TITLE: Neoadjuvant PD-1 Blockade in Patients with Stage IIB/C Melanoma	
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1.0 TRIAL SUMMARY

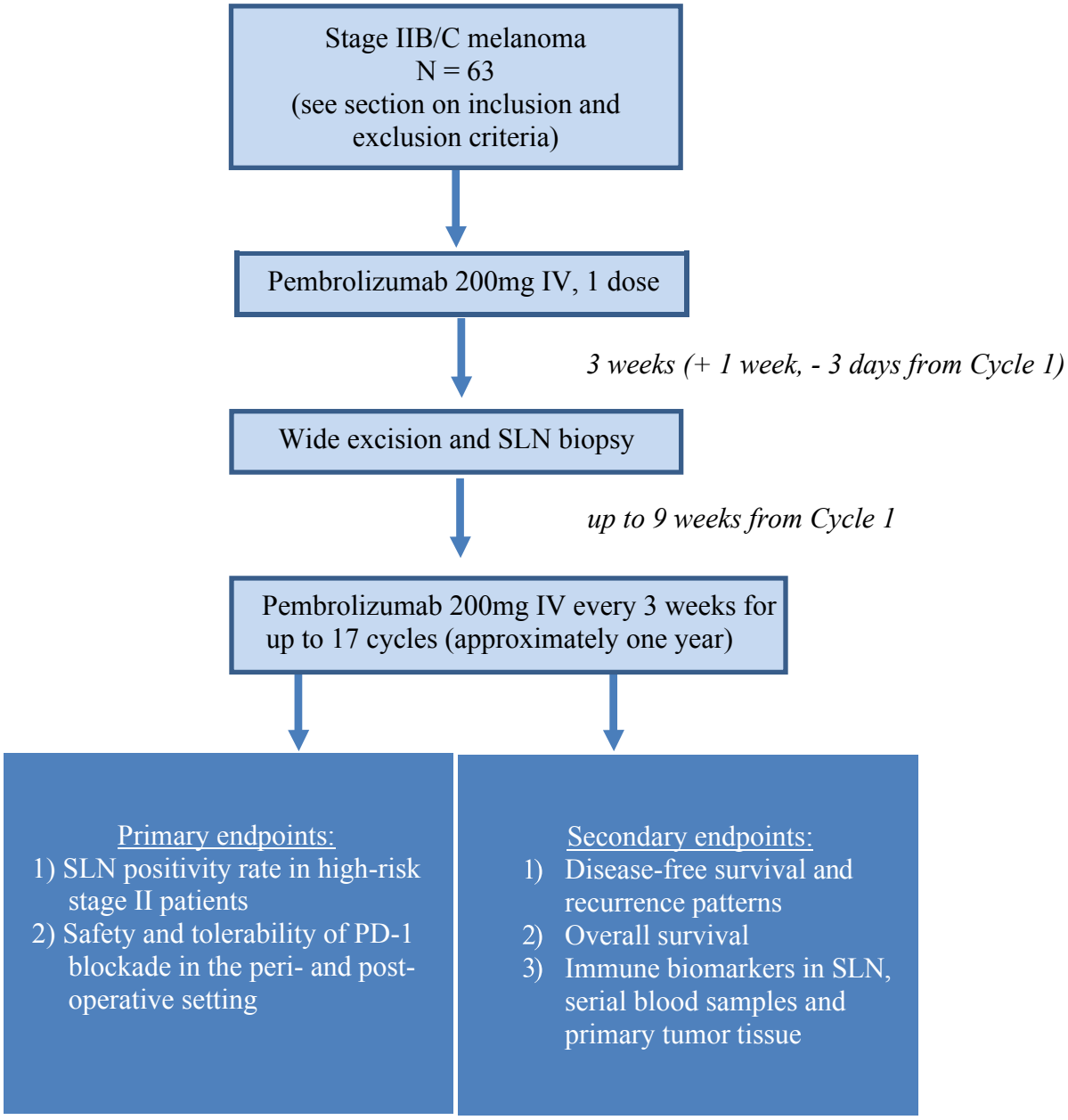
Abbreviated Title	**Neoadjuvant PD-1 blockade in patients with Stage IIB/C melanoma**
Abbreviated Phase	II
Clinical Indication	Patients with Stage IIB and IIC melanoma (MEL) undergoing sentinel lymph node biopsy
Trial Type	Clinical study
Type of control	None
Route of administration	IV
Trial Blinding	None
Treatment Groups	1
Number of trial participants	63
Estimated enrollment period	67 months
Estimated duration of trial	12 years
Duration of Participation	5 years
Estimated average length of treatment per patient	1 year

2.0 TRIAL DESIGN

2.1 Trial Design

Subjects with stage II melanoma will receive one dose of pembrolizumab 200 mg, then undergo standard definitive surgery with wide excision and sentinel lymph node (SLN) biopsy approximately 3 weeks after the initial dose of pembrolizumab. Post-operatively, subjects will receive up to 17 cycles (approximately 1 year) of adjuvant pembrolizumab 200 mg every 3 weeks.

2.2 Trial Diagram



3.0 OBJECTIVE(S) & HYPOTHESIS(ES)

3.1 Primary Objective(s) & Hypothesis(es)

Objectives:

(1) To determine the SLN positivity rate and test whether the SLN positivity rate is reduced in high risk stage II patients undergoing neoadjuvant PD-1 blockade.

Hypothesis: The SLN positivity rate will be reduced by 50% compared to historical (treatment naïve) patients.

(2) To evaluate safety, tolerability and adverse event rate of pembrolizumab in the peri- and post-operative settings.

Secondary **Hypothesis:** Safety profile of pembrolizumab administered in the peri- and post-operative settings for patients with clinically localized melanoma will be similar to the established safety profile of pembrolizumab with an infrequent occurrence of grade 3/4 related to the combination of peri-operative pembrolizumab and surgical procedure.

3.2 Objective(s) & Hypothesis(es)

Objectives:

(3) To evaluate and describe disease-free survival (DFS) and recurrence patterns in subjects with high risk localized melanoma receiving neoadjuvant and adjuvant pembrolizumab.

(4) To evaluate and describe overall survival (OS) in subjects with high risk localized melanoma receiving neoadjuvant and adjuvant pembrolizumab.

Hypothesis: DFS and OS will be estimated and described in this phase II study and may indicate improvement in these clinical outcomes. Recurrence patterns will also be described.

(5) To define the effect of PD-1 blockade with pembrolizumab on immune regulatory pathways, including T cell activation in SLN tissue samples from subjects with melanoma after treatment with a single dose of neoadjuvant pembrolizumab.

(6) To define the effect of PD-1 blockade with pembrolizumab on immune biomarkers in PBMC and serum from subjects before and at serial time points after treatment with pembrolizumab.

(7) To determine whether immune related changes in biomarkers in the primary tumor are associated with immune related changes in the SLN of patients receiving pembrolizumab pre-operatively.

Hypothesis: Collection of all relevant biosamples will be feasible. We hypothesize that various immune-related changes will be noted in the serum and SLN tissue of patients with clinically localized melanoma receiving pembrolizumab peri-operatively. We anticipate associations between immune related changes and clinical outcomes. Moreover, we hypothesize that immune-related biomarkers in the primary tumor will be associated with immune-related changes in the SLN after receipt of neoadjuvant pembrolizumab. This phase II study will enroll sufficient patient numbers to allow investigations of various pharmacodynamic effects and associations with clinical outcomes.

4.0 BACKGROUND & RATIONALE

4.1 Background

Patients with clinical Stage IIB/C melanoma have historically been high risk for recurrence and death (5-and 10-year overall survival rates 50-70% and 40-60%, respectively) with many stage IIC patients experiencing less favorable disease specific survival compared to patients with stage III disease. Moreover, incidence of sentinel lymph node (SLN) metastasis is high in these subgroups of patients ranging from approximately 30-40%.

As the SLN is the draining LN of the tumor and receives tumor antigen, it is the initial site of anti-tumor immune response even in the absence of clinically or radiographically detectable metastasis. As a result, we hypothesize that the draining lymph node is the site of earliest detectable immune response to anti-PD1 therapy and successful initiation of an immune response is associated with clinical cure while failure to initiate an immune response is associated with recurrence.

Toward this end, we have proposed a neoadjuvant anti-PD1 clinical trial in Stage IIB/C melanoma. The trial is novel and unique in that with the use of SLN mapping, we will be able to identify and study the immune response in the draining LN, which is the site of first contact between tumor and host immune response.

We have previously shown that the immune response in peripheral blood peaks at 3 weeks after 1 dose of pembrolizumab. Consistent with peripheral blood data, we have also shown that anti- tumor activity is evident in tumor tissue at 3 weeks after 1 dose of pembrolizumab. In the same manner, given the presence of detectable melanoma micrometastasis in the lymph nodes of 25-40% of this high-risk patient population, we would expect to observe evidence of T cell immune response in the LNs of patients treated with one dose of PD-1 blockade prior to SLN sampling, as well as a lower rate of SLN positive patients, which is the primary hypothesis of this trial. As we have now seen pathologic complete responses in patients with clinical nodal disease after 1 dose of pembrolizumab, we would expect similar anti-tumor activity in patients with clinically occult nodal disease

Pembrolizumab is a potent humanized immunoglobulin G4 (IgG4) monoclonal antibody (mAb) with high specificity of binding to the programmed cell death 1 (PD-1) receptor, thus inhibiting its interaction with programmed cell death ligand 1 (PD-L1) and programmed cell death ligand 2 (PD-L2). Based on preclinical in vitro data, pembrolizumab has high affinity and potent receptor blocking activity for PD-1. Pembrolizumab has an acceptable preclinical safety profile and is in clinical development as an intravenous (IV) immunotherapy for advanced malignancies.

Keytruda® (pembrolizumab) is indicated for the treatment of patients across a number of indications because of its mechanism of action to bind the PD-1 receptor on the T cell. For more details on specific indications refer to the Investigator brochure.

Despite accumulating knowledge on PD-1 inhibition, additional studies in human tumor tissue are needed to better define the mechanism of action of PD-1 inhibition. Defining the mechanism is an important step in developing predictors of response to treatment with PD-1 inhibition, which would allow for more strategic development of combination immune therapy strategies, ultimately with the goal of increasing objective response rates to immune therapy in melanoma and other tumor types. Importantly, this proposed study will generate a wealth of mechanism of action data in a group of high-risk patients but with low clinical disease burden, as well as additional neoadjuvant and adjuvant administration feasibility data.

Currently, standard options for patients with resected high-risk stage II melanoma include, most commonly, close observation, with high dose interferon (without definite benefit on overall survival) rarely used at this time. Therefore, application of effective therapies already in use in the advanced setting that could reduce the risk of recurrence and improve survival are needed in this patient population.

4.1.1 Pharmaceutical and Therapeutic Background

The importance of intact immune surveillance function in controlling outgrowth of neoplastic transformations has been known for decades [Disis, 2010]. Accumulating evidence shows a correlation between tumor-infiltrating lymphocytes in cancer tissue and favorable prognosis in various malignancies. In particular, the presence of CD8+ T-cells and the ratio of CD8+ effector T-cells/FoxP3+ regulatory T-cells (T-regs) correlates with improved prognosis and long-term survival in solid malignancies, such as ovarian, colorectal, and pancreatic cancer; hepatocellular carcinoma; malignant melanoma; and renal cell carcinoma. Tumor-infiltrating lymphocytes can be expanded ex vivo and reinfused, inducing durable objective tumor responses in cancers such as melanoma [Dudley et al., 2005; Hunder et al., 2008].

The PD-1 receptor-ligand interaction is a major pathway hijacked by tumors to suppress immune control. The normal function of PD-1, expressed on the cell surface of activated T-cells under healthy conditions, is to down-modulate unwanted or excessive immune responses, including autoimmune reactions. PD-1 (encoded by the gene *Pdcd1*) is an immunoglobulin (Ig) superfamily member related to cluster of differentiation 28 (CD28) and cytotoxic T-lymphocyte- associated protein 4 (CTLA-4) that has been shown to negatively regulate antigen receptor signaling upon engagement of its ligands (PD-L1 and/or PD-L2) [Greenwald et al., 2005; Okazaki et al., 2001].

The structure of murine PD-1 has been resolved [Zhang et al., 2004]. PD-1 and its family members are type I transmembrane glycoproteins containing an Ig-variable–type (IgV-type) domain responsible for ligand binding and a cytoplasmic tail responsible for the binding of signaling molecules. The cytoplasmic tail of PD-1 contains 2 tyrosine-based signaling motifs, an immunoreceptor tyrosine-based inhibition motif, and an immunoreceptor tyrosine-based switch motif. Following T-cell stimulation, PD-1 recruits the tyrosine phosphatases, SHP-1 and SHP-2, to the immunoreceptor tyrosine-based switch motif within its cytoplasmic tail, leading to the dephosphorylation of effector molecules such as CD3 zeta (CD3ζ), protein kinase C-theta (PKCθ), and zeta-chain-associated protein kinase (ZAP70), which are involved in the CD3 T-cell signaling cascade [Okazaki et al., 2001; Chemnitz et al., 2004; Sheppard et al., 2004; and Riley, 2009]. The mechanism by which PD-1 down-modulates T-cell responses is similar to, but distinct from, that of CTLA-4, because both molecules regulate an overlapping set of signaling proteins [Parry et al., 2005; Francisco, 2010]. As a consequence, the PD-1/PD-L1 pathway is an attractive target for therapeutic intervention in melanoma.

4.1.2 Preclinical and Clinical Trial Data

Refer to the Investigator's Brochure for Preclinical and Clinical data.

4.2 Rationale

Currently, standard options for patients with resected high risk localized (Stage IIB/IIC) melanoma include close observation (most commonly), high dose interferon, or clinical trials, as high dose interferon can have significant toxicity and has not demonstrated a definite overall survival benefit in this patient population. Effective therapies that reduce the risk of recurrence and improve survival in this population are needed. Pembrolizumab is a highly selective, humanized monoclonal antibody (IgG4/kappa isotype) that directly blocks the interaction between PD-1 and its ligands, PD-L1 and PD-L2, with safety and efficacy demonstrated in advanced melanoma and other advanced solid tumors. Unfortunately, little is known about the mechanism of action of pembrolizumab in tumor tissue and its interaction with other immune regulatory pathways.

Anti-cancer activities by anti- PD-1mAb seen in MEL patients thus far provide a strong rationale for evaluation of pembrolizumab in MEL patients with high risk resectable disease.

Pembrolizumab has been well tolerated with dose levels up to 200 mg and has shown a promising response rate of 41% in patients with advanced melanoma, which is much higher than the 10-15% response rate observed in ipilimumab registration trials. Existing data from adjuvant pembrolizumab study support the evaluation of the safety and efficacy of pembrolizumab in patients with resectable high risk stage III MEL, in which setting the drug is currently approved.

Additional studies in human tumor tissue are needed to better define the mechanism of action of PD-1 inhibition. Defining the mechanism is the first step in determining factors that predict response to treatment with PD-1 inhibition, which would allow for more strategic development of combination immune therapy strategies, ultimately with the goal of increasing objective response rates to immune therapy in melanoma and other tumor types. Based on other studies and our own peri-operative stage III clinical study, we do not anticipate any increased toxicity with pembrolizumab in the peri-operative period. This proposed study will generate further mechanism of action data, as well as, neoadjuvant and adjuvant administration feasibility data. Importantly, this study will provide the opportunity to identify novel findings in the SLN tissue (first draining lymph node tissue from the primary tumor) in patients with high risk of relapse but with low burden of disease in response to PD-1 inhibition.

4.2.1 Rationale for the Trial and Selected Population

Patients for inclusion would be patients with clinical stage IIB/C, who are at significant risk for relapse. Patients with clinical Stage IIB/C melanoma have historically been high risk for recurrence and death (5-and 10-year overall survival rates 50-70% and 40-60% or lower respectively) with many stage IIC patients experiencing less favorable disease specific survival compared to patients with stage III disease. Moreover, incidence of sentinel lymph node (SLN) metastasis is high in these stage II patients ranging from approximately 25-40%.

This patient population is selected as their risk of disease recurrence is appreciable, but there are no currently approved effective systemic therapy options that have shown prolonged survival in this population.

Tumor imaging is strongly preferred to confirm absence of any clinical metastases.

Computed tomography (CT) is preferred. PET/CT may be used. For the abdomen and pelvis, contrast-enhanced magnetic resonance imaging (MRI) may be used when CT with iodinated contrast

is contraindicated, or when local practice mandates it. For the regional nodal bed, ultrasound imaging may be used.

63 evaluable subjects are expected to be enrolled in this study and will be recruited from the Penn Medicine Abramson Cancer Center, The University of Texas MD Anderson Cancer Center and the University of Louisville James Graham Brown Cancer Center. We plan to consent sufficient subjects such that 63 evaluable subjects are enrolled. Subjects will be considered evaluable if they receive at least 1 dose of neoadjuvant pembrolizumab and undergo definitive surgery with SLN biopsy.

4.2.2 Justification for Dose

The planned dose of pembrolizumab for this study is 200 mg every 3 weeks (Q3W). Based on the totality of data generated in the Keytruda development program, 200 mg Q3W is the appropriate dose of pembrolizumab for adults across all indications and regardless of tumor type. As outlined below, this dose is justified by:

- Clinical data from 8 randomized studies demonstrating flat dose- and exposure-efficacy relationships from 2 mg/kg Q3W to 10 mg/kg every 2 weeks (Q2W)
- Clinical data showing meaningful improvement in benefit-risk including overall survival at 200 mg Q3W across multiple indications, and
- Pharmacology data showing full target saturation in both systemic circulation (inferred from pharmacokinetic [PK] data) and tumor (inferred from physiologically-based PK [PBPK] analysis) at 200 mg Q3W

Among the 8 randomized dose-comparison studies, a total of 2262 participants were enrolled with melanoma and non-small cell lung cancer (NSCLC), covering different disease settings (treatment naïve, previously treated, PD-L1 enriched, and all-comers) and different treatment settings (monotherapy and in combination with chemotherapy). Five studies compared 2 mg/kg Q3W versus 10 mg/kg Q2W (KN001 Cohort B2, KN001 Cohort D, KN002, KN010, and KN021), and 3 studies compared 10 mg/kg Q3W versus 10 mg/kg Q2W (KN001 Cohort B3, KN001 Cohort F2 and KN006). All of these studies demonstrated flat dose- and exposure- response relationships across the doses studied representing an approximate 5- to 7.5-fold difference in exposure. The 2 mg/kg (or 200 mg fixed-dose) Q3W provided similar responses to the highest doses studied. Subsequently, flat dose-exposure-response relationships were also observed in other tumor types including head and neck cancer, bladder cancer, gastric cancer and classical Hodgkin Lymphoma, confirming 200 mg Q3W as the appropriate dose independent of the tumor type. These findings are consistent with the mechanism of action of pembrolizumab, which acts by interaction with immune cells, and not via direct binding to cancer cells.

Additionally, pharmacology data clearly show target saturation at 200 mg Q3W. First, PK data in KN001 evaluating target-mediated drug disposition (TMDD) conclusively demonstrated saturation of PD-1 in systemic circulation at doses much lower than 200 mg Q3W. Second, a PBPK analysis was conducted to predict tumor PD-1 saturation over a wide range of tumor penetration and PD-1 expression. This evaluation concluded that pembrolizumab at 200 mg Q3W achieves full PD-1 saturation in both blood and tumor.

Finally, population PK analysis of pembrolizumab, which characterized the influence of body weight and other participant covariates on exposure, has shown that the fixed-dosing provides

similar control of PK variability as weight-based dosing, with considerable overlap in the distribution of exposures from the 200 mg Q3W fixed dose and 2 mg/kg Q3W dose. Supported by these PK characteristics, and given that fixed-dose has advantages of reduced dosing complexity and reduced potential of dosing errors, the 200 mg Q3W fixed-dose was selected for evaluation across all pembrolizumab protocols.

4.2.3 Rationale for Endpoints

4.2.3.1 Safety Endpoints

We will monitor closely for adverse events and toxicity grades per Section 5.2.2 to ensure that they do not exceed what would otherwise be expected for treatment with pembrolizumab in this earlier stage melanoma patient population.

4.2.3.2 Efficacy Endpoints

Primary Endpoint: SLN positivity rate is the percentage of patients who received neoadjuvant pembrolizumab and who have a positive SLN. The SLN positivity rate is associated with stage of disease (i.e., stage IIB or stage IIC). The historical SLN positivity rate for this study population is described in Section 8.6.

Secondary Endpoints:

Disease-free and overall survival. Disease-free survival (DFS) is defined as the time from date of surgery to date of first documented disease progression, death due to any cause or last date that patient was documented to be disease-free (i.e., a scan date). Site of recurrence will also be described. Overall survival (OS) is defined as the time from date of surgery to date of death due to any cause or last patient contact alive. Both DFS and OS outcomes will be measured from date of surgery to allow us to compare DFS and OS estimates with those of other adjuvant trials.

4.2.3.3 Immune Biomarker Endpoints and Assays

Biosamples (blood, SLN and primary tumor) obtained from subjects before and after neoadjuvant treatment and at serial time points during adjuvant treatment (blood only) will be analyzed. High priority assays will be performed as indicated below. Second priority assays will be performed as the budget allows.

High priority assays:

1 Peripheral blood mononuclear cells (PBMC) will be obtained at various time points at baseline and during this protocol, including before and after treatment and before and after surgery. PBMCs will be analyzed by flow cytometry to measure important T cell subsets. Expression of inhibitory receptors including PD-1 and others will be examined in PBMC populations to test whether changes in such pathways on T cells occurs following treatment.

2 A: We will perform multicolor immunofluorescence for CD8 and dendritic cell (DC) markers, as well as activation markers including Ki67. In the setting of an active immune response to anti-PD1, we would expect clusters of DC and CD8 T cell aggregates in the T cell zone of the LN and increase in Ki67 expressing CD8 T cells. In addition, we will assess for markers of the progenitor exhausted T cell population such as TCF-1, PD1, CXCR5, and BCL-6, and track

frequency and proliferation after therapy using Ki67. In addition, we will have the opportunity to gain information on the effect of anti-PD1 therapy on T follicular helper (TFH) and germinal center response.

B: We will perform targeted transcriptomic analysis on the sentinel LN using the Nanostring platform. We predict that there will be a distinct immune activation signature after one dose of anti-PD1 that will correlate with clinical outcomes.

C: Neoadjuvant treatment in the setting of early stage disease provides a unique opportunity to compare the fates of T Cell Receptor (TCR) clones in the setting of cure and recurrence. We will extract DNA from the primary tumor pre-treatment, the sentinel LN post treatment and peripheral blood at pre and post treatment time points. We will perform TCR beta sequencing on all samples through Adaptive Biotechnologies. We will then sort T cell populations (naïve, stem cell memory, central memory, exhausted, and effector populations) at 1 year or time of progression from patients with no recurrence or recurrence respectively. We will test the hypothesis that patients with good prognosis have tumor associated clones present in the primary tumor that are also: i) Found in the draining LN and ii) identified in the memory pool at 1 year. In contrast patients who recur do not have clones in the draining LN and upon progression do not have clones in the memory pool and rather in exhausted or effector pools.

Second priority assays:

1. Inflammatory cytokines/chemokines. Serum obtained at various times at baseline and during the protocol will be analyzed using Luminex bead array and Invitrogen 30-plex kits, which can simultaneously quantify/determine concentrations of 30 important factors, including IL-1, TNF-alpha, IL-6 and others.
2. Antigen-specific T-cell responses will be measured when possible using established assays in which PBMC pre and post therapy will be tested in vitro in T-cell responder assays. For patients who are HLA-A2+, functional responses and peptide-MHC quantification will be measured for a panel of melanoma associated antigens, as well as, viral antigens as a control. Functional measurements can include cytokine production, CD107a mobilization, and lysis against peptide-pulsed targets or tumor cells lines expressing the melanoma antigen. In addition, for functional assessments of T cells without regard to the patients' MHC, we will use autologous CD40-activated B cells electroporated with viral or tumor antigen mRNA as in vitro stimulators.

New antibodies with reactivity to defined antigens will be measured in serum collected before and during treatment. When possible, we will use the ProtoArray protein array, which uses chip based technology to profile 9,000 proteins. Analysis will be by Prospector software. The induction of high titer antibodies to defined tumor antigens not only reflects a humoral immune response to treatment but also a cellular response.

3. New blood based or tissue biomarkers showing significant promise may be investigated based on results of ongoing research investigations using samples otherwise procured in the context of the trial.

5.0 METHODOLOGY

5.1 Study Population

5.1.1 Participant Inclusion Criteria

Participants are eligible to be included in the study only if all of the following criteria apply:

1. The subject must have clinical stage IIB or IIC resectable MEL. Subjects may not have a diagnosis of uveal or mucosal melanoma.
2. Either the subject or the subject's legal representative must be willing and able to provide written informed consent for the trial.
3. The subject must be ≥ 18 years of age on day of signing informed consent.
4. The subject must have a performance status of 0 or 1 on the ECOG Performance Scale.
5. The subject must demonstrate adequate organ function as defined in Table 1; all screening labs must be performed within 21 days of treatment initiation.

Table 1. Adequate Organ Function Laboratory Values

System	Laboratory Value
Hematologic	
Absolute neutrophil count (ANC)	≥ 1500/mcL
Platelets	≥ 100,000/mcL
Hemoglobin	≥ 9 g/dL or ≥5.6 mmol/L
Renal	
Serum creatinine **OR** measured or calculated[a] creatinine clearance (GFR can also be used in place of creatinine or CrCl)	≤ 1.5 X upper limit of normal (ULN) **OR** ≥50 mL/min for subject with creatinine levels >1.5 X institutional ULN
Hepatic	
Serum total bilirubin	≤1.5 X ULN **OR** Direct bilirubin ≤ ULN for subjects with total bilirubin levels >1.5 ULN
AST (SGOT) and ALT (SPGT)	≤2.5 X ULN **OR** ≤5 X ULN for subjects with liver metastases
Coagulation	
International Normalized Ratio (INR) or Prothrombin Time (PT)	≤1.5 X ULN unless subject is receiving anticoagulant therapy as long as PT or PTT is within therapeutic range of intended use of anticoagulants
Activated Partial Thromboplastin Time (aPTT)	≤1.5 X ULN unless subject is receiving anticoagulant therapy as long as PT or PTT is within therapeutic range of intended use of anticoagulants
[a]Creatinine clearance should be calculated per institutional standard.	

6. Female participants:
    a. A female participant is eligible to participate if she is not pregnant (see Appendix 3), not breastfeeding, and at least one of the following conditions applies:

        a.) Not a woman of childbearing potential (WOCBP) as defined in Appendix 3 OR

        b.) A WOCBP who agrees to follow the contraceptive guidance in Appendix 3 during the treatment period and must be willing to use a highly effective method of contraception or abstain from heterosexual intercourse for at least 2 weeks prior to the time of first dose of study medication through 120 days after the last dose of study medication.

    b. Female subjects of childbearing potential must have a negative urine or serum pregnancy test within 72 hours prior to receiving the first dose of study medication.

    c. Female subjects of childbearing potential are defined as those women who have not been surgically sterilized or have not been free from menses for >1 year.

7. Male participants:

    Male subjects must agree to follow the contraceptive guidance in Appendix 3 starting with the first dose of study medication, while on study, through 120 days after the last dose of study medication.

5.1.2 Participant Exclusion Criteria

Participants are excluded from the study if any of the following criteria apply:

1. Subject has unresectable disease; i.e. in the opinion of the surgical oncologist, all of the subject's melanoma cannot be completely removed with a clear margin.
2. A woman of child bearing potential who has a positive urine pregnancy test within 72 hours prior to first dose of study drug (see Appendix 3). If the urine test is positive or cannot be confirmed as negative, a serum pregnancy test will be required. Note: In the event that 72 hours have elapsed between the screening pregnancy test and the first dose of study treatment, another pregnancy test (urine or serum) must be performed and must be negative in order for subject to start receiving study medication.
3. Has received prior therapy with an anti-PD-1, anti-PD-L1, or anti-PD-L2 agent or with an agent directed to another stimulatory or co-inhibitory T-cell receptor (e.g., CTLA-4, OX-40, CD137), interferon, high dose IL-2 or any other antibody or drug specifically targeting T-cell co-stimulation or checkpoint pathways.
4. Has received systemic anti-cancer therapy including investigational agents within 4 weeks prior to starting study treatment. Note: Participants must have recovered from all AEs due to previous therapies to ≤Grade 1 or baseline. Participants with ≤ Grade 2 neuropathy are an exception to this criterion.
5. If participant received major surgery, they must have recovered adequately from the toxicity and/or complications from the intervention prior to starting study treatment.
6. Subject has received transfusion of blood products (including platelets or red blood cells) or administration of colony stimulating factors (including G-CSF, GM-CSF or recombinant erythropoietin) within 4 weeks prior to study Day 1.

7. Has received a live vaccine within 30 days prior to the first dose of study drug. Examples of live vaccines include, but are not limited to, the following: measles, mumps, rubella, varicella/zoster (chicken pox), yellow fever, rabies, Bacillus Calmette–Guérin (BCG), and typhoid vaccine. Seasonal influenza vaccines for injection are generally killed virus vaccines and are allowed; however, intranasal influenza vaccines (e.g., FluMist®) are live attenuated vaccines and are not allowed.
8. Is currently participating in or has participated in a study of an investigational agent or has used an investigational device within 4 weeks prior to the first dose of study treatment.
9. Has a diagnosis of immunodeficiency or is receiving chronic systemic steroid therapy (in dosing exceeding 10 mg daily of prednisone equivalent) or any other form of immunosuppressive therapy within 7 days prior to the first dose of study drug.
10. Has a known additional malignancy that is progressing or requires active treatment. Note: Participants with basal cell carcinoma of the skin, squamous cell carcinoma of the skin, or carcinoma in situ (e.g. breast carcinoma, cervical cancer in situ) that have undergone potentially curative therapy are not excluded.
11. Has severe hypersensitivity (≥Grade 3) to pembrolizumab and/or any of its excipients.
12. Has active autoimmune disease that has required systemic treatment in the past 3 months (i.e. with use of disease modifying agents, corticosteroids or immunosuppressive drugs). Replacement therapy (e.g., thyroxine, insulin, or physiologic corticosteroid replacement therapy for adrenal or pituitary insufficiency, etc.) is not considered a form of systemic treatment.
13. Has evidence of active interstitial lung disease or a history of (non-infectious) pneumonitis that required steroids or has current pneumonitis.
14. Has an active infection requiring systemic therapy.
15. Has a known history of Human Immunodeficiency Virus (HIV). (HIV 1/2 antibodies) as determined by medical record review.
16. Has a known history of Hepatitis B (defined as Hepatitis B surface antigen [HBsAg] reactive) or known active Hepatitis C virus (defined as HCV RNA is detected) infection as determined by medical record review.
17. Has a known history of active TB (Bacillus Tuberculosis).
18. Has a history or current evidence of any condition, therapy, or laboratory abnormality that might confound the results of the study, interfere with the subject's participation for the full duration of the study, or is not in the best interest of the subject to participate, in the opinion of the treating investigator.
19. Has known psychiatric or substance abuse disorders that would interfere with cooperation with the requirements of the trial.
20. Is pregnant or breastfeeding, or expecting to conceive or father children within the projected duration of the study, starting with the screening visit through 120 days after the last dose of investigational drug.
21. Has severe cardiovascular disease, i.e. poorly controlled arrhythmias, congestive heart failure (NYHA Class III or IV) or symptomatic ischemic heart disease.

22. Has hepatic decompensation (Child-Pugh score > 6 [class B and C]).
23. Has uncontrolled thyroid dysfunction.
24. Has uncontrolled diabetes mellitus.

5.1.3 Lifestyle Restrictions

5.1.3.1 Meals and Dietary Restrictions

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

5.1.3.2 Contraception

Pembrolizumab may have adverse effects on a fetus in utero. Refer to Appendix 3 for approved methods of contraception.

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

5.1.4 Pregnancy

If a participant inadvertently becomes pregnant while on treatment with pembrolizumab, the participant will be immediately discontinued from study treatment. The site will contact the participant at least monthly and document the participant's status until the pregnancy has been completed or terminated. The outcome of the pregnancy will be reported within 24 hours to the Sponsor-Investigator and to Merck within 2 working days 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). 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 Merck. If a male participant impregnates his female partner, the study personnel at the site must be informed immediately and the pregnancy must be reported to the Sponsor-Investigator and to Merck and followed as described in Section 7.3.2.

5.1.5 Use in Nursing Women

It is unknown whether pembrolizumab is 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, participants who are breast-feeding are not eligible for enrollment.

5.2 Trial Treatments

The treatment to be used in this trial is outlined below in Table 1

Table 1: Trial Treatment

Drug	Dose/Potency	Dose Frequency	Route of Administration	Regimen/ Treatment Period	Use
Pembrolizumab	200 mg	Q3W +/- 5 days	IV infusion	Day 1 of each 3-week cycle	Experimental
The pembrolizumab dosing interval may be increased to Q4W due to toxicity.					

5.2.1 Timing of Dose Administration

Trial treatment should be administered on Day 1 of each cycle after all procedures/assessments have been completed as detailed on the Trial Flow Chart (Section 6.1). Trial treatment may be administered up to 5 days before or after the scheduled Day 1 of each cycle due to administrative reasons.

All trial treatments will be administered on an outpatient basis.

Pembrolizumab 200 mg will be administered as a 30-minute IV infusion every 3 weeks. Sites should make every effort to target infusion timing to be as close to 30 minutes as possible. However, given the variability of infusion pumps from site to site, a window of -5 minutes and +10 minutes is permitted (i.e., infusion time is 30 minutes: -5 min/+10 min).

5.2.2 Dose Modification and Toxicity Management <u>for Immune-Related AEs Associated with Pembrolizumab</u>

AEs associated with pembrolizumab exposure may represent an immunologic etiology. These immune-related AEs (irAEs) may occur shortly after the first dose or several months after the last dose of pembrolizumab treatment and may affect more than one body system simultaneously. Therefore, early recognition and initiation of treatment is critical to reduce complications. Based on existing clinical study data, most irAEs were reversible and could be managed with interruptions of pembrolizumab, administration of corticosteroids and/or other supportive care. For suspected irAEs, ensure adequate evaluation to confirm etiology or exclude other causes.

Additional procedures or tests such as bronchoscopy, endoscopy, skin biopsy may be included as part of the evaluation. Based on the severity of irAEs, withhold or permanently discontinue pembrolizumab and administer corticosteroids. Dose modification and toxicity management guidelines for irAEs associated with pembrolizumab are provided in Table 3.

Dose Limiting Toxicity (DLT): Dose Limiting toxicity will be defined as Grade 3 or higher non-hematologic, and Grade 4 or higher hematologic toxicity that is at least possibly related to pembrolizumab and is not expected for surgery alone or pembrolizumab alone. DLTs will be monitored for up to 6 weeks post-surgery. **A termination rule for a DLT rate >33% is presented in Section 5.8.**

Pembrolizumab will be withheld for drug-related Grade 4 hematologic toxicities, non- hematological toxicity ≥ Grade 3 including laboratory abnormalities, and severe or life- threatening AEs as per Table 3 below.

Table 3: Dose Modification and Toxicity Management Guidelines (Immune-Related AEs Associated with Pembrolizumab)

General instructions:

1. Corticosteroid taper should be initiated upon AE improving to Grade 1 or less and continue to taper over at least 4 weeks.
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 ≤10 mg prednisone or equivalent per day within 12 weeks.
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.

Immune-related AEs	**Toxicity grade or conditions (CTCAE v. 5.0)**	**Action taken to pembrolizumab**	**irAE management with corticosteroid and/or other therapies**	**Monitor and follow-up**
Pneumonitis	Grade 2	Withhold	• Administer corticosteroids (initial dose of 1-2 mg/kg prednisone or equivalent) followed by taper	• Monitor participants for signs and symptoms of pneumonitis • Evaluate participants with suspected pneumonitis with radiographic imaging and initiate corticosteroid treatment • Add prophylactic antibiotics for opportunistic infections
	Grade 3 or 4, or recurrent Grade 2	Permanently discontinue		

Immune-related AEs	**Toxicity grade or conditions (CTCAE v. 5.0)**	**Action taken to pembrolizumab**	**irAE management with corticosteroid and/or other therapies**	**Monitor and follow-up**
Diarrhea / Colitis	Grade 2 or 3	Withhold	• Administer corticosteroids (initial dose of 1-2 mg/kg prednisone or equivalent) followed by taper	• Monitor participants for signs and symptoms of enterocolitis (i.e., diarrhea, abdominal pain, blood or mucus in stool with or without fever) and of bowel perforation (i.e., peritoneal signs and ileus). • Participants with ≥ Grade 2 diarrhea suspecting colitis should consider GI consultation and performing endoscopy to rule out colitis. • 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.
	Grade 4	Permanently discontinue		
AST / ALT elevation or Increased bilirubin	Grade 2	Withhold	• Administer corticosteroids (initial dose of 0.5- 1 mg/kg prednisone or equivalent) followed by taper	• Monitor with liver function tests (consider weekly or more frequently until liver enzyme value returned to baseline or is stable)
	Grade 3 or 4	Permanently discontinue	• Administer corticosteroids (initial dose of 1-2 mg/kg prednisone or equivalent) followed by taper	

Immune-related AEs	**Toxicity grade or conditions (CTCAE v. 5.0)**	**Action taken to pembrolizumab**	**irAE management with corticosteroid and/or other therapies**	**Monitor and follow-up**
Type 1 diabetes mellitus (T1DM) or Hyperglycemia	Newly onset T1DM or Grade 3 or 4 hyperglycemia associated with evidence of β-cell failure	Withhold	• Initiate insulin replacement therapy for participants with T1DM • Administer anti-hyperglycemic in participants with hyperglycemia	• Monitor participants for hyperglycemia or other signs and symptoms of diabetes.
Hypophysitis	Grade 2	Withhold	• Administer corticosteroids and initiate hormonal replacements as clinically indicated.	• Monitor for signs and symptoms of hypophysitis (including hypopituitarism and adrenal insufficiency)
	Grade 3 or 4	Withhold or permanently discontinue[1]		
Hyperthyroidism	Grade 2	Continue	• Treat with non-selective beta-blockers (e.g., propranolol) or thionamides as appropriate	• Monitor for signs and symptoms of thyroid disorders.
	Grade 3 or 4	Withhold or permanently discontinue[1]		
Hypothyroidism	Grade 2-4	Continue	• Initiate thyroid replacement hormones (e.g., levothyroxine or liothyroinine) per standard of care	• Monitor for signs and symptoms of thyroid disorders.
Nephritis and Renal dysfunction	Grade 2	Withhold	• Administer corticosteroids (prednisone 1-2 mg/kg or equivalent) followed by taper.	• Monitor changes of renal function
	Grade 3 or 4	Permanently discontinue		
Myocarditis	Grade 1 or 2	Withhold	• Based on severity of AE	• Ensure adequate evaluation to

Immune-related AEs	**Toxicity grade or conditions (CTCAE v. 5.0)**	**Action taken to pembrolizumab**	**irAE management with corticosteroid and/or other therapies**	**Monitor and follow-up**
	Grade 3 or 4	Permanently discontinue	administer corticosteroids	confirm etiology and/or exclude other causes
All other immune-related AEs	Intolerable/ persistent Grade 2	Withhold	• Based on type and severity of AE administer corticosteroids	• Ensure adequate evaluation to confirm etiology and/or exclude other causes
	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 ≤ Grade 2 and is controlled with hormonal replacement therapy or achieved metabolic control (in case of T1DM).				

5.2.1 Dose Modification and Toxicity Management of Infusion-Reactions Related to Pembrolizumab

Pembrolizumab may cause severe or life-threatening infusion-reactions including severe hypersensitivity or anaphylaxis. Signs and symptoms usually develop during or shortly after drug infusion and generally resolve completely within 24 hours of completion of infusion. Dose modification and toxicity management guidelines on pembrolizumab associated infusion reaction are provided in Table 4.

Table 4: Pembrolizumab Infusion Reaction Dose modification and Treatment Guidelines

NCI CTCAE Grade	Treatment	Premedication at Subsequent Dosing
Grade 1 Mild reaction; infusion interruption not indicated; intervention not indicated	Increase monitoring of vital signs as medically indicated until the participant is deemed medically stable in the opinion of the investigator.	None
Grade 2 Requires therapy or infusion interruption but responds promptly to symptomatic treatment (e.g., antihistamines, NSAIDs, narcotics, IV fluids); prophylactic medications indicated for ≤ 24 hrs	**Stop Infusion.** 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 participant is deemed medically stable in the opinion of the investigator. If symptoms resolve within 1 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 participant should be premedicated for the next scheduled dose. **Participants who develop Grade 2 toxicity despite adequate premedication should be permanently discontinued from further study drug treatment**	Participant may be premedicated 1.5 h (± 30 minutes) prior to infusion of Pembrolizumab with: • Diphenhydramine 50 mg po (or equivalent dose of antihistamine). • Acetaminophen 500-1000 mg po (or equivalent dose of analgesic).

NCI CTCAE Grade	Treatment	Premedication at Subsequent Dosing
Grades 3 or 4 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	**Stop Infusion.** Additional appropriate medical therapy may include but is not limited to: • Epinephrine** • IV fluids • Antihistamines • NSAIDs • Acetaminophen • Narcotics • Oxygen • Pressors • Corticosteroids Increase monitoring of vital signs as medically indicated until the participant is deemed medically stable in the opinion of the investigator. Hospitalization may be indicated. **In cases of anaphylaxis, epinephrine should be used immediately. **Participant is permanently discontinued from further study drug treatment.**	No subsequent dosing
Appropriate resuscitation equipment should be available at the bedside and a physician readily available during the period of drug administration. For further information, please refer to the Common Terminology Criteria for Adverse Events v5.0 (CTCAE) at https://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm#ctc_50		

5.2.2 Other Allowed Dose Interruption for Pembrolizumab

Pembrolizumab may be interrupted for situations other than treatment-related AEs such as medical / surgical events or logistical reasons not related to study therapy. Participants should be placed back on study therapy within 12 weeks of the scheduled interruption, unless otherwise discussed with the Sponsor-Investigator. The reason for interruption should be documented in the patient's study record.

5.3 Randomization or Treatment Allocation

This is not a randomized study design. Eligible subjects defined in Sections 5.1.1 and 5.1.2 will be included in the study to receive pembrolizumab.

5.4 Stratification

There is no stratification of study participants.

5.5 Concomitant Medications/Vaccinations (allowed & prohibited)

Medications or vaccinations specifically prohibited in the exclusion criteria are not allowed during the ongoing trial. If there is a clinical indication for one of these or other medications or vaccinations specifically prohibited during the trial, discontinuation from trial therapy or vaccination may be required. The investigator must discuss any questions regarding this with the Sponsor-Investigator and medical monitor. The final decision on any supportive therapy or vaccination rests with the investigator and/or the participants' primary physician.

However, the decision to continue the subject on trial therapy or vaccination schedule requires the mutual agreement of the Sponsor-Investigator, the treating investigator, medical monitor and the subject.

5.5.1 Acceptable Concomitant Medications

All treatments that the investigator considers necessary for a participant's welfare may be administered at the discretion of the investigator in keeping with the community standards of medical care.

All concomitant medication will be recorded on the case report form (CRF) including all prescription, over-the-counter (OTC), herbal supplements, and IV medications and fluids. If changes occur during the trial period, documentation of drug dosage, frequency, route, and date may also be included on the CRF.

All concomitant medications received within 28 days before the first dose of trial treatment and 30 days after the last dose of trial treatment should be recorded. Concomitant medications administered after 30 days after the last dose of trial treatment should be recorded for SAEs and ECIs as defined in Section 7.3.

Additional surgery may be allowed at the investigator's discretion. Adjuvant radiation therapy to the regional nodal bed may be allowed at the investigator's discretion.

5.5.2 Prohibited Concomitant Medications

Participants are prohibited from receiving the following therapies during the Screening and Treatment Phase of this trial:

- Antineoplastic systemic chemotherapy or biological therapy
- Immunotherapy not specified in this protocol
- Chemotherapy not specified in this protocol
- Investigational agents other than pembrolizumab
- Radiation therapy
    - Note: Adjuvant radiation therapy to the regional nodal bed may be allowed at the investigator's discretion.
- Live vaccines within 30 days prior to the first dose of study treatment and while participating in the study. Examples of live vaccines include, but are not limited to, the following: measles, mumps, rubella, varicella/zoster, yellow fever, rabies, BCG, and typhoid vaccine. Seasonal influenza vaccines for injection are generally killed virus vaccines and are allowed; however, intranasal influenza vaccines (e.g., FluMist®) are live attenuated vaccines and are not allowed.
- Systemic glucocorticoids for any purpose other than to modulate symptoms from an event of clinical interest of suspected immunologic etiology. The use of physiologic doses of corticosteroids may be approved after consultation with the Sponsor-Investigator.

Participants who, in the assessment by the investigator, require the use of any of the aforementioned treatments for clinical management should be removed from the study. All treatments that the Investigator considers necessary for a participant's welfare may be administered at the discretion of the Investigator in keeping with the community standards of medical care. NOTE: See Administrative Clarification Letter dated August 27, 2024, regarding continued monitoring of participants following discontinuation of protocol treatment (clarification that participants will not be removed from the study for the reasons listed above).

The Exclusion Criteria describes other medications which are prohibited in this trial.
There are no prohibited therapies during the Post-Treatment Follow-up Phase.

5.5.3 Rescue Medications & Supportive Care

Participants should receive appropriate supportive care measures as deemed necessary by the treating investigator. Suggested supportive care measures for the management of AEs with potential immunologic etiology are outlined along with the dose modification guidelines in Section 5.2.2, [Table 3]. Where appropriate, these guidelines include the use of oral or IV treatment with corticosteroids, as well as additional anti-inflammatory agents if symptoms do not improve with administration of corticosteroids. Note that several courses of steroid tapering may be necessary as symptoms may worsen when the steroid dose is decreased. For each disorder, attempts should be made to rule out other causes such as metastatic disease or bacterial or viral infection, which might require additional supportive care. The treatment guidelines are intended to be applied when the Investigator determines the events to be related to pembrolizumab.

Note: If after the evaluation of the event, it is determined not to be related to pembrolizumab, the Investigator does not need to follow the treatment guidance. Refer to [Table 3] in Section 5.2.2 for guidelines regarding dose modification and supportive care.

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

5.6 Participant Withdrawal/Discontinuation Criteria

Participants may discontinue study treatment at any time for any reason or be dropped from the study treatment at the discretion of the investigator should any untoward effect occur. In addition, a participant may be discontinued from study treatment by the investigator or the Sponsor-Investigator if study treatment is inappropriate, the trial plan is violated, or for administrative and/or other safety reasons. Specific details regarding procedures to be performed at study treatment discontinuation are provided in Section 7.2.7 – Other Procedures.

A participant must be discontinued from study treatment but continue to be monitored in the study for any of the following reasons:

- The participant or participant's legally acceptable representative requests to discontinue study treatment
- Confirmed radiographic disease recurrence outlined in Section 7.2.5.13
- Any progression or recurrence of any malignancy, or any occurrence of another malignancy that requires active ongoing systemic treatment
- Unacceptable adverse experiences as described in Section 5.2.2.
- The participant has a medical condition or personal circumstance which, in the opinion of the investigator and/or Sponsor-Investigator, placed the participant at unnecessary risk from continued administration of study treatment.
- The participant has a confirmed positive serum pregnancy test
- Noncompliance with study treatment or procedure requirements
- Recurrent Grade 2 pneumonitis
- The participant is lost to follow-up
- Administrative reasons

After the end of treatment, each subject will be followed for 30 days for adverse event monitoring (serious adverse events will be collected for 90 days after the end of treatment as described in Section 7.3). Subjects will have post-treatment follow-up for disease status until disease recurrence, withdrawing consent or becoming lost to follow-up. After documented disease progression, survival information will be extracted from the medical record or obtained via telephone contact until death, withdrawal of consent, becoming lost to follow-up or the end of the study, whichever occurs first. Survival follow-up will continue for up to 5 years.

5.7 Participant Replacement Strategy

All patients who receive neoadjuvant pembrolizumab and undergo definitive surgery with SLN biopsy are evaluable. We will continue to enroll until 63 evaluable patients have been enrolled. Non-evaluable patients can be replaced.

5.8 Bayesian Early Termination Rules for Toxicity

The study will be terminated early if there is evidence that the rate of dose limiting toxicity is greater than 33%. A DLT is defined as any grade 3 or higher non-hematologic or grade 4 hematologic toxicity that is at least possibly related to pembrolizumab and is not an expected adverse event for pembrolizumab or surgery and which occurs anytime from the initiation of study therapy to 6 weeks post-surgery.

Early termination rules will be used to monitor DLTs during the enrollment and treatment of patients. Toxicity data will likely be reviewed after groups of 5 patients have been treated and followed for 6 weeks post-surgery. Enrollment will be terminated if the data indicate that the rate of DLTs, as defined above, is >33%. There are published data on the safety of pembrolizumab in other populations of melanoma patients. Thus, we will assume a modestly informative beta (.5,1.5) prior which is equivalent to information on 2 treated patients. Given the prior and the observed data, the Bayesian posterior probability that the toxicity rate exceeds 33%, is calculated. If the number of patients with a DLT **equals or exceeds** the number in the table below, then it is likely that the toxicity rate is unacceptable, as noted by Bayesian posterior probabilities. NOTE: See Administrative Clarification Letter dated August 27, 2024, regarding discontinuation of 6-week post-operative toxicity data review (no additional meetings will be scheduled).

Patients treated	**5**	**10**	**15**	**20**	**25**	**30**
Patients with DLT	3	5	7	9	11	13
Posterior Prob [DLT rate >33%]	0.81	0.81	0.82	0.83	0.84	0.85
Patients treated	**35**	**40**	**45**	**50**	**55**	**60**
Patients with DLT	15	16	18	20	22	24
Posterior Prob [DLT rate >33%]	0.86	0.80	0.81	0.83	0.84	0.85

5.9 Clinical Criteria for Early Trial Termination

Early trial termination will be the result of the criteria specified below:

1. Quality or quantity of data recording is inaccurate or incomplete
2. Poor adherence to protocol and regulatory requirements
3. Incidence or severity of adverse drug reaction in this or other studies indicates a potential health hazard to participants
4. Plans to modify or discontinue the development of the study drug

In the event of Merck decision to no longer supply study drug, ample notification will be provided so that appropriate adjustments to participant treatment can be made.

6.0 TRIAL FLOW CHART

6.1 Study Flow Chart

Trial Period:	**Pre-Treatment**	**Treatment[1]**																		**Post-Treatment Follow-up**	
Timepoint	**Screening**	**C1**	**Surgery**	**C2[1]**	**C3**	**C4**	**C5**	**C6**	**C7**	**C8**	**C9**	**C10**	**C11**	**C12**	**C13**	**C14**	**C15**	**C16**	**C17**	**Safety[2]**	**Survival[5]**
Administrative Procedures																					
Informed Consent[3]	X																				
Inclusion/Exclusion Criteria	X																				
Demographics and Medical History[4]	X																				
Prior / Concomitant Medication Review[4]	X	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
Clinical Procedures/Assessments																					
Review Adverse Events[6]		X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
Full Physical Examination	X																				
Directed Physical Examination		X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
Vital Signs and Weight[7]	X	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
ECOG Performance Status	X	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
Trial Treatment Administration[1]		X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X		
Post-treatment anticancer therapy status																				X	
Survival Status[5]																				X	X

Trial Period:	Pre-Treatment	Treatment[1]																		Post-Treatment Follow-up	
Timepoint	**Screening**	**C1**	**Surgery**	**C2[1]**	**C3**	**C4**	**C5**	**C6**	**C7**	**C8**	**C9**	**C10**	**C11**	**C12**	**C13**	**C14**	**C15**	**C16**	**C17**	**Safety[2]**	**Survival[5]**
Laboratory Procedures/Assessments: analysis performed by LOCAL laboratory[8]																					
Pregnancy Test – Urine or Serum β- HCG[9]	X																				
PT/INR and aPTT	X																				
CBC with Differential	X	X[18]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
Comprehensive Serum Chemistry Panel[10]	X	X[18]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
TSH[11]	X	X[18]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
HLA type[12]		X																			
Collection of Primary Tumor Specimen[15]			X																		
Archival Tumor Tissue[13]	X																				
Efficacy Measurements																					
Tumor Imaging[14]	X[14]						X[14]				X[14]				X[14]				X[14]		X[14]
Wide Excision and SLN Biopsy/Correlative Studies Blood																					
Wide Excision and SLN Biopsy[15]			X																		
Blood collection for DNA (at Penn only; up to 10 ml)[16]		X																			
Correlative Studies Blood Collection for PBMC/plasma (four (4) EDTA tubes; up to 120 ml per draw)[17]		X[17]	X	X	X	X												X[17]			

[1] Cycle 2 pembrolizumab may occur up to 9 weeks after Cycle 1 to allow for full surgical recovery. All subsequent cycles will be every 21 days (+/-5 days) up to 17 cycles (approximately 1 year).

[2] The mandatory Safety Follow-Up visit will be conducted within 30 days after the last dose of study drug or before the initiation of a new antineoplastic treatment, whichever comes first. Patients with an AE of Grade >1 will be further followed until the resolution of the AE to Grade 0-1 or until beginning of a new antineoplastic therapy, whichever occurs first.

Trial Period:	**Pre-Treatment**	**Treatment[1]**																		**Post-Treatment Follow-up**	
Timepoint	**Screening**	**C1**	**Surgery**	**C2[1]**	**C3**	**C4**	**C5**	**C6**	**C7**	**C8**	**C9**	**C10**	**C11**	**C12**	**C13**	**C14**	**C15**	**C16**	**C17**	**Safety[2]**	**Survival[5]**

[3] Written consent must be obtained prior to performing any protocol specific procedure. Results of a test performed as part of routine clinical management are acceptable in lieu of a screening test if performed within the specified time frame (e.g. within 28 days prior to Cycle 1, Day 1).

[4] Includes history of treatment for the primary diagnosis, including prior systemic, radiation treatment and surgical treatment, and best response to prior systemic treatments. Date of last prior cancer treatment must be documented. Radiographic studies performed prior to study entry may be collected for review by the investigator. Report complete medication history for 28 days prior to the screening visit (Visit 1).

[5] Once a subject completes 12 months of pembrolizumab therapy, experiences disease recurrence or starts a new antineoplastic therapy, the subject will complete the below noted safety follow up and move into the survival follow-up phase. Survival information will be extracted from the medical record or obtained via telephone contact approximately every 12 weeks until death, withdrawal of consent, becoming lost to follow-up, or the end of the study, whichever occurs first. Survival follow-up will continue for up to 5 years.

[6] Adverse experiences and laboratory safety measurements will be graded per NCI CTCAE version 5.0. All adverse experiences, whether gradable by CTCAE or not, will also be evaluated for seriousness. AEs occurring within 30 days after the last dose of study drug should be recorded. After this time, record only AEs that are considered related to study drug. SAEs that occur within 90 days of the end of treatment or before initiation of a new anti-cancer treatment should also be followed and recorded.

[7] Vital signs to include temperature, pulse, respiratory rate and blood pressure.

[8] Routine laboratory tests (serum chemistry; hematology) for screening should be performed within 21 days of Cycle 1 Day 1. After cycle 1, pre-treatment labs can be up to 72 hours prior to treatment.

[9] For women of reproductive potential, a urine or serum pregnancy test will be performed within 72 hours of the first dose. If the urine test is positive or cannot be confirmed as negative, a serum pregnancy test will be required. Women with amenorrhea for <1 years and who have not had a hysterectomy and oophorectomy will only be considered not to be of reproductive potential if they have a documented FSH value (2 times) in the postmenopausal range.

[10] Comprehensive serum chemistry panel must include sodium, potassium, BUN, Creatinine, AST, ALT, alkaline phosphatase, and total bilirubin.

[11] TSH is recommended at screening and with each cycle; T3 and T4 as indicated.

[12] HLA can be drawn once, at the time of any scheduled blood draw, or may be omitted, per PI decisions.

[13] Pre-treatment tumor sample (archival tissue) may be obtained at any point during the subject's participation; one (1) digital hematoxylin and eosin (H&E) slide and twenty (20) 5 µm unstained slides from the pre-treatment biopsy.

[14] Tumor imaging will be performed at time points in accordance with standard of care which is within 30 days prior to enrollment and every 3-6 months on and post-treatment.

[15] Wide excision and SLN biopsy for treatment of patients' melanoma will occur 3 weeks + 1 week, - 3 days after the first dose of pembrolizumab. **Post-treatment tumor and SLN sample**, if present, will be obtained at time of Wide excision and SLN biopsy after thorough clinical diagnostics performed on the sample. **Surgical specimen**: ten (10) unstained slides from the primary tumor, 25 unstained slides from the sentinel lymph node, twenty (20) unstained slides from the positive nodes, twenty (20) unstained slides from up to three (3) negative nodes, one (1) digital hematoxylin and eosin (H&E) slide from each node

[16] DNA sample for analysis should be obtained pre-dose at Cycle 1 (or with any subsequent scheduled blood draw).

[17] Whole blood samples should be obtained pre-dose at Cycle 1, Date of Surgery, Cycles 2, 3 and 4, with a final sample at Cycle 16 or at the time of recurrence, if available. Four 10 ml EDTA tubes (or the equivalent) should be collected at each timepoint.

[18] Screening labs can be used if done within 21 days of Cycle 1.

7.0 TRIAL PROCEDURES

7.1 Trial Procedures

The Trial Flow Chart - Section 6.1 summarizes the trial procedures to be performed at each visit. Individual trial procedures are described in detail below. It may be necessary to perform these procedures at unscheduled time points if deemed clinically necessary by the investigator.

Furthermore, additional evaluations/testing may be deemed necessary by the Sponsor-Investigator and/or Merck for reasons related to participant safety. In some cases, such evaluation/testing may be potentially sensitive in nature (e.g., HIV, Hepatitis C, etc.), and thus local regulations may require that additional informed consent be obtained from the participant. In these cases, such evaluations/testing will be performed in accordance with those regulations.

7.2 Administrative Procedures

7.2.1 Informed Consent

The Investigator or qualified designee must obtain documented consent from each potential participant prior to participating in a clinical trial.

7.2.1.1 General Informed Consent

Consent must be documented by the participant's dated signature or by the participant's legally acceptable representative's dated signature on a consent form along with the dated signature of the person conducting the consent discussion.

A copy of the signed and dated consent form should be given to the participant before participation in the trial.

The initial informed consent form, any subsequent revised written informed consent form and any written information provided to the participant must receive the IRB/ERC's approval/favorable opinion in advance of use. The participant or his/her legally acceptable representative should be informed in a timely manner if new information becomes available that may be relevant to the participant's willingness to continue participation in the trial. The communication of this information will be provided and documented via a revised consent form or addendum to the original consent form that captures the participant's dated signature or by the participant's legally acceptable representative's dated signature.

Specifics about a trial and the trial population will be added to the consent form template at the protocol level.

The informed consent will adhere to IRB/ERC requirements, applicable laws and regulations and Sponsor requirements.

7.2.2 Inclusion/Exclusion Criteria

All inclusion and exclusion criteria will be reviewed by the investigator or qualified designee to ensure that the participant qualifies for the trial.

7.2.3 Medical History

A medical history will be obtained by the investigator or qualified designee. Medical history will include all active conditions, and any condition diagnosed within the prior 10 years that are considered to be clinically significant by the Investigator. Details regarding the disease for which the participant has enrolled in this study will be recorded separately and not listed as medical history.

7.2.4 Prior and Concomitant Medications Review

7.2.4.1 Prior Medications

The investigator or qualified designee will review prior medication use, including any protocol-specified washout requirement, and record prior medication taken by the participant within 28 days before starting the trial. Treatment for the disease for which the participant has enrolled in this study will be recorded separately and not listed as a prior medication.

7.2.4.2 Concomitant Medications

The investigator or qualified designee will record medication, if any, taken by the participant during the trial. All medications related to reportable SAEs and ECIs should be recorded as defined in Section 7.3.

7.2.5 Disease Details and Treatments

7.2.5.1 Disease Details

The investigator or qualified designee will obtain prior and current details regarding disease status.

7.2.5.2 Prior Treatment Details

The investigator or qualified designee will review all prior cancer treatments including systemic treatments, radiation and surgeries.

7.2.5.3 Subsequent Anti-Cancer Therapy Status

The investigator or qualified designee will review all new anti-neoplastic therapy initiated after the last dose of trial treatment. If a participant initiates a new anti-cancer therapy within 30 days after the last dose of trial treatment, the 30-day Safety Follow-up visit must occur before the first dose of the new therapy. Once new anti-cancer therapy has been initiated the participant will move into survival follow-up.

7.2.5.4 Assignment of Screening Number

Patients will be assigned a screening number upon enrollment.

7.2.5.5 Assignment of Randomization Number

There will be no randomization; not relevant for this protocol.

7.2.5.6 Trial Compliance (Medication/Diet/Activity/Other)

Trial monitoring and adherence will be performed per institutional protocol.

7.2.5.7 Clinical Procedures/Assessments

Wide excision and sentinel lymph node biopsy and pathologic assessment of tissue will be performed per standard of care.

7.2.5.8 Adverse Event (AE) Monitoring

The investigator or qualified designee will assess each participant to evaluate for potential new or worsening AEs as specified in the Trial Flow Chart and more frequently if clinically indicated. Adverse experiences will be graded and recorded throughout the study and during the follow-up period according to NCI CTCAE Version 5.0 (see Appendix 2). Toxicities will be characterized in terms regarding seriousness, causality, toxicity grading, and action taken with regard to trial treatment.

Please refer to section 7.3 for detailed information regarding the assessment and recording of AEs.

7.2.5.9 Full Physical Exam

The investigator or qualified designee will perform a complete physical exam during the screening period. Clinically significant abnormal findings should be recorded as medical history. A full physical exam should be performed during screening.

7.2.5.10 Directed Physical Exam

For cycles that do not require a full physical exam per the Trial Flow Chart, the investigator or qualified designee will perform a directed physical exam as clinically indicated prior to trial treatment administration.

7.2.5.11 Vital Signs

The investigator or qualified designee will take vital signs at screening, prior to the administration of each dose of trial treatment and at treatment discontinuation as specified in the Trial Flow Chart (Section 6.1). Vital signs should include temperature, pulse, respiratory rate, weight and blood pressure.

7.2.5.12 Eastern Cooperative Oncology Group (ECOG) Performance Scale

The investigator or qualified designee will assess ECOG status (see Appendix 1) at screening, prior to the administration of each dose of trial treatment and discontinuation of trial treatment as specified in the Trial Flow Chart.

7.2.5.13 Tumor Imaging and Assessment of Disease

Tumor imaging is strongly preferred to be acquired by computed tomography (CT). PET/CT may be used. For the abdomen and pelvis, contrast-enhanced magnetic resonance imaging (MRI) may be used when CT with iodinated contrast is contraindicated, or when local practice mandates it. For the regional nodal bed, ultrasound imaging may be used.

Participant eligibility will be determined using local assessment (Investigator assessment). In addition, images (including via other modalities) that are obtained at an unscheduled time point to determine disease progression, as well as imaging obtained for other reasons, but which demonstrate radiologic progression, should also be used to determine progression.

When the Investigator identifies progression, treatment may be continued until PD has been verified.

7.2.5.13.1 Initial Tumor Imaging

Initial imaging at screening must be performed within 30 days prior to the date of enrolment. The site study team must review screening images to confirm the participant is eligible.

Tumor imaging performed as part of routine clinical management is acceptable for use as screening tumor imaging if performed within 30 days prior to the date of enrolment.

7.2.5.13.2 Tumor Imaging During the Study

The first on-study imaging assessment should be performed 3 to 6 months from the first day of protocol treatment. Subsequent tumor imaging should be performed every 3 to 6 months as per standard clinical care or more frequently if clinically indicated. Imaging timing should follow calendar days and should not be adjusted for delays in cycle starts. Imaging should continue to be performed until disease progression is identified by the Investigator.

7.2.5.13.3 End of Treatment and Follow-up Tumor Imaging

In participants who discontinue study treatment, tumor imaging should be performed per clinical standard of care. In participants who discontinue study treatment due to documented disease progression, this is the final required tumor imaging.

In participants who discontinue study treatment without documented disease progression, every effort should be made to continue monitoring their disease status by tumor imaging using the same imaging schedule used while on treatment (every 3 to 6 months) to monitor disease status until the start of a new anticancer treatment, disease progression, pregnancy, death, withdrawal of consent, or the end of the study, whichever occurs first.

7.2.5.14 Tumor Tissue Collection and Correlative Studies Blood Sampling

Biosamples (blood and tumor tissue) obtained from subjects before and at serial time points during treatment will be analyzed. Tissue will be collected according to the Trial Flow Chart - Section 6.1. Biosamples will be used for assessment as per Section 4.2.3.3.

7.2.6 Laboratory Procedures/Assessments

Laboratory tests for screening should be performed within 21 days prior to the first dose of treatment. After Cycle 1, pre-dose laboratory procedures can be conducted up to 72 hours prior to dosing. Results must be reviewed by the investigator or qualified designee and found to be acceptable prior to each dose of trial treatment.

Details regarding specific laboratory procedures/assessments to be performed in this trial are provided below.

Laboratory tests for hematology, chemistry, and others are specified in Table 6.

Table 6: Laboratory Tests

Hematology	**Chemistry**	**Other**
Hematocrit	Albumin	Serum or urine β-human chorionic gonadotropin†
Hemoglobin	Alkaline phosphatase	(β-hCG)†
Platelet count	Alanine aminotransferase (ALT)	PT (INR)
WBC (total and differential)	Aspartate aminotransferase (AST)	aPTT
Red Blood Cell Count	Carbon Dioxide ‡	Total thriiodothyronine (T3)
Absolute Neutrophil Count	*(CO_2 or bicarbonate)*	Free thyroxine (T4)
Absolute Lymphocyte Count	Calcium	Thyroid stimulating hormone (TSH)
	Chloride	
	Glucose	
	Potassium	Blood for correlative studies
	Sodium	
	Total Bilirubin	
	Total protein	
	Blood Urea Nitrogen	

7.2.7 Other Procedures

7.2.7.1 Withdrawal/Discontinuation

When a participant discontinues/withdraws prior to trial completion, all applicable activities scheduled for the final trial visit should be performed at the time of discontinuation. Any adverse events which are present at the time of discontinuation/withdrawal should be followed in accordance with the safety requirements outlined in Section 7.3 - Assessing and Recording Adverse Events. After discontinuing treatment, participants should return to the site for a Safety Follow-up Visit (described in Section 7.2.8.3.1) and then proceed to the Follow-Up Period of the study (described in Section 7.2.8.3.2).

7.2.7.2 Blinding/Unblinding

This is not relevant to the current study.

7.2.8 Visit Requirements

Visit requirements are outlined in Section 6.1 - Trial Flow Chart. Specific procedure-related details are provided above in Section 7.1 - Trial Procedures.

7.2.8.1 Screening

Screening requirements are outlined in Section 6.1 - Trial Flow Chart. If the subject meets all eligibility requirements, the subject can be enrolled in the study

7.2.8.1.1 Screening Period

Subjects may be rescreened after initially failing to meet the inclusion/exclusion criteria. Results from assessments performed during the initial screening period are acceptable in lieu of a repeating a screening test if performed within the specified time frame and the results meet the inclusion/exclusion criteria.

7.2.8.2 Treatment Period

Subjects who have signed informed consent and are eligible for the study can be enrolled on the study and can proceed to receive the week 0 (cycle 1) treatment with pembrolizumab.

Subjects must then undergo a wide excision and sentinel lymph node biopsy within 3 weeks (up to 1 week later, or up to 3 days earlier) after C1 of pembrolizumab treatment date.

If the surgery is delayed by more than one week after the 3-week window (more than 28 days after the first dose of pembrolizumab), the patient may continue with surgery and pembrolizumab afterwards. Patients are expected to initiate adjuvant pembrolizumab within 9 weeks of cycle 1 to allow for post-surgical recovery. Patients will be evaluated by both the surgeon and the oncologist or qualified designee to determine adequate surgical recovery before initiation of study drug. In the event of delays, there is no timeframe restriction in terms of when adjuvant pembrolizumab is initiated.

Up to 17 cycles (approximately 1 year) of pembrolizumab are given every 3 weeks after C2 per the Trial Flow Chart, Section 6.1.

7.2.8.3 Post-Treatment Visits

Subjects are evaluated within 4 weeks of their last dose of pembrolizumab and then according to the Trial Flow Chart, Section 6.1.

7.2.8.3.1 Safety Follow-Up Visit

The mandatory Safety Follow-Up Visit should be conducted approximately 30 days after the last dose of study treatment or before the initiation of a new anti-cancer treatment, whichever comes first. All AEs that occur prior to the Safety Follow-Up Visit should be recorded.

Participants with an AE of Grade > 1 will be followed until the resolution of the AE to Grade 0-1 or until the beginning of a new anti-cancer therapy, whichever occurs first. SAEs that occur within 90 days of the end of treatment or before initiation of a new anti-cancer treatment should also be followed and recorded.

7.2.8.3.2 Follow-up Visits

Participants who discontinue study treatment for a reason other than disease progression will move into the Follow-Up Phase and should be assessed according to standard of care.

7.2.8.3.3 Survival Follow-up

Participants who experience confirmed disease progression or start a new anticancer therapy, will move into the Survival Follow-Up Phase. Survival information will be extracted from the medical record or obtained via telephone contact approximately every 12 weeks until death, withdrawal of consent, becoming lost to follow-up or the end of the trial, whichever occurs first. Survival follow-up will continue for up to 5 years.

7.3 Assessing and Recording Adverse Events

An adverse event is defined as any untoward medical occurrence in a patient or clinical investigation participant administered a pharmaceutical product and which does not necessarily have to have a causal relationship with this treatment. An adverse event can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding, for example), symptom, or disease temporally associated with the use of a medicinal product or protocol-specified procedure, whether or not considered related to the medicinal product or protocol-specified procedure. Any worsening (i.e., any clinically significant adverse change in frequency and/or intensity) of a preexisting condition that is temporally associated with the use of the Merck's product, is also an adverse event.

Changes resulting from normal growth and development that do not vary significantly in frequency or severity from expected levels are not to be considered adverse events.

Examples of this may include, but are not limited to onset of menses or menopause occurring at a physiologically appropriate time.

Merck product includes any pharmaceutical product, biological product, device, diagnostic agent or protocol-specified procedure, whether investigational (including placebo or active comparator medication) or marketed, manufactured by, licensed by, provided by or distributed by Merck for human use.

Adverse events may occur during the course of the use of Merck product in clinical trials, or as prescribed in clinical practice, from overdose (whether accidental or intentional), from abuse and from withdrawal.

- All AEs, SAEs and other reportable safety events that occur after the consent form is signed but before treatment allocation/randomization must be reported by the investigator if the event causes the participant to be excluded from the study, or is the result of a protocol-specified intervention, including but not limited to washout or discontinuation of usual therapy, diet, or a procedure.
- All AEs from the time of treatment allocation/randomization through 30 days following cessation of study treatment must be reported by the investigator.
- All AEs meeting serious criteria, from the time of treatment allocation/randomization through 90 days following cessation of study treatment, or 30 days following cessation of study treatment if the participant initiates new anticancer therapy, whichever is earlier must be reported by the investigator.
- All pregnancies and exposure during breastfeeding, from the time of treatment allocation/randomization through 120 days following cessation of study treatment, or 30 days following cessation of study treatment if the participant initiates new anticancer therapy must be reported by the investigator.
- Additionally, any SAE brought to the attention of an investigator at any time outside of the time period specified above must be reported immediately by the investigator if the event is considered to be drug-related.

Investigators are not obligated to actively seek AE or SAE or other reportable safety events in former study participants. However, if the investigator learns of any SAE, including a death, at any time after a participant has been discharged from the study, and he/she considers the event to be reasonably related to the study treatment or study participation, the investigator must promptly notify the Sponsor-Investigator who will notify Merck.

7.3.1 Definition of an Overdose for This Protocol and Reporting of Overdose to the Sponsor and to Merck

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

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

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

All reports of overdose with and without an adverse event must be reported within 24 hours to the Sponsor-Investigator and within 2 working days to Merck Global Safety. (Attn: Worldwide Product Safety; FAX 215-661-6229)

7.3.2 Reporting of Pregnancy and Lactation to the Sponsor and to Merck

Although pregnancy and infant exposure during breast feeding are not considered adverse events, it is the responsibility of investigators or their designees to report any pregnancy or lactation in a participant (spontaneously reported to them) that occurs during the study.

Pregnancies and infant exposures during breastfeeding that occur after the consent form is signed but before treatment allocation/randomization must be reported by the investigator if they cause the participant to be excluded from the trial, or are the result of a protocol- specified intervention, including but not limited to washout or discontinuation of usual therapy, diet, placebo treatment or a procedure.

Pregnancies and infant exposures during breastfeeding that occur from the time of treatment allocation/randomization through 120 days following cessation of study treatment, or 30 days following cessation of treatment if the participant initiates new anticancer therapy, whichever is earlier, must be reported by the investigator. All reported pregnancies 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.

Such events must be reported within 24 hours to the Sponsor-Investigator and within 2 working days to Merck Global Safety. (Attn: Worldwide Product Safety; FAX 215-661- 6229)

7.3.3 Immediate Reporting of Adverse Events to the Sponsor and to Merck

7.3.3.1 Serious Adverse Events

A serious adverse event is any adverse event occurring at any dose or during any use of Merck's product that:

- Results in death;
- Is life threatening;
- Results in persistent or significant disability/incapacity;
- Results in or prolongs an existing inpatient hospitalization;
- Is a congenital anomaly/birth defect;
- Is another important medical event
- **Note:** In addition to the above criteria, adverse events meeting either of the below criteria, although not serious per ICH definition, are reportable to the Merck in the same timeframe as SAEs to meet certain local requirements. Therefore, these events are considered serious by Merck for collection purposes.
- Is a new cancer (that is not a condition of the study);
- Is associated with an overdose.

Refer to Table 7 for additional details regarding each of the above criteria.

For the time period beginning when the consent form is signed until treatment allocation/randomization, any serious adverse event, or follow up to a serious adverse event, including death due to any cause that occurs to any participant must be reported within 24 hours to the Sponsor-Investigator and within 2 working days to Merck Global Safety if it causes the participant 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 participant initiates new anticancer therapy, whichever is earlier, any serious adverse event, or follow up to a serious adverse event, including death due to any cause whether or not related to the Merck product, must be reported within 24 hours to the Sponsor-Investigator and within 2 working days to Merck Global Safety.

Additionally, any serious adverse event, considered by an investigator or qualified designee to be related to Merck product that is brought to the attention of the investigator at any time following consent through the end of the specified safety follow-up period specified in the paragraph above, or at any time outside of the time period specified in the previous paragraph also must be reported immediately to the Sponsor-Investigator who will notify Merck Global Safety.

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

SAE reports and any other relevant safety information are to be forwarded to the Merck Global Safety facsimile number: +1-215-661-6229

A copy of all 15 Day Reports and Annual Progress Reports is submitted as required by FDA, European Union (EU), Pharmaceutical and Medical Devices agency (PMDA) or other local regulators. Investigators will cross reference this submission according to local regulations to the Merck Investigational Compound Number (IND, CSA, etc.) at the time of submission.

Additionally, investigators will submit a copy of these reports to Merck & Co., Inc. (Attn: Worldwide Product Safety; FAX 215-661-6229) at the time of submission to FDA.

7.3.3.2 Events of Clinical Interest

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-Investigator and within 2 working days to Merck Global Safety. (Attn: Worldwide Product Safety; FAX 215-661-6229).

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 participant must be reported within 2 working days to the Sponsor-Investigator and to Merck Global Safety if it causes the participant 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 participant

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-Investigator and within 2 working days to Merck Global Safety.

Events of clinical interest for this trial include:

1. an overdose of Merck product, as defined in Section 7.3.1 - Definition of an Overdose for This Protocol and Reporting of Overdose to the Sponsor-Investigator, that is not associated with clinical symptoms or abnormal laboratory results.

2. 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.

7.3.4 Evaluating Adverse Events

An investigator or qualified designee will evaluate all adverse events according to the NCI Common Terminology for Adverse Events (CTCAE), version 5.0. Any adverse event which changes CTCAE grade over the course of a given episode will have each change of grade recorded on the adverse event case report forms/worksheets.

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

Table 7: Evaluating Adverse Events

An investigator or qualified designee, will evaluate all adverse events as to:

V5.0 CTCAE Grading	**Grade 1**	**Mild; asymptomatic or mid symptoms; clinical or diagnostic observations only; intervention not indicated.**
	Grade 2	**Moderate; minimal, local or noninvasive intervention indicated; limiting age-appropriate instrumental ADL.**
	Grade 3	**Severe or medically significant but not immediately life-threatening; hospitalization or prolongation or hospitalization indicated; disabling; limiting self-care ADL.**
	Grade 4	**Life threatening consequences; urgent intervention indicated.**
	Grade 5	**Death related to AE**
Seriousness	A serious adverse event is any adverse event occurring at any dose or during any use of Merck product that:	
	†**Results in death**; or	
	†**Is life threatening; or** places the participant, in the view of the investigator, at immediate risk of death from the event as it occurred (Note: This does not include an adverse event that, had it occurred in a more severe form, might have caused death.); or	
	†**Results in a persistent or significant disability/incapacity** (substantial disruption of one's ability to conduct normal life functions); or	
	†**Results in or prolongs an existing inpatient hospitalization** (hospitalization is defined as an inpatient admission, regardless of length of stay, even if the hospitalization is a precautionary measure for continued observation. (Note: Hospitalization for an elective procedure to treat a pre-existing condition that has not worsened is not a serious adverse event. A pre-existing condition is a clinical condition that is diagnosed prior to the use of a Merck product and is documented in the patient's medical history.); or	
	†**Is a congenital anomaly/birth defect** (in offspring of participant taking the product regardless of time to diagnosis);or	
	Is a new **cancer** (that is not a condition of the study) **(**although not serious per ICH definition, is reportable to the Sponsor-Investigator within 24 hours and to Merck within 2 working days to meet certain local requirements); **or**	

	Is an overdose (whether accidental or intentional). Any adverse event associated with an overdose is considered a serious adverse event for collection purposes. An overdose that is not associated with an adverse event is considered a non-serious event of clinical interest and must be reported within 24 hours to the Sponsor-Investigator and to Merck within 2 working days..	
	Other important medical events that may not result in death, not be life threatening, or not require hospitalization may be considered a serious adverse event when, based upon appropriate medical judgment, the event may jeopardize the participant and may require medical or surgical intervention to prevent one of the outcomes listed previously (designated above by a †).	
	Record the start and stop dates of the adverse event. If less than 1 day, indicate the appropriate length of time and units	
Duration	Did the adverse event cause Merck product to be discontinued?	
Action taken	Did Merck product cause the adverse event? The determination of the likelihood that Merck product caused the adverse event will be provided by an investigator or qualified designee. The investigator's signed/dated initials on the source document or worksheet that supports the causality noted on the AE form, ensures that a medically qualified assessment of causality was done. This initialed document must be retained for the required regulatory time frame. The criteria below are intended as reference guidelines to assist the investigator in assessing the likelihood of a relationship between the test drug and the adverse event based upon the available information.	
Relationship to Merck Product	**The following components are to be used to assess the relationship between Merck product and the AE**; the greater the correlation with the components and their respective elements (in number and/or intensity), the more likely Merck product caused the adverse event (AE):	
	Exposure	Is there evidence that the participant was actually exposed to Merck product such as: reliable history, acceptable compliance assessment (pill count, diary, etc.), expected pharmacologic effect, or measurement of drug/metabolite in bodily specimen?
	Time Course	Did the AE follow in a reasonable temporal sequence from administration of Merck product? Is the time of onset of the AE compatible with a drug-induced effect (applies to trials with investigational medicinal product)?
	Likely Cause	Is the AE not reasonably explained by another etiology such as underlying disease, other drug(s)/vaccine(s), or other host or environmental factors

Relationship to Merck Product (continued)	**The following components are to be used to assess the relationship between the test drug and the AE: (continued)**	
	Dechallenge	Was Merck product discontinued or dose/exposure/frequency reduced? If yes, did the AE resolve or improve? If yes, this is a positive dechallenge. If no, this is a negative dechallenge. (Note: This criterion is not applicable if: (1) the AE resulted in death or permanent disability; (2) the AE resolved/improved despite continuation of the study treatment; or (3) the trial is a single-dose drug trial); or (4) investigational product(s) is/are only used one time.)
	Rechallenge	Was the participant re-exposed to Merck product in this study? If yes, did the AE recur or worsen? If yes, this is a positive rechallenge. If no, this is a negative rechallenge. (Note: This criterion is not applicable if: (1) the initial AE resulted in death or permanent disability, or (2) the trial is a single-dose drug trial); or (3) investigational product(s) is/are used only one time). NOTE: IF A RECHALLENGE IS PLANNED FOR AN ADVERSE EVENT WHICH WAS SERIOUS AND WHICH MAY HAVE BEEN CAUSED BY MERCK PRODUCT, OR IF REEXPOSURE TO MERCK PRODUCT POSES ADDITIONAL POTENTIAL SIGNIFICANT RISK TO THE PARTICIPANT, THEN THE RECHALLENGE MUST BE APPROVED IN ADVANCE BY THE SPONSOR-INVESTIGATOR AS PER DOSE MODIFICATION GUIDELINES IN THE PROTOCOL.
	Consistency with Trial Treatment Profile	Is the clinical/pathological presentation of the AE consistent with previous knowledge regarding Merck product or drug class pharmacology or toxicology?

The assessment of relationship will be reported on the case report forms /worksheets by an investigator or qualified designee according to his/her best clinical judgment, including consideration of the above elements.	
Record one of the following	**Use the following scale of criteria as guidance (not all criteria must be present to be indicative of Merck product relationship).**
Yes, there is a reasonable possibility of Merck product relationship.	There is evidence of exposure to Merck product. The temporal sequence of the AE onset relative to the administration of Merck product is reasonable. The AE is more likely explained by Merck product than by another cause.
No, there is not a reasonable possibility of Merck product relationship	Participant did not receive the Merck product OR temporal sequence of the AE onset relative to administration of Merck product is not reasonable OR the AE is more likely explained by another cause than the Merck product. (Also entered for a participant with overdose without an associated AE.)

7.3.5 Sponsor Responsibility for Reporting Adverse Events

All Adverse Events will be reported to regulatory authorities, IRB/IECs and investigators in accordance with all applicable global laws and regulations.

8.0 STATISTICAL CONSIDERATIONS

Design

This a single arm Phase II trial in resectable high risk stage II melanoma patients. Patients receive one dose of pembrolizumab and then undergo definitive surgery and SLN biopsy. After approximately 9 weeks for post-surgical recovery, patients receive adjuvant pembrolizumab for up to 17 cycles (approximately 1 year) as long as subjects are receiving benefit from treatment and have not had disease progression or met any criteria for study withdrawal. SLN, primary tumor tissue and blood samples will be evaluated for immune changes. Sixty-three evaluable patients will be enrolled.

8.1 Objectives

The primary objectives are to:

1. To determine the SLN positivity rate and test whether the SLN positivity rate is reduced in high risk stage II patients undergoing neoadjuvant PD-1 blockade.
2. To evaluate safety, tolerability and adverse events of pembrolizumab when given as neoadjuvant and adjuvant therapy in high risk stage II patients.

The secondary objectives are to:

1. To evaluate the disease-free-survival (DFS) and overall survival (OS) in patients receiving pembrolizumab as adjuvant therapy.
2. To describe the patterns of recurrence and time to recurrence in patients receiving pembrolizumab as adjuvant therapy.
3. To examine the effects of pembrolizumab on immune regulatory pathways in SLN tissue after treatment with a single dose of neoadjuvant pembrolizumab.
4. To examine the effects of pembrolizumab on immune biomarkers in PBMC and serum at serial time points during and up to approximately 1 year of treatment with pembrolizumab.
5. To examine the effects of pembrolizumab on immune biomarkers in the primary tumor and determine whether immune related changes in the primary tumor are associated with similar changes in the SLN.

8.2 Endpoints

Evaluability. A patient will be deemed evaluable if the patient received one pre-operative dose of pembrolizumab and underwent definitive surgery with SLN biopsy.

Toxicity. CTCAE version 5.0 grades will be employed. Any patient who receives any amount of pembrolizumab will be included in the toxicity analysis.

Dose-limiting toxicity: All grade 4 or higher hematologic or grade 3 or higher non- hematologic toxicities that are at least possibly related to pembrolizumab and not an expected adverse event for either pembrolizumab or surgery and which occur anytime from the initiation of study therapy to 6 weeks post-surgery are considered DLTs and will be described separately.

Disease-free and overall survival. Disease-free survival (DFS) is defined as the time from date of surgery to date of first documented disease progression, death due to any cause or last date that patient was documented to be disease-free (i.e., a scan date). Overall survival (OS) is defined as the time from date of surgery to date of death due to any cause or last patient contact alive. Both DFS and OS outcomes will be measured from date of surgery to allow us to compare DFS and OS estimates with those of other adjuvant trials.

Patterns of recurrence will be characterized by location(s) of disease recurrence. Time to recurrence is defined as the time from date of surgery to date of first documented disease progression for patients who recurred.

Immune biomarkers. Priority assays will include characterization of T cell subsets, cytokines, Tregs, CD8 T cells, exhausted T cells and TCR clones. A full description of all immunologic outcomes and hypotheses is included in Section 4.2.3.3.

8.3 Monitoring Toxicity

Although there is a growing literature on the safety of pembrolizumab in high risk stage II melanoma from ongoing clinical trials, this study will closely monitor toxicity and dose- limiting toxicity. The study will be terminated early if there is evidence that the rate of dose limiting toxicity is greater than 33%. Early termination rules are defined in Section 5.8.

8.4 Plans for Data Analysis

Patient Evaluability. Evaluable patients must receive one dose of neoadjuvant pembrolizumab and undergo definitive surgery and SLN biopsy. Only evaluable patients will contribute to the primary analysis that tests a hypothesis about the SLN positivity rate. All patients who receive any amount of pembrolizumab will contribute to safety analyses. Patients who undergo definitive surgery and receive any amount of adjuvant pembrolizumab will be analyzed for disease-free and overall survival. Patients who undergo definitive surgery, receive any amount of adjuvant pembrolizumab and contribute tissue & serial blood samples, will be included in the secondary and exploratory analyses of immunologic effects of pembrolizumab in LN tissue and peripheral blood.

SLN positivity: The SLN positivity rate and 95% confidence interval will be computed. The rate will be compared to an historical control rate using a one sample chi-square test. The historical rate will be selected, as shown in Section 8.6.

Toxicity: All observed adverse events which occur anytime from the initiation of study therapy to 30 days after the final dose of pembrolizumab will be graded and tabled.

Disease-free and overall survival: will be estimated by the method of Kaplan and Meier. Median DFS and OS rates and 95% CIs will be estimated. As patients will be followed for up to 5 years after enrollment, milestone DFS and OS rates (i.e., 2-yr, 5-yr) will also be estimated. Time to recurrence in months will be calculated for patients who recur.

Immune biomarkers: Immunologic biomarkers will be described using scatter and box plots and descriptive statistics (e.g., mean, median, SD, range). Biomarkers from paired SLN and primary tissue samples will be compared by paired Student's t-test or Wilcoxon signed ranks test, as appropriate. Cox regression modeling will investigate associations between biomarker values and DFS & OS. With several post-treatment blood samples intended, trends over time will be

described using scatter and box plots, descriptive statistics (e.g., fold changes from baseline). Longitudinal modeling using linear mixed effects models will also be examined, in order to extend this investigation to characterized trends over time and could include modeling jointly the inter-dependency among several longitudinally measured immunologic outcomes.

8.5 Sample Size

The study will enroll 63 evaluable patients. The primary objective is to detect a 50% reduction in the SLN positivity rate at the time of surgery, after a single dose of neoadjuvant pembrolizumab.

The percentage of patients who have a positive SLN is known to vary by stage of disease. We have examined two datasets to determine the historical rates of SLN positivity in stage IIB and stage IIC patients who will enroll on this study. According to the National Cancer Database (NCDB) (2004 to 2012), the SLN positivity rate for stage IIB is 24% (4,713/19,683 cases) and the rate for stage IIC is 39% (3,340/8,510 cases). According to the Penn Medicine database (1997 to 2014), the SLN positivity rate for stage IIB is 27.5% (66/240 cases) and the rate for stage IIC is 45% (48/107 cases).

We have therefore computed power to detect a 50% reduction in the percentage of patients who have a positive SLN, assuming that the historical SLN positivity rate in stage IIB patients is 25% and the SLN positivity rate in stage IIC patients is 40%. The distribution of stage IIB and stage IIC patients is not controlled in this study. Therefore, we have computed power for a range of possible enrollment distributions by stage. We have allowed the percentage of stage IIB patients to vary from 30% to 60%. The overall SLN positivity rates of the null hypothesis based on these varying distributions, can vary from 35.5% to 31.0%.

The table below displays power to detect a 50% reduction in the percentage of treated patients who have a positive SLN at surgery, for a one sample chi-square test based on 63 patients, at a 2-sided 5% type I error rate. With 63 patients, the power for the chi-square test will be acceptable, at a range of 81 – 88%.

Percentage stage IIB patients	Percentage stage IIC patients	SLN positive rate stage IIB patients	SLN positive rate stage IIC patients	Null Hypothesis SLN^+ rate	Alternative Hypothesis SLN^+ rate	Power chi-square test
30%	70%	25%	40%	35.5%	17.8%	88%
35%	65%	25%	40%	34.8%	17.4%	88%
40%	60%	25%	40%	34.0%	17.0%	86%
45%	55%	25%	40%	33.3%	16.6%	85%
50%	50%	25%	40%	32.5%	16.3%	84%
55%	45%	25%	40%	31.8%	15.9%	83%
60%	40%	25%	40%	31.0%	15.5%	81%

For exploratory analyses of immunological effects of pembrolizumab in SLN, tumor tissue and blood, we anticipate conducting many paired comparisons. Due to multiple parameters tested, we will control the false positive rate by calculating adjusted p values using the Bonferroni-Holm method.

Considering a stricter type I error rate, with 63 evaluable patients, there is 90% power to detect an effect size of $1/2s_d$ at a reduced 2-sided 1% error rate using a paired Student's t- test.

8.6 Study Duration

With an estimated accrual of 30-35 patients per year, it is anticipated that accrual will continue for nearly 7 years. We plan to follow patients for an additional 5 years, in order to better estimate DFS and OS. This study is expected to be active for 12 years.

9.0 LABELING, PACKAGING, STORAGE AND RETURN OF CLINICAL SUPPLIES

9.1 Investigational Product

The 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.

Pembrolizumab will be provided by Merck as summarized in Table 8.

Table 8 Product Descriptions

Product Name & Potency	**Dosage Form**
Pembrolizumab 100 mg/ 4mL	Solution for Injection

9.2 Packaging and Labeling Information

Supplies will be labeled in accordance with regulatory requirements.

9.3 Clinical Supplies Disclosure

This trial is open-label; therefore, the participant, the trial site personnel, the Sponsor-Investigator and/or designee are not blinded to treatment. Drug identity (name, strength) is included in the label text; random code/disclosure envelopes or lists are not provided.

9.4 Storage and Handling Requirements

Clinical supplies must be stored in a secure, limited-access location under the storage conditions specified on the label.

Receipt and dispensing of trial medication must be recorded by an authorized person at the trial site.

Clinical supplies may not be used for any purpose other than that stated in the protocol.

9.5 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 participants 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.

10.0 ADMINISTRATIVE AND REGULATORY DETAILS

10.1 Confidentiality

Information about study subjects will be kept confidential and managed according to the requirements of the Health Insurance Portability and Accountability Act (HIPAA). Those regulations require a signed subject authorization informing the subject of the following:

- What protected health information (PHI) will be collected from subjects in this study

- Who will have access to that information and why

- Who will use or disclose that information

- The rights of a research subject to revoke their authorization for use of their PHI.

In the event that a subject revokes authorization to collect or use PHI, the investigator, by regulation, retains the ability to use all information collected prior to the revocation of subject authorization. For subjects that have revoked authorization to collect or use PHI, attempts should be made to obtain permission to collect at least vital status (i.e. that the subject is alive) at the end of their scheduled study period.

10.2 Compliance with Trial Registration and Results Posting Requirements

Under the terms of the Food and Drug Administration Modernization Act (FDAMA) and the Food and Drug Administration Amendments Act (FDAAA), the Sponsor-Investigator of the trial is solely responsible for determining whether the trial and its results are subject to the requirements for submission to the Clinical Trials Data Bank, https://www.clinicaltrials.gov/.

Information posted will allow subjects to identify potentially appropriate trials for their disease conditions and pursue participation by calling a central contact number for further information on appropriate trial locations and trial site contact information.

10.3 Quality Management System

The Data Safety and Monitoring Committee (DSMC) of the University of Pennsylvania's Abramson Cancer Center will monitor the data quality and adherence to safety rules.

Additionally, the DSMC will review all safety/toxicity data for the trial and recommend trial suspension or termination as needed. Specific details of monitoring and audit frequency will be included in the Monitoring Plan, but will be at least every 6 months.

Medical Monitor

This study will be monitored in accordance with the DSMC monitoring plan. Dr. Vivek Narayan, an independent clinician in the Department of Medicine, Division of Hematology Oncology, will serve as medical monitor and will consult on decisions made as a part of this trial as noted above. The medical monitor will perform a regular review and assessment of the number and type of serious adverse events (at minimum yearly, but may occur more frequently as needed).

It is the responsibility of the Principal Investigator to oversee the safety of the study. This safety monitoring will include careful assessment and appropriate reporting of adverse events, as noted above, as well as the construction and implementation of a site data and safety monitoring plan.

Notifying the Data Safety Monitoring Committee (DSMC)

Adverse Events

1. All grade 3 or higher events regardless of attribution or expectedness within 10 business days of knowledge.
2. All unexpected deaths within two business days of knowledge.
3. All others deaths within 30 days of knowledge. Deaths of subjects greater than 90 days from the last study treatment/intervention are not reportable

Deviations

A deviation is a one time, **unintentional** action or process that departs from the IRB and DSMC approved study protocol, involving one incident and **identified retrospectively**, after the event occurred. If the impact on the protocol disrupts the study design, may affect the outcome (endpoints) or compromises the safety and welfare of the subjects, the deviation must be reported to the DSMC and the IRB within 10 days from the time the event becomes known to the study team.

Exceptions

A one time, intentional action (planned prospectively) or process that departs from the IRB and CTSRMC approved study protocol, intended for one occurrence. Advance documented IRB and DSMC approval is required. Approval must be obtained from the Medical Monitor prior to submitting an exception request to the DSMC.

10.4 Data Management

Data collection and management will be carried out by the staff of the Clinical Research Unit at Penn's Abramson Cancer Center. The study case report form (CRF) is the primary data collection instrument for the study. All data required on the CRF must be recorded. All entries will be entered into an electronic data capture system (EDC) via PennCTMS (VELOS).

10.5 Compliance with Auditing and Inspecting

The investigator will permit the Cancer Center's Administrative Director of Compliance and Auditing or her designee, and the Abramson Cancer Center Data Safety and Monitoring Committee (DCOM), to review records, data and facilities at mutually agreeable times in accordance with the DSMC monitoring plan.

The investigator also agrees to allow IRB review and regulatory authority (FDA) inspection of trial-related documents and procedures and provide for direct access to all trial-related source data and documents.

The investigator shall prepare and maintain complete and accurate trial documentation in compliance with Good Clinical Practice standards and applicable federal, state and local laws, rules and regulations; and, for each subject participating in the trial, provide all data, and, upon completion or termination of the clinical trial, submit any other reports to the Sponsor-Investigator and Merck as required by this protocol.

10.6 Compliance with Trial Registration and Results Posting Requirements

Under the terms of the Food and Drug Administration Modernization Act (FDAMA) and the Food and Drug Administration Amendments Act (FDAAA), the Sponsor of the trial is solely responsible for determining whether the trial and its results are subject to the requirements for submission to the Clinical Trials Data Bank, https://www.clinicaltrials.gov/. Information posted will allow subjects to identify potentially appropriate trials for their disease conditions and pursue participation by calling a central contact number for further information on appropriate trial locations and trial site contact information.

10.7 Ethical Considerations

This study is to be conducted according to US and international standards of Good Clinical Practice (FDA Title 21 part 312 and International Conference on Harmonization guidelines), applicable government regulations and Institutional research policies and procedures. This protocol and any amendments will be submitted to the University of Pennsylvania IRB and CTSRMC for formal approval of the study conduct. The decision of the IRB concerning the conduct of the study will be made in writing to the investigator and a copy of this decision will be provided before commencement of this study.

All subjects for this study will be provided a consent form describing this study and providing sufficient information for subjects to make an informed decision about their participation in this study. This consent form will be submitted with the protocol for review and approval by the IRB and CTSRMC for the study. The formal consent of a subject, using the IRB-approved consent form, must be obtained before that subject is submitted to any study procedure. This consent form must be signed by the patient or legally acceptable surrogate, and the investigator-designated research professional obtaining the consent.

11.0 REFERENCES

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12.0 APPENDICES

Appendix 1: ECOG Performance Status

Grade	Description
0	Normal activity. Fully active, able to carry on all pre-disease performance without restriction.
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).
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.
3	In bed >50% of the time. Capable of only limited self-care, confined to bed or chair more than 50% of waking hours.
4	100% bedridden. Completely disabled. Cannot carry on any self-care. Totally confined to bed or chair.
5	Dead.
*As published in Am. J. Clin. Oncol.: *Oken, M.M., Creech, R.H., Tormey, D.C., Horton, J., Davis, T.E., McFadden, E.T., Carbone, P.P.: Toxicity And Response Criteria Of The Eastern Cooperative Oncology Group. Am J Clin Oncol 5:649-655, 1982.* The Eastern Cooperative Oncology Group, Robert Comis M.D., Group Chair.	

Appendix 2: Common Terminology Criteria for Adverse Events V5.0 (CTCAE)

The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 5.0 will be utilized for adverse event reporting. (https://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm#ctc_50)

Appendix 3: Contraceptive Guidance and Pregnancy Testing

Woman of Childbearing Potential (WOCBP)

A woman is considered fertile following menarche and until becoming post-menopausal unless permanently sterile (see below)

Women in the following categories are not considered WOCBP:

- Premenarchal
- Premenopausal female with 1 of the following:
  - Documented hysterectomy
  - Documented bilateral salpingectomy
  - Documented bilateral oophorectomy

  Note: Documentation can come from the site personnel's review of the participant's medical records, medical examination, or medical history interview.

- Postmenopausal female
  - A postmenopausal state is defined as no menses for 12 months without an alternative medical cause.
    - A high follicle stimulating hormone (FSH) level in the postmenopausal range may be used to confirm a postmenopausal state in women not using hormonal contraception or hormonal replacement therapy (HRT). However, in the absence of 12 months of amenorrhea, confirmation with two FSH measurements in the postmenopausal range is required.
  - Females on HRT and whose menopausal status is in doubt will be required to use one of the non-hormonal highly effective contraception methods if they wish to continue their HRT during the study. Otherwise, they must discontinue HRT to allow confirmation of postmenopausal status before study enrollment.

Contraception Requirements

Male Participants:

Male participants with female partners of childbearing potential are eligible to participate if they agree to one of the following during the protocol defined time frame in section 6.0:

- Be abstinent from penile-vaginal intercourse as their usual and preferred lifestyle (abstinent on a long term and persistent basis) and agree to remain abstinent
- Use a male condom plus partner use of a contraceptive method with a failure rate of <1% per year as described in Table 9 when having penile-vaginal intercourse with a woman of childbearing potential who is not currently pregnant.
  - Note: Men with a pregnant or breastfeeding partner must agree to remain abstinent from penile-vaginal intercourse or use a male condom during each episode of penile penetration.

Female Participants:
Female participants of childbearing potential are eligible to participate if they agree to use a highly effective method of contraception consistently and correctly as described in Table 10 during the protocol-defined time frame in Section 6.0.

Table 10 Highly Effective Contraception Methods

Highly Effective Contraceptive Methods That Are User Dependent [a] *Failure rate of <1% per year when used consistently and correctly.*
● Combined (estrogen- and progestogen- containing) hormonal contraception [b] ○ Oral ○ Intravaginal ○ Transdermal ○ Injectable
● Progestogen-only hormonal contraception [b] ○ Oral ○ Injectable
Highly Effective Methods That Have Low User Dependency *Failure rate of <1% per year when used consistently and correctly.*
● Progestogen- only contraceptive implant [b] ● Intrauterine hormone-releasing system (IUS) ● Intrauterine device (IUD) ● Bilateral tubal occlusion
● **Vasectomized partner** A vasectomized partner is a highly effective contraception method provided that the partner is the sole male sexual partner of the WOCBP and the absence of sperm has been confirmed. If not, an additional highly effective method of contraception should be used.
● **Sexual abstinence** Sexual abstinence is considered a highly effective method only if defined as refraining from heterosexual intercourse during the entire period of risk associated with the study treatment. The reliability of sexual abstinence needs to be evaluated in relation to the duration of the study and the preferred and usual lifestyle of the participant.)
Notes: Use should be consistent with local regulations regarding the use of contraceptive methods for participants of clinical studies. a) Typical use failure rates are lower than perfect-use failure rates (i.e. when used consistently and correctly). b) If locally required, in accordance with Clinical Trial Facilitation Group (CTFG) guidelines, acceptable hormonal contraceptives are limited to those which inhibit ovulation.

Pregnancy Testing

WOCBP should only be included after a negative highly sensitive urine or serum pregnancy test.

Pregnancy testing will be performed whenever an expected menstrual cycle is missed or when pregnancy is otherwise suspected.