and  $1 \times 10^8$  vaccine cells. An additional 5 patients received  $5 \times 10^8$  vaccine cells. Study patients were jointly enrolled in an adjuvant chemoradiation protocol for 6 months. Following the completion of adjuvant chemoradiation, patients were re-assessed and those who were still in remission were treated with 3 additional vaccinations given one month apart at the same original dose that they received for the first vaccination. Toxicities were limited to grade I/II local reactions at the vaccine site, and self-limited systemic rashes, including one documented case of Grover's syndrome. Systemic GM-CSF levels were evaluated as an indirect measure of the longevity of vaccine cells at the immunizing site. As was observed in pre-clinical studies, GM-CSF levels peaked at 48 hours following vaccination. In addition, serum GM-CSF levels could be detected for up to 96 hours following vaccination. The vaccine sites were also evaluated as a measure of the local immune reaction to the vaccine. Eleven of 14 patients demonstrated a similar local inflammatory response to what has been observed in pre-clinical models and autologous GM-CSF vaccine clinical trials. Post-vaccination DTH responses to autologous tumor cells have been used in previously reported vaccine studies as a surrogate to identify and characterize specific immune responses that are associated with vaccination. In the pancreatic cancer vaccine trial, post-vaccination DTH responses to autologous tumor cells were observed in 1 of 3 patients receiving  $1 \times 10^8$  and in 2 of 5 patients receiving  $5 \times 10^8$  vaccine cells.

### **Follow-up Phase II Study Integrating the Whole Cell Vaccine with Chemoradiation for Resected Pancreatic Adenocarcinoma**

The follow-up phase II study of 60 patients with resected pancreatic adenocarcinoma based on the results of their phase I experience was completed [17]. The highest dose of vaccine from the phase I study ( $5 \times 10^8$  vaccine cells) was used. The common toxicities associated with the vaccine in this study included: local vaccine site skin reactions and systemic rashes similar in severity (grade 1-2) to what was observed in the phase I trial. The results from this study include the following:

- The administration of the whole cell vaccine is safe and well-tolerated. Treatment related side effects included transient vaccine injection site reactions. There has been no incidence of anaphylaxis secondary to the vaccine reported thus far.
- Systemic GM-CSF levels were evaluated as an indirect measure of the longevity of vaccine cells at the immunizing site. As was observed in the phase I study, GM-CSF levels peaked at 48 hours following the first and second vaccination but peaked earlier following the 3<sup>rd</sup> and 4<sup>th</sup> vaccination with diminution in amplitude. Serum GM-CSF levels following vaccine 5 peaked again at 48 hours and returned to vaccine 1 serum levels. The results would suggest the possibility that the potency of an

allogeneic vaccine is diminished with repeated monthly vaccinations, but returns to pre-treatment levels with an extended time interval between boosts.

- Post-Immunotherapy induction of mesothelin-specific CD8<sup>+</sup> T cells with higher avidity and increased mesothelin epitope recognition (T cell repertoire expansion) correlates with disease free survival (DFS).

### **Phase I and II Trials of Combining GM-CSF Secreting Allogeneic Vaccines in Sequence with Immune Modulating Doses of Cyclophosphamide (CY)**

Immune tolerance remains a major barrier to effective vaccine therapies. In particular, regulatory T cells (CD4<sup>+</sup>CD25<sup>+</sup>) have been shown to play a role in inducing CD8<sup>+</sup> T cell tolerance. Manipulating the regulatory T cells may result in more effective vaccine strategies. In mouse models, immune modulating doses of CY in combination with GM-CSF based cell vaccines have been shown to improve tumor rejection from 0% to 10 - 30% in the HER-2/neu transgenic mouse model. The addition of CY allowed the activation of high-avidity RNEU<sub>420-429</sub>-specific CD8<sup>+</sup> T cells in the mice, which rejected tumor. This effect was abrogated by CD4<sup>+</sup>CD25<sup>+</sup> T cells derived from neu-N transgenic mice suggesting that CY before vaccination may block T regulatory cells allowing for recruitment of latent high-avidity neu-specific CD8<sup>+</sup> T cells [23].

A feasibility study of the GM-CSF-secreting, allogeneic vaccine administered alone or in sequence with CY in patients with stage 4 pancreatic cancer, has been completed. This study consisted of two cohorts: Cohort A- 30 patients administered a maximum of six doses of vaccine using our two pancreatic cancer cell lines ( $2.5 \times 10^8$  of each cell line) intradermally at 21 day intervals; Cohort B- 20 patients administered CY 250 mg/m<sup>2</sup> IV one day prior to each vaccination (administered as in Cohort A). Results from this study represent the first demonstration that integrating immunomodulatory doses of CY with a GM-CSF-secreting vaccine in patients with advanced pancreatic cancer is safe and feasible to administer. These data suggest that the vaccine given in sequence with CY results in anti-tumor activity, where median survivals in Cohort A and Cohort B were 2.3 months and 4.3 months respectively in a patient population that had received > 2 prior chemotherapies. In addition, mesothelin-specific CD8<sup>+</sup> T cell responses can be detected in stage 4 patients treated with the vaccine and may correlate with time to progression and overall survival [24].

A phase I dose ranging study was completed that evaluated the safety and immunogenicity of administering an allogeneic HER2-positive granulocyte-macrophage colony-stimulating factor (GM-CSF) secreting tumor vaccine in combination with low-dose CY and doxorubicin in patients with metastatic breast cancer. Patients received three monthly immunizations with a booster vaccine 6-8 months from study entry. This study found that low dose CY can augment the magnitude of vaccine-induced humoral immunity, but the therapeutic window for enhancing immune responses was narrow. Specifically, 200 mg/m<sup>2</sup> of CY augmented the magnitude of HER2-specific humoral immunity, whereas doses above 200 mg/m<sup>2</sup> were more likely to suppress both delayed type hypersensitivity and antibody responses. The addition of CY did not potentiate vaccine-related toxicity [25]. Thus, these findings provide the scientific rationale to continue to test combinations of vaccine with other more potent immune modifying agents.

## **Safety and Feasibility Study of an Allogeneic Colon Cancer Cell Vaccine Administered with a GM-CSF Secreting Bystander Cell line in Combination with Cyclophosphamide in Patients with Metastatic Colon Cancer**

Nine patients were treated in this dose-escalating safety and feasibility study. Prior to treatment, six patients underwent curative metastasectomy for hepatic metastases of colorectal adenocarcinoma and the other three had nonresectable metastatic colorectal adenocarcinoma. The investigational colon cancer vaccine consists of three components: a) SW837 cell line derived from a primary colorectal adenocarcinoma, b) SW620 cell line derived from a lymph node metastasis of colorectal adenocarcinoma, and c) K562/GM-CSF cell line, which is a K562 cell line derived from a CML patient in a blast crisis and subsequently transfected with a plasmid vector encoding human GM-CSF. A classical '3+3' dose escalation design was adopted in this trial. Three patients were treated at dose level 1 ( $5 \times 10^7$  colon cancer cells +  $4 \times 10^7$  K562). No dose limited toxicities (DLT) were observed in the first 3 patients, therefore six patients were treated at dose level 2 ( $5 \times 10^8$  colon cancer cells +  $2 \times 10^8$  K562). No DLTs were observed at dose level 2. Patients received up to four monthly vaccinations. Patients received an immunomodulatory dose of cyclophosphamide  $200 \text{ mg/m}^2$  intravenously one day prior to each vaccine administration.

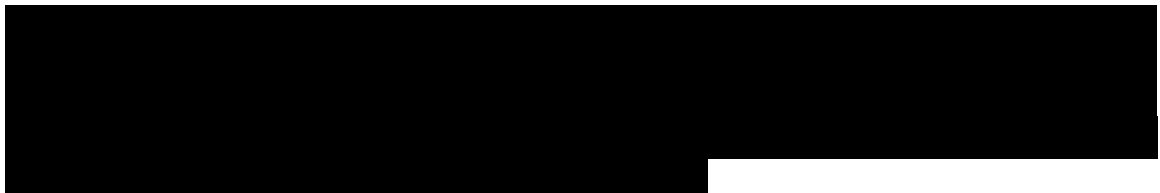
Local reactions at vaccination sites including erythema, induration, pruritis, and tenderness were observed with every vaccination in all patients, similar to those reported in patients who received GM-CSF producing tumor vaccine of other disease types. Minimal systemic toxicities were observed. The most common side-effects are transient, low grade fever, chill, nausea and headache. Fever and chill are likely attributive to the vaccine treatment; and nausea is likely attributive to the cyclophosphamide treatment. Headache may be attributive to ondansetron, which was given as an optional anti-emetic prophylactic treatment prior to the administration of cyclophosphamide. There has been no incidence of anaphylaxis secondary to the vaccine reported thus far. Grade 3 lymphopenia was transiently observed in one patient, presumably attributive to the cyclophosphamide treatment. Thus, the higher dose level comprised of  $2.5 \times 10^8$  cells per colon tumor cell lines and  $2 \times 10^8$  GM-CSF-producing K562 cells is considered to have an acceptable degree of safety. In addition, cyclophosphamide at  $200 \text{ mg/m}^2$  intravenously was well tolerated.

### **2.3 Anti-PD-1/PD-L1 therapy (Pembrolizumab (MK-3475 or Keytruda))**

The importance of intact immune surveillance in controlling outgrowth of neoplastic transformation has been known for decades [26]. Accumulating evidence shows a correlation between tumor-infiltrating lymphocytes (TILs) in cancer tissue and favorable prognosis in various malignancies. In particular, the presence of  $\text{CD8}^+$  T-cells and the ratio of  $\text{CD8}^+$  effector T-cells /  $\text{FoxP3}^+$  regulatory T-cells seems to correlate with improved prognosis and long-term survival in solid malignancies such as ovarian, colorectal and pancreatic cancer, hepatocellular carcinoma, malignant MEL and RCC. TILs can be expanded *ex vivo* and re-infused, inducing durable objective tumor responses in cancers such as melanoma [27, 28].

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. The ligands for PD-1 (PD-L1 and PD-L2) are constitutively expressed or can be induced in various tumors [29-32]. 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 [29]. Although healthy organs express little (if any) PD-L1, a variety of cancers were demonstrated to express abundant levels of this T-cell inhibitor. High expression of PD-L1 on tumor cells (and to a lesser extent of PD-L2) has been found to correlate with poor prognosis and survival in various cancer types, including renal cell carcinoma (RCC) [33], pancreatic carcinoma [34], hepatocellular carcinoma [35], ovarian carcinoma [36]. Furthermore, PD-1 has been suggested to regulate tumor-specific T-cell expansion in patients with malignant melanoma [37]. The observed correlation of clinical prognosis with PD-L1 expression in multiple cancers 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 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. Keytruda™ (pembrolizumab) has recently been approved in the United States for the treatment of patients with unresectable or metastatic non-small-cell lung cancer (NSCLC) and melanoma.



[REDACTED]

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## **2.4 Preclinical and Clinical Trial Data**

Refer to the Investigator's Brochure [IB] for Preclinical and Clinical Data

## **2.5 Rationale**

PD-1 checkpoint inhibitors have shown durable responses in a wide variety of tumors. However, in colorectal cancer, single agent pd-1 blockade with pembrolizumab has little or no activity in the majority of patients. A small subset of advanced colorectal carcinoma (<5%) is MMR-deficient (MMR-d) colorectal carcinoma. MMR-d, also referred to as microsatellite instability (MSI), is biologically marked by genomic instability with the potential of high numbers of neo-antigens and well described

increases in tumor-infiltrating lymphocytes (TILs) compared to other subtypes. In exploratory clinical trials, single agent pembrolizumab has been shown durable objective responses in many patients with MMR-d cancers. However, MMR-p colorectal cancer is the largest subtype of colorectal cancer, estimated to be between 90-95% of advanced disease. How to overcome the ineffectiveness of pembrolizumab in MMR-p colorectal cancer is therefore an important objective.

The preclinical/clinical researchers at Johns Hopkins with GVAX in pancreatic cancer and colon cancer have suggested that GVAX may prime an immune checkpoint inhibitor insensitive cancer into a sensitive one. We recently reported the results of a novel study of GVAX given as both neo-adjuvant and adjuvant therapy, either alone or with immune modulating doses of cyclophosphamide (Cytosan, Cy) to deplete regulatory T cells (Tregs) [38]. Pathological examination of PDA tumor tissue resected just two weeks following a single neoadjuvant dose of GVAX identified the formation of novel vaccine-induced, immunologically active, tertiary lymphoid aggregates, organized lymph node-like structures that are not observed in tumor tissue resected from unvaccinated patients. Our study showed for the first time that a vaccine-based immunotherapy can reprogram an immunologically quiescent tumor microenvironment (TME) into an immunologically active TME. However, activated T cells secrete interferon- $\gamma$ , which in turn upregulates the PD-1/PD-L1 pathway. These data support an emerging concept that vaccines are required to induce a T cell response that is capable of infiltrating the TME. However, vaccination is just the first step toward establishing an effective antitumor immune response, converting the TME into an environment similar to what is observed in MMR-d colon cancer exhibiting infiltrating but immunosuppressed T cells prior to immunotherapy treatment. Thus, we hypothesize that treatment with GVAX primes the TME for anti-PD-1/PD-L1-targeted therapy [39].

To support this hypothesis, we showed in a preclinical model of PDA that combining anti-PD-1 and anti-PD-L1 antibodies with GVAX+Cy enhances the infiltration of effector T cells into PDA tumors as well as the cure rate in PDA tumor-bearing mice [40]. Others have showed the synergistic effect of anti-PD-1 antibody and colon GVAX in colon tumor-bearing mice [41]. We found that GVAX treatment can also induce the membranous expression of PD-L1 in colon tumors in the mouse model. Therefore, we propose a clinical trial concept to examine the objective response rate of the combination of pembrolizumab and GVAX in MMR-p, advanced colorectal cancer.

### **3. PATIENT SELECTION**

#### **3.1 Eligibility Criteria**

3.1.1 Pathologically confirmed, mismatch repair-proficient adenocarcinoma of colorectum, who have received at least two prior lines of therapy in the metastatic setting.

3.1.1.1 Mismatch repair proficiency can be assessed for eligibility by immunohistochemistry (intact expression of MLH1, MSH2, PMS2, and MSH6) or by molecular testing in a CLIA-certified laboratory for microsatellite instability (0 or 1 microsatellites unstable)

- 3.1.2 Measurable disease by RECIST v1.1.
- 3.1.3 Age  $\geq 18$  years
- 3.1.4 ECOG performance status 0-1 (**Appendix A**)
- 3.1.5 Estimated life expectancy of greater than 3 months
- 3.1.6 Adequate hematologic, renal, and liver function as defined below:

Absolute neutrophil count	$\geq 1,500$ cells/mm <sup>3</sup>
Hemoglobin	$\geq 9$ g/dL (transfusion allowed but must demonstrate stability after transfusion)
Platelets	$\geq 75$ K/mm <sup>3</sup>
Serum creatinine	$\leq 2.0$ mg/dL
AST and ALT	$\leq 3$ x ULN
Total bilirubin	$\leq 1.5$ x ULN*

\*Subjects with Gilbert's Syndrome should have direct bilirubin within normal institutional limits

- 3.1.7 Women of childbearing potential (WOCBP) must have a negative urine or serum pregnancy test for the patient to be eligible for trial enrolment. In a case of a positive HCG test, a vaginal ultrasound must be used to confirm a lack of pregnancy.
- 3.1.8 WOCBP must be willing to use either two adequate barrier methods *or* a barrier method plus a hormonal method of contraception to prevent pregnancy, or to abstain from heterosexual activity (complete abstinence) throughout the study, starting with visit 1 through 120 days after the last dose of study therapy. Approved contraceptive methods include for example; intrauterine device, diaphragm with spermicide, cervical cap with spermicide, male condoms, female condoms with spermicide, or oral contraceptives. Spermicides alone are not an acceptable method of contraception. Non-pregnant, non-breast-feeding women may be enrolled if they are considered highly unlikely to conceive. Highly unlikely to conceive is defined as 1) surgically sterilized, or 2) postmenopausal (a woman who is  $\geq 45$  years of age and has not had menses for greater than 2 years will be considered postmenopausal), or 3) amenorrheic for  $< 2$  years without a hysterectomy and oophorectomy and with a documented FSH value in the postmenopausal range, or 4) not heterosexually active for the duration of the study, or 5) heterosexually active and willing to use 2 methods of birth control (which is also required for the female partners of male patients).  
  
Male patients must agree to use an adequate method of contraception, or to abstain from heterosexual activity (complete abstinence), starting with the first dose of study drug through 120 days after the last dose of study therapy.
- 3.1.9 Ability to understand and the willingness to sign a written informed consent



document.

- 3.2.10 Willing to undergo tumor biopsy at baseline and during treatment (during week 6 or 7). Please note that tumor biopsy is not needed in subjects where the tumor is not accessible or if tumor biopsy is considered not in patient's best interest.

## **3.2 Exclusion Criteria**

- 3.2.1 Has a known additional malignancy that is progressing or requires active treatment. Exceptions include basal cell carcinoma of the skin or squamous cell carcinoma of the skin that has undergone potentially curative therapy or in situ cervical cancer.
- 3.2.2 Has known central nervous system (CNS) metastases and/or carcinomatous meningitis.
- 3.2.3 Patients with malignant small bowel obstruction within the last 6 months, on parenteral nutrition, clinically significant ascites (palpable on physical exam and/or causing symptoms) or ascites requiring fluid removal more than twice in the last 6 weeks.
- 3.2.4 Has active autoimmune disease that has required systemic treatment in the past 2 years (i.e. with use of disease modifying agents, corticosteroids or immunosuppressive drugs). Replacement therapy (eg., thyroxine, insulin, or physiologic corticosteroid replacement therapy for adrenal or pituitary insufficiency, etc.) is not considered a form of systemic treatment.
- 3.2.5 Has a diagnosis of immunodeficiency or is receiving systemic steroid therapy or any other form of immunosuppressive therapy within 7 days prior to the first dose of trial treatment.
- 3.2.6 Has an active infection requiring systemic therapy.
- 3.2.7 Has a known history of active TB (Bacillus Tuberculosis)
- 3.2.8 Has a known history of Human Immunodeficiency Virus (HIV) (HIV 1/2 antibodies).
- 3.2.9 Has known history of Hepatitis B or Hepatitis C
- 3.2.10 Has history of (non-infectious) pneumonitis that required steroids, evidence of interstitial lung disease or active, non-infectious pneumonitis.
- 3.2.11 Must not require supplemental oxygen or have a pulse oximetry < 92% on room air.
- 3.2.12 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.

- 3.2.13 Has known psychiatric or substance abuse disorders that would interfere with cooperation with the requirements of the trial.
- 3.2.14 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. Women of childbearing potential must have a negative urine HCG (refer to **Section 3.1.7**)
- 3.2.15 Is currently participating and receiving study therapy or has participated in a study of an investigational agent and received study therapy or used an investigational device within 4 weeks of the first dose of treatment.
- 3.2.16 Has had prior chemotherapy, targeted small molecule therapy, or radiation therapy within 2 weeks prior to study Day 1 or who has not recovered (i.e.,  $\leq$  Grade 1 or at baseline) from adverse events due to a previously administered agent.

Note: Subjects with  $\leq$  Grade 2 neuropathy are an exception to this criterion and may qualify for the study.

- 3.2.17 Patients who have had surgery within 4 weeks of dosing of investigational agent, excluding minor procedures (dental work, skin biopsy, etc), celiac plexus block, and biliary stent placement.
- 3.2.18 Has had a prior anti-cancer monoclonal antibody (mAb) within 4 weeks prior to study Day 1 or who has not recovered (i.e.,  $\leq$  Grade 1 or at baseline) from adverse events due to agents administered more than 4 weeks earlier.
- 3.2.19 Has received a live vaccine within 30 days of planned start of study therapy.

Note: Seasonal influenza vaccines for injection are generally inactivated flu vaccines and are allowed; however intranasal influenza vaccines (e.g., Flu-Mist®) are live attenuated vaccines, and are not allowed.

- 3.2.20 Has received prior therapy with any other systemic immunotherapy treatment, including but not limited to: IL-2, interferon, anti-PD-1 antibodies, anti-PD-L2 antibodies, anti-CD137 antibodies, anti-OX-40 antibodies, anti-CD40 antibodies, anti-CTLA-4 antibodies, therapeutic anticancer vaccines, cellular immunotherapies including chimeric antigen receptor–modified T cells, or bi-specific CD3 antibodies.
- 3.2.21 Patients receiving growth factors including, but not limited to, granulocyte-colony stimulating factor (G-CSF), GM-CSF, erythropoietin, within 14 days of study drug administration. Use of such agents while on study is also prohibited.

- 3.2.22 Hypersensitivity to pembrolizumab or any of its excipients.
- 3.2.23 Patient has a known or suspected hypersensitivity to GM-CSF, hetastarch, corn, dimethyl sulfoxide, fetal bovine serum, trypsin (porcine origin), yeast or any other component of GVAX vaccine.
- 3.2.24 Patient is unwilling or unable to follow the study schedule for any reason.
- 3.2.25 Presence of any tissue or organ allograft, regardless of need for immunosuppression, including corneal allograft. Patients with a history of allogeneic hematopoietic stem cell transplant will also be excluded.

### 3.3 Inclusion of Women and Minorities

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

## 4. TREATMENT PLAN

### 4.1 Agent Administration

Treatment will be administered on an outpatient basis. Reported adverse events and potential risks are described in **Section 6**. Appropriate dose modifications are described in **Section 5**. No investigational or commercial agents or therapies other than those described below may be administered with the intent to treat the patient's malignancy.

**Table 1: Study Regimen**

<i>Agent</i>	<i>Premedications, Precautions</i>	<i>Dose</i>	<i>Route</i>
Cyclophosphamide	Subjects may be pre-medicated with anti-emetics	200 mg/m <sup>2</sup> in 100 mL NS	IV infusion over 30 minutes
GVAX	Lidocaine-based topical anesthetic (approximately 2.5 grams per site, at least 1 hour prior to vaccination)	5x10 <sup>8</sup> (colon cancer cells) + 5x10 <sup>7</sup> (GM-CSF secreting cells)	8-9 Intradermal injections
Pembrolizumab	No prophylactic pre-medications unless indicated by previous experience in an individual subject	200 mg	IV over 30 minutes

Pembrolizumab should be administered after cyclophosphamide, with a minimum of 30 min wait time between infusions. Infusion times are approximate (+/- 10 min) and may need to be adjusted based on subject tolerability.

Please see **Section 5.2** for guidance regarding dosing delays.

**4.1.1** *GVAX* GVAX is administered through a total of 8-9 intradermal injections. Because the spatial distribution of vaccine cells among three or more lymph node regions primes a more potent anti-tumor immune response distributed equally among the right and left thighs, and the non-dominant arm. In the event that the specified limb is contraindicated, the dominant arm may be used.

No pre-medications will be administered except a lidocaine-based topical anesthetic (may include but is not limited to EMLA or ELA-MAX) cream, which will be applied to the injection site at least 1 hour prior to vaccination to diminish the discomfort associated with intradermal injections. The patient must be observed in the clinic for at least 60 minutes after the first vaccine and 30 minutes for subsequent vaccinations. Acute reactions will be managed using standard therapy for acute drug reactions as per institutional standard of care and reported to the sponsor.

**4.1.2** *Cyclophosphamide (CY, Cytoxan<sup>R</sup>)* Cy is a FDA-approved and widely used chemotherapy agent. Subjects may be pre-medicated prior to administration with anti-emetics per institutional guidelines. Acute reactions will be managed using standard therapy for acute drug reactions as per institutional standard of care and reported to the sponsor.

**4.1.3** *Pembrolizumab* (KEYTRUDA®, MK-3475) 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 FDA-approved for treating metastatic melanoma and metastatic PDL1 positive non-small cell lung cancer. Pembrolizumab will be administered IV over 30 minutes at 200 mg. Antiemetic medications should not be routinely administered prior to dosing except as indicated by patient's prior reaction. Acute reactions will be managed as described in Section 4.2.3.1 below.

## **4.2 General Concomitant Medication and Supportive Care Guidelines**

In general, concomitant medications and therapies deemed necessary for the supportive care and safety of the subject are allowed, provided their use is documented in the subject records and on the appropriate case report form. If toxicity occurs, the appropriate treatment will be used to ameliorate signs and symptoms (including antiemetics for nausea and vomiting, anti-diarrheals for diarrhea and anti-pyretics and anti-histamines for drug fever). All supportive measures for optimal medical care will be given during the period of study.

### **4.2.1 Cyclophosphamide (Cy)**

Cyclophosphamide is commonly used in chemotherapy to treat a variety of cancer diseases. Although cyclophosphamide is administered at low doses, subjects may still experience the side effects below, but generally the side

effects tend to be milder than standard dose cyclophosphamide. Side effects that have reported with cyclophosphamide, when it is given at a standard dose, include:

- **Common toxicities:** leucopenia/neutropenia, low appetite, nausea, vomiting, fatigue, and hair loss.
- **Less common toxicities:** hyponatremia, early menopause, hematuria, and pain due to inflammation in the bladder, liver, lungs, or veins
- **Rare but clinical important toxicities:** hives, blurry vision, numbness and tingling of the mouth and throat, pulmonary fibrosis. Treatment with Cytoxan can also increase the risk of other cancers (bladder and skin cancers, leukemia).

Acute reactions will be managed using standard therapy for acute drug reactions as per institutional guidelines

#### 4.2.2 GVAX





Local skin reactions at vaccine sites may be treated with cold packs, topical lotions (e.g. aloe vera or vitamin E). Pruritus can be managed with topical or systemic benadryl. Significant local inflammation leading to severe pain may be treated with oral analgesics. Local ulceration should be managed with local wound care, with or without antibiotics, and should be evaluated on a case-by-case basis.

#### **4.2.3 Pembrolizumab (KEYTRUDA®, MK-3475)**

Pembrolizumab is a humanized monoclonal Ab. Subjects should be closely monitored for potential adverse reactions during antibody infusion and potential adverse events throughout the study.

Unexpected Grade 3 or greater laboratory abnormalities should be repeated within 24-72 hours if clinically indicated and monitored as necessary to determine if event meets toxicity criteria.

##### **4.2.3.1 Infusion Reactions**

Pembrolizumab infusion reactions may consist of fever, chills/rigor, nausea, pruritus, angioedema, hypotension, headache, bronchospasm, urticaria, rash, vomiting, myalgia, dizziness or hypertension. Severe reactions may include acute respiratory distress syndrome, myocardial infarction, ventricular fibrillation, and cardiogenic shock. Patients should be closely monitored for such reactions. Guidelines for patients who experience an infusion related or allergic reaction during or after infusion with pembrolizumab are shown below.

**Table 2: Guidance on Infusion and Hypersensitivity Reactions**

NCI CTCAE Grade	Treatment	Premedication at subsequent dosing
<u>Grade 1</u> 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
<u>Grade 2</u> Requires infusion interruption but responds promptly to symptomatic treatment (e.g., antihistamines, NSAIDs, narcotics, IV fluids); prophylactic medications indicated for <=24 hrs	<b>Stop Infusion and monitor symptoms.</b> Additional appropriate medical therapy may include but is not limited to: IV fluids Antihistamines NSAIDs Acetaminophen Narcotics Increase monitoring of vital signs as medically indicated until the subject is deemed medically stable in the opinion of the investigator. 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. <b>Subjects who develop Grade 2 toxicity despite adequate premedication should be permanently discontinued from further trial treatment administration.</b>	Subject may be premedicated 1.5h (± 30 minutes) prior to infusion of pembrolizumab (MK-3475) with:  Diphenhydramine 50 mg po (or equivalent dose of antihistamine).  Acetaminophen 500-1000 mg po (or equivalent dose of antipyretic).
<u>Grades 3 or 4</u> Grade 3: 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) Grade 4: Life-threatening; pressor or ventilatory support indicated	<b>Stop Infusion.</b> Additional appropriate medical therapy may include but is not limited to: IV fluids Antihistamines NSAIDs Acetaminophen Narcotics Oxygen Pressors Corticosteroids Epinephrine  Increase monitoring of vital signs as medically indicated until the subject is deemed medically stable in the opinion of the investigator. Hospitalization may be indicated. <b>Subject is permanently discontinued from further trial treatment administration.</b>	No subsequent dosing
Appropriate resuscitation equipment should be available in the room and a physician readily available during the period of drug administration.		

#### 4.2.3.2 Immune-Related Adverse Events (IRAEs)

Blocking PD-1 function may permit the emergence of auto-reactive T cells and resultant clinical autoimmunity. Rash/pruritis, diarrhea/colitis, pneumonitis, hepatitis, and hypothyroidism were drug-related, presumptive autoimmune events, now termed IRAEs.

For the purposes of this study, an IRAE is defined as an AE of unknown etiology, associated with drug exposure and is consistent with an immune phenomenon. Efforts should be made to rule out neoplastic, infectious, metabolic, toxin or other etiologic causes prior to labeling an AE an IRAE. Serological, immunological, and histological (biopsy) data should be used to support the diagnosis of an immune-mediated toxicity. Suspected IRAEs must

be documented on an AE or SAE CRF. Identification and treatment of IRAEs can be found in **Appendix B**.

### 4.3 Prohibited and Restricted Therapies

Patients may not use any of the following agents during the study:

- Any non-study anticancer or immunotherapy treatments (investigational or non-investigational)
- Any other cancer immunotherapy treatments, including but not limited to: IL-2, interferon, anti-PD-1 antibodies, anti-PD-L1 antibodies, anti-PD-L2 antibodies, anti-CD137 antibodies, anti-OX-40 antibodies, anti-CD40 antibodies, anti-CTLA-4 antibodies, therapeutic anticancer vaccines, cellular immunotherapies including chimeric antigen receptor–modified T cells, or bi-specific CD3 antibodies.
- Systemically active steroids can be used but should be reported to the Principal Investigator and IND Sponsor. Steroid treatment should be titrated down to physiologic dosing prior to resuming study-related treatments (See **Section 5.2** for dosing delays for steroids)
- Filgrastim (Neupogen® or G-CSF) or sargramostim (Leukine® or GM-CSF)
- Prophylactic vaccines (e.g., influenza, pneumococcal, Td/Tdap) within 28 days prior to or after dosing combination immunotherapy
- Mistletoe

### 4.4 Dosing Criteria

Dosing will be delayed for the following laboratory criteria:

- AST, ALT > 3.0 x ULN
- Total bilirubin >2.0 (patients with diagnosed Gilbert’s Syndrome, direct bilirubin should be within normal institutional limits)
- Hemoglobin < 8 g/dL
- ANC < 1000/uL
- Platelets < 75 x 10<sup>3</sup>/uL

### 4.5 Definition of an Overdose for this Protocol

Overdose of pembrolizumab is defined as:

The patient has taken (accidentally or intentionally) a dose of pembrolizumab of ≥1000mg (≥5 times the indicated dose). No specific information is available on the treatment of overdose of pembrolizumab. In the event of overdose, pembrolizumab should be discontinued and the subject should be observed closely for signs of toxicity. Appropriate supportive treatment should be provided if clinically indicated.



If an adverse experience(s) is associated with (“results from”) the overdose of test drug or vaccine, the adverse experience(s) is reported as a serious adverse experience, even if no other criteria for serious are met.

If a dose of test drug or vaccine 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 experience must be reported within 24 hours to the Sponsor and Merck Global Safety. Merck Global Safety (GS) contact information can be found in **Section 6.5.1**.

## **4.6 Contraception, Use in Pregnancy, Use in Nursing**

### 4.6.1 Contraception

The investigational agents may have adverse effects on a fetus *in utero*. Furthermore, it is not known if the investigational agents have transient adverse effects on the composition of sperm. Non-pregnant, non-breast-feeding women may be enrolled if they are considered highly unlikely to conceive. Highly unlikely to conceive is defined as 1) surgically sterilized, or 2) postmenopausal (a woman who is  $\geq 45$  years of age and has not had menses for greater than 2 years will be considered postmenopausal), or 3) amenorrheic for  $< 2$  years without a hysterectomy and oophorectomy and with a documented FSH value in the postmenopausal range, or 4) not heterosexually active for the duration of the study, or 5) heterosexually active and willing to use 2 methods of birth control (which is also required for the female partners of male patients). The 2 birth control methods can be 2 barrier methods *or* a barrier method plus a hormonal method to prevent pregnancy, used throughout the study starting with Visit 1 through 120 days after the last dose of study medication. Male patients enrolled in this study must also agree to use an adequate method of contraception starting with Visit 1 through 120 days after the last dose of study drug.

The following are considered adequate barrier methods of contraception: diaphragm, condom (by the partner), copper intrauterine device, sponge, or spermicide. Appropriate hormonal contraceptives will include any registered and marketed contraceptive agent that contains an estrogen and/or a progestational agent (including oral, subcutaneous, intrauterine, or intramuscular agents).

Patients 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, they must adhere to the contraception requirement (described above) for the duration of the study. If there is any question that a patient will not reliably comply with the requirements for contraception, that patient should not be entered into the study.

### 4.6.2 Use in Pregnancy

The investigational agents may have adverse effects on a fetus; therefore, women with a positive pregnancy test at screening will not be eligible for enrollment. If a patient inadvertently becomes pregnant while on treatment with combination immunotherapy, the patient will immediately be removed from the study. The site will contact the patient at least monthly and document the patient's status until the pregnancy has been completed or terminated. The outcome of the pregnancy will be reported to the Sponsor and to Merck Global Safety without delay. The outcome must be reported to the Sponsor within 24 hours and to Merck Global Safety if the outcome is a serious adverse experience (e.g., death, abortion, congenital anomaly, or other disabling or life-threatening complication to the mother or newborn). If a male patient's partner becomes pregnant on study the pregnancy must be reported to the Sponsor and to Merck Global Safety as described in **Section 6.5.1**. The study Investigator will make every effort to obtain permission to follow the outcome of the pregnancy and report the condition of the fetus or newborn to the Sponsor and to Merck Global Safety.

#### 4.6.3 Use in Nursing Women

It is unknown whether the investigational agents are excreted in human milk. Since many drugs are excreted in human milk, and because of the potential for serious adverse reactions in the nursing infant, patients who are breast-feeding are not eligible for enrollment.

### 4.7 Dose Limiting Toxicity

Unless otherwise specified below, unacceptable toxicities are defined as:

- Treatment-related  $\geq$  grade 4 AEs, or
- Treatment-related grade 3 AEs that do not improve to  $\leq$  grade 2 under supportive therapy within 3 weeks.
- Death within 30 days of receiving investigational agent that is considered related to the agent as a study stopping criterion

#### Hematologic toxicity

The only adverse hematologic events that will be considered dose limiting are:

- Grade 4 anemia related to study therapy
- Grade 4 neutropenia that lasts  $>$  than 7 days or febrile neutropenia
- Grade 4 thrombocytopenia that lasts greater than 7 days or has clinically significant bleeding

#### Electrolyte aberrations

- Unexpected Grade 3 or greater laboratory abnormalities should be repeated within 24-72 hours if clinically indicated and monitored as necessary to determine if event meets toxicity criteria.
- Patients may continue therapy without interruption for any electrolyte abnormality that can be corrected with intervention to  $<$  grade 2.

#### Diarrhea/Colitis

Grade 1	Asymptomatic pathologic or radiographic findings only	Consider endoscopy to confirm
Grade 2	Abdominal pain; mucus or blood in stool	Endoscopy is indicated to confirm the etiology of the patient's symptoms. Hold therapy, start lomotil and budesonide 9mg daily and start systemic steroids. Therapy may be resumed only when patient returns to $\leq$ grade 1. If symptoms are not controlled within 7 days of initiating budesonide, contact Dr. Azad or Dr. Yarchoan.
Grade 3	Abdominal pain, fever, change in bowel habits with ileus; peritoneal signs	Endoscopy is indicated to confirm the etiology of the patient's symptoms. Hold therapy, start lomotil and budesonide 9mg daily and start systemic steroids. Continue steroids until symptoms improve to grade 1, then taper. Therapy may be resumed only when patient returns to $\leq$ grade 1. If symptoms are not controlled within 7 days of initiating steroids, contact Dr. Azad or Dr. Yarchoan. Patients should be taken off study if symptoms are not controlled within 14 days.
Grade 4	Life-threatening consequences (perforation, bleeding, ischemia, necrosis, toxic megacolon)	Permanently discontinue therapy

#### Hepatitis

Grade 1	ALT/AST $>$ ULN – 2.5 x ULN; Bilirubin $>$ ULN – 1.5 x ULN	Maintain treatment, but increase frequency of liver function testing
Grade 2	ALT/AST $>$ 2.5 – 5.0 x ULN; Bilirubin $>$ 1/5 – 3.0 x ULN	Maintain treatment, increase frequency of liver function testing, consider systemic steroids until liver function tests return to $\leq$ grade 1.
Grade 3	ALT/AST $>$ 5.0 – 20.0 x ULN; Bilirubin $>$ 3.0 – 10.0 x ULN	Permanently discontinue therapy. Hepatology consultation is indicated to confirm the etiology of the patient's symptoms. Start systemic steroids. Rule out infectious causes of hepatitis.
Grade 4	AST/ALT 20.0 x ULN; Bilirubin $>$ 10.0 x ULN	Permanently discontinue therapy.

For patients with liver metastasis who begin treatment with Grade 2 AST or ALT, if AST or ALT increases by greater than or equal to 50% relative to baseline and lasts for at least 1 week then patients should be discontinued.

Pneumonitis

Grade 1	Asymptomatic; clinical or diagnostic observations only; intervention not needed	Hold MK-3475 but may continue Cy/GVAX. Monitor symptoms every 2-3 days. Consider pulmonary consult and/or systemic steroids. May resume MK-3475 when symptoms near baseline.
Grade 2	Symptomatic; medical intervention indicated limited instrumental ADLs	Hold MK-3475 but may continue Cy/GVAX. Monitor symptoms every 2-3 days. Obtain pulmonary consult and initiate systemic steroids (1/2 mg/kg/day prednisone or equivalent). Consider hospitalization and bronchoscopy. Hold MK-3475 and continue systemic steroids until symptoms near baseline. If symptoms persist for 3 weeks or re-develop upon re-challenge, permanently discontinue MK-3475.
Grade 3	Severe symptoms; limiting self-care ADL; oxygen indicated	Permanently discontinue therapy and admit to the hospital. Obtain pulmonary and ID consults, and treat with systemic steroids (1/2 mg/kg/day prednisone or equivalent) and prophylactic antibiotics. Recommend bronchoscopy and consideration of lung biopsy. Consider addition of additional immunosuppressant (eg anti-tumour necrosis factor alpha, IVIG, etc.)
Grade 4	Life-threatening respiratory compromise; urgent intervention indicated (e.g. tracheostomy or intubation).	

Toxicity	Hold Treatment For Grade	Timing for Restarting Treatment	Treatment Discontinuation
Type 1 diabetes mellitus (if new onset) or Hyperglycemia	T1DM or 3-4	Hold therapy for new onset Type 1 diabetes mellitus or Grade 3-4 hyperglycemia associated with evidence of beta cell failure.	May resume therapy when patients are clinically and metabolically stable.

Toxicity	Hold Treatment For Grade	Timing for Restarting Treatment	Treatment Discontinuation
Hypophysitis	2-4	Toxicity resolves to Grade 0-1. Therapy can be continued while endocrine replacement therapy is instituted	Toxicity does not resolve within 12 weeks of last dose or inability to reduce corticosteroid to 10 mg or less of prednisone or equivalent per day within 12 weeks.
Hyperthyroidism	3	Toxicity resolves to Grade 0-1	Toxicity does not resolve within 12 weeks of last dose or inability to reduce corticosteroid to 10 mg or less of prednisone or equivalent per day within 12 weeks.
	4	Permanently discontinue	Permanently discontinue
Hypothyroidism		Therapy can be continued while thyroid replacement therapy is instituted	Therapy can be continued while thyroid replacement therapy is instituted.
Infusion Reaction	3-4	Permanently discontinue	Permanently discontinue
Renal Failure or Nephritis	2	Toxicity resolves to Grade 0-1	Toxicity does not resolve within 12 weeks of last dose or inability to reduce corticosteroid to 10 mg or less of prednisone or equivalent per day within 12 weeks.
	3-4	Permanently discontinue	Permanently discontinue
All Other Drug-Related Toxicity <sup>2</sup>	3 or Severe	Toxicity resolves to Grade 0-1	Toxicity does not resolve within 12 weeks of last dose or inability to reduce corticosteroid to 10 mg or less of prednisone or equivalent per day within 12 weeks.
	4	Permanently discontinue	Permanently discontinue

The proportion of unacceptable toxicities will be monitored. If the toxicity levels are unacceptable (high probability that it is >45% of subjects), then enrollment will be suspended until further review and consideration by the IND Sponsor and MEC.

#### 4.8 Duration of Therapy

In the absence of treatment delays due to adverse event(s), treatment may continue indefinitely until one of the following criteria applies:

- Continued radiographic evidence of disease progression on two subsequent imaging scans 6 weeks apart,
- Clinical progression or deterioration per **Section 4.10**,
- Intercurrent illness that prevents further administration of treatment,
- Unacceptable adverse event(s) as described in **Section 4.7**,
- Patient decides to withdraw from the study, or
- General or specific changes in the patient's condition render the patient unacceptable for further treatment in the judgment of the investigator, or
- Achieved a complete response to therapy, and received 6 weeks of therapy beyond the date when the initial complete response was declared without evidence of disease recurrence, or
- Completed 24 months of study medication or 35 administrations of Cy/GVAX/pembrolizumab, whichever is later. Note: 24 months of study medication is calculated from the date of first dose. Subjects who stop therapy after 24 months may be eligible for up to one year of additional study treatment if they progress after stopping study treatment provided they meet the initial study requirements. See **Section 4.9** below.

#### **4.9 Off Treatment Evaluation and Retreatment Criteria**

If a patient is discontinued from study medication after completion of 24 months of treatment, a mandatory Off Treatment Evaluation should be performed 30 days (+/- 7 days) after the last treatment as described in the Study Calendar (**Section 9**). Patients should continue to be monitored for disease status by radiologic imaging and tumor marker(s) every 12 weeks (+/- 2 weeks). In addition, we will also continue to collect peripheral blood, serum, and plasma samples every 12 weeks (+/- 2 weeks) as described in the Study Calendar (**Section 9**). Patients will also be monitored for AEs up to the Off Treatment Evaluation or to resolution of drug-related AEs to  $\leq$  Grade 1, whichever occurs later. SAEs that occur within 90 days of the last treatment or before initiation of a new antineoplastic treatment should also be followed and recorded.

##### Retreatment Criteria

Patients who stop therapy with SD or better may be eligible for up to one year of additional therapy with Cy/GVAX/pembrolizumab if they progress after stopping therapy and they meet the following criteria:

EITHER:

- Stopped initial treatment with Cy/GVAX/pembrolizumab after attaining an investigator determined confirmed CR according to RECIST 1.1
  - Was treated for at least 24 weeks with Cy/GVAX/pembrolizumab before

- discontinuing therapy
- Received at least two treatments with Cy/GVAX/pembrolizumab beyond the date when the initial CR was declared

OR:

- Subject had SD, PR or CR and stopped Cy/GVAX/pembrolizumab treatment after 24 months of study therapy for reasons other than disease progression or intolerability

AND:

- Experienced an investigator-determined confirmed radiographic disease progression after stopping their initial treatment with Cy/GVAX/pembrolizumab
- Did not receive any anti-cancer treatment since the last dose of Cy/GVAX/pembrolizumab
- Meets the initial study requirements as detailed in **Section 3.1**
- Female subject of childbearing potential should have a negative urine or serum pregnancy test within 72 hours prior to receiving retreatment with study medication.

Subjects who restart treatment should resume at the same dose and cycle interval which they were receiving prior to discontinuation. Study visit requirements are outlined in the Study Calendar (**Section 9**).

#### **4.10 Duration of Follow Up**

Patients will be followed for 30 days after their last dose of study drug or until death, whichever occurs first. SAEs that occur within 90 days of the last infusion of MK-3475 or before initiation of a new antineoplastic treatment should also be followed and recorded. Patients removed from study for unacceptable adverse event(s) will be followed until resolution or stabilization of the adverse event.

Patients who discontinue from treatment should be contacted every three months to monitor overall survival. Information of other cancer therapies after discontinuation from the study treatment will be collected.

At the end of the study, subjects will be required to enroll in a long-term follow-up protocol, an FDA requirement for genetically modified products, in which they will be followed for at least 5 years (or until death) after receiving the last dose of any investigational agent for disease progression, survival, and potential long term toxicity of gene therapy. The subjects will be asked to enroll in the study entitled, “Long-Term Follow-Up of Patients who Received the Allogeneic Colon Cancer Cell Vaccine Administered with a GM-CSF Producing Bystander Cell Line.”

#### **4.11 Criteria for Removal from Study Treatment**

Patients will be removed from study treatment when any of the criteria listed in **Section 4.8** applies. The reason for study removal and the date the patient was removed must be documented in the Case Report Form.

Patients with disease progression by radiographic imaging or laboratory parameters during the 12-week evaluation but without rapid clinical deterioration or significant change in performance status that requires additional immediate therapy may continue to receive treatment on study. Subjects that meet the above criteria and continue on study therapy must discontinue treatment upon documentation of disease progression on the second scan six weeks later, with the date of progression being backdated to the first RECIST 1.1 progression time point.

NOTE: For patients who progress and remain on study, at the time of the next restaging, stable disease with clinical stability as determined by their treating physician/team can remain on study. Specifically, these patients do not have to have regression to <20% growth from baseline. If they do not have 20% increase from the last restaging scan, they may remain on study.

## **5. DOSING DELAYS/DOSE MODIFICATIONS**

### **5.1 Dose Modifications**

Dose reduction or dose increase of CY, GVAX, and pembrolizumab will not be permitted in individual patients.

### **5.2 Dosing Delays**

5.2.1 All scheduled cycles within a course are to be given approximately 3 weeks apart. If necessary, a cycle may be delayed for up to 1 week. In this case, subsequent cycles should continue so that a subject can still receive all cycles given that the cycles are a minimum of 3 weeks apart and they have not experienced an AE necessitating discontinuation. If delayed more than 1 week, the Principal Investigator must be contacted for further instructions on continued treatment. Additional delays or modifications to the treatment schedule must be approved by the Principal Investigator or IND Sponsor.

If a delay occurs between Day 1 and 2 in a cycle:

- Pembrolizumab-related infusion reactions must resolve to baseline prior to administration of GVAX.
- Resume Day 2 treatment and assessment schedule without repeating Day 1 study treatments if the delay is within 72 hours.
- If the delay is longer than 72 hours, repeat Day 1 and Day 2 (if applicable) study treatments/assessments with a minimum of 1 week from the previous Day 1 treatment. This includes steroid treatment requiring at least a 14 day washout prior to resuming study-related treatments.

Pembrolizumab will be withheld for drug-related Grade 4 hematologic toxicities, non-hematological toxicity  $\geq$  Grade 3 including laboratory



abnormalities, and severe or life-threatening AEs as per **Table 5**.

**Table 5: Pembrolizumab Dose Delay and Discontinuation Criteria**

<b>General instructions:</b>				
<ol style="list-style-type: none"> <li>1. Corticosteroid taper should be initiated upon AE improving to Grade 1 or less and continue to taper over at least 4 weeks.</li> <li>2. For situations where pembrolizumab has been withheld, pembrolizumab can be resumed after AE has been reduced to Grade 1 or 0 and corticosteroid has been tapered. Pembrolizumab should be permanently discontinued if AE does not resolve within 12 weeks of last dose or corticosteroids cannot be reduced to <math>\leq 10</math> mg prednisone or equivalent per day within 12 weeks.</li> <li>3. For severe and life-threatening irAEs, IV corticosteroid should be initiated first followed by oral steroid. Other immunosuppressive treatment should be initiated if irAEs cannot be controlled by corticosteroids.</li> </ol>				
<b>Immune-related AEs</b>	<b>Toxicity grade or conditions (CTCAEv4.0)</b>	<b>Action taken to pembrolizumab</b>	<b>irAE management with corticosteroid and/or other therapies</b>	<b>Monitor and follow-up</b>
Pneumonitis	Grade 2	Withhold	<ul style="list-style-type: none"> <li>• Administer corticosteroids (initial dose of 1-2 mg/kg prednisone or equivalent) followed by taper</li> </ul>	<ul style="list-style-type: none"> <li>• Monitor participants for signs and symptoms of pneumonitis</li> <li>• Evaluate participants with suspected pneumonitis with radiographic imaging and initiate corticosteroid treatment</li> <li>• Add prophylactic antibiotics for opportunistic infections</li> </ul>
	Grade 3 or 4, or recurrent Grade 2	Permanently discontinue		
Diarrhea / Colitis	Grade 2 or 3	Withhold	<ul style="list-style-type: none"> <li>• Administer corticosteroids (initial dose of 1-2 mg/kg prednisone or equivalent) followed by taper</li> </ul>	<ul style="list-style-type: none"> <li>• Monitor participants for signs and symptoms of enterocolitis (ie, diarrhea, abdominal pain, blood or mucus in stool with or without fever) and of bowel perforation (ie, peritoneal signs and ileus).</li> <li>• Participants with <math>\geq</math> Grade 2 diarrhea suspecting colitis should consider GI consultation and performing endoscopy to rule out colitis.</li> <li>• 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.</li> </ul>
	Grade 4	Permanently discontinue		
AST / ALT elevation or Increased bilirubin	Grade 2	Withhold	<ul style="list-style-type: none"> <li>• Administer corticosteroids (initial dose of 0.5- 1 mg/kg prednisone or equivalent) followed by taper</li> <li>• Administer corticosteroids (initial dose of 1-2 mg/kg prednisone or equivalent) followed by taper</li> </ul>	<ul style="list-style-type: none"> <li>• Monitor with liver function tests (consider weekly or more frequently until liver enzyme value returned to baseline or is stable)</li> </ul>
	Grade 3 or 4	Permanently discontinue		

Type 1 diabetes mellitus (T1DM) or Hyperglycemia	Newly onset T1DM or Grade 3 or 4 hyperglycemia associated with evidence of $\beta$ -cell failure	Withhold	<ul style="list-style-type: none"> <li>Initiate insulin replacement therapy for participants with T1DM</li> <li>Administer anti-hyperglycemic in participants with hyperglycemia</li> </ul>	<ul style="list-style-type: none"> <li>Monitor participants for hyperglycemia or other signs and symptoms of diabetes.</li> </ul>
Hypophysitis	Grade 2	Withhold	<ul style="list-style-type: none"> <li>Administer corticosteroids and initiate hormonal replacements as clinically indicated.</li> </ul>	<ul style="list-style-type: none"> <li>Monitor for signs and symptoms of hypophysitis (including hypopituitarism and adrenal insufficiency)</li> </ul>
	Grade 3 or 4	Withhold or permanently discontinue <sup>1</sup>		
Hyperthyroidism	Grade 2	Continue	<ul style="list-style-type: none"> <li>Treat with non-selective beta-blockers (eg, propranolol) or thionamides as appropriate</li> </ul>	<ul style="list-style-type: none"> <li>Monitor for signs and symptoms of thyroid disorders.</li> </ul>
	Grade 3 or 4	Withhold or permanently discontinue <sup>1</sup>		
Hypothyroidism	Grade 2-4	Continue	<ul style="list-style-type: none"> <li>Initiate thyroid replacement hormones (eg, levothyroxine or liothyronine) per standard of care</li> </ul>	<ul style="list-style-type: none"> <li>Monitor for signs and symptoms of thyroid disorders.</li> </ul>
Nephritis and Renal dysfunction	Grade 2	Withhold	<ul style="list-style-type: none"> <li>Administer corticosteroids (prednisone 1-2 mg/kg or equivalent) followed by taper.</li> </ul>	<ul style="list-style-type: none"> <li>Monitor changes of renal function</li> </ul>
	Grade 3 or 4	Permanently discontinue		
Myocarditis	Grade 1 or 2	Withhold	<ul style="list-style-type: none"> <li>Based on severity of AE administer corticosteroids</li> </ul>	<ul style="list-style-type: none"> <li>Ensure adequate evaluation to confirm etiology and/or exclude other causes</li> </ul>
	Grade 3 or 4	Permanently discontinue		
All other immune-related AEs	Intolerable/persistent Grade 2	Withhold	<ul style="list-style-type: none"> <li>Based on type and severity of AE administer corticosteroids</li> </ul>	<ul style="list-style-type: none"> <li>Ensure adequate evaluation to confirm etiology and/or exclude other causes</li> </ul>
	Grade 3	Withhold or discontinue based on the type of event. Events that require discontinuation include and not limited to: Guillain-Barre Syndrome, encephalitis		
	Grade 4 or recurrent Grade 3	Permanently discontinue		

1. Withhold or permanently discontinue pembrolizumab is at the discretion of the investigator or treating physician.

**NOTE:**

For participants with Grade 3 or 4 immune-related endocrinopathy where withhold of pembrolizumab is required, pembrolizumab may be resumed when AE resolves to  $\leq$  Grade 2 and is controlled with hormonal replacement therapy or achieved metabolic control (in case of T1DM).

In case toxicity does not resolve to Grade 0-1 within 12 weeks after last infusion, trial treatment should be discontinued after consultation with the Sponsor. With Sponsor and Merck agreement, subjects with a laboratory adverse event still at Grade 2 after 12 weeks may continue treatment in the trial only if asymptomatic and controlled.

In the case that a participant develops intolerance or side effect that is attributable to one agent (for example, intolerance only to Cy/GVAX or intolerance only to pembrolizumab), the participant may, at his or her discretion, continue to receive the other active agent on study as otherwise indicated as long as they have received at least 1 full dose of both Cy/GVAX and pembrolizumab.

## 6. ADVERSE EVENTS: LIST AND REPORTING REQUIREMENTS

This study will use the descriptions and grading scales found in the revised National Cancer Institute Common Terminology Criteria for Adverse Events (CTCAE) Active version for adverse event reporting that can be found at [http://ctep.cancer.gov/protocolDevelopment/electronic\\_applications/ctc.htm](http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm).

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, recorded, and followed as appropriate.

The PI has the primary responsibility for continuous internal monitoring for safety, protocol compliance, and identification, grading, coding, and required reporting of all anticipated and unanticipated adverse events and protocol problems. Although this responsibility is usually shared among the PI, research nurse, and data manager, the PI is ultimately responsible for grading and attribution of all events.

All adverse events experienced by subjects will be collected and reported from the first dose of the investigational agent, throughout the study, and will only be followed for 90 days unless related to the investigational agent. All Serious Adverse Events (SAEs) will be collected for 90 days after the end of treatment.

Subjects who have an ongoing adverse event related to the study procedures and/or medication(s) may continue to be periodically contacted by a member of the study staff until the event is resolved or determined to be irreversible by the investigator.

Patients who experience a Grade 2 or higher irAE should be discussed with the IND Sponsor. In addition, irAEs listed in **Section 6.1.3** must be reported as an Event of Clinical Interest (ECI) within 24 hours to the Sponsor and to Merck Global Safety even if no Serious Adverse Event Criteria are met.

**Laboratory abnormalities:** Laboratory abnormalities present at the screening visit will be recorded as pre-treatment signs and symptoms. After study treatment administration, all grade 3 and 4 clinical laboratory results that represent an increase in severity from baseline will be reported as adverse events. A grade 1 or 2 clinical laboratory abnormality should be reported as an adverse event only if it is considered clinically significant by the investigator.

## 6.1 Definitions

### 6.1.1 Adverse Event (AE)

Adverse event is defined as any undesirable sign, symptom or medical condition occurring after starting the study drug (or therapy) even if the event is not considered to be related to the study. An undesirable medical condition can be symptoms (e.g., nausea, chest pain), signs (e.g., tachycardia, enlarged liver) or the abnormal results of an investigation (e.g., laboratory findings, electrocardiogram). Medical conditions/diseases present before starting the study treatment are only considered adverse events if they worsen after starting the study treatment (any procedures specified in the protocol). Adverse events occurring before starting the study treatment but after signing the informed consent form will not be recorded. Abnormal laboratory values or test results constitute adverse events only if they induce clinical signs or symptoms or require therapy.

### 6.1.2 Serious Adverse Event (SAE)

A serious adverse event is an undesirable sign, symptom or medical condition which:

- Results in death
- Is life threatening (defined as an event in which the subject was at risk of death at the time of the event; it does not refer to an event which hypothetically might have caused death if it were more severe)
- Requires inpatient hospitalization or causes prolongation of existing hospitalization (see note below for exceptions) > 24 hours
- Results in persistent or significant disability/incapacity
- Is a congenital anomaly/birth defect (note: reports of congenital anomalies/birth defects must also be reported on the Pregnancy Supplemental Form)
- Is an important medical event (defined as a medical event(s) that may not be immediately life-threatening or result in death or hospitalization but, based upon appropriate medical and scientific judgment, may jeopardize the subject or may require intervention [eg, medical, surgical] to prevent one of the other serious outcomes listed in the definition above.). Examples of such events include, but are not limited to, intensive treatment in an emergency room or at home for allergic bronchospasm; blood dyscrasias or convulsions that do not result in hospitalization.)
- Is a new cancer (that is not a condition of the study)
- Is associated with an overdose

Events **not** considered to be serious adverse events are hospitalizations for the:

- Admissions as per protocol for a planned medical/surgical procedure or to facilitate a procedure
- Routine health assessment requiring admission for baseline/trending of health status (eg, routine colonoscopy)

- Medical/surgical admission for purpose other than remedying ill health state and was planned prior to entry into the study. Appropriate documentation is required in these cases
- Admission encountered for another life circumstance that carries no bearing on health status and requires no medical/surgical intervention (eg, lack of housing, economic inadequacy, care-giver respite, family circumstances, administrative).

### 6.1.3 Adverse Events of Clinical Interest (ECI) for pembrolizumab

Selected non-serious and serious adverse events are also known as Events of Clinical Interest (ECI) and must be reported within 24 hours to the Sponsor and within 2 working days to Merck Global Safety using the form found in **Appendix C**. (Attn: Worldwide Product Safety; [REDACTED]).

For the time period beginning when the consent form is signed until treatment allocation/randomization, any ECI, or follow up to an ECI, that occurs to any subject must be reported within 24 hours to the Sponsor and within 2 working days to Merck Global Safety 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 allocation/randomization 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 Merck product, must be reported within 24 hours to the Sponsor and within 24 hours to Merck Global Safety.

Events of clinical interest for this trial include:

- An overdose of Merck's product, as defined in **Section 4.5**, 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.*

## 6.2 Relationship

The relationship of an AE to the administration of the study drug is to be assessed by the investigator according to the following definitions:

- *No (unrelated, not related, no relation):* The time course between the administration of study drug and the occurrence or worsening of the adverse event rules out a causal relationship and another cause (concomitant drugs, therapies, complications, etc.) is suspected.
- *Yes (related):* The time course between the administration of study drug and the occurrence or worsening of the adverse event is consistent with a causal relationship and no other cause (concomitant drugs, therapies, complications, etc.) can be identified.

The following factors should also be considered:

- The temporal sequence from study drug administration - The event should occur after the study drug is given. The length of time from study drug exposure to event should be evaluated in the clinical context of the event.
- Underlying, concomitant, intercurrent diseases - Each report should be evaluated in the context of the natural history and course of the disease being treated and any other disease the subject may have.
- Concomitant medication - The other medications the subject is taking or the treatment the subject receives should be examined to determine whether any of them might be recognized to cause the event in question.
- Known response pattern for this class of study drug - Clinical and/or preclinical data may indicate whether a particular response is likely to be a class effect.
- Exposure to physical and/or mental stresses - The exposure to stress might induce adverse changes in the recipient and provide a logical and better explanation for the event.

The pharmacology and pharmacokinetics of the study drug - The known pharmacologic properties (absorption, distribution, metabolism, and excretion) of the study drug should be considered.

Assessment of Grade:

The investigator will make an assessment of grade for each AE and SAE reported during the study, which will be recorded in the CRF. The assessment will be based on the National Cancer Institute's CTCAE (Version 4.03) and graded as shown below:

- Grade 1: Mild; asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated
- Grade 2: Moderate; minimal, local or noninvasive intervention indicated; limiting age-appropriate instrumental activities of daily living
- Grade 3: Severe or medically significant but not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care activities of daily living
- Grade 4: Life-threatening consequences; urgent intervention indicated
- Grade 5: Death related to AE

Any AE that changes in grade during its course will be recorded in the CRF at the highest level experienced by the subject.

### 6.3 Expectedness

Unexpected adverse event: An adverse event, which varies in nature, intensity or frequency from information on the investigational drug/agent provided in the Investigator's Brochure, package insert or safety reports. Any adverse event that is not included in the informed consent is considered "unexpected".

Expected (known) adverse event: An adverse event, which has been reported in the Investigator's Brochure. An adverse event is considered "expected", only if it is included in the informed consent document as a risk.

### 6.4 Handling of Expedited Safety Reports

In accordance with local regulations, the Sponsor will notify investigators of all SAEs that are unexpected (i.e. not previously described in the Investigator Brochure), and definitely, probably, or possibly related to pembrolizumab, cyclophosphamide, or GVAX. This notification will be in the form of an expedited safety report (ESR) that is to be faxed to the investigators and the study coordinators within 48 hours. Upon receiving such notices, the investigator must review and retain the notice with the Investigator's Brochure and where required by local regulations, the investigator will submit the ESR to the appropriate IRB. The investigator and IRB will determine if the informed consent requires revision. The investigator should also comply with the IRB procedures for reporting any other safety information. Where required, submission of ESRs by the investigator to Health Authorities should be handled according to local regulations.

### 6.5 Reporting

#### 6.5.1 General

All adverse events (both expected and unexpected) will be captured on the appropriate study-specific case report forms (CRFs).

In addition, all serious adverse events, regardless of causality to study drug, will be reported promptly to the IND Sponsor (Dr. Lei Zheng, [REDACTED]) within 24 hours of recognition of the adverse event using the form found in **Appendix D**. If this falls on a weekend or holiday, an email notification is acceptable but must be followed by an SAE reporting form on the next business day.

All serious adverse events, regardless of causality to study drug and/or administration will be forwarded to Merck's Global Safety ("Merck GS") group within 24 hours of learning of the event.

After the initial SAE report, the investigator is required to proactively follow each subject and provide further information to the safety department in regards to the subject's condition.

All SAE(s) will be followed until:

- Resolution
- The condition stabilizes
- The event is otherwise explained
- The subject is lost to follow-up

Within 24 hours of receipt of follow-up information, a follow-up SAE report will be submitted to the IND Sponsor, Merck GS.

Non-serious Events of Interest will be forwarded to Merck GS and will be handled in the same manner as SAEs.

Although pregnancy and lactation are not considered adverse events, it is the responsibility of investigators or their designees to report any pregnancy or lactation in a subject (spontaneously reported to them), including the pregnancy of a male subject's female partner who has provided written consent to provide information regarding pregnancy, that occurs during the trial or within 120 days of completing the trial. All subjects who become pregnant must be followed to the completion/termination of the pregnancy. Pregnancy outcomes of spontaneous abortion, missed abortion, benign hydatidiform mole, blighted ovum, fetal death, intrauterine death, miscarriage and stillbirth must be reported as serious events (Important Medical Events). If the pregnancy continues to term, the outcome (health of infant) must also be reported to Merck GS.

**SAE reports and any other relevant safety information are to be forwarded to Merck GS facsimile number: [REDACTED].**

#### 6.5.2 Institutional Review Board (IRB) and Institutional Biosafety Committee (IBC)

All serious adverse events will be reported to the IRB and IBC per institutional standards. Follow-up information will be given to the IRB and IBC as soon as relevant information is available, per institutional standards.

#### 6.5.3 Food and Drug Administration (FDA)

All reporting to the FDA will be completed by the sponsor.

##### 6.5.3.1 Expedited IND Safety Reports:

###### *7 Calendar-Day Telephone or Fax Report:*

The Sponsor is required to notify the FDA of any fatal or life-threatening adverse event that is unexpected and assessed by the investigator to be possibly related to the investigational agent. Such reports are to be telephoned or faxed ([REDACTED]) to the FDA within 7 calendar days of first learning of the event.

###### *15 Calendar-Day Written Report:*

The Sponsor is required to notify the FDA of any serious adverse event that is unexpected and possibly related to the investigational agent in a written IND Safety Report.



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

Written IND safety reports with Analysis of Similar Events are to be submitted to the FDA within 15 calendar days of first learning of the event.

#### 6.5.3.2 IND Annual Reports

In accordance with the regulation 21 CFR § 312.33, the Sponsor shall within 60 days of the anniversary date that the IND went into effect submit a brief report of the adverse events and progress of the investigation. Please refer to Code of Federal Regulations, 21 CFR § 312.33 for a list of the elements required for the annual report. All IND annual reports will be submitted to the FDA by the Sponsor-Investigator.

#### 6.5.4 Recombinant DNA Advisory Committee (RAC)

Unexpected SAEs believed to be related to GVAX will be reported to RAC by email if fatal or life-threatening within 7 calendar days or by written report if related and unexpected to GVAX within 15 calendar days. SAEs that are unrelated or related and expected with GVAX will be reported to RAC in the Annual Report. Follow-up information will be submitted to the RAC as soon as relevant information is available.

## 7. PHARMACEUTICAL INFORMATION

### 7.1 Cyclophosphamide (Cytosan<sup>®</sup>, CY)

#### 7.1.1 Agent Accountability

The IND Sponsor or the Sponsor's representative shall take responsibility for and shall take all steps to maintain appropriate records and ensure appropriate supply, storage, handling, distribution and usage of investigational product in accordance with the protocol and any applicable laws and regulations.

### 7.1.2 Mode of Action

CY is a synthetic antineoplastic drug chemically related to the nitrogen mustards. CY is biotransformed principally in the liver to active alkylating metabolites by a mixed function microsomal oxidase system. These metabolites interfere with the growth of susceptible rapidly proliferating malignant cells. The mechanism of action is thought to involve cross-linking of tumor cell DNA.

### 7.1.3 Description

CY (CYTOXAN®; cyclophosphamide for injection, USP) is a sterile, white powder containing cyclophosphamide monohydrate and is supplied in vials for single-dose use.

### 7.1.4 Packaging and Labeling Information

CY is commercially available.

### 7.1.5 Preparation

Parenteral drug products should be inspected visually for particulate matter and discoloration prior to administration, whenever solution and container permit. Add the diluent to the vial and shake it vigorously to dissolve. If the powder fails to dissolve immediately and completely, it is advisable to allow the vial to stand for a few minutes. Use the quantity of diluent shown below to constitute the product:

<b>Dosage Strength</b>	<b>CYTOXAN Contains Cyclophosphamide Monohydrate</b>	<b>Quantity of Diluent</b>
500 mg	534.5 mg	25 mL
1 g	1069.0 mg	50 mL
2 g	2138.0 mg	100 mL

CY may be prepared for parenteral use by infusion using any of the following methods:

1. CY constituted with 0.9% sterile sodium chloride may be infused without further dilution.
2. CY constituted with 0.9% sterile sodium chloride may be infused following further dilution in the following:
  - Dextrose Injection, USP (5% dextrose)
  - Dextrose and Sodium Chloride Injection, USP (5% dextrose and 0.9% sterile sodium chloride)
  - 5% Dextrose and Ringer's Injection
  - Lactated Ringer's Injection, USP
  - Sodium Chloride Injection, USP (0.45% sterile sodium chloride)
  - Sodium Lactate Injection, USP (1/6 molar sodium lactate)

### 7.1.6 Storage

Store vials at or below 77° F (25° C).

#### 7.1.7 Stability

CY (prepared for either direct injection or infusion) is chemically and physically stable for 24 hours at room temperature or for 6 days in the refrigerator; it does not contain any antimicrobial preservative and thus care must be taken to assure the sterility of prepared solutions.

#### 7.1.8 Route of Administration

CY is administered by IV injection over 30 minutes.

#### 7.1.9 Subject Care Implications

During treatment, the subject's hematologic profile (particularly neutrophils and platelets) should be monitored regularly to determine the degree of hematopoietic suppression.

The rate of metabolism and the leukopenic activity of CY reportedly are increased by chronic administration of high doses of phenobarbital. The physician should be alert for possible combined drug actions, desirable or undesirable, involving CY even though CY has been used successfully concurrently with other drugs, including other cytotoxic drugs. CY treatment, which causes a marked and persistent inhibition of cholinesterase activity, potentiates the effect of succinylcholine chloride. If a subject has been treated with CY within 10 days of general anesthesia, the anesthesiologist should be alerted.

CY may interfere with normal wound healing.

#### 7.1.10 Returns and Reconciliation

N/A

## 7.2 GVAX

#### 7.2.1 Agent Accountability

The IND Sponsor or the Sponsor's representative shall take responsibility for and shall take all steps to maintain appropriate records and ensure appropriate supply, storage, handling, distribution and usage of investigational product in accordance with the protocol and any applicable laws and regulations.

#### 7.2.2 Mode of Action

GM-CSF secreting whole cell vaccines recruit and activate tumor specific T-cells and induce a cytotoxic response through two mechanisms: 1. they deliver a range of peptide antigens (without the need for specific knowledge of the relevant target

antigens), and 2. GM-CSF is an important growth and differentiation factor for dendritic cells, which are potent antigen-presenting cells.

### 7.2.3 Description

[REDACTED]

[REDACTED]

### 7.2.4 Storage

[REDACTED]

### 7.2.5 Preparation

[REDACTED]

### 7.2.6 Stability

[REDACTED]

### 7.2.7 Route of Administration

[REDACTED]

[REDACTED]

### 7.2.8 Subject Care Implications

[REDACTED]

[REDACTED]

[REDACTED]

[REDACTED]

[REDACTED]

[REDACTED]

[REDACTED]

### 7.2.9 Returns and Reconciliation

The investigator is responsible for keeping accurate records of the clinical supplies, 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 unused 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.

### 7.3 Pembrolizumab

#### 7.3.1 Agent Accountability

The sponsor/investigator shall take responsibility for and shall take all steps to maintain appropriate records and ensure appropriate supply, storage, handling, distribution and usage of investigational product in accordance with the protocol and any applicable laws and regulations.

#### 7.3.2 Mode of Action

Pembrolizumab is a highly selective humanized monoclonal antibody designed to block the interaction between PD-1 and its ligands, PD-L1 and PD-L2. Pembrolizumab is an IgG4/kappa isotype with a stabilizing sequence alteration in the Fc region.

#### 7.3.3 Description

[REDACTED]

[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]

#### [REDACTED] Packaging and Labeling Information

[REDACTED]

#### 7.3.5 Preparation

Refer to Procedures Manual for preparation instructions.

#### 7.3.6 Storage

[REDACTED]

[REDACTED]

### 7.3.7 Stability

Refer to Procedures Manual for Stability information.

### 7.3.8 Route of Administration

The reconstituted product is intended for IV administration.

### 7.3.9 Patient Care Implications

Based on results from the nonclinical studies, there are currently no specific safety

[REDACTED]

[REDACTED]

[REDACTED]

### 7.3.10 Agent Ordering

[REDACTED]

[REDACTED]

### 7.3.11 Returns and Reconciliation

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 unused 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.

## 8. CORRELATIVE/SPECIAL STUDIES

Sample collection, processing, and storage instructions will be provided in the Laboratory Manual. The specimens below will be stored and evaluated in Dr. Zheng's laboratory.

### 8.1 Tumor Tissue Studies

Tumor biopsies will be performed on subjects with accessible tumor at baseline and during treatment (during week 6 or 7). However, tumor biopsy will not be performed either at baseline and/or during treatment if the tumor is not accessible or if biopsy is considered not in patient's best interest. An additional biopsy at the time of response and/or progression is optional. Tumor samples will be collected by core biopsy from accessible tumor tissue. A maximum of 6 core biopsies (or fine needle aspirates, if cores are considered unsafe) will be obtained from each subject.

Attempts will be made to obtain archived tissue samples from all subjects

### 8.2 Whole Blood Peripheral Blood Lymphocytes (PBLs) and Circulating Tumor Cells (CTCs)

Up to 180 cc of whole blood will be collected for isolation of PBL at baseline (C1D1) and at every 6 weeks (with every other cycle of treatment) up to and including C5D1. After C5D1, blood will then be collected every 12 weeks (every 4<sup>th</sup> cycle; C9D1) and at the off study visit. In addition, up to 20 cc of whole blood may be collected/aliquoted for circulating tumor cell analyses.

### 8.3 Serum and Plasma Marker Studies

Whole blood for serum (20 cc) and plasma (20 cc) will be collected at baseline (C1D1) and at every 6 weeks (with every other cycle of treatment) up to and including C5D1. After C5D1, blood will then be collected every 12 weeks (every 4<sup>th</sup> cycle; C9D1) and at the off study visit to identify potential therapeutic targets, biomarkers, circulating DNA analyses, and predictors of response and autoimmune toxicity through proteomic approaches.

### 8.4 Diagnostic Tissue Samples



Tissue, fluid, or blood may be collected from standard of care procedures used to treat or diagnoses immune related toxicities.

## 9. STUDY CALENDAR

	Screening	Cycles 1-4		Week 6	Week 12	Cycles 5 and Above <sup>21</sup>		Off Treatment <sup>16</sup>	Off Study <sup>19</sup>
		Day 1	Day 2			Every Cycle	Every 4 <sup>th</sup> cycle		
Visit Window <sup>1</sup>	-14 to -1	+/- 3		+/-7	+/-7	+/-7	+/-7	+/-7	+/- 7
<b>TREATMENT</b>									
Cyclophosphamide		X					X <sup>21</sup>		
GVAX			X				X <sup>21</sup>		
Pembrolizumab		X				X			
<b>CLINICAL ASSESSMENTS/TESTS</b>									
Informed consent	X								
Inclusion/exclusion criteria	X								
Demographics	X								
Medical History <sup>2</sup>	X								
Concurrent medications	X	X				X		X	X
Physical Exam <sup>3</sup>	X	X				X			X
Vital Signs and pulse ox <sup>4</sup>	X	X	X			X			X
Height	X								
Weight	X	X				X			X
Performance Status	X	X				X			X
CBC with differential <sup>5,6</sup>	X	X				X		X	X
Complete Metabolic Profile <sup>5-7</sup>	X	X				X		X	X
TSH <sup>5,8</sup>	X	X				X		X	X
Serum or Urine Preg <sup>9</sup>	X	X				X			
Urinalysis and microscopic exam <sup>5</sup>	X								
INR and pTT <sup>5</sup>	X								
MRI brain with contrast	X <sup>20</sup>								
Adverse event evaluation <sup>10</sup>		X				X		X	X
Vaccine Site Assessment <sup>11</sup>		X				X		X	X
<b>ASSESSMENT OF RESPONSE AND CORRELATIVE STUDIES</b>									
Archival tumor sample	X								
CEA <sup>5,6</sup>	X	X		X	X	X		X	X
Tumor measurements <sup>12</sup>	X				X		X	X <sup>17</sup>	X
Research Blood <sup>13</sup>		X <sup>15</sup>					X <sup>15</sup>	X <sup>18</sup>	X
Tumor Biopsy <sup>14</sup>	X			X					X

- 1) Longer delays to be approved by the sponsor
- 2) Includes history of lung disease, HIV, hepatitis B or C infection, and complete cancer history, including primary site of cancer, gross location of primary tumor, secondary sites of cancer, histology, histologic grade, date of initial diagnosis, date of metastatic diagnosis, and prior cancer therapy regimens.
- 3) Complete physical exam will be completed at baseline; focused physical examinations will be conducted thereafter. Exams, concomitant medication, AE assessments can be made up to 3 days prior to infusion.
- 4) Temperature, respiration rate, blood pressure, pulse, and pulse ox should be taken prior to the administration of each of the drugs: CY, GVAX, and pembrolizumab. Vitals will also be collected post-GVAX.
- 5) Labs may be collected within a window of up to 4 days prior to dosing. Unexpected Grade 3 or greater laboratory abnormalities should be repeated within 24-72 hours if clinically indicated and monitored as necessary to determine if event meets toxicity criteria.
- 6) Screening bloodwork does not need to be repeated if performed within 7 days of C1D1
- 7) Albumin, alkaline phosphatase, total bilirubin, bicarbonate, BUN, calcium, chloride, creatinine, glucose, potassium, total protein, SGOT [AST], SGPT [ALT], sodium.
- 8) Free T3/T4 to be checked reflexively if TSH is abnormal
- 9) Serum or urine pregnancy for women of childbearing potential

- 10) Baseline adverse event evaluation may be performed either at the screening visit or at C1D1 prior to the administration of therapy.
- 11) Not applicable for first cycle, but applicable for day 1 of all subsequent cycles
- 12) Tumor measurement will be performed with contrast CT chest/abdomen/pelvis. Noncontrast CT chest/abdomen/pelvis or CT Chest and MRI Abdomen/pelvis will be performed in subjects with contrast allergies. Baseline scans may be performed within 28 days prior to first treatment. If patients remain on study after initial progression, a repeat CT scan will be obtained at 6 weeks after progression (+/- 7 day window), 12 weeks after progression (+/- 7 days), and every 12 weeks thereafter (+/- 7 days). If the off study visit occurs early, scans do not need to be repeated if one has been performed within 6 weeks.
- 13) Baseline serum and plasma collection should be performed at C1D1 prior to the administration of therapy. Please see section on correlative studies (section 8).
- 14) Only for subjects with tumor that is safely accessible for biopsy, a biopsy should be performed prior to initiation of therapy, and again at 6-7 weeks after initiation of therapy, and (optional) at the time of progression. Refer to section on correlative studies. Optional tumor biopsies may be performed at the time of clinical response as well.
- 15) Follow-up peripheral blood collection will be performed at every other cycle of immunotherapy, (e.g. at C1D1, C3D1, C5D1) prior to the administration of therapy, up to and including C5D1 and then only on cycles where the patient receives GVAX therapy (e.g. every 4<sup>th</sup> cycle).
- 16) Patients will undergo the Off Treatment Evaluation if they completed 24 months of study medication (**Section 4.9**).
- 17) Patients should continue to be monitored for disease status by radiologic imaging and tumor marker(s) every 12 weeks (+/- 2 weeks).
- 18) Peripheral blood, serum, and plasma samples should continue to be collected every 12 weeks (+/- 2 weeks).
- 19) 30 days after their last dose of study drug or within 7 days prior to initiation of a new anti-cancer treatment, whichever comes first. Patients who discontinue from treatment should be contacted every three months to monitor overall survival. Information of other cancer therapies after discontinuation from the study treatment will be collected. SAEs that occur within 90 days of the last infusion of pembrolizumab or before initiation of a new antineoplastic treatment should also be followed and recorded.
- 20) MRI brain with contrast performed in the 8 weeks prior to day 1 can be used to fulfill these criteria.
- 21) After cycle 4, cyclophosphamide and GVAX will be administered with every 4th cycle (12 weeks), restarting with cycle 8. Similar to cycles 1-4, pembrolizumab will administered on cycle day 1. For cycles that include cyclophosphamide and GVAX, cyclophosphamide will also be administered on day 1 and GVAX on day 2. The treatment window for cycles beyond 4 is +/- 7 days.

## 10. MEASUREMENT OF EFFECT

### 10.1 Antitumor Effect – Solid Tumors

#### 10.1.1 Definitions

Evaluable for toxicity. All subjects are evaluable for toxicity after receiving first dose of combined immunotherapy.

Evaluable for objective response. All patients who have received at least two doses of immunotherapy and have had their disease re-evaluated with imaging will be considered evaluable for response. Response criteria will be classified by RECIST and irRECIST criteria (**Appendix E and F**)

#### 10.1.2 Methods for Evaluation of Measurable Disease

All measurements should be taken and recorded in metric notation using a ruler or calipers. All baseline evaluations should be performed as closely as possible to the beginning of treatment and never more than 14 days before the beginning of the treatment. Subjects will be evaluated for anti-tumor effect by follow-up imaging (CT chest/abdomen/pelvis with IV contrast; noncontrast CT chest/abdomen/pelvis or CT Chest and MRI Abdomen/pelvis will be performed in subjects with contrast allergies, intolerance, elevated creatinine, or other contraindication). All subsequent scans (post-treatment) will be compared to the same pretreatment scan that was used prior to initiating of study treatment.

The same method of assessment and the same technique should be used to characterize each identified and reported lesion at baseline and during follow-up. Imaging-based evaluation is preferred to evaluation by clinical examination unless the lesion(s) being followed cannot be imaged but are assessable by clinical exam.

CT Chest/Abdomen/Pelvis: This guideline has defined measurability of lesions on CT scan based on the assumption that CT slice thickness is 5 mm or less. If CT scans have slice thickness greater than 5 mm, the minimum size for a measurable lesion should be twice the slice thickness.

Use of MRI remains a complex issue. MRI has excellent contrast, spatial, and temporal resolution; however, there are many image acquisition variables involved in MRI, which greatly impact image quality, lesion conspicuity, and measurement. Furthermore, the availability of MRI is variable globally. As with CT, if an MRI is performed, the technical specifications of the scanning sequences used should be optimized for the evaluation of the type and site of disease. Furthermore, as with CT, the modality used at follow-up should be the

same as was used at baseline and the lesions should be measured/assessed on the same pulse sequence. It is beyond the scope of the RECIST guidelines to prescribe specific MRI pulse sequence parameters for all scanners, body parts, and diseases. Ideally, the same type of scanner should be used and the image acquisition protocol should be followed as closely as possible to prior scans. Body scans should be performed with breath-hold scanning techniques, if possible.

#### 10.1.3 Overall Survival (OS)

OS is defined as the duration of time from start of study treatment to time of death. Individuals will be censored at the date of the last study visit if no event occurs.

## 11. DATA REPORTING / REGULATORY REQUIREMENTS

Adverse event guidelines and instructions for AE reporting can be found in **Section 6 (Adverse Events: List and Reporting Requirements)**.

### 11.1 Data Management

All information will be collected on study-specific case report forms (CRFs) by study staff. These data will be reviewed for completeness and accuracy by the Principal Investigator.

### 11.2 Safety Meetings

Scheduled meetings will take place weekly and will include the protocol principal investigator, study coordinator(s), research nurse (s), and sub-investigators (as appropriate). During these meetings matters related to the following will be discussed: safety of protocol participants, validity and integrity of the data, enrollment rate relative to expectation, characteristics of participants, retention of participants, adherence to protocol (potential or real protocol violations), data completeness, and progress of data for objectives.

Quarterly teleconferences will be scheduled to include the Investigator and Merck representatives. During these meetings, the Investigator shall provide Merck with study progress updates. The Investigator will provide a summary of key points from the weekly meetings with a focus on safety of the protocol participants, enrollment status, and progress of data for objectives. In addition, Merck will provide safety and applicable program updates to the Sponsor.

### 11.3 Monitoring

This is a DSMP Level III study under the Johns Hopkins Sidney Kimmel Comprehensive Cancer Center (SKCCC). Data monitoring of this protocol will occur on a regular basis with the frequency dependent on the rate of subject accrual and the progress of the study. The protocol will be monitored internally by the Principal Investigator. The protocol will be monitored externally by the SKCCC CRO in accordance with SKCCC guidelines. Additional data and safety monitoring oversight will also be performed by the SKCCC Safety Monitoring Committee (SMC - as defined in the DSMP) and a Medical Expert Committee (MEC) as detailed below.

The Medical Expert Committee (MEC) for this clinical study contains three medical oncologists from other disciplines who are not affiliated with this clinical trial protocol. The MEC will review safety data on at least a semi-annual basis. The MEC will provide a written summary of each assessment to the IND Sponsor after each meeting. In turn, the study team will forward these summaries to the JHU and other participating site IRBs and JHU SKCCC SMC. The operating plan of the MEC will be as follows:

- Meetings will be held at least semi-annually, and potentially more frequently if needed.
- Meetings will be conducted in-person or via video/teleconference, with a participant sign-in sheet collected at each meeting.
- Approximately one week prior to each MEC meeting, the study team will submit the following items to MEC personnel for review and discussion at the meeting (The PI may join the MEC meeting in order to answer any questions the MEC might have):
  - A summary of the clinical trial's progress to date;
  - The latest IRB-approved consent document;
  - A summary of all adverse events, serious adverse events, deaths, and withdrawals to date;

Note that the MEC reserves the right to halt trial accrual or all study activity if, after review, serious safety concerns warrant this action. If the MEC halts study accrual or all study activity, then the study team must notify the JHU SKCCC SMC, JHU IRB, JHU IBC, RAC and the FDA immediately.

Dr. Lei Zheng will be holding the IND for this study. He will comply with all regulated reporting requirements to the FDA.

## **12. STATISTICAL CONSIDERATIONS**

### **12.1 Overview**

The proposed study is an open-label, single-arm phase II study of GVAX in combination with pembrolizumab in patients with metastatic colorectal cancer who have progressed at least two lines of therapies. Subjects with microsatellite instability

(MSI) would not be eligible for this study. The primary objective of the trial is to determine whether the combination of GVAX with pembrolizumab administered in this patient population yields a clinically compelling antitumor activity measured as objective response rate (ORR, assessed by RECIST 1.1). Secondary endpoints include safety, duration of response (DOR), progression-free survival (PFS), overall survival (OS), and immunologic correlates. The study is planned with 25 evaluable subjects based on a two-stage design that allows early termination for lack of efficacy. A safety run-in phase will be included to ensure that the combination of GVAX and pembrolizumab is safe. The study will be continuously monitored for adverse events after the safety run-in phase.

At the beginning of the study, GVAX plus pembrolizumab will be tested in the first 6 patients in the safety run-in portion of the study. These subjects will be monitored for unacceptable toxicities (defined in Section 4.7) through Cycle 1 of treatment before continuation with further accrual. The study will continue without pause to complete the accrual goal for the first stage if no more than 1 of the first 6 subjects experiences unacceptable toxicities (as defined as dose-limiting toxicities in cycle one in section 4.7) in the run-in. If 2 of the 6 subjects experience unacceptable toxicities, the study will be paused for a review. If 3 or more of the first 6 subjects experience unacceptable toxicities with the combination therapy during the run-in, the study will be discontinued. The probability of observing 2 or more patients with DLT among the first 6 subjects is greater than 64% if the true incidence rate is greater than 33%.

## **12.2 Sample Size and Accrual**

Approximately 25 patients with metastatic colorectal cancer who have progressed at least two lines of therapies will be enrolled according to this single-arm, Phase 2 trial, with ORR as the primary endpoint. Enrollment will be carried out in 2 stages so that the study can terminate early if GVAX in combination with pembrolizumab is not sufficiently effective. A recently published phase III study showed that TAS-102 resulted in an ORR of 1.6% (95% confidence interval 0.7-3%) and the agent had an overall survival benefit vs. placebo in a similar patient population [42]. Simon's minimax two-stage design will be employed to test the null hypothesis that the true ORR is 3% or less (not considered clinically compelling for this combination). In the first stage, 15 subjects will be accrued. If none of these 15 subjects respond, the study will be terminated. Otherwise, 10 additional subjects will be accrued to target a total of 25 treated and response evaluable subjects. If 3 or more responses are observed in these 25 subjects, we will conclude the combination of pembrolizumab with GVAX is promising and warrant further investigation. The study could also be terminated early as soon as 3 responses with the combination treatment are confirmed. The probability of stopping the trial early for futility is 63% if the true ORR is 3% or less. This design yields 89% power at a one-sided type I error rate of 5% when the true response rate is 20%. Subjects who are withdrawn from the study prior to receiving one cycle of the combination treatment of cyclophosphamide, GVAX, and

pembrolizumab for reasons other than disease progression will be replaced. The study may enroll 28-30 subjects to ensure 25 would be evaluable for the primary efficacy endpoint.

More than 300 colorectal cancer patients seek primary oncology care at JHH and an additional 200 patients or more come to JHH for consultation each year. The majority of these patients have MMR-p metastatic disease. We estimate that approximately a third of the colorectal cancer patients seen at JHH would be potentially eligible for enrollment for the proposed study. We expect to enroll 2-3 patients per month. We estimate that we could reach the accrual goal within 1 year.

## **12.3 Statistical analysis**

### **12.3.1 Analysis of Primary Endpoint**

The objective response rate defined as the proportion of response evaluable subjects who have a complete response (CR) or partial response (PR) using RECIST 1.1 criteria at any time during the study. The evaluable population includes all subjects who have completed at least two dose of combination therapy and have received at least one follow-up scan. It is known that this crude proportion underestimates the true ORR due to the adaptive nature of Simon's two stage design. Thus we will also report the bias-reduced estimator for ORR due to Whitehead [43] and the adjusted 95% confidence intervals that account for the adaptiveness of the study design [44].

### **12.3.2 Analysis of Secondary Efficacy Endpoints**

Overall survival is defined as the duration of time from start of study treatment to time of death. For subjects lost to follow-up or whose vital status is unknown, every effort will be made to determine the date such subjects were last known to be alive. Such efforts may include phone calls, certified mail, and the checking of public records. Subjects who are alive or lost to follow-up as of the data analysis cutoff date will be right-censored. The censoring date will be determined from the date the subject was last known to be alive.

Progression-free Survival (PFS) is defined as the time from cycle 1, day 1 of immunotherapy until first documented local progression or death due to any cause. Disease progression will be assessed using RECIST (version 1.1). RECIST evaluation will be performed by the radiology staff designated for completing RECIST reads for GI Oncology clinical trials conducted at the SKCCC at Johns Hopkins University. Due to the expectation that some patients may experience delayed clinical responses to therapy, patients with disease progression by radiographic imaging or laboratory parameters during a 12 week evaluation period but without rapid clinical deterioration or significant change in performance status that requires additional immediate therapy may continue to receive treatment on study. Subjects that meet the above criteria and



continue on study therapy must discontinue treatment upon documentation of disease progression on the second scan. The date of progression will be backdated to the time of first RECIST criteria progression. Tumor assessments will be made using RECIST 1.1 and immune-related response criteria (irRC). Local Progression-Free Survival (LPFS) is defined as the duration of time from start of treatment to time of first documented local progression or death due to any cause, whichever occurs first. Individuals will be censored at the date of the last scan for PFS and LPFS if no event occurs.

Duration of response (ROR) will be calculated for subjects who achieve a best overall response of CR or PR with GVAX in combination with pembrolizumab. For such subjects, duration of response is defined as the number of weeks from the start date of PR or CR (whichever response is recorded first) and subsequently confirmed to the first date that recurrent or progressive disease or death is documented. In such cases, recurrent or progressive disease will be assessed relative to the smallest tumor measurements recorded since the start of study treatment. The primary analysis of duration of response will be based on the time point responses, best overall response, and PD status that are determined by the review of the investigator.

OR, PFS, LPFS, and DOR will be summarized descriptively using the Kaplan-Meier method. Analysis of PFS, LPFS, and DOR will be restricted to evaluable individuals who complete at least two doses of combination therapy and have at least one follow-up scan, while the analysis of OS will be performed on all evaluable patients as well as all the individuals who complete at least one dose of pembrolizumab/GVAX. Median event-free survival will be reported and the associated 95% confidence interval will be estimated using the method of [45].

### 12.3.3 Analysis of Safety Endpoints

The safety analysis will be performed in all subjects who receive any amount of study drug. A baseline measurement and at least 1 laboratory or other safety-related measurement obtained after at least 1 dose of study treatment may be required for inclusion in the analysis of a specific safety parameter (e.g., lab shifts from baseline). A complete list of all AE data will be provided along with an assessment of NCI CTCAE grade and relationship to study drug. The incidence of AEs will be tabulated by subgroups of interest (e.g. grade 3 or higher, organ class, relationship to study drug). For analyses at the individual level, the highest grade and relationship to study drug will be assumed if multiple events have occurred. Toxicity will be tabulated by type and grade and will be summarized with descriptive statistics. Other safety data will be assessed in terms of physical examination, clinical chemistry, hematology, vital signs, and ECGs. Negative binomial regression and Cox proportional hazards models will be used to assess the rate of AE and time to first toxicity, respectively.

#### 12.3.4 Biomarker Analysis

Intratumoral changes in pre- and post-treatment core biopsy specimens at week 6 and at the time of progression, if available, will be studied using immunohistochemistry (IHC) and transcriptional analysis. The immune parameters include suppressive pathways (including PD-L2, LAG3, BTLA, TIM3, IDO1, CTLA-4, and Tregs), activation pathways (including CD137, OX40, CD40, CD40L), cytokines/chemokines or their receptors (including CCL12, CXCR4, CCL2, CCL5, CCR2, CCR5). Change standard protein biomarkers such as CEA will be evaluated after each pembrolizumab/GVAX treatment. Descriptive statistics for both pre and post administration of each immunotherapy and the differences in immune parameters will be computed. Plots will be used to show the changes in immune response over time both for each individual and for each arm. Continuous variables will be summarized with means or medians and standard deviations. Dichotomous and categorical variables will be summarized using proportions with exact 95% confidence intervals and counts. For each immunotherapy, comparisons in the pre and 28 day post-immunotherapy responses will be compared using paired t-tests (or Wilcoxon signed-rank tests if appropriate) for continuous variables and McNemar's tests for dichotomous variables. Associations between immune parameters will be explored graphically (e.g. scatterplots, boxplots) and numerically (e.g. correlations,  $\chi^2$  tests). Regression techniques (linear, logistic, linear mixed effects models) will be used to explore the differences between the treatment arms cross-sectionally as well as longitudinally. The associations between immune parameters and clinical outcomes (OS and DFS) will be evaluated using univariate and multivariate Cox regression models. Analyses will be performed using data from all evaluable patients and from the subgroup of patients who receive at least one immunotherapies. Sensitivity analyses will be carried out to evaluate the extent to which results of data analyses can be affected by early dropouts. The significance level is set at 0.05 for all tests in this phase 2 study.

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**APPENDIX A: Performance Status Criteria**

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

**APPENDIX B: Guidance for Identification and Treatment of IRAEs (Version 5.0, Date:  
18-Dec-2014)**



## **APPENDIX C: Adverse Event of Clinical Interest (ECI) Reporting Form**

## Adverse Event of Clinical Interest (ECI) Reporting Form

Please notify: IND Sponsor within 24 hours ( [REDACTED] ),  
Merck within 24 hours ( [REDACTED] )

<b>Protocol Title:</b>	A Phase 2 Study of GVAX Colon Vaccine (with Cyclophosphamide) and Pembrolizumab in Patients with Mismatch Repair-Proficient (MMR-p) Advanced Colorectal Cancer		
<b>Protocol Number</b> J16154 (MK-3475-485)	<b>Signature of PI:</b>	<b>Principal Investigator:</b>  Dr. Nilofer Azad	<b>Date:</b>
<b>Report Type:</b>  <input type="checkbox"/> Initial <input type="checkbox"/> Follow-up <input type="checkbox"/> Final Follow-up <input type="checkbox"/> Addendum to:			
<b>Section A: Subject Information</b>			
<b>Subject ID:</b>	<b>Subject Initial:</b>		<b>Subject Gender:</b> <input type="checkbox"/> Male <input type="checkbox"/> Female
<b>Section B: Event Information</b>			
<b>Event diagnosis or symptoms:</b>	<b>Date of First Dose:</b>		<b>Action taken with the study drug:</b>  <input type="checkbox"/> None <input type="checkbox"/> Interrupted <input type="checkbox"/> Discontinued <input type="checkbox"/> Delayed
	Pembrolizumab	CY      GVAX	
	<b>Date of Last Dose Prior to Event:</b>		
	Pembrolizumab	CY      GVAX	
	<b>Number of Total Doses:</b>		
	Pembrolizumab	CY      GVAX	
<b>Event Onset Date:</b>	<b>Event End Date:</b>		<b>Date Event Discovered:</b>

<b>Relationship to:</b>	<b>Pembrolizumab</b>	<b>CY</b>	<b>GVAX</b>	<b>Underlying Disease</b>		
<b>Unrelated</b>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>		
<b>Related</b>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>		
<b>Section C: Brief Description of the Event: (please include relevant procedures and laboratory values)</b>						
<b>Section D: Relevant Medical History</b>						
<b>Section E: Concomitant Drug (Not related to ECI)</b>						
<b>Name of the Drug</b>	<b>Start Date</b>		<b>Stop Date</b>	<b>Route</b>	<b>Dose</b>	<b>Frequency</b>
<b>Section F: Comments</b>						
<b>Additional Documents:</b> <input type="checkbox"/> Please specify						

## **APPENDIX D: Serious Adverse Event (SAE) Reporting Form**

## Serious Adverse Event Reporting Form

Please notify: Dr. Zheng within 24 hours ( [REDACTED] )  
 Merck within 24 hours ( [REDACTED] )

<b>Protocol Title:</b>	A Phase 2 Study of GVAX Colon Vaccine (with Cyclophosphamide) and Pembrolizumab in Patients with Mismatch Repair-Proficient (MMR-p) Advanced Colorectal Cancer			
<b>Protocol Number:</b> J16154 (MK-3475-485)	<b>Signature of PI:</b>	<b>Principal Investigator:</b>  Dr. Nilofer Azad	<b>Date:</b>	
<b>Report Type:</b> <input type="checkbox"/> Initial <input type="checkbox"/> Follow-up <input type="checkbox"/> Final Follow-up <input type="checkbox"/> Death <input type="checkbox"/> Addendum to:	<b>Serious Criteria (check all that apply):</b> <input type="checkbox"/> Death <input type="checkbox"/> Life-threatening <input type="checkbox"/> Hospitalization or Elongation of Existing Hospitalization <input type="checkbox"/> Persistent or Significant Disability <input type="checkbox"/> Congenital Anomaly <input type="checkbox"/> Other Important Medical Event <input type="checkbox"/> Cancer <input type="checkbox"/> Overdose	<b>Hospital Admission Date:</b>          <b>Hospital Discharge Date:</b>	<b>Date Event Discovered:</b>	
<b>Section A: Subject Information</b>				
<b>Subject ID:</b>	<b>Subject Initial:</b>	<b>Subject Gender:</b> <input type="checkbox"/> Male <input type="checkbox"/> Female		
<b>Section B: Event Information</b>				
<b>Event diagnosis or symptoms:</b>	<b>Date of First Dose:</b>		<b>Action taken with the study drug:</b> <input type="checkbox"/> None <input type="checkbox"/> Interrupted <input type="checkbox"/> Discontinued <input type="checkbox"/> Delayed	
	Pembrolizumab	CY		GVAX
	<b>Date of Last Dose Prior to Event:</b>			
	Pembrolizumab	CY		GVAX
	<b>Number of Total Doses:</b>			
Pembrolizumab	CY	GVAX		

<b>Event Onset Date:</b>			<b>Event End Date:</b>		
<b>Relationship to:</b>	<b>Pembrolizumab</b>	<b>CY</b>	<b>GVAX</b>	<b>Underlying Disease</b>	
<b>Unrelated</b>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	
<b>Related</b>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	
<b>Section C: Brief Description of the Event:</b>					
<b>Section D: Relevant Medical History</b>					
<b>Section E: Concomitant Drug (Not related to SAE)</b>					
<b>Name of the Drug</b>	<b>Start Date</b>	<b>Stop Date</b>	<b>Route</b>	<b>Dose</b>	<b>Frequency</b>
<b>Section F: Comments</b>					
<b>Additional Documents:</b> <input type="checkbox"/> Please specify					

## APPENDIX E: Response Evaluation Criteria in Solid Tumors (RECIST) 1.1 Criteria for Evaluating Response in Solid Tumors

RECIST version 1.1 will be used in this study for assessment of tumor response. While either CT or MRI may be used, as per RECIST 1.1, CT is the preferred imaging technique in this study.

### Disease Parameters

Measurable disease: Measurable lesions are defined as those that can be accurately measured in at least one dimension (longest diameter to be recorded) as  $\geq 20$  mm by chest x-ray, as  $\geq 10$  mm with CT scan, or  $\geq 10$  mm with calipers by clinical exam. All tumor measurements must be recorded in millimeters (or decimal fractions of centimeters).

Note: Tumor lesions that are situated in a previously irradiated area might or might not be considered measurable unless there is evidence of progression in the irradiated site. Malignant lymph nodes. To be considered pathologically enlarged and measurable, a lymph node must be  $\geq 15$  mm in short axis when assessed by CT scan (CT scan slice thickness recommended to be no greater than 5 mm). At baseline and in follow-up, only the short axis will be measured and followed.

Non-measurable disease: All other lesions (or sites of disease), including small lesions (longest diameter  $< 10$  mm or pathological lymph nodes with  $\geq 10$  to  $< 15$  mm short axis), are considered non-measurable disease. Bone lesions, leptomeningeal disease, ascites, pleural/pericardial effusions, lymphangitis cutis/pulmonitis, inflammatory breast disease, and abdominal masses (not followed by CT or MRI), are considered as non-measurable.

Note: Cystic lesions that meet the criteria for radiographically defined simple cysts should not be considered as malignant lesions (neither measurable nor non-measurable) since they are, by definition, simple cysts.

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

Target lesions: All measurable lesions up to a maximum of 2 lesions per organ and 5 lesions in total, representative of all involved organs, should be identified as **target lesions** and recorded and measured at baseline. Target lesions should be selected on the basis of their size (lesions with the longest diameter), be representative of all involved organs, but in addition should be those that lend themselves to reproducible repeated measurements. It may be the case that, on occasion, the largest lesion does not lend itself to reproducible measurement in which circumstance the next largest lesion which can be measured reproducibly should be selected. A sum of the diameters (longest for non-nodal lesions, short axis for nodal lesions) for all target lesions will be calculated and reported as the baseline sum diameters. If lymph nodes are to be

included in the sum, then only the short axis is added into the sum. The baseline sum diameters will be used as reference to further characterize any objective tumor regression in the measurable dimension of the disease.

Non-target lesions: All other lesions (or sites of disease) including any measurable lesions over and above the 5 target lesions should be identified as **non-target lesions** and should also be recorded at baseline. Measurements of these lesions are not required, but the presence, absence, or in rare cases unequivocal progression of each should be noted throughout follow-up.

### **Evaluation of Target Lesions**

Complete Response (CR): Disappearance of all target lesions. Any pathological lymph nodes (whether target or non-target) must have reduction in short axis to <10 mm.

Partial Response (PR): At least a 30% decrease in the sum of the diameters of target lesions, taking as reference the baseline sum diameters.

Progressive Disease (PD): At least a 20% increase in the sum of the diameters of target lesions, taking as reference the smallest sum on study (this includes the baseline sum if that is the smallest on study). In addition to the relative increase of 20%, the sum must also demonstrate an absolute increase of at least 5 mm. (Note: the appearance of one or more new lesions is also considered progressions).

Stable Disease (SD): Neither sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD, taking as reference the smallest sum diameters while on study.

### **Evaluation of Non-Target Lesions**

Complete Response (CR): Disappearance of all non-target lesions and normalization of tumor marker level. All lymph nodes must be non-pathological in size (<10 mm short axis).

Note: If tumor markers are initially above the upper normal limit, they must normalize for a patient to be considered in complete clinical response.

Non-CR/Non-PD: Persistence of one or more non-target lesion(s) and/or maintenance of tumor marker level above the normal limits.

Progressive Disease (PD): Appearance of one or more new lesions and/or *unequivocal progression* of existing non-target lesions. *Unequivocal progression* should not normally trump target lesion status. It must be representative of overall disease status change, not a single lesion increase.

Although a clear progression of “non-target” lesions only is exceptional, the opinion of the



treating physician should prevail in such circumstances, and the progression status should be confirmed at a later time by the review panel (or Principal Investigator).

### Evaluation of Best Overall Response

The best overall response is the best response recorded from the start of the treatment until disease progression/recurrence (taking as reference for progressive disease the smallest measurements recorded since the treatment started). The patient's best response assignment will depend on the achievement of both measurement and confirmation criteria.

#### For Patients with Measurable Disease (i.e., Target Disease)

Target Lesions	Non-Target Lesions	New Lesions	Overall Response	Best Overall Response when Confirmation is Required*
CR	CR	No	CR	$\geq 4$ wks. Confirmation**
CR	Non-CR/Non-PD	No	PR	$\geq 4$ wks. Confirmation**
CR	Not evaluated	No	PR	
PR	Non-CR/Non-PD/not evaluated	No	PR	
SD	Non-CR/Non-PD/not evaluated	No	SD	Documented at least once $\geq 4$ wks. from baseline**
PD	Any	Yes or No	PD	no prior SD, PR or CR
Any	PD***	Yes or No	PD	
Any	Any	Yes	PD	
<p>* See RECIST 1.1 manuscript for further details on what is evidence of a new lesion.</p> <p>** Only for non-randomized trials with response as primary endpoint.</p> <p>*** In exceptional circumstances, unequivocal progression in non-target lesions may be accepted as disease progression.</p> <p><u>Note:</u> Patients with a global deterioration of health status requiring discontinuation of treatment without objective evidence of disease progression at that time should be reported as “<i>symptomatic deterioration.</i>” Every effort should be made to document the objective progression even after discontinuation of treatment.</p>				

## Reference

E.A. Eisenhauer, P. Therasse, J. Bogaerts, L.H. Schwartz, D. Sargent, R. Ford, J. Dancey, S. Arbuck, S. Gwyther, M. Mooney, L. Rubinstein, L. Shankar, L. Dodd, R. Kaplan, D. Lacombe, J. Verweij. New response evaluation criteria in solid tumours: Revised RECIST guideline (version 1.1). *Eur J Cancer*. 2009 Jan;45(2):228-47.

## APPENDIX F: Immune Related Response Criteria

For all patients who experience disease progression on study, the date noted for of disease progression is the time of the scan where it is originally detected, and not the following date of the confirmatory scan.

### Definitions of measurable and non-measurable disease

**Measurable disease:** Neoplastic masses that can be precisely measured in 2 in-plane perpendicular diameters. Both its longest diameter and its longest perpendicular must be greater than or equal to 10 mm. Lymph nodes must have a short-axis line-length of  $\geq 15$  mm. Malignant lymph nodes must be measurable in 2 perpendicular diameters. Both its longest diameter and its longest perpendicular must be greater than or equal to 15 mm. The quantitative endpoint will be defined as the product of the longest diameter with its longest perpendicular.

**Non-measurable disease:** Non-measurable lesions are those that are not suitable for quantitative assessment over time. These include:

- 1) Neoplastic masses that are too small to measure, because their longest uninterrupted diameter or longest perpendicular are less than 10 mm.
- 2) Neoplastic masses whose boundaries cannot be distinguished. This includes masses which cannot be demarcated from surrounding tissue because of inadequate contrast, masses with overly complex morphology, or those with highly heterogeneous tissue composition.
- 3) Other types of lesions that are confidently felt to represent neoplastic tissue, but difficult to quantify in a reproducible manner. These include bone metastases, leptomeningeal metastases, malignant ascites, pleural/pericardial effusions, inflammatory breast disease, lymphangitis cutis/pulmonis, cystic lesions, ill-defined abdominal masses, etc.

**For irRC, only target lesions selected at baseline and measurable new lesions are taken into account.**

At the baseline tumor assessment, the sum of the products of the two largest perpendicular diameters (SPD) of all **index lesions** (five lesions per organ, up to 10 visceral lesions and five cutaneous index lesions) is calculated.

At each subsequent tumor assessment, the SPD of the index lesions and of new, measurable lesions ( $\geq 5 \times 5$  mm; up to 5 new lesions per organ: 5 new cutaneous lesions and 10 visceral lesions) are added together to provide the total time-point **tumor burden**.

### **Overall response using irRC:**

- **Complete Response (irCR):** Complete disappearance of all tumor lesions (whether measurable or not, and no new lesions). CR must be confirmed by repeated, consecutive assessments made no less than 4 weeks from the date first documented.
- **Partial Response (irPR):** Decrease in SPD of 50% or greater by a consecutive assessment at least 4 weeks after first documentation.
- **Stable Disease (irSD):** Failure to meet criteria for irCR or irPR, in absence of irPD.
- **Progressive Disease (irPD):** At least 25% increase in SPD relative to nadir (minimum recorded tumor burden) Confirmation by a repeat, consecutive assessment no less than 4 weeks from the data first documented.

### **Please note other key differences between irRC and the original WHO criteria:**

New measurable lesions will be incorporated into the SPD

New non measurable lesions do not define progression but preclude irCR

Non-index lesions contribute to defining irCR (complete disappearance required).

**See the Investigators Imaging Operations Manual (IIOM) for more details.**

### **REFERENCE**

IrRC for the current protocol is adopted from the following reference:

Wolchok, JD, Hoos, A, O'Day S, et al., Guidelines for the Evaluation of Immune Therapy Activity in Solid Tumors: Immune-Related Response Criteria. Clinical Cancer Research, 2009 Dec 1;15(23):7412-20. Epub 2009 Nov 24.