Study Title: **Neoadjuvant Immunotherapy in Resectable Non-Small-Cell Lung Cancer NCT02259621**

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Contents

1. Summary

Host immunity is fundamental to the suppression of human cancer, and conversely host immune evasion by tumor cells is an essential feature in the development and progression of human cancer. PD-1 is a co-inhibitory receptor expressed on the surface of activated and exhausted T-cells, B-cells and certain myeloid cells[1]. PD-L1 (programmed death – ligand-1), one of two ligands for PD-1, is highly expressed in certain human tumors and expression has been associated with a poor prognosis[2,3]. Little is known about expression of the second ligand PD-L2 in solid cancers, nor its role in immune evasion.

An ongoing early phase clinical trial of the anti-PD-1 antibody, nivolumab, has demonstrated durable responses in heavily pretreated patients with advanced melanoma, renal cell (RCC) and non-small-cell lung cancer (NSCLC), a tumor which was previously thought to be non-immunogenic[4]. In an interim analysis of this study, 10% of NSCLC patients were free of tumor progression at 6 months, and 22 of 129 patients (17%) had an objective tumor response by RECIST 1.0 with a median duration of response lasting 17 months[5]. Patients with both squamous and non-squamous lung cancers showed durable objective responses. Median overall survival was 9.6 months, and the one-year landmark survival rate was 42%. These are promising results, given that most of these patients had exhausted standard treatment options: 53% of those with NSCLC had received 3-5 prior lines of treatment.

More recently, in two phase 3 clinical trials, nivolumab has demonstrated a survival advantage over second line chemotherapy with docetaxel in platinum-pretreated metastatic squamous and non-squamous NSCLC [6, 7]. In both of these studies nivolumab was associated with a lower incidence of both grade 3-4 and any grade toxicity than docetaxel. In the first-line treatment of metastatic NSCLC, nivolumab has also shown promise in a large phase 1 study with an objective response rate (ORR) of 23% in patients unselected by PD-L1 status[8].

Given the potentially harmful effects of multiple chemotherapy treatment regimens on the immune system, it is quite possible that application of PD-1 pathway blockade in lung cancer patients prior to receiving chemotherapy may significantly enhance its ability to induce immune-mediated cancer regression. In addition, it is postulated that PD-L1 and other immune biomarkers may be induced by prior anti-cancer therapy therefore interrogating the tumor immune micro-environment in non-pretreated disease may help to better define the immune signature of developing tumors.

In total, 306 patients with advanced solid tumors (129 with NSCLC) received nivolumab every two weeks continuously for up to 2 years[77]. Treatment was generally well tolerated, with grade 3-4 drug-related toxicities occurring in 17% of patients. As of March 2013, there were 3 nivolumab-related deaths on this trial (1%) associated with pneumonitis (two patients with NSCLC, one with colorectal cancer). This incidence of high-grade pneumonitis is similar to that

seen with standard cytotoxic chemotherapies and kinase inhibitors. Early diagnosis of nivolumab-related pneumonitis with administration of immunosuppression including steroids and other agents has mitigated pulmonary toxicity. However, the mechanisms causing pneumonitis require elucidation in order to develop more effective detection and treatment algorithms.

Exploratory analysis of archived pretreatment tumor biopsies from a limited number of NSCLC patients treated with nivolumab indicates that PD-L1 expression on tumor cells may be a candidate biomarker of sensitivity to anti-PD-1 therapy[4](and Taube & Topalian, unpublished). However, this preliminary finding remains to be validated in larger cohorts.

The proposed study will evaluate the safety and feasibility of preoperative administration nivolumab +/- ipilimumab in patients with high-risk resectable NSCLC, and will facilitate a comprehensive exploratory characterization of the tumor immune milieu and circulating immune cells and soluble factors in these patients. Data obtained in this study will provide valuable information for planning further prospective clinical trials of anti-PD-1 and other immunotherapies in NSCLC, both in the peri-operative and advanced disease setting. Ultimately, it is highly desirable to discover prospective biomarkers of response and toxicity to allow patients with NSCLC who are most likely to derive benefit to receive anti-PD-1 treatment, and conversely to minimize the risk of toxicity and ineffective treatment for patients who are unlikely to benefit.

An earlier amendment to this study allowed evaluation of the combination of nivolumab and the anti-CTLA4 antibody, ipilimumab in the neoadjuvant setting for the treatment of resectable NSCLC. In a large, multicohort, phase 1 trial, the ORR to combination ipilimumab and nivolumab therapy in patients unselected by PD-L1 status ranged from 39-47%[9]. Incidence of grade 3-4 toxicity ranged from 33-37% across the combination ipilimumab and nivolumab cohorts which compares favorably with the rates of toxicity due to platinum doublet chemotherapy in this disease setting.[9]

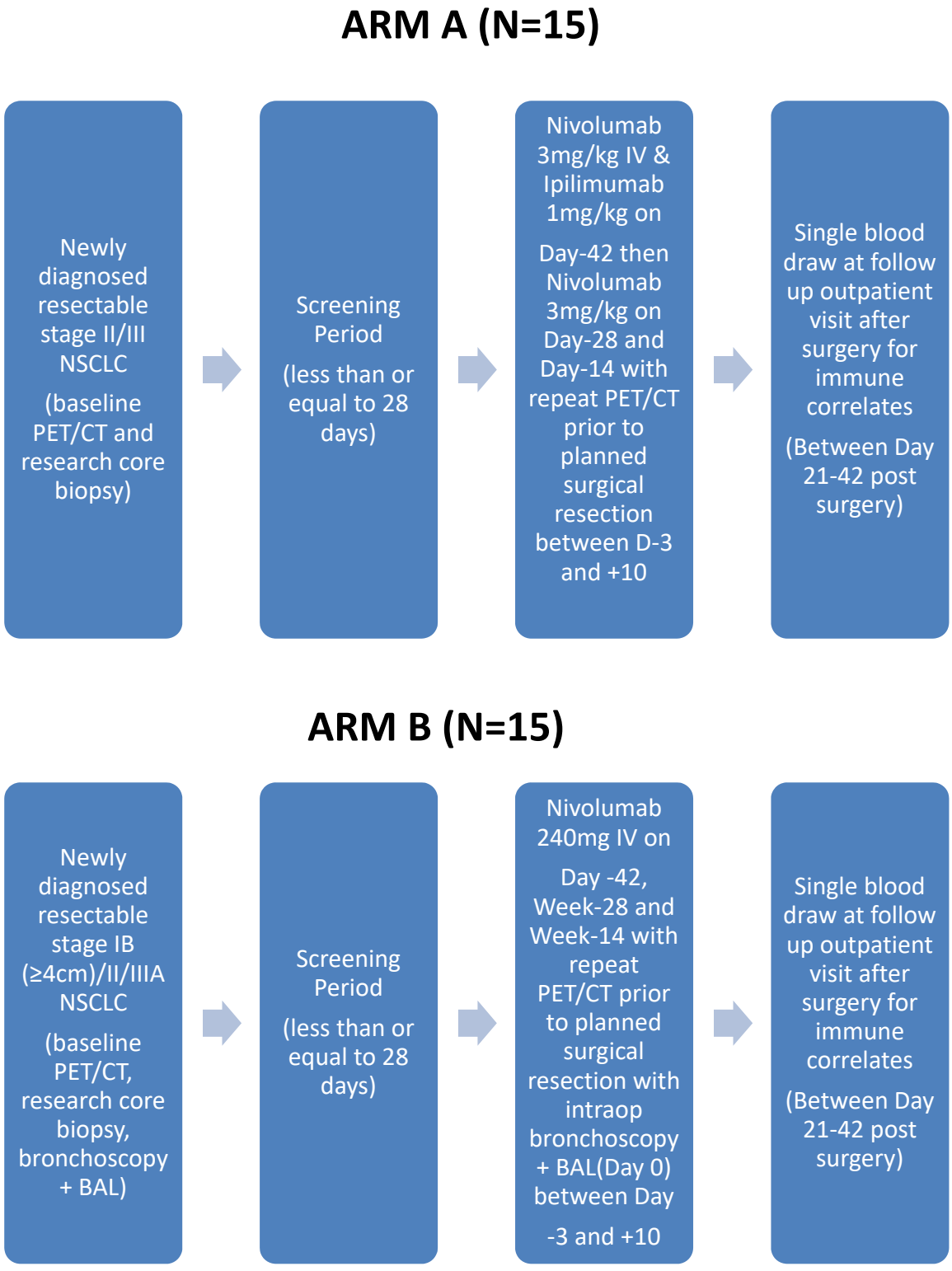
Since this amendement, preliminary data from other studies has demonstrated similar rates of major pathologic response (mPR) between neoadjuvant nivolumab and nivolumab + ipilimumab (Cascone et al., ESMO 2018). In addition, a large phase III study of neoadjuvant immunotherapy in NSCLC recently closed their nivolumab + ipilimumab arm leading to a situation where further clinical development of the combination in early stage lung cancer is unlikely to occur. Because of this, in conjunction with encouraging results of neoadjuvant single agent PD-1 therapy +/- chemotherapy, an amendent was made to this study to close the nivolumab + ipilimumab arm (Arm A) prior to complete accrual and move on to the arm of the study with extended doses (3 doses) of preoperative nivolumab (Arm B).

A previous amendment to this study allowed for bronchoscopy with bronchoalveolar lavage (BAL) in patients pre- and post-treatment to enable genomic and molecular characterization of bronchial and nasal epithelium. Through genomic and molecular characterizations of these samples, and comparison between responders and non-responders to neoadjuvant checkpoint blockade, this data will help to deepen our understanding of NSCLC pathogenesis, elucidate the mechanisms of checkpoint blockade, and aid in creation of predictive biomarkers of response.

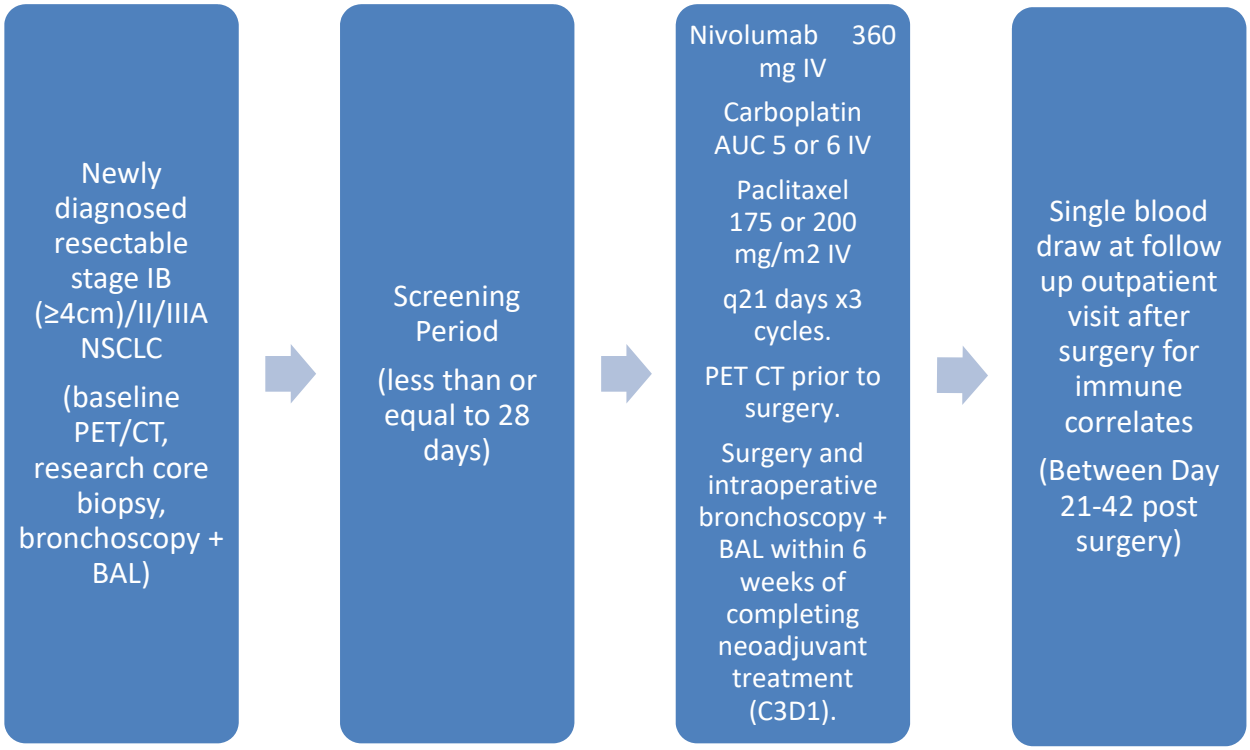
The current amendment includes addition of a third arm (Arm C) combining nivolumab and platinum-doublet chemotherapy in the neoadjuvant setting. Since this trial began, PD-1 and PD-L1 inhibitors have transformed the treatment of NSCLC and are now approved as part of first line treatment for locally-advanced unresectable and metastatic NSCLC in the absence of a targetable driver mutation, including concurrent chemo-immunotherapy for metastatic NSCLC [Gandhi et al. NEJM 2018][97]. However, PD-1 pathway blockade is not yet incorporated in the treatment of resectable NSCLC, and we still require predictive biomarkers to to identify patients likely to experience long-term disease control with checkpoint inhibitors. The expansion to include combined chemotherapy with immunotherapy in the preoperative setting is based on high-level evidence from metastatic NSCLC where combined pembrolizumab and chemotherapy is approved as a safe and effective first line therapy with improved overall survival compared to chemotherapy alone, as well as recent early phase trials in resectable NSCLC demonstrating high rates of major pathologic and complete pathologic responses with preoperative combination therapy [Shu et al. Lancet Oncol 2020][104].

To further investigate the synergy of chemotherapy and PD-1 checkpoint blockade and dynamics of the tumor immune response, this neoadjuvant trial provides an ideal platform for detailed molecular analysis and biomarker discovery. Investigators at SKCCC have developed advanced techniques for integrated immunogenomic analysis of tumor and liquid biopsies as well as single cell transcriptomics.

2. Schema


ARM A (N=15)
Newly diagnosed resectable stage II/III NSCLC
(baseline PET/CT and research core biopsy)
Screening Period
(less than or equal to 28 days)
Nivolumab 3mg/kg IV & Ipilimumab 1mg/kg on
Day-42 then Nivolumab 3mg/kg on Day-28 and Day-14 with repeat PET/CT prior to planned surgical resection between D-3 and +10
Single blood draw at follow up outpatient visit after surgery for immune correlates
(Between Day 21-42 post surgery)
ARM B (N=15)
Newly diagnosed resectable stage IB (≥4cm)/II/IIIA NSCLC
(baseline PET/CT, research core biopsy, bronchoscopy + BAL)
Screening Period
(less than or equal to 28 days)
Nivolumab 240mg IV on
Day -42, Week-28 and Week-14 with repeat PET/CT prior to planned surgical resection with intraop bronchoscopy + BAL(Day 0) between Day
-3 and +10
Single blood draw at follow up outpatient visit after surgery for immune correlates
(Between Day 21-42 post surgery)

ARM C (N=15)



3. Hypotheses

3.1 Anti-PD-1 (nivolumab) administration with or without anti-CTLA4 (ipilimumab) in the pre-operative setting will be safe and feasible in patients with resectable NSCLC.

3.2 Neoadjuvant administration of nivolumab with or without ipilimumab or nivolumab with chemotherapy will change cellular and molecular characteristics of the tumor microenvironment that can be quantitatively measured.

3.3 Failure to respond to immune checkpoint inhibition in NSCLC results from either pre-existing or compensatory (i.e. adaptive) up-regulation of additional immune "checkpoint" pathways in the tumor, draining lymph nodes, and/or peripheral blood that inhibit immune recognition and killing of tumor cells. Characterization of these pathways (i.e. ligands and receptors) in patients receiving preoperative therapy, and comparison with a cohort of patients who proceed to surgical resection without preoperative therapy, will illuminate mechanisms of

adaptation and immune resistance to directly guide future therapeutic development of anti-PD-1 as monotherapy and in combination with other immunomodulators in NSCLC.

3.4 Neoadjuvant combination platinum-based chemotherapy with nivolumab will result in a high rate of pathologic complete response (pCR) in resectable high-risk NSCLC.

3.5 There will be quantitative differences in the molecular and immune characteristics of pre- and post-treatment lung cancer among responders and non-responders following combined nivolumab and chemotherapy, which can be elucidated using integrated immunogenomic analysis of tumor and liquid biopsies and single cell transcriptomics and MANAFEST.

3.6. There will be quantitative differences in the immune response among patients treated with nivolumab alone, nivolumab + ipilimumab, and nivolumab + chemotherapy that are demonstrable using single cell transcriptomics and MANAFEST.

4. Objectives

Primary

4.1.1 To investigate the safety and feasibility of neoadjuvant nivolumab + ipilimumab administration in subjects with resectable high-risk NSCLC [stage IB, II and IIIA], including squamous and non-squamous histologies.

4.1.2 To investigate the safety and feasibility of 3 doses of neoadjuvant nivolumab in subjects with resectable high-risk NSCLC [stage IB, II and IIIA], including squamous and non-squamous histologies.

4.1.3 Arm C - To investigate the pathologic response to 3 doses of neoadjuvant nivolumab with carboplatin and paclitaxel in subjects with high-risk NSCLC [stage IB, II and IIIA], including squamous and non-squamous histologies.

4.2 Secondary

4.2.1 To assess radiographic response to neoadjuvant nivolumab, nivolumab plus ipilimumab, and nivolumab plus chemotherapy using RECIST 1.1.

4.2.2 To assess safety for Arm C.

4.3 Exploratory

4.3.1 To determine changes in expression of selected immune markers compared to baseline, in the blood, primary tumor tissue and draining lymph nodes from patients receiving neoadjuvant therapy; to determine changes in the quality and quantity of tumor infiltrating lymphocytes; and to compare findings in tumor and draining lymph

nodes from treated patients, to findings in a parallel stage-matched cohort of untreated patients on a companion tissue collection protocol.

4.3.2 To evaluate the potential effects of neoadjuvant therapy on normal lung tissue, by comparing tissues obtained on this study to those obtained from untreated patients undergoing lung tumor resection on a parallel tissue collection protocol.

4.3.3 To compare immunologic markers in squamous versus non-squamous lung tumors.

4.3.4 To explore genomic and molecular characteristics of bronchial and nasal epithelium from patients receiving neoadjuvant immune checkpoint blockade and compare findings between patients who do and do not achieve a major pathologic response. In addition, these genomic and molecular changes will be compared to those observed in the primary tumor and peripheral circulation.

4.3.5 To explore the association between nivolumab +/- ipilimumab exposure or nivolumab + chemotherapy and selected pharmaco-dynamic markers in the peripheral blood and in the tumor microenvironment, including measurement of PD-1 receptor occupancy on tumor infiltrating lymphocytes.

4.3.6 To explore features of the gut microbiota of NSCLC patients before and after neoadjuvant nivolumab +/- ipilimumab, or nivolumab + chemotherapy, that may correlate with clinical response.

4.3.7 To assess recurrence-free survival in patients receiving preoperative therapy in this study

4.3.8 To assess overall survival in high-risk patients with NSCLC receiving neoadjuvant therapy.

5. Background and Rationale

Lung cancer is the most common invasive cancer and cause of cancer death worldwide. In 2008, the most recent year for which global statistics are available, there were an estimated 1.61 million cases and 1.38 million deaths[12]. In the United States, the estimated number of new lung cancer cases for 2012 is 226,160 (116,470 men and 109,690 women) while 160,340 people will die of the condition[13]. Adjuvant systemic chemotherapy improves disease free and overall survival for patients with stage II NSCLC and for some patients with Stage IB NSCLC and is likely to remain an integral part of therapy[14]. However, many patients still suffer recurrences and die of their disease emphasizing the need for new treatments or approaches.

Host immunity is fundamental to the suppression of human cancer and conversely host immune evasion by tumor cells is an essential pathway in the development of human cancer. The concept of cancer immune editing is well described in animal models whereby tumors are capable of subverting host immunity despite developing frequent genetic aberrations with the potential to generate immunogenic neo-antigens[15]. The three phases of immune editing are as follows: elimination (host immune system responds to tumor neo-antigens and destroys tumor cells), equilibrium (immune evasive tumor cells persist; however, growth and metastasis is restrained by residual host immunity) and escape (tumor cells overcome immune control and can develop into clinically evident cancers). The development of clinically apparent tumors indicates failure of the host immune system to recognize and destroy incipient cancers. This is due to induction of immune tolerance among tumor-specific T cells as well as expression of immune inhibitory ligands termed checkpoints. These ligands bind to receptors on T cells that signal to down-modulate effector functions such as cytokine production and killing activity. Consequently, strategies aimed at augmenting host anti-tumor immunity are attractive with potential for long-term tumor control or even cure if persistent immune responsiveness can be engendered particularly in earlier stages of disease.

5.1 Cancer immunotherapy

Attempts have been made over many years to potentiate the host immune response to human cancer with limited success until recently and, in some cases, significant toxicity[16-18]. While occasional dramatic tumor responses have been seen with interleukin-2 treatment in particular, these responses are difficult to predict based on clinical criteria and a dependable biological marker of response in the patient or tumor has yet to be described[19-21]. Since the initial approval of this protocol, immunotherapy has transformed the treatment landscape in lung cancer and is now standard of care in metastatic NSCLC and locally-advanced unresectable NSCLC, and is now under investigation in the adjuvant and neoadjuvant settings.

5.2 Rational Immunotherapy Targets – CTLA-4

Recent breakthroughs in bringing years of preclinical work on the adaptive immune response to tumor to the clinic have led to the regulatory approval of two immune-modulatory anti-cancer therapies for advanced disease, the autologous dendritic cell vaccine, sipuleucel-T, for castration-resistant prostate cancer and the anti-cytotoxic T lymphocyte antigen- 4 (anti-CTLA-4) immune checkpoint inhibitor, ipilimumab, for metastatic melanoma[22,23]. CTLA-4, first described in 1987, is the prototypical immune checkpoint, a molecule whose expression is induced by T cell activation leading to down-regulation of T cell responses and consequent suppression of the innate response to foreign tumor neo-antigen[24,25]. The subsequent discovery of multiple non-redundant intrinsic T cell molecules that act to limit immune responses have led to the advent of antibodies to block these inhibitory checkpoints as a strategy to enable anti-tumor immune responses and elicit durable clinical benefit in cancer patients. Ipilimumab,

which inhibits CTLA-4, thus releasing the block on antitumor immunity, has become the first systemic treatment to demonstrate a durable overall survival advantage in a phase III study for metastatic melanoma with responses in 10 – 20% of patients treated[23]. Despite these promising results, many patients do not respond to treatment, and toxicity can be serious or fatal in certain cases, illustrating the need for prospective biomarkers of immune response and sensitivity.

5.3 Programmed death-1 – Molecular Biology

Programmed death-1 (PD-1 or CD279), primarily expressed on activated T cells, B cells and myeloid cells[1], is a 55 kD type I transmembrane protein that is a member of the CD28 family of T-cell co-stimulatory receptors that also includes CD28, CTLA-4, ICOS and BTLA2[26]. Two ligands specific for PD-1 have been identified: PD-L1 (also known as B7-H1 or CD274) and PD-L2 (also known as B7-DC or CD273), each of which are primarily expressed on antigen presenting cells. PD-L1 and PD-L2 have been shown to downregulate T-cell activation upon binding to PD-1 in both murine and human systems[27-29]. PD-1 has been shown to inhibit CD28-mediated upregulation of IL-2, IL-10, IL-13, IFN-γ and Bcl-xL. PD-1 expression has also been noted to inhibit T cell activation and expansion of previously activated cells. PD-1 contains an intracellular membrane proximal immunoreceptor tyrosine inhibitory motif (ITIM) and a membrane distal immunoreceptor tyrosine based switch motif (ITSM). Both Src homology region 2 domain-containing phosphatase (SHP) -1 and -2 have been found to bind to the cytoplasmic tail of PD-1 and mediate its signaling. Once this signaling has occurred, PD-1 binds to the phosphorylated tyrosine residue in the ITSM in its cytoplasmic region and mediates the suppressive effects of PD-1[30,31].

5.4 Programmed death-1 – Preclinical Studies

Further evidence for a negative regulatory role of PD-1 comes from studies of PD-1-deficient mice. PD-1-deficient mice develop various autoimmune phenotypes, including dilated cardiomyopathy, a lupus-like syndrome with arthritis and nephritis, and accelerated diabetes mellitus[32-35]. The emergence of these autoimmune phenotypes is dependent upon the genetic background of the mouse strain and many of these phenotypes emerge at different times (almost always after 6 months of age) and show variable penetrance. Thus PD-1 plays a more subtle regulatory role than CTLA-4. In addition to the phenotypes of null mutations, PD-1 inhibition by antibody-mediated blockade in several murine models has been found to play a role in the development of autoimmune diseases such as encephalomyelitis[32], graft-versus-host disease[37], and type I diabetes[34]. Taken together, these results suggest that PD-1 modulates immune responses in tissues undergoing inflammatory responses and PD-1 blockade has the potential to enhance inflammatory (including “anti-self”) responses in tissue, but these

responses are variable and dependent upon various host genetic factors. Thus, PD-1 deficiency or inhibition is not accompanied by a universal loss of tolerance to self-antigens. Preclinical animal models of tumors have shown that blockade of PD-1 by monoclonal antibodies (mAbs) can enhance the anti-tumor immune response and result in tumor rejection. The effects of anti-PD-1 blockade in combination with a variety of chemotherapeutic agents were tested in several murine tumor models (MC38, SA1/N and PAN02).

5.5 Programmed Death Ligand -1 – Expression in Humans

In humans, constitutive PD-L1 expression is normally limited to macrophage-lineage cells, although expression of PD-L1 can be induced on other hematologic cells as well, including activated T cells. Aberrant expression of PD-L1 by tumor cells (retrospectively detected by immunohistochemistry, IHC) has been reported in a number of human malignancies[38-44]. In renal cell carcinoma, high surface expression levels of PD-L1 on tumor cells is related to tumor aggressiveness[41,45] and subjects with high tumor and/or lymphocyte PD-L1 levels are 4.5 times more likely to die from cancer than subjects exhibiting low levels of PD-L1 expression. These findings may be explained by the notion that high PD-L1 expression leads to immune evasion. This hypothesis is supported by separate studies demonstrating that PD-L1 expressed by tumor cells enhances apoptosis of activated tumor-specific T cells in vitro[27] and that the expression of PD-L1 protects tumor cells from the induction of apoptosis by effector T cells[45]. Preclinical data suggests that antitumor activity by PD-1 blockade functions in both PD-L1-expressing and -negative tumors suggesting that it may act in immune priming as well as at the tumor microenvironment level[2,46-50]. This suggests that host mechanisms (i.e. expression of PD-L1 on antigen-presenting cells) limit the antitumor response. Consequently, it is possible that both PD-L1 positive and negative tumors may be targeted using this approach.

5.6 Early stage non-small-cell lung cancer – Background and treatments

Approximately 80% of lung cancer cases are NSCLC with most patients presenting with late stage disease. Of patients with NSCLC, 20% present with stage I or II disease, whereas 30% present with stage III disease and 50% with stage IV disease[9]. A standard TNM staging system is used to determine the staging for NSCLC (Appendix A). Patients with pathologic stage I NSCLC have approximately a 60% 5-year survival. Stage II NSCLC patients have approximately 25% to 40% 5-year survival. Surgical resection remains the mainstay of treatment for stage I and II patients. However, despite apparently curative surgery approximately 50% of stage IB and 70% of stage II NSCLC patients will relapse and eventually die of their disease[13]. A rational approach to eradicate micrometastatic disease and minimize the risk of relapse is treatment with adjuvant or neoadjuvant chemotherapy. Many adjuvant studies have been performed and these trials are summarized in **Table 1**. Although there are some conflicting results, the overall evidence from these studies suggests that adjuvant platinum doublet chemotherapy is

beneficial for good performance status patients with stage II disease. The benefit for stage IB patients is less clear and may depend on the size of the primary tumor and other risk factors[51-56]. The LACE meta-analysis of modern adjuvant and neoadjuvant trials, all of which used cisplatin-based chemotherapy, suggested a 5% survival advantage at 5 years from adjuvant chemotherapy with the benefit being greatest for stage II and IIIA patients. A 2010 meta-analysis including both older and more recent trials confirmed the survival benefit shown in the LACE meta-analysis and also suggested a benefit of adjuvant chemotherapy for stage IB disease patients[57,58]. This led to the adoption of adjuvant platinum doublet chemotherapy for high risk stage IB - IIIA patients. Despite the high risk of relapse and disease-related death in these patients, no new systemic therapies have been shown to prolong survival for this large group of patients.

Table 1 - Selected Adjuvant NSCLC Studies

Trial	Stage	Treatment	Pt No	5 yr OS	HR	p value
ALPI	I - III	Surg MVP	603 601	45% 50%	0.96	0.59
IALT	I – III	Surg Cis-based	935 932	40% 44.5%	0.86	<0.03
ANITA	IB - IIIA	Surg Cis-Vin	433 407	43% 51%	0.80	0.017
BLT	I - IIIA	Surg Cis-based	189 192	58% 60%	1.02	0.90
NCIC/JBR10	IB – II	Surg Cis-Vin	240 242	54% 69%	0.69	0.03
CALGB	IB	Surg Carb-pac	171 173	57% 59%	0.80	0.10

Abbreviations – OS, overall survival; HR, hazard ratio; ALPI, Adjuvant Lung Cancer Project Italy; IALT, International Adjuvant Lung Cancer Trial; ANITA, Adjuvant Navelbine International Triallist Association; BLT, Big Lung Trial; NCI-C, National Cancer Institute-Canada; CALGB, Cancer and Leukemia Group B; MVP, mitomycin, vindesine, cisplatin; Cis-Vin, cisplatin-vinorelbine; Carb-pac, carboplatin-paclitaxel.

5.7 Rationale for preoperative systemic therapy in NSCLC

Traditionally, surgery has preceded systemic therapy in patients with resectable NSCLC. New systemic approaches are generally investigated in the metastatic setting and later in large adjuvant clinical trials. However, patients in the metastatic setting may have received multiple

previous treatments, may have poor performance status and compromised organ function which makes new drug investigation challenging. Furthermore, randomization and follow-up of adjuvant trials may require decades until a new treatment can be introduced into clinical care. Preoperative chemotherapy has been assessed in a number of trials for patients with resectable NSCLC, though most were closed early when the adjuvant chemotherapy data revealed a survival advantage. A meta-analysis based upon seven trials involving 988 patients suggested that neoadjuvant chemotherapy improved overall survival when given preoperatively (five-year survival 20 versus 14 percent without neoadjuvant chemotherapy), this improvement in survival being similar to that noted in the meta-analyses of predominantly adjuvant chemotherapy[59]. Furthermore, preoperative systemic therapy offers the possibility for the identification of surrogate clinical and biological markers that may correlate with response to therapy and, in some cases, long term outcome. In addition, preoperative therapy may be a useful platform for the development of new targeted therapies. Efficient strategies to screen promising agents in early phase development are essential for rapid progress in lung cancer treatment and prevention.

Several studies have shown preoperative systemic therapy to be safe prior to surgical resection of NSCLC with no difference in extent of surgical procedures performed, operative morbidity and mortality[60-62].

5.8 Immunology of NSCLC – Preclinical Findings

Cancer cells characteristically have six intrinsic phenomena that lead to oncogenesis. These include self-sufficiency of growth signals, insensitivity to growth-inhibitory signals, avoidance of apoptosis, limitless replicative potential, development and sustaining of angiogenesis, and tissue invasion and metastasis. An additional phenomenon, avoidance of immunosurveillance, has been proposed since lung cancer cells escape innate and adaptive immune responses[63,64]. This concept is based upon the idea that the immune system can recognize precursors of cancer and destroy them. One strategy of eluding a T-cell-mediated immune response is through the downregulation or loss of expression of HLA class I molecules that is noted to be common in lung cancer. It has been postulated that only immunoselected tumor cells that lack HLA class I expression can escape immune attack and develop into cancers. Tumors often down-modulate antigen processing molecules such as transporter associated with antigen processing 1 (TAP1), low-molecular-mass protein (LMP) 2, LMP7, and tapasin. The overexpression of the serine-protease inhibitor P19 by tumor cells blocks the granzyme-B-perforin pathway of target cell lysis. However, silencing of these genes in lung cancer is reversible and they are usually upregulated by IFN-γ, suggesting that, in the presence of a T cell or NK response, this escape mechanism can be overcome. Additional outcomes of immunoselection include down regulation or mutation of death receptors, methylation or mutation of the gene encoding

caspase-8, and overexpression of FLIP (caspase-8 (FLICE)-like inhibitory protein) or decoy receptors for TRAIL. All of these cause resistance to CTL-induced killing of tumor cells[65]. Key findings that led to greater understanding of immunosurveillance included the discovery that endogenously produced interferon-γ(IFN-γ) protected hosts against transplanted, spontaneous, or chemically induced tumors in mice. Another finding showed that C57BL/6 mice lacking perforin (perforin -/-) were more prone to methylcholanthrene-induced tumor formation compared with their wild-type counterparts. It was noted that the perforin -/- mice lack functional cytotoxic T cells and NK cells[65,66]. Additional research has shown an overlap between the tumor suppressor pathways that depend upon IFN-γ and lymphocyte function. 129/SvEv mice lacking IFN-γ responsiveness (IFNGR1 receptor -/- or STAT1 -/- mice) were compared with mice lacking recombination activating gene (RAG-2) (which fail to rearrange lymphocyte antigen receptors and completely lack natural killer T cells, T and B cells) and RAG-2 and STAT1 negative mice. No difference was noted in the tumor development in the mice as compared with wild-type mice. It was also felt that the IFN-γ and lymphocyte function overlapped with one another[65,66].

5.9 Immunology of NSCLC – Clinical Findings

Data have been reported regarding a correlation between the presence of tumoral lymphocytes and patient survival. The presence of tumor-infiltrating lymphocytes (TILs) has been noted in cancer cell nests and central cancer stroma[67-69]. TILs are stimulated in the presence of IFN-γ and tumor-necrosis factor-α (TNF-α). TILs attempt to regulate the proliferation and metastatic activity of the tumor and interrupt angiogenesis, in addition to their function in host immunity against cancers. The activity of the TILs is noted to be increased in early stage NSCLC, but fails to control tumor cell growth in later stages of cancer[69,71]. Anti-tumor immune system activity may become suppressed by the expansion of regulatory T (Treg) cells in the tumor and the draining N1 and N2 lymph nodes with a compensatory reduction in natural killer (NK) cells6 [70], stimulation of mature and immature dendritic cells providing immunosuppressive cytokines like interleukin-10 (IL-10) or transforming growth factor-β (TGF-β), and further inhibition of dendritic cell maturation by VEGF, IL-6, IL-10, TGF-β, macrophage colony-stimulating factor, NOS2, arginase-1, IDO, PGE2, COX2 and gangliosides[71,72]. PD-1 expression is upregulated on TILs in patients with a variety of tumors including NSCLC[38]. Zhang et al[73] analyzed peripheral blood mononuclear cells (PBMCs) and TILs and noted that PD-1 levels were highest on CD8+ TILs and over 2-fold higher on PBMCs of patients with NSCLC than on control patients. Other inhibitory receptors, such as CTLA-4, were not expressed on the TILs. In vitro, blocking the interaction between PD-1 and its ligands PD-L1 and PD-L2 resulted in TIL proliferation and increased production of IFN-γ. Aberrant expression of PD-L1 by tumor cells has been reported in NSCLC. It is notable that fewer TILs were found in tumors that expressed B7-H1/PD-L1,

though it remains to be seen whether some of the PD-L1 staining procedures reported in the literature are valid[38,74,75].

A study has been performed using the anti-CTLA-4 monoclonal antibody ipilimumab in combination with carboplatin/paclitaxel chemotherapy (CA184041) in advanced NSCLC. This showed a statistically significant improvement in PFS and OS when immunotherapy was added to chemotherapy[76]. Further randomized studies of ipilimumab in squamous NSCLC are ongoing at this time.

5.10 Rationale for Assessment of Bronchial & Nasal Epithelium

Over the past decade, there has been increased molecular and genomic understanding of NSCLC pre-malignant lesions (PMLs), and how these lesions evolve and transform into invasive disease. This stepwise nature of tumorigenesis has been demonstrated in both squamous and non-squamous NSCLC. In squamous NSCLC, increased expression of multiple genes have been demonstrated in PMLs, including SLC2A1, CEACAM5, and PTBP3, in addition to increased activation of MYC (Ooi et al., Cancer Prev Res 2014). In addition, coordinated loss and gain of expression was observed in chromosome 3p and 3q regions, respectively. More recent work has also identified subclasses of gene expression in these pre-malignant lesions, with certain groups demonstrating enrichment of genes associated with T-cell mediated immunity. Similar genomic alterations have been observed in PMLs of adenocarcinoma, with common driver mutations such as KRAS, TP53, and EGFR identified in the earliest stages of cytologic atypia (Izumchenko et al., Nat Commun. 2015). Of significance, alterations associated with DNA repair were identified in most early lesions, lending support to the theory that early spatial heterogeneity and clonal expansion is integral to tumorigenesis. Furthermore, characterization of progression-associated muations located in both PMLs and frank adenocarcinoma identified these mutations and their derived neoepitopes as being associated with the greatest level of CD8+ T-cell infiltration. This suggests immune recognition of neoepitopes occurs even at the earliest stages of lung adenocarcinoma development, and highlights the potential of immunoprevention for lung cancer interception.

These cancer-associated molecular alterations have not only been seen in PMLs, but also in normal appearing bronchial mucosa. mRNA analysis of bronchial epithelial cells has identified changes in key molecular pathways such as JAK/STAT, DNA damage repair, and mitochondrial transport in patients with PMLs compared to those without them (Beane et al., Clin Cancer Res. 2017). Using RNA sequencing, differential expression of many cancer-associated genes has been observed in both the nasal and bronchial epithelium of patients with invasive NSCLC compared to those without invasive cancer (Beane et al., Cancer Prev Res 2011; Perez-Rogers et al., J Natl Cancer Inst 2017). Based off this differential gene expression, novel diagnostic gene-expression arrays have been created that are predictive of lung cancer diagnosis and improve

diagnostic accuracy when combined with bronchoscopy (Spira et al., Nat Med. 2007; Whitney et al., BMC Med Genomics 2015; Silvestri et al., N Engl J Med 2015). However, little is known about how these genomic and molecular alterations evolve in response to cancer treatment. This information could prove vital in identifying novel predictive and prognostic biomarkers for immunotherapy response. Furthermore, when combined with information taken from blood and pathologic tissue, this data will exponentially increase our mechanistic understanding of these paradigm-shifting therapies.

5.11 Development of nivolumab

Nivolumab (BMS-936558, ONO-5438, MDX-1106) is a fully human, IgG4 (kappa) monoclonal antibody that binds PD-1 with high affinity blocking its interactions with its ligands PD-L1 (B7-H1) and PD-L2 (B7-DC) and increasing tumor antigen specific T cell proliferation and cytokine secretion[63].

Nivolumab is currently FDA-approved for the treatment of metastatic NSCLC in patients with progression on or after platinum-based chemotherapy and in the first-line treatment of recurrent or metastatic NSCLC in combination with chemotherapy and/or ipilimumab. Nivolumab, in combination with ipilimumab, is indicted for the 1L treatment of adult patients with metastatic NSCLC whose tumors express PD-L1 (≥1%) as determined by an FDA-approved test, with no EGFR or ALK genomic tumor aberrations. Nivolumab, in combination with ipilimumab and 2 cycles of platinum-doublet chemotherapy, is indicated for the 1L treatment of adult patients with metastatic or recurrent NSCLC, with no EGFR or ALK genomic tumor aberrations. Nivolumab is also indicated for the treatment of patients with metastatic NSCLC with progression on or after platinum-based chemotherapy. Patients with EGFR or ALK genomic tumor aberrations should have disease progression on FDA-approved therapy for these aberrations prior to receiving nivolumab. Many studies are ongoing of nivolumab in combination with other agents in the first-line setting. Nivolumab is also undergoing clinical trial evaluation for unresectable stage III NSCLC and as adjuvant therapy for resected NSCLC. .

In the proposed study, the safety and feasibility of preoperative nivolumab will be assessed as well as the impact of preoperative anti-PD-1 blockade on anti-tumor immunity.

5.12 Clinical experience with nivolumab

A phase 1 single dose, dose-escalation study of nivolumab in 39 heavily pretreated patients with solid tumors (CA209-001, NCT00441337) demonstrated good tolerance and signals of efficacy[11]. Patients with advanced NSCLC, melanoma, RCC, metastatic castration-resistant prostate cancer (mCRPC) and metastatic colorectal cancer (CRC) in this study received a single IV infusion of nivolumab over 1 hour in escalating cohorts of 0.3, 1, 3 or 10 mg/kg. Restaging was performed radiologically at 8 and 12 weeks, and patients with no adverse event (AE) ≥3

and stable disease or response by RECIST received additional doses of nivolumab at weeks 12 and 16 followed by further restaging at 3 months. Those with continued clinical benefit could receive two more doses, spaced by 4 weeks. Treatment could continue for up to 2 years.

Nivolumab was in general well-tolerated with most frequent adverse events being hematologic (notably a grade 3 reduction in CD4 count in 17.9% of patients), fatigue and mild musculoskeletal symptoms. A maximum tolerated-dose (MTD) was not reached.

Immune-related colitis, well described with CTLA-4 inhibition, occurred in 1 patient and resolved with treatment with infliximab and steroids.

Grade 2 hypothyroidism and grade 2 polyarthritis were noted in 1 and 2 patients respectively. Efficacy was promising with a durable complete response (CR) in a CRC patient, 2 partial responses (PRs) in RCC and melanoma patients, and a transient response not meeting PR criteria in a NSCLC patient. PD-L1 (B7-H1) expression was assessed by immunohistochemistry in pretreatment tumor specimens from 9 patients. Of these, 3 of 4 patients with membranous (cell surface) tumor cell expression of PD-L1 experienced tumor regression; none of 5 patients without expression of PD-L1 experienced a tumor response, suggesting a marker for further investigation. Pharmacodynamic analyses suggested that high level occupancy of the PD-1 receptor on circulating T cells persisted for up to 85 days after a single dose of nivolumab.

Safety, efficacy and immune correlative data from a multicenter phase 1b expansion study of single agent nivolumab in a multi-dose regimen were published in 2012 and updated at the ASCO Annual Meeting in 2013[4,77]. Nivolumab was administered as an intravenous infusion every 2 weeks of an 8 week treatment cycle, to patients with advanced treatment-refractory NSCLC, melanoma, RCC, CRC or mCRPC. Patients with partial response or stable disease received treatment for up to 2 years (12 cycles), and after 2 years of treatment, patients were followed for up to 1 year and offered retreatment for an additional year in the event of disease progression. MTD was not reached in this study, and five expansion cohorts of 16 patients each were enrolled at the 10mg/kg dose for each tumor type. After initial assessment of activity, pharmacokinetics and receptor occupancy, additional expansion cohorts of 16 patients each were enrolled for melanoma (0.1, 0.3, 1.0, 3.0mg/kg) NSCLC (squamous and non-squamous, 1.0, 3.0, 10.0mg/kg) and RCC (1.0mg/kg). This study enrolled 306 patients between 10/2008 and 1/2012. The cohort was heavily pretreated with 47% having received ≥3 prior treatments.

Tolerance in general was good with grade 3 or grade 4 treatment related adverse events (most commonly fatigue, diarrhea) noted in 17% of patients. Of particular importance, drug-related pneumonitis occurred in 3% of patients with three (1%) drug-related deaths associated with pneumonitis. Subsequent care in excluding patients with underlying pulmonary inflammatory processes, careful monitoring of lung function and routine institution of systemic steroids upon

altered lung function or radiologic changes appears to have mitigated pneumonitis-related mortality. Objective tumor response or prolonged stabilization of disease was seen in 31% and 7 % of melanoma patients, respectively; 29% and 27% of kidney cancer patients; and 17% and 10% of NSCLC patients. In addition, at the time of report several other patients had unconventional response patterns consistent with "immune-related" responses[78]. No responses to nivolumab were noted in patients with CRC or mCRPC.

In a subgroup of 42 patients for whom PD-L1 expression in pretreatment tumor biopsy was evaluated, 9 of 25 patients responded to nivolumab treatment in the PD-L1-positive group whereas none of 17 patients with PD-L1-negative tumors had a response, suggesting tumor PD-L1 expression as a candidate predictive marker for future investigation. However, only 10 NSCLC patients were included in this evaluation and thus, the consequences of PD-L1 expression in lung cancer in the context of therapeutic PD-1 pathway blockade remain to be fully elucidated.

PD-1 receptor occupancy was assessed on circulating CD3+ T cells and median receptor occupancy was 64 to 70% among the various dose levels. However, the level of PD-1 occupancy among tumor-infiltrating T cells, an important pharmacodynamic measurement, has never been assessed.

Despite early indications that PD-L1 may be a biomarker for response to anti-PD-1, recently reported data using different immunohistochemical assays for PD-L1 have suggested that some patients without PD-L1 expression on their tumors may indeed still respond to modulation of the PD-1 /PD-L1 axis[79-81]. Many questions remain for this issue, including the quality of biopsy sample and the antibody and immunohistochemistry assay used.

In two phase 3 clinical trials, nivolumab has demonstrated a survival advantage over second line chemotherapy with docetaxel in platinum-pretreated metastatic squamous and non-squamous NSCLC[6,7]. In both of these studies nivolumab was associated with a lower incidence of both grade 3-4 and any grade toxicity than docetaxel. In the first-line treatment of metastatic NSCLC, nivolumab has also shown promise in a large phase 1 study with an objective response rate (ORR) of 23% in patients unselected by PD-L1 status[8].

Further information on nivolumab is available in the current version of the investigator's brochure.

5.13 Development of Ipilimumab

Ipilimumab is a fully human monoclonal IgG1κ that binds to the CTLA-4 antigen expressed on a subset of T cells from human and nonhuman primates. CTLA-4 is a negative regulator of T-cell activity. Ipilimumab is a monoclonal antibody that binds to CTLA-4 and blocks the interaction of

CTLA-4 with its ligands, CD80/CD86. Blockade of CTLA-4 has been shown to augment T-cell activation and proliferation, including the activation and proliferation of tumor infiltrating T-effector cells. Inhibition of CTLA-4 signaling can also reduce T-regulatory cell function, which may contribute to a general increase in T-cell responsiveness, including the anti-tumor response. Ipilimumab is currently approved in combination with nivolumab for first-line treatment of metastatic or recurrent NSCLC.

5.14 Clinical Experience with Ipilimumab

BMS and Medarex (acquired by BMS in Sep-2009) have co-sponsored an extensive clinical development program for ipilimumab, encompassing more than 19,500 subjects (total number of subjects enrolled in ipilimumab studies) in several cancer types in completed and ongoing studies, including a compassionate use. The focus of the clinical program is in melanoma, prostate cancer, and lung cancer, with advanced melanoma being the most comprehensively studied indication. Ipilimumab is being investigated both as monotherapy and in combination with other modalities such as chemotherapy, radiation therapy, and other immunotherapies.

Phase 3 programs are ongoing in melanoma, prostate cancer, and lung cancer. In melanoma, 2 completed Phase 3 studies (MDX010-20 and CA184024) have demonstrated a clinically meaningful and statistically significant survival benefit in pretreated advanced melanoma and previously untreated advanced melanoma with a manageable safety profile, respectively. A recent phase 3 study in melanoma demonstrated an improvement in recurrence-free survivial for ipilimumab as adjuvant monotherapy for high-risk Stage III melanoma (CA184029). [82] In addition, a Phase 3 study comparing the safety and efficacy of 3 versus 10 mg/kg ipilimumab monotherapy in pretreated or treatment-naïve subjects with unresectable or metastatic melanoma is ongoing (CA184169).

Toxicities and efficacy of ipilimumab both as a single agent and in combination with nivolumab and other agents are detailed in the current version of the ipilimumab investigator's brochure.

5.15

Rationale for Peri-operative Systemic Immunotherapy in NSCLC

Several studies have shown preoperative cytotoxic chemotherapy to be safe prior to surgical resection of NSCLC with no difference in extent of surgical procedures performed, operative morbidity and mortality[60-62]. Immune checkpoint inhibition has the potential to provide benefits in early stage disease and the anti-CTLA-4 antibody, ipilimumab, is currently in phase III trial investigation in the adjuvant setting for melanoma[82]. Patients with early stage bladder cancer have been treated with preoperative ipilimumab in a phase I study which utilized a similar

dosing schedule and design to our study[86]. Preoperative ipilimumab was found to have a tolerable safety profile without an increase in perioperative complications after 1 or 2 doses of preoperative therapy. Valuable information on the immunological effects of ipilimumab was gleaned from studying the resected bladder tumors in these patients[86].

Subjects with advanced stages of disease on immunotherapy trials rarely undergo surgical biopsies or procedures; therefore, there have been limitations in accessing sufficient tumor tissues for phenotypic and functional immunologic studies. Laboratory studies from these prior trials focused primarily on assessing immune responses in peripheral blood and expression of PD-L1 in archived pretreatment tumor specimens; however, these studies have not yet led to the identification of immunologic markers that clearly predict clinical outcomes and could guide future investigation of immune checkpoint inhibition in NSCLC. The primary aim of our study is to establish the safety and feasibility of using neoadjuvant immunotherapy in the preoperative setting for resectable stage IB, II and IIIA NSCLC. Data obtained from this study will facilitate potential further investigation of immunotherapy as a possible therapy in early stage NSCLC by establishing safety and feasibility, and will also provide a comprehensive data set on the immune response to immunotherapy in serial peripheral blood samples and tumor and biopsies obtained pre/post treatment. Because the patient population to be included in this trial has a high risk of tumor relapse, it is possible that a large effect of neoadjuvant immunotherapy on relapse rate may be observed, this would provide further impetus for future randomized studies evaluating the efficacy of neoadjuvant immunotherapy.

In the initial portion of this study, treatment with two doses of of nivolumab 3mg/kg IV at 4 and 2 weeks prior surgical resection was demonstrated to be safe and feasible in 21 patients with no treatment-related delays to surgery. One patient experienced an SAE, which was assessed by the investigator as being possibly related to nivolumab therapy. This patient proceeded to surgery after a single dose of nivolumab and the rest of his course was uncomplicated. Twenty patients went on to complete resection and this regimen resulted in high pathologic response rates with MPR of 45% and pCR of 10%. Data from this portion of the study were published in the New England Journal of Medicine in 2018 [Forde 2018][103].

Based on these initial results, two further arms were added to the protocol to evaluate the safety and feasibility of a longer course of neoadjuvant nivolumab therapy (3 doses of 240 mg over 6 weeks prior to surgery) and the combination of nivolumab with ipilimumab in the neoadjuvant setting. In expansion Arm A, we explored the combination of ipilimumab and nivolumab. While this combination was safe and feasible per protocol, there did not appear to be evidence of additional efficiacy compared to nivolumab alone and the arm was closed early. Data for this arm were published recently in the journal of Immunotherpy for cancer [Reuss et al. JITC 2020][96]. At the time of this amendment, Arm B (nivolumab alone for three doses

preoperatively) has accrued 13 of a planned 15 patients who underwent full resection. Following accrual to Arm B, we plan to proceed to Arm C which will explore the combination of nivolumab and chemotherapy.

Rationale for combination nivolumab and chemotherapy in NSCLC

The combination of traditional chemotherapy and immunotherapy has demonstrated synergy in NSCLC in phase III studies of metastatic disease. KEYNOTE-189 confirmed prior phase 2 trial KEYNOTE 021 demonstrating higher rates of response and longer progression-free survival with chemotherapy plus pembrolizumab compared to chemotherapy alone [Gandhi et al, Awad et al.][97,98]. This randomized, placebo-controlled study of 616 patients demonstrated one year OS of 69% in the combination arm compared to 49% in the chemotherapy only arm ($p<0.001$). The benefit of adding pembrolizumab was seen across all tumor PD-L1 expression groups (<1%, 1-49%, >=50%). Two trials, IMpower130 and IMpower150, demonstrated that the addition of atezolizumab to carboplatin and paclitaxel (plus bevacizumab in IMpower150), resulted in improved progression free and overall survival in first line treatment of metastatic non-squamous NSCLC [West et al, Socinski et al][99,100]. Most recently, CheckMate 9LA studied the combination of chemotherapy plus nivolumab and ipilimumab versus chemotherapy alone in 719 patients with untreated metastatic NSCLC [Reck et al NEJM 2018][101]. This trial showed longer median overall survival of 15.6 vs 10.9 months with the addition of dual immunotherapy. Notably, the treatment discontinuation rate due to toxicity in the combination immunotherapy arm was 24%, far exceeding the rates of toxicity with single agent immunotherapy. Based on these trials, pembrolizumab, atezolizumab, and nivolumab/ipilimumab are recommended in combination with platinum-doublet chemotherapy as first line treatment for patients with metastatic disease without a targetable driver mutation.

The success of combination chemotherapy-immunotherapy in advanced disease has spurred interest in this combination in patients with resectable NSCLC, and some early phase studies have shown promising responses. In the phase II NADIM study 46 patients with resectable stage IIIA NSCLC received neoadjuvant nivolumab, carboplatin and paclitaxel, and 41 had complete surgical resection, demonstrating the feasibility of this approach [Provencio et al Lancet Oncol 2020][102]. Furthermore, they reported impressive response rates of MPR of 85% and pCR of 61%. These results are striking compared to high response rates with neoadjuvant nivolumab alone in Arm B, with MPR of 45% and pCR of 10% [Forde et al NEJM 2018] [103] or historical response rates with chemotherapy alone of MPR 20% and pCR <5% [Hellmann 2014][105]. Another recent phase 2 trial treated 30 patients with resectable NSCLC with atezolizumab, carboplatin, and nab-paclitaxel for up to four cycles [Shu et al Lancet Oncol 2020] [104]. This approach yielded MPR in 57% of patients and pCR in 33%. These findings suggest that nivolumab plus platinum doublet chemotherapy, may improve both survival and pathologic

response relative to platinum doublet chemotherapy and nivolumab alone, while providing an acceptable safety profile in the neoadjuvant treatment of NSCLC.

Finally, the mechanisms underlying chemo-immunotherapy synergy and biomarkers to predict response have yet to be elucidated and will have clinical application across nearly all stages of NSCLC. In addition to the known cytotoxic effects of chemotherapy, there is growing evidence that traditional chemotherapy reduces tumor immune evasion through multiple pathways and putative mechanisms, including promoting tumor antigen delivery to antigen-presenting cells as well as modulation of tumor-infiltrating T cells, T regulatory cells, and myeloid derived suppressor cells. The addition of Arm C will help characterize the functional immune response to PD-1 blockade with and without chemotherapy at the cellular and molecular level.

Rationale for Dosing and Schedule

The planned schedule in arm B of 3 preoperative doses of nivolumab, flat dose 240mg, given once every 2 weeks (+/- 3 days) with surgery scheduled for approximately 2 weeks after the third dose. The rationale for flat dosing is noted below.. The duration of preoperative treatment of approximately 4 weeks was initially chosen as a short time period unlikely to allow significant progression of disease which might preclude complete surgical resection. Given the excellent tolerability and signals of antitumor efficacy of the initial regimen, an amendment to this protocol explores longer durations of nivolumab treatment (arm B), which may lead to greater pathologic response, and also the addition of a single dose of ipilimumab in combination with nivolumab (Arm A). The combination of ipilimumab (1mg/kg) given every 6 weeks and nivolumab 3mg/kg given every 2 weeks has been shown to be safe and associated with a promising ORR in early phase clinical trials in NSCLC and other solid tumors. This combination therapy is being actively evaluated in phase 3 clinical trials in advanced NSCLC hence the interest in developing this regimen for early stage disease. Due to incomplete accrual and the closing of the nivolumab + ipilimumab in a phase III neoadjuvant study in NSCLC, the current amendment to this protocol has resulted in closing the nivolumab + ipilimumab arm prior to complete accrual and advancing to the 3 preoperative doses of nivolumab arm (Arm B).

Flat (standardized) dosing of nivolumab:

Nivolumab monotherapy has been extensively studied in a number of tumor types including NSCLC, MEL, RCC, and CRC with body weight normalized dosing (mg/kg). Nivolumab pharmacokinetics (PK) and exposures of subjects in these studies have been characterized by population pharmacokinetic (PPK) analysis of data collected in these studies, together with PK data from several phase 1, 2, and 3 clinical studies of nivolumab monotherapy in solid tumors. Population PK (PPK) analyses have shown that the PK of nivolumab are linear, with dose

proportional exposures over a dose range of 0.1 mg/kg to 10 mg/kg, and are similar across tumor types. Nivolumab clearance and volume of distribution were found to increase with increasing body weight, but the increase was less than proportional, indicating that a mg/kg dose represents an over-adjustment for the effect of body weight on nivolumab PK. Given the relationship between nivolumab PK and body weight, a flat dose is expected to lead to lower exposures in heavier patients, relative to the exposures in lighter patients.

Using the PPK model, nivolumab steady-state trough, peak and time-averaged concentration (Cminss, Cmaxss, and Cavgss, respectively) were predicted for a flat nivolumab dose of 240 mg Q2W and compared to those following administration of 3 mg/kg Q2W in NSCLC subjects. A dose of 240 mg nivolumab is identical to a dose of 3 mg/kg for subjects weighing 80 kg, which is the approximate median body weight of NSCLC subjects in the 3 Phase 2 and 3 BMS clinical studies of nivolumab monotherapy. The geometric mean values of Cminss, Cmaxss, and Cavgss with flat dosing are slightly (< 15%) higher than that produced by a 3 mg/kg dose, and the coefficient of variation (cv%) in these measures of exposure are only slightly (< 10%) greater than that of 3 mg/kg dosing.

Across the various tumor types in the BMS clinical program, nivolumab has been shown to be be safe and well tolerated up to a dose level of 10 mg/kg, and the relationship between nivolumab exposure produced by 3 mg/kg and efficacy has been found to be relatively flat. Taken together, the PK, safety, and efficacy data indicate that the safety and efficacy profile of 240 mg nivolumab will be similar to that of 3 mg/kg nivolumab.

Thus a flat dose of 240 mg every 2 weeks is recommended for investigation in arm B of this study.

Dosing and schedule for concurrent nivolumab and chemotherapy (Arm C)

The proposed Arm C dosing regimen and schedule of nivolumab 360 mg, carboplatin AUC 5 or 6, paclitaxel 175 or 200 m/m2 every 21 days (+/- 3 days) for three doses is based on extensive clinical use of these agents.

Safety of nivolumab in combination with platinum doublet chemotherapy was explored in the multichohort phase 1 study CheckMate 012, including 56 chemotherapy-naïve patients with advanced NSCLC treated with concurrent nivolumab (5 or 10 mg/kg) and platinum-doublet chemotherapy every 21 days for four cycles, including carboplatin-paclitaxel, gemcitabine-cisplatin, and pemetrexed-cisplatin [Rizvi JCO 2016]. Twenty-five of 56 patients (45%) reported serious adverse events including 7% with grade 3-4 pneumonitis (n=4). The safety profile of this combination reflected additive toxicities of the individual agents, which were manageable using established safety guidelines. No dose limiting toxicities were observed during the first 6 weeks of treatment. The frequency of most immune-related select AEs was higher for the

combination than what has been observed for nivolumab monotherapy. However, these treatment-related AEs, including pneumonitis, were effectively managed and did not lead to any deaths. The overall response rate across all the nivolumab and chemotherapy cohorts ranged from 33-47% and median duration of response was 27.3 weeks. In the 15 participants that received nivolumab 10 mg/kg plus pemetrexed and cisplatin, 47% achieved a PR or CR. In the 12 participants that received nivolumab 10 mg/kg plus gemcitabine and cisplatin, 33% achieved a CR or PR. In the 15 participants that received nivolumab 10 mg/kg plus paclitaxel and carboplatin, 47% achieved a PR or CR. In the 14 participants that received nivolumab 5 mg/kg plus paclitaxel and carboplatin, 43% achieved a PR or CR. The 1-year survival rate was 87%.

Subsequent studies utilized flat-dosing of nivolumab with chemotherapy, for example Checkmate 227 included an arm with patients receiving nivolumab 360 mg with platinum-doublet chemotherapy every 21 days in patients with <1% PD-L1 expression [Borghei ASCO 2018]. In this study PFS was improved with nivolumab plus chemotherapy vs chemotherapy (mPFS: 5.6m vs 4.7m; HR=0.74 [95% CI: 0.58 to 0.94]). ORR was 36.7% in nivolumab plus chemotherapy arm, 23.1% in chemotherapy arm, and 25.1% in nivolumab plus ipilimumab arm. The rate of grade 3 or 4 treatment-related adverse events was 52% with nivolumab plus chemotherapy, 35% with chemotherapy, and 25% with nivolumab plus ipilimumab, overall, for nivolumab plus chemotherapy, the safety profile and efficacy are consistent with previously reported data as well as data from other PD-(L)1 blockades in combination with chemotherapy.

More recently, in the phase II NADIM study, 46 patients with stage IIIA received the same proposed regimen of nivolumab 360 mg, carboplatin AUC 6, and paclitaxel 200 mg/m2 every three weeks [Provencio 2019] [102]. In this neoadjuvant population, no patients withdrew from the study prior to surgery due to toxicity or progression, and they recorded a grade 3-5 adverse event rate of 15%. Similarly, in a phase II trial of atezolizumab 1200 mg and carboplatin AUC 5 every three weeks with paclitaxel 100 mg/m2 weekly, three of 30 patients (10%) reported a serious treatment-related adverse event [Shu 2020] [104]. Together, the experience with combined chemotherapy and immunotherapy demonstrates acceptable safety profile.

Rationale for nivolumab flat dosing is described previously in this section. A flat dose of 360 mg every 3 weeks is expected to produce the equivalent average exposure to 3 mg/kg every 2 weeks at the median body weight of ~80 kg in nivolumab-treated subjects. Given the longer dosing interval in Arm C, dose delays for nivolumab will be permitted according to criteria in Section 7.6.2.1.

The choice of carboplatin and paclitaxel as the chemotherapy backbone for both squamous and non-squamous histologies is based on the promising data of using this doublet as backbone to combine with nivolumab in neoadjuvant therapy [Provencio][102]. Meta-analysis of individual

participant data from neoadjuvant trials did not identify evidence of a difference in effect of chemotherapy by whether regimens were cisplatin or carboplatin-based (interaction p=0.48) [Meta-analysis 2014-58]. The carboplatin-paclitaxel chemotherapy regimen every 21 days has a well-described safety profile, characterized by myelosuppression, peripheral neuropathy, nausea/vomiting, and infusion-reactions.

Rationale for Main Inclusion and Exclusion Criteria

The choice of resectable stage IB, II and IIIA NSCLC was made as these patients have a high risk of tumor relapse and death with current standard therapy, including surgery with preoperative or postoperative chemotherapy. There is an urgent need for improved, novel therapies in this group of patients. Patients with stage IB NSCLC have been included as these patients are also at high risk of tumor relapse and may be considered candidates for standard adjuvant chemotherapy[53, 54][57,58].

This study initially included patients with primary tumors greater than 4cm in diameter and did not include patients with resectable stage IIIA NSCLC if they had N2 nodal involvement. Data from phase 3 clinical trials of nivolumab versus standard of care docetaxel chemotherapy reported subsequent to this trial commencing accrual, have demonstrated a significant survival advantage for nivolumab over chemotherapy in platinum-pretreated advanced squamous and non-squamous NSCLC[59,60]. In addition, there was approximately a 5 fold reduction in grade 3-4 toxicity with nivolumab compared with docetaxel. These data have led to the approval by the FDA of nivolumab for the treatment of patients with platinum-pretreated metastatic NSCLC. We have also now refined the immunologic analyses (proposed in the exploratory aims of this study) and are confident we can perform them successfully on smaller primary tumors (≥2cm diameter). Discussions with our co-investigators and colleagues at MSKCC have also led to a consensus to include patients with stage IIIA N2 node positive patients that are deemed resectable at the time of diagnosis in this study. Given these new positive data and in conjunction with our thoracic surgical, radiation oncology and medical oncology colleagues at JHU and MSKCC, we have expanded eligibility for this study to include resectable stage IB, II and IIIA NSCLC.

It is anticipated that the majority of patients enrolled on this study will require adjuvant platinum-based chemotherapy. This will commence, if deemed clinically indicated in the postoperative period, according to standard schedules.

Patients who were assessed clinically by their surgeon as possibly requiring a pneumonectomy to obtain complete surgical resection of their primary tumor were initially excluded from this study, as these patients have a significantly higher risk of postoperative complications including

respiratory distress. Given the good tolerability of the regimen in patients enrolled to date, it is now the consensus of surgical, medical oncology and radiation oncology colleagues at JHU and MSKCC to include patients who may require pneumonectomy in this study. This will allow the population enrolled to reflect a real-life cohort of patients with NSCLC rather than a select group.

6. Patient Population

6.1 Subjects

6.1.1 Inclusion Criteria

6.1.1.1 Men and women aged ≥ 18 years old.

6.1.1.2 Histologically proven non-small-cell lung cancer (core biopsy required).

- Squamous or non-squamous histology.
- Diagnostic core biopsy specimens must be reviewed by a faculty pathologist at SKCCC, McGill, SCI, or MSKCC.
- Either a formalin fixed paraffin block that has been confirmed by a pathologist to contain tumor or a minimum of twenty 5-micron tissue sections (slides) of tumor biopsy sample must be available for biomarker evaluation (study pathologist must review for adequacy of sampling). This can be obtained from archived tissues if adequate, or from a new biopsy as needed. (For details of handling of new tissue biopsies, please refer to the lab manual).

6.1.1.3 Stage - NSCLC with primary resection option for potential cure, as assessed by a faculty surgeon at SKCCC, McGill, SCI, or MSKCC. This may include clinical stage IB (≥4cm), II and IIIA(see Appendix A). Subjects with N3 nodal involvement are not included.

6.1.1.4 For patients enrolled at Johns Hopkins only, a bronchoscopy will be required after consent (prior to initiation of treatment)

6.1.1.5 ECOG performance status 0-1 (see Appendix B).

6.1.1.6 Adequate organ function as follows:

- Leukocytes ≥ 2,000/mm^3
- Absolute neutrophil count (ANC) ≥ 1000/mm^3
- Platelet count ≥ 100,000/mm^3
- Hemoglobin ≥ 9 g/dL

- Creatinine ≤ 1.5 x ULN or creatinine clearance (CrCl) ≥40 mL/min (if using the Cockcroft-Gault formula below):

$$\textit{Female}\ \text{CrCl} = \frac{(140 - \text{age in years}) \times \text{weight in kg} \times 0.85}{72 \times \text{serum creatinine in mg/dL}}$$

$$\textit{Male}\ \text{CrCl} = \frac{(140 - \text{age in years}) \times \text{weight in kg} \times 1.00}{72 \times \text{serum creatinine in mg/dL}}$$

- Total Bilirubin ≤ 1.5 x ULN (except subjects with Gilbert Syndrome, who can have total bilirubin < 3.0 mg/dL)
- AST(SGOT), ALT(SGPT), and alkaline phosphatase ≤ 3 times the upper limit of normal
- Subjects must have adequate lung function to permit surgical resection determined by pre-enrollment pulmonary function tests to include DLCO

6.1.1.7 The effects of nivolumab on the developing human fetus are unknown. Paclitaxel and carboplatin are pregnancy category D. For this reason, women of child-bearing potential (WOCBP) and men must agree to use adequate contraception (hormonal or barrier method of birth control; abstinence) prior to study entry and for the duration of study participation and for up to 23 weeks after the last dose of nivolumab and/or chemotherapy. Should a woman become pregnant or suspect she is pregnant while she or her partner is participating in this study, she should inform her treating physician immediately. Sexually active fertile men must use effective barrier birth control if their partners are WOCBP for up to 31 weeks after the last dose of nivolumab. WOCBP must have a negative serum or urine pregnancy test (minimum sensitivity 25 IU/L or equivalent units of HCG) within two weeks of registration. Women must not be breastfeeding.

6.1.1.8 Patient understands the study regimen, its requirements, risks and discomforts and is able and willing to sign the informed consent form. Voluntary signed and dated IRB/IEC approved written informed consent form in accordance with regulatory and institutional guidelines must be obtained before the performance of any protocol related procedures that are not part of normal patient care. Subjects must be competent to report AEs, understand the drug dosing schedule and use of medications to control AEs.

6.1.2 Exclusion Criteria

6.1.2.1 Subjects are excluded if they have an active, known or suspected autoimmune disease. Subjects are permitted to enroll if they have vitiligo, type I diabetes mellitus, residual

hypothyroidism due to autoimmune condition only requiring hormone replacement, psoriasis not requiring systemic treatment, or conditions not expected to recur in the absence of an external trigger.

6.1.2.2 Subjects are excluded if they have a condition requiring systemic treatment with either corticosteroids (> 10 mg daily prednisone equivalents) or other immunosuppressive medications within 14 days of study drug administration. Inhaled or topical steroids and adrenal replacement doses > 10 mg daily prednisone equivalents are permitted in the absence of active autoimmune disease. As there is potential for hepatic toxicity with nivolumab or nivolumab/ipilimumab combinations, drugs with a predisposition to hepatoxicity should be used with caution in patients treated with nivolumab-containing regimen.

6.1.2.3 Administration of chemotherapy or any other cancer therapy in the pre-operative period other than that indicated by treatment Arm C.

6.1.2.4 Subjects with **active** concurrent malignancies are excluded i.e. cancers other than NSCLC (except non melanoma skin cancers, in situ bladder, gastric, breast, colon or cervical cancers/dysplasia).

6.1.2.5 Subjects with brain metastasis are excluded from this study, and all patients should have brain imaging (either MRI brain or CT brain with contrast) prior to enrollment.

6.1.2.6 Subjects with a history of symptomatic interstitial lung disease.

6.1.2.7 Active systemic infection requiring therapy, positive tests for Hepatitis B surface antigen or Hepatitis C Antibody.

6.1.2.8 Known positive history or positive test for Human Immunodeficiency Virus or Acquired ImmunoDeficiency Syndrome (AIDS).

6.1.2.9 History of allergy to study drug components.

6.1.2.10 Women who are pregnant or nursing.

6.1.2.11 Men with female partners (WOCBP) that are not willing to use contraception.

6.1.2.12 Prior therapy with an anti-PD-1, anti-PD-L1, anti-PDL-2, or anti-CTLA-4 antibody (or any other antibody targeting T cell co-regulatory pathways).

6.1.2.13 Underlying medical conditions that, in the Investigator's opinion, will make the administration of study drug hazardous or obscure the interpretation of toxicity or adverse events.

6.1.2.16 Prisoners or subjects who are involuntarily incarcerated or compulsorily detained for treatment of either a psychiatric or physical (e.g. infectious disease) illness.

6.1.2.17 History of allergy or hypersensitivity to study drug components.

6.1.2.18 > Grade 2 peripheral neuropathy (Arm C only)

6.2 Inclusion of Genders and Minorities

Individuals of all races and ethnic groups are eligible for this trial. There is no bias towards age, gender or race in the clinical trial outlined. This trial is open to the accrual of men and women who meet the inclusion/exclusion criteria outlined.

7. Overview of Study Design and Treatment Plan

7.1 Recruitment

Patients will be recruited through the thoracic oncology clinics at Sidney Kimmel Comprehensive Cancer Center at Johns Hopkins (SKCCC), McGill University Health Center (McGill), Swedish Cancer Institute (SCI), and at Memorial Sloan-Kettering Cancer Center (MSKCC). Each enrolled subject will be assigned a participant sequential subject number at the time of consent e.g., 1-001, 1-002, etc.

7.2 Determination of Eligibility

After eligibility is established, the study staff will register participants. The following are required to be submitted for successful registration:

- Registration forms
- Copy of subject consent
- If an archival tumor sample is being used to satisfy eligibility then it should be evaluated by a study pathologist to confirm it meets the study requirements (see section 7.4.2) prior to the patient commencing therapy.
- Copies of the following documents:
    - Diagnostic pathology report(s)
    - PET/CT scan report, MRI brain or CT brain with contrast report
    - Laboratory reports including:

- Complete blood count (CBC) with differential (including absolute lymphocyte count) and direct platelet count.
- Chemistry: Albumin, SGOT (AST), SGPT (ALT), Bilirubin (direct and total), Calcium, Creatinine, Glucose, Total protein, Urea nitrogen, Electrolytes (including sodium, potassium, chloride and bicarbonate).
- Baseline thyroid immune safety assay: Thyroid Stimulating Hormone (TSH). Abnormal endocrine results should be followed up per standard of care, and may require an endocrine consult and additional testing.

- Other documents, if requested.

Study treatment cannot begin until the patient is registered and randomized to either Arm A (nivolumab + ipilimumab) or Arm B (nivolumab). At the time of the current amendment, Arm B is completing enrollment. All subsequent additional patients will enroll on Arm C (nivolumab + chemotherapy).

Subjects who sign a consent form but do not initiate protocol treatment for any reason (e.g., subjects who are screen failures), and patients who do not undergo surgical resection will be replaced and will not count towards our accrual goal.

7.3 Study Design and Toxicity Assessments

This is a two arm study that will be conducted at SKCCC , SCI, McGill, and MSKCC.

7.3.1 Screening - Eligible subjects will be consented to receive the investigational treatments (nivolumab +/- ipilimumab or nivolumab + chemotherapy). The initial planned study sample size for the protocol was to consist of 15 patients in each Arm A and B. Arm A was closed prior to complete accrual. Arm B and Arm C will sequentially accrue 15 resected patients to each arm. The staff at the treating center (SKCCC , McGill, SCI, or MSKCC) will arrange drug supply and treatment. Nivolumab and ipilimumab will be supplied by Bristol-Myers Squibb Pharmaceuticals. Carboplatin and paclitaxel are standard of care treatment for resectable NSCLC.

7.3.2 Treatment and collection of biological specimens—For Arm A 15 patients with resectable stage IB(≥4cm), II or IIIA NSCLC (squamous and non-squamous) were planned to be enrolled and receive preoperative nivolumab, 3mg/kg IV, on Day -42, -28 and Day -14 (+/- two days for each timepoint) + ipilimumab 1mg/kg IV on Day -42 prior to planned surgery on Day 0 (to allow for scheduling surgery may take place between Day -3 and Day +10). See section 9 for details on nivolumab and ipilimumab administration. A prior amendment to the protocol moved forward with enrollment of Arm B prior to completion of accrual to Arm A. For Arm B, 15 patients with resectable stage IB (≥4cm), II or IIIA NSCLC will be enrolled and receive nivolumab 240mg IV on Day -42, -28 and -14

(+/- 2 days for each timepoint) prior to planned surgery on Day 0 (to allow for scheduling delays surgery may take place between Day -3 and Day +10).

For Arm C, 15 patients with resectable stage IB, II or IIIA NSCLC will be enrolled and receive nivolumab at a flat dose of 360 mg as 30-minute IV infusion on Day 1, followed by paclitaxel 175 or 200 mg/m2 IV over 180 minutes or per institutional standard and carboplatin AUC 5 or 6 IV over 30 minutes or per institutional standard of a 3-week treatment cycle, for up to 3 cycles. When study drugs (nivolumab and chemotherapy) are to be administered on the same day, nivolumab is to be administered first. Nivolumab infusion must be promptly followed by a saline flush to clear the line of nivolumab before starting the chemotherapy infusion. The time in between infusion of nivolumab and chemotherapy is expected to be approximately 30 minutes but may be more or less depending on the situation. Participants in Arm C may be dosed no less than 18 days between treatments. Surgery will be completed on Day 0, within 6 weeks after the final dose of neoadjuvant systemic therapy.

Serial peripheral blood samples for exploratory analyses will be collected prior to each dose of study drug(s), once within the 3 days prior to surgery, and at 3-6 weeks after surgery. To explore gut microbial correlates of response to neoadjuvant nivolumab +/- ipilimumab, stool samples will be collected at baseline (within 48h of D -42), prior to surgery (D -3 to Day 0), and follow-up visits. Subjects will also be asked to fill out a medical and dietary questionnaire at these times that will be used to assess whether antibiotic use or dietary patterns correlate with features of the gut microbiome.

Postoperatively subjects will receive standard of care treatment and will be followed every 3-6 months with clinic visits and research blood draws. Preoperative core biopsies of the primary tumor, and optional preoperative mediastinal lymph node biopsies, will be obtained. Archived (if sufficient tissue is available) or new tissue specimens will fulfill these criteria. Patients will have a PET/CT scan during the 7 days prior to surgery to assess response to treatment.

7.3.3 Toxicity assessments (Arms A & B) – Safety will be monitored continuously by the study investigators for all patients through day 100 following the last dose of study drug. The initial six patients to be enrolled in Arm A (ipilimumab + nivolumab) will be monitored continually through day 100 following the last dose of study drug (or day 30 post surgery, whichever is longer). Safety will be monitored on a continuous basis by the study investigators. A detailed statistical analysis plan for safety and feasibility is contained in section 14 of this protocol.

Dose Delays due to Toxicity – No dose delays due to toxicity will be permitted for patients enrolled on Arms A and B of this study. For example, if patients, after receiving the first dose of nivolumab (Day -42) are unable to receive the second dose on Day -28 (+/-2 days) due to treatment-related toxicity, they will be discontinued from study drug and will proceed to surgery after standard preoperative evaluation. Prior to surgery, any treatment-related toxicity should have resolved to ≤grade 1.

General Management Algorithms for potential nivolumab (and/or ipilimumab)-related toxicities are contained in Appendix D of this protocol and in the Investigators Brochure.

7.3.4 Dose-Limiting toxicity (DLT) (Arms A & B) is defined as any of the items listed below that occur through day 100 following the last dose of nivolumab (+/- ipilimumab) (or day 30 post surgery, whichever is longer).
Any patient in Arm A &B who experiences a DLT will be permanently discontinued from treatment and will proceed to surgery after standard preoperative evaluation by a surgeon and anesthesiologist. Prior to surgery any treatment-related toxicity should have resolved to ≤grade 1.

- Any ≥ Grade 2 drug-related pneumonitis or interstitial lung disease that does not resolve to Grade 0 or 1 within 2 weeks with systemic steroids. The management algorithm for pneumonitis or pulmonary toxicity can be found in the appendix of current Investigator Brochure for nivolumab
- Any ≥ Grade 2 drug-related uveitis or eye pain that does not respond to topical therapy and does not improve to Grade 1 severity within the re-treatment period OR requires systemic treatment. If uveitis occurs during nivolumab treatment, workup and treatment should follow the nivolumab uveitis toxicity treatment algorithm located in the appendix of the nivolumab Investigators Brochure.
- Any Grade 3 non-skin drug-related adverse event lasting ≥ 7 days with the exception of asymptomatic laboratory abnormalities.
- Grade 3 drug-related bronchospasm, allergic reaction, or infusion-related reaction will be recorded and treated as per the guidelines in 7.4.3; however, it will not count as a dose-limiting toxicity for study purposes.
- Any Grade 3 drug-related diarrhea that does not respond to dose delay and the use of systemic steroids within 2 weeks. If diarrhea occurs during nivolumab treatment, workup and treatment should follow the nivolumab diarrhea toxicity treatment algorithm located in the appendix of the nivolumab Investigators Brochure.

- Any Grade 4 drug-related adverse event, including laboratory abnormalities apart from isolated Grade 4 electrolyte imbalances/abnormalities that are not associated with clinical sequelae and are corrected with appropriate management within 72 hours of their onset.
- Any of the following drug-related hepatic function laboratory abnormalities or potential Drug Induced Liver Injury (DILI):
  − AST or ALT > 5-10x ULN for > 2 weeks
  − AST or ALT >10x ULN
  − Total bilirubin > 5 x ULN
  − Concurrent AST or ALT >3 x ULN and total bilirubin >2 x ULN
- If suspected DILI occurs, workup and treatment should follow the nivolumab DILI treatment algorithm located in the appendix of the nivolumab Investigators Brochure.
- Any other toxicity which is assessed by the principal investigator as having directly led to a delay in surgical resection more than 40 days past the planned Day 0.
- Failure to complete all protocol specified treatment doses due to toxicity (at the discretion of the PI).
- **Note** - Adverse events of special interest are nivolumab-related events with potential immune-mediated causalities. For example, this may include cutaneous toxicities, colitis, liver function abnormalities (AST, ALT, total bilirubin, alkaline phosphatase), endocrine abnormalities (hyperthyroidism, hypothyroidism, hypophysitis and secondary adrenal insufficiency), interstitial pneumonitis and nephritis. These events will be noted. However, it may not constitute DLT's unless they fulfill the previously outlined DLT criteria in section 7.3.3 above

7.3.5 Toxicity Assessment for Arm C: Nivolumab + Carboplatin/Paclitaxel

No dose modifications for nivolumab flat dose are permitted, but dose can be delayed or discontinued for patients in Arm C based on criteria in Section 7.6.2.1. Doses of paclitaxel and/or carboplatin may be modified, delayed, or discontinued depending on how well the participant tolerates the treatment. Dose modifications for chemotherapy toxicity will be performed according to Section 7.6.2.2. Dose delay criteria for chemotherapy can be found in Section 7.6.2.3, and discontinuation criteria can be found in Section 7.6.2.4. Criteria to resume chemotherapy can be found in Section 7.6.2.5.

Dosing of all drugs should be delayed if any criteria in Section 7.4.2.1 (nivolumab) or Section 7.6.2.3 (platinum doublet chemotherapy) are met. That is, nivolumab should be delayed if criteria for delay of platinum doublet chemotherapy are met, and platinum doublet chemotherapy should be delayed if criteria for delay of nivolumab are met.

7.3.5.1 Nivolumab toxicity guidelines for Arm C

Nivolumab dose modifications: Treatments will be delayed or discontinued as below.

Nivolumab dose delays: Administration should be delayed for the following:

- Grade 2 non-skin, drug-related adverse event, with the exception of fatigue
- Grade 2 drug-related creatinine, AST, ALT and/or Total Bilirubin abnormalities
- Grade 3 skin, drug-related adverse event
- Grade 3 drug-related laboratory abnormality, with the following exceptions:
  - Grade 3 lymphopenia or asymptomatic amylase or lipase does not require dose delay
  - Grade ≥ 3 AST, ALT, Total Bilirubin will require dose discontinuation (see Section 7.6.1.2)
- Any adverse event, laboratory abnormality, or intercurrent illness which, in the judgment of the investigator, warrants delaying the dose of study medication.

Participants who require delay of nivolumab should be re-evaluated weekly or more frequently if clinically indicated and resume nivolumab dosing when re-treatment criteria are met.

Nivolumab dose discontinuation: Nivolumab will be discontinued for any DLT listed above in Section 7.6.1.2.

Nivolumab criteria to resume treatment: Participants may resume treatment with study drug when the drug-related AE(s) resolve to Grade ≤ 1 or baseline value, with the following exceptions:

- Participants may resume treatment in the presence of Grade 2 fatigue
- Participants who have not experienced a Grade 3 drug-related skin AE may resume treatment in the presence of Grade 2 skin toxicity
- For participants with Grade 2 AST, ALT and/or Total Bilirubin abnormalities, dosing may resume when laboratory values return to baseline and management with corticosteroids, if needed, is complete.
- Participants with combined Grade 2 AST/ALT AND total bilirubin values meeting discontinuation parameters (Section 7.6.1.2) should have treatment permanently discontinued.
- Drug-related pulmonary toxicity, diarrhea or colitis must have resolved to baseline before treatment is resumed. Participants with persistent Grade 1 pneumonitis after completion of a steroid taper over at least 1 month may be eligible for retreatment if discussed with and approved by BMS Medical Monitor.

7.3.5.2 Dose modification criteria for platinum doublet chemotherapy

Dose reductions of platinum doublet chemotherapy may be required and will be performed as below. For any drug-related toxicity/AE that requires dose reduction, chemotherapy should be delayed until recovery and dose modification should be applied as described in Tables 2, 3 and 4. These serve as a guide and do not replace investigator judgment and applicable local label recommendations if more stringent. Chemotherapy dose reductions are permanent; once the dose of any chemotherapy agent is reduced, it may not be re-escalated in subsequent cycles. The dose reductions for each agent are not linked and may be adjusted independently per institutional standards. Any participants with 2 prior dose reductions for 1 agent who experiences a toxicity that would cause a third dose reduction must be discontinued from that agent.

If chemotherapy must be delayed due to drug-related toxicity/AE, investigators will withhold both chemotherapy and nivolumab until chemotherapy retreatment criteria are met and restart both at the same time. If chemotherapy retreatment criteria are not met or toxicity does not resolve within 3 weeks (before next cycle is due), nivolumab can be continued on schedule provided the retreatment criteria for nivolumab are met.

Table 2: Recommended Dose Modifications/Discontinuation for Carboplatin and Paclitaxel

Dose level	**Carboplatin**	**Paclitaxel**
Dose level 0	AUC 5 or 6	175 or 200 mg/m2
Dose level -1	AUC 4 or 5	150 mg/m2
Dose level -2	AUC 3 or 4	100 mg/m2
Dose level -3	Discontinue	Discontinue

Dose reductions for hematologic toxicity

Dose modifications for hematologic toxicities (according to CTCAE version 4) are summarized in Table 3. Dose adjustments are based on nadir blood counts (assessed as per local standards) since the preceding drug administration. Dose level adjustments for platinum doublet chemotherapy are relative to that of the preceding administration. Generally, both chemotherapy agents in the platinum doublet chemotherapy regimen should be dose reduced together for hematologic toxicity. After the first cycle, growth factors may be used to assist hematologic recovery. Use local standards of care in the use of these supportive measures. Additionally, prophylactic antibiotics may be used according to local standards of care. Investigators will report any antibiotic or growth factor use.

Table 3: Recommended Dose Modifications for Chemotherapy Hematological Toxicity

Toxicity	**Carboplatin**	**Paclitaxel**
ANC < 0.5 (Grade 4)	Reduce one dose level and consider prophylactic G-CSF in subsequent cycles	Reduce one dose level and consider prophylactic G-CSF in subsequent cycles
Platelets 25 to < 50k (Grade 3)	Reduce one dose level	Reduce one dose level
Platelets < 25k (Grade 4)	Reduce one dose level	Reduce one dose level
Hemoglobin <10.0 to 8.0 (Grade 2)	Reduce one dose level	Reduce one dose level
Hemoglobin < 8.0 (Grade 3)	Reduce one dose level	Reduce one dose level
Hemoglobin low with life threatening consequences (Grade 4)	Hold Drug	Hold Drug

Dose reductions for non-hematologic toxicities

Dose adjustments for platinum doublet chemotherapy for non-hematologic toxicities during treatment are described in Table 4. All dose reductions should be made based on the worst grade toxicity. Participants experiencing any of the toxicities detailed in Table 4 during the previous cycle should have their chemotherapy delayed until retreatment criteria are met (per Section 7.6.2.5) and then reduced for all subsequent cycles by 1 dose level or discontinued as appropriate. Dose levels for the carboplatin and paclitaxel are not linked and may be reduced independently, as summarized in Table 4.

Table 4: Recommended Dose Modifications for Chemotherapy Non-Hematological Toxicity

Toxicity	**CTC Grade**	**Carboplatin**	**Paclitaxel**
Febrile Neutropenia	Grade ≥ 3	Reduce one dose level	Reduce one dose level
Diarrhea	Grade ≥ 3	No change	Reduce one dose level
Acute hypersensitivity or infusion reaction	Grade ≥ 3	Discontinue	Discontinue

Neuropathy	Grade 2	No change	Reduce one dose level
	Grade ≥ 3	Discontinue	Discontinue
Calculated Creatinine Clearance < 20 ml/min		Discontinue	No change
Other toxicity (except for fatigue and transient arthralgia and myalgia)	Grade ≥ 3	Adjust as medically indicated	Adjust as medically indicated

7.3.5.3 Dose Delay Criteria for Platinum Doublet Chemotherapy

If any agent is delayed > 7 days, the dose should be skipped, and the participant should resume treatment at the next scheduled dose if criteria to resume treatment (Section 7.6.2.5) are met.

Dosing of both drugs in the platinum doublet chemotherapy regimen selected should be delayed for any of the following on the Day 1 of each cycle:

- Absolute neutrophil count (ANC) < 1500/µL
- Platelets < 100,000/mm3
- Any Grade ≥ 2 non-skin, non-hematologic, drug-related adverse event (excluding Grade 2 alopecia, Grade 2 fatigue, and Grade 2 laboratory abnormalities)
- Any Grade ≥ 3 skin, drug-related adverse event
- Any Grade ≥ 3 drug-related laboratory abnormality, with the following exceptions for lymphopenia, AST, ALT, or total bilirubin:
    - Grade 3 lymphopenia does not require dose delay.
    - If a participant has a baseline AST, ALT or total bilirubin that is within normal limits, delay dosing for drug-related Grade ≥ 2 toxicity.
    - If a participant has baseline AST, ALT, or total bilirubin within the Grade 1 toxicity range, delay dosing for drug-related Grade ≥ 3 toxicity.

Any adverse event, laboratory abnormality, or intercurrent illness which, in the judgment of the investigator, warrants delaying the dose of study medication. Investigators should consult local labeling for the chemotherapy drugs being administered to any given participant for additional guidance on dose delays.

If both drugs in the platinum doublet chemotherapy regimen are delayed, then the participant should be re-evaluated weekly or more frequently if clinically indicated until re-treatment criteria are met (as per Section 7.3.5.5).

7.3.5.4 Dose Discontinuation Criteria for Platinum Doublet Chemotherapy

Except where specified below, both chemotherapy drugs in the platinum doublet chemotherapy regimen should be discontinued for any of the following:

- Any Grade ≥ 3 peripheral neuropathy
- Grade ≥ 3 drug-related thrombocytopenia associated with clinically significant bleeding
- Any drug-related liver function test (LFT) abnormality that meets the following criteria requires discontinuation:
  - AST or ALT > 5-10x ULN for > 2 weeks
  - AST or ALT > 10x ULN
  - Total bilirubin > 5 x ULN
  - Concurrent AST or ALT > 3 x ULN and total bilirubin > 2 x ULN
- Any drug-related adverse event which recurs after 2 prior dose reductions for the same drug-related adverse event (as specified in Section 7.6.2.2) requires discontinuation of the drug(s) which was/were previously dose reduced.
- Any Grade ≥ 3 drug-related hypersensitivity reaction or infusion reaction requires discontinuation of the drug(s) felt to be causing the reaction. The drug not felt to be related to the hypersensitivity reaction or infusion reaction may be continued.
- Any Grade 4 drug-related adverse event which the investigator deems is inappropriate to be managed by dose reduction(s) requires discontinuation of the drug(s) felt to be causing the event. The drug not felt to be related to the event may be continued.
- Any event that leads to delay in dosing of any study drug(s) for > 6 weeks from the previous dose requires discontinuation of that drug(s) with the following exception:
  - Dosing delays lasting > 6 weeks from the previous dose that occur for non-drug-related reasons may be allowed if approved by the BMS medical monitor. Prior to re-initiating treatment in a participant with a dosing delay lasting > 6 weeks, the BMS Medical Monitor must be consulted. Periodic study visits to assess safety and laboratory studies should also continue every 6 weeks or more frequently if clinically indicated during such dosing delays.
- Any adverse event, laboratory abnormality, or intercurrent illness which, in the judgment of the Investigator, presents a substantial clinical risk to the participant with continued platinum doublet chemotherapy dosing. Investigators should consult local labeling for the chemotherapy drugs being administered to any given participant for additional guidance on dose discontinuation.

For participants in Arms C, if the investigator is unable to determine whether a serious adverse event is due to nivolumab or to platinum doublet chemotherapy, then all drugs must be discontinued.

7.3.5.5 Criteria to Resume Platinum Doublet Chemotherapy Dosing

- Participants may resume treatment with platinum doublet chemotherapy when the ANC returns to ≥ 1500/µL, the platelet count returns to ≥ 100,000/mm3, and all other drug-related toxicities have returned to baseline or Grade ≤ 1 (or Grade ≤ 2 for alopecia and fatigue)
- If a participant fails to meet criteria for reinitiating treatment, then treatment should be delayed, and the participant should be re-evaluated weekly or more frequently as clinically indicated
- When resuming platinum doublet chemotherapy treatment, follow the dose reduction recommendations in Section 7.6.2.2.

7.4 Diagnostic and surgical evaluation of participants

7.4.1 Diagnostic evaluation and pre-surgical workup

All patients enrolled on this protocol must be surgical candidates with clinical stage IB, II or IIIA NSCLC. Patients will have undergone radiographic evaluation indicating no evidence of distant disease and no evidence of unresectable loco-regional tumor extension before surgical resection. Any further preoperative testing that is recommended by the surgeon or anesthesiologist will be performed as part of standard of care. Surgery for patients enrolled on this protocol will be according to generally accepted standards of care. It is advised that patients have at least 3 mediastinal and hilar lymph node stations sampled during surgery. Patients should have a PET/CT scan and CT of chest with IV contrast performed after completing 3 doses of neoadjuvant therapy and within the 7 days prior to planned surgery to assess response.

7.4.2 Bronchial airway brushing collection (mandatory for patients enrolled at Johns Hopkins only)

Endobronchial brushings will be obtained using bronchoscope. Samples will be obtained at the level of the right upper lobe take-off within the right mainstem bronchus unless this area is grossly involved with disease, in which case brushings will be obtained from the contralateral side. Four separate brushings will be obtained using one brush per cryovial. Each cytology brush will be rubbed vigorously against the mucosa for 10-20 strokes rotating the brush along its axis to obtain cells from the 360-degree surface of the brush. A visible abrasion should be at the site afterwards, which ensures appropriate brush-mucosa contact has been obtained. After obtaining brushings, brush will be cut and placed into the appropriate reagent-containing cryovial and shaken vigorously.

7.4.3 Nasal Epithelium collection (for patients enrolled at Johns Hopkins only)

Patient will blow nose to remove debris. Patient will be offered option of lidocaine for numbing prior to procedure. A speculum will be inserted into left nostril and, using penlight for visual inspection, a cytopak soft brush will be inserted just past the inferior turbinate and pressed against the outside of the nose, opposite the septum. Brush will be rotated while in contact with nasal epithelium for ~3 seconds. Brush will then be removed and placed into vial containing RNAprotect cell reagent. This will be repeated with a 2nd brush in the same nostril.

7.4.4 Tumor sample acquisition

Patients enrolled on this study will be required to have pretreatment primary tumor biopsy material available for diagnosis and exploratory immunological studies. This may consist of diagnostic biopsies that have been previously performed, or biopsies conducted by the study team in the case of inadequate pre-existing material. Excisional biopsies, or ideally 8 (minimum 6) core needle biopsies (≤19 gauge diameter) of the primary tumor are required; the first 3 core needle biopsies obtained from a research biopsy will be placed in formalin and paraffin embedded while any subsequent core needle biopsies obtained should be flash frozen; fine needle aspirates will not be adequate. A minimum of twenty 5-micron paraffin tissue sections or a paraffin-embedded tumor block is required. If an archival paraffin embedded block is used the presence of tumor in the block must be confirmed by a pathologist. If archival slides are being used they must have been cut with DNA precautions. Biopsies may be obtained by the following approaches: transbronchial, radiographically-guided transthoracic approach, or video-assisted thoracoscopy. Biopsies that are formalin-fixed and paraffin embedded (FFPE) are required. Fresh frozen biopsy specimens may be analyzed in addition to, but not in place of, FFPE specimens. Pretreatment biopsies of draining lymph nodes are desirable but not required; fine-needle aspirations of lymph nodes (>21 gauge diameter) are acceptable.

Primary tumor, draining lymph nodes and normal lung specimens will be collected from patients who undergo surgical resection, after receiving nivolumab. After removal of tissue necessary for clinical assessment, remaining tissue specimens for research purposes will be divided in surgical pathology into 1)fresh tissue that will be transported to the laboratory for viable cell isolation, 2)fixation and paraffin embedding (FFPE), and

3) flash frozen for DNA and RNA analysis. If additional tissue remains, it will be flash frozen.

Specific procedures for accessioning specimens are outlined in detail in the laboratory manual.

7.5 Postoperative treatment of participants

7.5.1 Adjuvant chemotherapy

Postoperative chemotherapy will be administered at the discretion of the treating oncologist based on established standard indications. Postoperative chemotherapy will start at a time based on the standard of care approach at the institution taking into account postoperative recovery time for the subject. Postoperative chemotherapy should not commence until treatment-related toxicity due to study drug(s) has resolved to <grade 2.

7.5.2 Postoperative radiation therapy

Postoperative radiation therapy (PORT) will be administered based on a standard of care at the discretion of the treating oncologist. Subjects who are recommended PORT should have a CT scan of thorax (non-contrast or contrast) to exclude subclinical changes suggestive of pneumonitis prior to commencing PORT. In the event that subjects have radiological findings suggestive of pneumonitis on this scan, then further assessment +/- treatment may be required prior to commencing PORT.

7.6 Evaluation of peri-operative safety

The subject's medical record will be reviewed on a weekly basis from the start of therapy until 100 days after the last dose of study treatment or 30 days following surgery, whichever is longer, for information regarding operative complications including delay in planned surgery and in particular potential immune related toxicities (Note: Only those subjects who initiate protocol treatment will be followed). Toxicities will be reviewed at regular meetings of study investigators and minutes of these meetings will be documented by the clinical research staff. In the event that a subject does not continue his or her peri-operative care at the institution, every attempt will be made to collect this information either by direct contact or through communication with the subjects outside physician(s).

7.7 Discontinuation, withdrawal and replacement of subjects

All patients who receive at least one dose of study treatment (including those who do not undergo surgical resection of their tumor) will be included in the overall evaluation of safety (intention-to-treat analysis). All reasons for discontinuation of therapy should

be documented clearly in the medical record. If a subject discontinues or withdraws from the study, every attempt will be made to obtain an off-study blood collection if the subject is able and willing to do so.
Subjects who do not undergo surgical resection of their tumor, while evaluable for safety, will be replaced and will not count toward the 15 patient per arm accrual target.

7.7.1 Discontinuation of Treatment

The reasons for discontinuation of protocol treatment include:

- Evidence of significant disease progression during the preoperative phase at the discretion of the treating investigator.
- Non-compliance with the study protocol; including, but not limited to not attending the majority of scheduled visits. The principal investigator will determine when non-compliance should lead to removal from study. Note: The patients will still be included in the overall evaluation of safety (intent-to-treat analysis).
- Unacceptable toxicity. Note: The patients will still be included in the overall evaluation of safety (intent-to-treat analysis).
- Intercurrent illness or condition that would, in the judgment of the treating investigator, affect assessment of clinical status to a significant degree or require discontinuation of study treatment.
- At subject's own request. Note: The reason for discontinuation from the study must be documented. The patients will be included in the overall evaluation of safety (intent-to-treat analysis) if any protocol therapy was administered prior to withdrawal.
- Study is closed for any reason (e.g. new information shows that the patient's welfare would be at risk if he or she continued study treatment).

7.7.2 Withdrawal from study

The reasons for withdrawal from the study include:

- Subject withdraws consent.
- Subject is lost to follow-up.
- Study is terminated for any reason.

8. Study Assessments and Procedures**

Table 2: Screening procedural outline		
Procedure	**Screening Visit[a]**	**Notes**
Eligibility Assessments		
Informed Consent	X	
Inclusion/Exclusion Criteria	X	
Medical History	X	
Safety Assessments		
Physical Examination	X	
Smoking History	X	
Con-medication Review	X	
Vital Signs	X	
Assessment of Signs and Symptoms	X	
Laboratory Tests	X	CBC with differential, serum chemistry (BUN or serum urea level, serum creatinine, albumin, sodium, potassium, chloride, bicarbonate, and glucose levels),AST, ALT, total bilirubin, Alk phosphatase, T3/T4/TSH and urinalysis, hepatitis B and C Antibody.
Pregnancy Test	X	This is only for WOCBP. Serum or Urine pregnancy test is required to be conducted within 2 weeks prior to registration
Physical measurements including ECOG status, Height and Weight	X	
Archived Tumor or Repeat Research Tumor Core Biopsy	X	This is mandatory for study entry. Sample should be received prior to first dose of study treatment. Submit a copy of the original pathology report along with the sample.
Post consent/Pre-treatment bronchoscopy	X (Johns Hopkins only)	Mandatory for patients enrolled at Johns Hopkins only, a bronchoscopy will be performed prior to nivolumab treatment and intraoperatively to enable pre- and post-treatment comparison
Imaging (this is standard of care and may have been performed prior study enrollment)	X	PET/CT Scan *and* CT of chest with IV contrast And MRI brain or CT brain with IV contrast

Table 3: Study Calendar						
Procedure	**Baseline (Day of first neoadjuvant therapy +/- 2 days)**	**Day of 2nd dose of study drugs (+/- 2 days)**	**Day of 3rd dose of study drug (+/-2 days)**	**Once within the 72 hours prior to planned date of surgery (+/- 7 days)**	**Post Surgery (Once during postoperative period day 21-42)**	**Follow up for RFS and OS (H+P and research blood draw every 3-6 months for 2 years and then every 6-12 months until 5 years)**
Clinical Assessments						
Physical Exam	X	X		X	X	X
Vital Signs	X	X		X	X	X
ECOG PS	X	X		X	X	X
Laboratory Tests						
CBC with diff	X	X	X	X	X	
Chemistry including LFTs	X	X	X	X	X	
TSH (TSH + T4/T3 at screening only)	X		X		X	
PT/INR, PTT				X		
Pregnancy Test (WOCBP only)[a]	X		X		X	
Treatment						
Nivolumab	X	X	X			
Ipilimumab (only for subjects enrolled to Arm A)	X					
Carboplatin (only for subjects enrolled to Arm C)	X	X	X			
Paclitaxel (only for subjects enrolled to Arm C)	X	X	X			
Correlative Blood/Tissue/Stool Studies						
PBMCs, Serum, plasma	X	X		X	X	X
Tumor biopsy/sample	X				X (i.e. resection specimen)	

Stool Sample	X		X			X[B]
Initial Sociodemographic Questionnaire	X					
Follow-up Sociodemographic questionnaire *			X			X
Other Assessments						
PET/CT	X (may have been performed during screening or at any time prior to first dose)			X		
CT Chest with IV contrast	X (may have been performed during screening or at any time prior to first dose)			X	X (only required if postoperative radiation is planned)	
RECIST Read	X			X		
Bronchoscopy (Johns Hopkins only)	X (to be performed after consent is signed but before commencement of nivolumab)			X (intraoperative)		
Con meds	X				X	
Symptom/ Toxicity Assessment	X	X		X	X	X
Follow up					X	X

WOCBP, women of child-bearing potential. OS, Overall Survival. RFS, recurrence-free survival
a = Repeat pregnancy test 4-10 weeks post nivolumab for WOCBP,
PBMCs, peripheral blood mononuclear cells
*See Appendix E; Samples are required for JHU participants and optional for MSKCC, SCI, and McGill particpants
B= Every 3 months for the first year as feasible and after the first year every 6 months as feasible

**In order to minimize the need for research-only in-person visits, telemedicine visits may be substituted for inperson clinical trial visits or portions of clinical trial visits where determined to be appropriate and where determined by the investigator not to increase the participants risks. Prior to initiating telemedicine for study visits the study team will explain to the participant, what a telemedicine visit entails and confirm that the study participant is in agreement and able to proceed with this method. Telemedicine acknowledgement will be obtained in accordance with the Guidance for Use of

Telemedicine in Research. In the event telemedicine is not deemed feasible, the study visit will proceed as an in-person visit. Telemedicine visits will be conducted using HIPAA compliant method approved by the Health System and within licensing restrictions.

9. Pharmacology, safety and administration of investigational study drugs

9.1 Availability

Nivolumab and Ipilimumab will be supplied by Bristol-Myer Squibb Pharmaceuticals free of charge to subjects for the duration of participation in this study via the Investigational Drug Service Pharmacies at SKCCC, McGill, SCI, and MSKCC **(Table 3)**. Carboplatin and paclitaxel are standard of care drugs.

9.1.1 Nivolumab – Clinical Pharmacology Summary

Following single dose, maximum concentration (Cmax), and area under the curve (AUC) of nivolumab were found to be dose proportional within the studied dose range of 0.3-10 mg/kg[6]. The terminal half-life of nivolumab from single dose was 17-25 days. Following multiple doses of 0.1 to 10 mg/kg administered every 2 weeks, Cmax and AUC of nivolumab were found dose proportional. The steady-state was reached by the sixth dose.

9.1.2 Safety Summary and Adverse effects of Nivolumab +/- Ipilimumab

General Management Algorithms for potential immune-related toxicities are contained in Appendix D of this protocol.
Please refer to the current version of the Investigators Brochure for detailed information on safety and adverse events.

9.2 Nivolumab and Ipilimumab administration

9.2.1 In arms A and B, three doses of nivolumab will be administered to enrolled patients on Day -42 and Day-28 and Day-14 prior to planned surgery on Day 0 or up to -3 or +10 days. Subjects enrolled in Arm A of this protocol will also receive ipilimumab on day -42. In arm C, nivolumab will be administered on day 1 of each of 3 cycles of neoadjuvant carboplatin and paclitaxel chemotherapy.

When study drugs (ipilimumab or nivolumab) are to be administered on the same day, separate infusion bags and filters must be used for each infusion. It is recommended that nivolumab be administered first. The second infusion will always be ipilimumab, and will start approximately 30 minutes after completion of the nivolumab infusion.

Nivolumab is to be administered as a 30 minute IV infusion. Ipilimumab should be administered as a 30 minute infusion following.

Ipilimumab and nivolumab may be diluted in 0.9% Sodium Chloride Solution or 5% Dextrose solution.

The dosing calculations for Arm A (nivolumab + ipilimumab) should be based on the body weight. If the subject's weight on the day of dosing differs by > 10% from the weight used to calculate the dose, the dose must be recalculated. All doses should be rounded up or to the nearest milligram per institutional standard. Subjects in Arm B will receive nivolumab at a flat dose of 240mg IV. **(Table 3)**.

Table 3 Product Description

Product Description and Dosage Form	Potency	Primary Packaging (Volume)/ Label Type	Secondary Packaging (Qty) /Label Type	Appearance	Storage Conditions (per label)
Nivolumab BMS-936558-01 Solution for Injection[a]	100 mg (10 mg/mL)	10 mL viall	5 vials per carton/ Open-label	Clear to opalescent colorless to pale yellow liquid. May contain particles	2 to 8 C. Protect form light and freezing
Ipilimumab Solution for Injection	200 mg (5 mg/mL)	40 mL vial	4 vials per carton/Open-label	Clear, colorless to pale yellow liquid. May contain particles	2 to 8°C. Protect from light and freezing.
If stored in a glass front refrigerator, vials should be stored in the carton. Recommended safety measures for preparation and handling of nivolumab and ipilimumab include laboratory coats and gloves.					

9.2.1 Investigational product

An investigational product, also known as investigational medicinal product in some regions, is defined as follows: A pharmaceutical form of an active substance or placebo being tested or used as a reference in a clinical study, including products already with a marketing authorization but used or assembled (formulated or packaged) in a way different from the authorized form, or used for an unauthorized indication, or when used to gain further information about the authorized form[87].

The investigational product should be stored in a secure area according to local regulations. It is the responsibility of the investigator to ensure that investigational product is only dispensed to study subjects. The investigational product must be

dispensed only from official study sites by authorized personnel according to local regulations.
In this protocol the investigational products are nivolumab and ipilimumab.

9.2.2 Handling and dispensing of nivolumab and ipilimumab

If stored in a glass front refrigerator, vials should be stored in the carton. Recommended safety measures for preparation and handling of nivolumab and ipilimumab include laboratory coats and gloves.
For additional details on prepared drug storage and use time of nivolumab or ipilimumab under room temperature/light and refrigeration, please refer to the BMS-936558 (nivolumab) and Ipilimumab Investigator Brochure section for "Recommended Storage and Use Conditions". There will be no dose escalations or reductions of study drugs allowed. There are no premedications recommended for nivolumab or ipilimumab on the first dose.

9.2.3 Treatment of Nivolumab or Ipilimumab-Related Infusion Reactions

- Since nivolumab and ipilimumab contain only human immunoglobulin protein sequences, it is unlikely to be immunogenic and induce infusion or hypersensitivity reactions. However, if such a reaction were to occur, it might manifest with fever, chills, rigors, headache, rash, pruritis, arthralgias, hypo- or hypertension, bronchospasm, or other symptoms.
- All Grade 3 or 4 infusion reactions should be reported as an SAE if criteria are met. Infusion reactions should be graded according to NCI CTCAE (Insert version e.g.: 4.0) guidelines.
- Treatment recommendations are provided below and may be modified based on local treatment standards and guidelines as appropriate:
- For Grade 1 symptoms: (Mild reaction; infusion interruption not indicated; intervention not indicated)

  Remain at bedside and monitor subject until recovery from symptoms. The following prophylactic premedications are recommended for future infusions: diphenhydramine 50 mg (or equivalent) and/or paracetamol 325 to 1000 mg (acetaminophen) at least 30 minutes before additional nivolumab administrations.

- For Grade 2 symptoms: (Moderate reaction requires therapy or infusion interruption but responds promptly to symptomatic treatment [eg, antihistamines, non-steroidal anti-inflammatory drugs, narcotics, corticosteroids, bronchodilators, IV fluids]; prophylactic medications indicated for 24 hours).

- Stop the nivolumab or ipilimumab infusion, begin an IV infusion of normal saline, and treat the subject with diphenhydramine 50 mg IV (or equivalent) and/or paracetamol 325 to 1000 mg (acetaminophen); remain at bedside and monitor subject until resolution of symptoms. Corticosteroid or bronchodilator therapy may also be administered as appropriate. If the infusion is interrupted, then restart the infusion at 50% of the original infusion rate when symptoms resolve; if no further complications ensue after 30 minutes, the rate may be increased to 100% of the original infusion rate. Monitor subject closely. If symptoms recur then no further nivolumab or ipilimumab will be administered at that visit. Administer diphenhydramine 50 mg IV, and remain at bedside and monitor the subject until resolution of symptoms. The amount of study drug infused must be recorded on the electronic case report form (eCRF). The following prophylactic premedications are recommended for future infusions: diphenhydramine 50 mg (or equivalent) and/or paracetamol 325 to 1000 mg (acetaminophen) should be administered at least 30 minutes before additional nivolumab or ipilimumab administrations. If necessary, corticosteroids (recommended dose: up to 25 mg of IV hydrocortisone or equivalent) may be used. For Grade 3 or Grade 4 symptoms: (Severe reaction, Grade 3: prolonged [ie, not rapidly responsive to symptomatic medication and/or brief interruption of infusion]; recurrence of symptoms following initial improvement; hospitalization indicated for other clinical sequelae [eg, renal impairment, pulmonary infiltrates]). Grade 4: (life threatening; pressor or ventilatory support indicated).
- Immediately discontinue infusion of nivolumab or ipilimumab. Begin an IV infusion of normal saline, and treat the subject as follows. Recommend bronchodilators, epinephrine 0.2 to 1 mg of a 1:1,000 solution for subcutaneous administration or 0.1 to 0.25 mg of a 1:10,000 solution injected slowly for IV administration, and/or diphenhydramine 50 mg IV with methylprednisolone 100 mg IV (or equivalent), as needed. Subject should be monitored until the investigator is comfortable that the symptoms will not recur. Nivolumab or ipilimumab will be permanently discontinued. Investigators should follow their institutional guidelines for the treatment of anaphylaxis. Remain at bedside and monitor subject until recovery from symptoms. In the case of late-occurring hypersensitivity symptoms (eg, appearance of a localized or generalized pruritis within 1 week after treatment), symptomatic treatment may be given (eg, oral antihistamine, or corticosteroids).

9.3 Carboplatin and paclitaxel administration with nivolumab

Carboplatin will be administered at a dose of AUC 5 or 6 as a 30-minute or as per institutional standard IV infusion, on Day 1 of each 3-week cycle. Carboplatin should be given following

paclitaxel on Day 1 of each cycle, and the carboplatin dose will be calculated using the Calvert formula as follows: Carboplatin dose (mg) = Target AUC x [(CrCl (mL/min) + 25]. Creatinine clearance (CrCl) calculation is based on the Cockroft-Gault formula) and should include the most recent serum creatinine and most recent weight. NOTE: If calculation of the CrCl by the Cockroft-Gault formula yields a result of > 125 mL/min, then a CrCl should be calculated by an alternative formula per institutional standards or capped at 125 mL/min. The dose of carboplatin may be capped per local standards.

Paclitaxel will be administered at a dose of 175 or 200 mg/m2 as a 180 minute or per institutional standard IV infusion and carboplatin will be administered at a dose of AUC 5 or 6 as a 30-minute or per institutional standard IV infusion on Day 1 of a 3-week treatment cycle for up to 3 cycles. Dosing calculations for paclitaxel should be based on the body surface area calculation and may be capped per local standards. The dose may remain the same if the participant's weight is within 10% of the baseline weight or prior dose weight.

Premedications for use with paclitaxel: Oral or IV corticosteroid should be given prior to paclitaxel according to local standard. Such premedication may consist of oral dexamethasone 20 mg 12 hours and 6 hours prior to paclitaxel administration. Oral or IV diphenydramine 50 mg (or its equivalent) and an H2-blocker (per local standard of care) should be administered 30 to 60 minutes prior to paclitaxel infusion. Antiemetic premedication will be administered according to local standards. Recommended antiemetic treatments are dexamethasone (dosing according to local standards; an equivalent dose of another corticosteroid may be substituted) and a 5-HT3 receptor antagonist (type per investigator discretion and local standards of care). Additional use of antiemetic premedications may be employed at the discretion of the investigator per local standards of care.

All participants should be carefully monitored for infusion reactions during the paclitaxel administration. Participants should be treated in a facility with the necessary medical-resuscitation equipment and medications on hand to manage serious acute infusion reactions.

Doses of paclitaxel and/or carboplatin may be modified, delayed, or discontinued depending on how well the participant tolerates the treatment. Dose modifications for toxicity will be performed according to Section 7.6.2.2. Dose delay criteria can be found in Section 7.6.2.3, and discontinuation criteria can be found in Section 7.6.2.4. Criteria to resume treatment can be found in Section 7.6.2.5. If a dose is delayed for any reason, participants should be dosed no later than 7 days following a planned dose. If more than 7 days delay is needed for any reason, the intended dose should be skipped.

9.3.2 Treatment Compliance

Treatment compliance will be monitored by drug accountability as well as the subject's medical record and eCRF.

9.3.3 Destruction of Study Drug

The investigator will ensure that arrangements are made for the disposal of study drug according to applicable regulations, guidelines and institutional procedures, and appropriate records of the disposal have been documented.

10. Exploratory immunologic studies

10.1 Immunologic correlates

All patients will undergo the same laboratory correlate studies on tumor biopsy, resection specimen and serum samples as subsequently enrolled patients.

10.2 Tumor Tissue Samples

10.2.1 Collection of pretreatment tumor and lymph node biopsies

Archived FFPE specimens from the original diagnostic lung tumor biopsy may be utilized. If they do not provide sufficient material for study, then new biopsies will be performed. Ideally 8 (minimum of 6 core) needle biopsies of the primary tumor will be required at the time of diagnosis (prior to first dose of study drug); fine needle aspiration biopsy is sufficient for hilar or mediastinal lymph node sampling, but not for primary tumor biopsy.

Where possible, and after a consent form has been signed, attempts will be made to coordinate diagnostic and study biopsies.

10.2.2 Pretreatment Biopsy Handling, Transportation, Storage, and Processing

Please see flow diagram in appendix C for overview of tissue collection and the laboratory manual for detailed procedures. The study staff will be notified when a biopsy is taking place. The following procedures will be followed:

If a core needle biopsy is being performed specifically for entry to the study, then at least six (and ideally 8) core-biopsy specimens will be obtained, the first 2-3 of these cores obtained will be suspended in 10% buffered formalin while subsequent cores will be flash frozen in liquid nitrogen (see lab manual). After approximately 24 hours of suspension in formalin, the cores will be embedded in paraffin. As required for correlative analyses slides will be cut for the appropriate studies listed below. For fine needle aspiration biopsies of draining lymph nodes, cells will be collected by centrifugation, fixed in formalin and embedded in a paraffin block using standard pathology procedures.

The study coordinator will keep a log with the study number, the patient's study number, the date and time, and a consecutive sample number; thus, the samples will be numbered serially and will not contain identifying information.

10.2.3 Operative specimens (tumor, normal lung, draining lymph nodes)

Tissue specimens obtained at the time of surgery will be dissociated enzymatically into single cell suspensions and will be viably cryopreserved according to a protocol provided in a companion laboratory manual. Additional specimens will fixed in formalin and embedded in paraffin blocks, for routine pathologic studies and immunohistochemistry. Tissue will also be flash-frozen at -80°C for subsequent RNA/DNA analysis. If there is additional tissue available, it will be embedded in OCT (Optimal Cutting Tissue) compound for analysis of frozen sections.

10.3 Bronchial & Nasal Epithelium Samples (Johns Hopkins only)

Bronchoscopy for bronchial and nasal epithelial sampling will occur prior to commencement of study treatment (after signing consent) and intra-operatively. Bronchial and epithelial samples will be obtained utilizing techniques described in sections 7.42 and 7.43. Bronchial samplings will be placed in 3 separate vials containing 1mL each of RNA protect cell reagent, PBS solution for proteomic analysis, and PBS solution for DNA extraction, respectively. Tubes containing RNA protect for cell reagent and PBS for proteomic analysis will be stored at -80◦C. Tube containing PBS solution for DNA extraction will be centrifuged at 1500g for 10min prior to storing at -80◦C. Nasal samplings will be placed in 2 separate vials each containing 2mL of RNAprotect cell reagent and will be stored at -80◦C.

10.4 Blood Samples

10.4.1 Collection schedule

Blood samples will be drawn at the time points identified on the study calendar (section 8, **Table 3**).

Time points include:

- Day of first neoadjuvant treatment (prior to nivolumab +/- ipilimumab or chemotherapy administration): 90 ml whole blood or local protocol for collection accepted for peripheral blood mononuclear cell (PBMC) isolation, 20ml of whole blood for plasma isolation and 10 ml whole blood for serum isolation
- Day of second neoadjuvant treatment (+/- 1 day) (prior to nivolumab +/- nivolumab dose 2 administration): 90 ml, 20ml and 10 ml whole blood, for PBMCs, plasma and serum, respectively.

- Once between Day -3 to D0 (+/- 7 days; must be prior to surgery): 90 ml, 20ml and 10 ml whole blood, for PBMCs, plasma and serum, respectively.
- Once between Week 3-6 postoperatively (postoperative visit, prior to chemotherapy or radiation therapy): 90 ml whole blood or local protocol for collection accepted for PBMC isolation, 20ml whole blood for plasma isolation and 10 ml whole blood for serum preparation
- At every 3-6 month visit during follow up: 90ml, 20ml and 10ml whole blood, for PBMCs, plasma and serum, respectively.

10.4.2 Specimen handling, transportation, storage, processing (Appendix D)

- Serum samples: Whole blood will be collected in serum separator tubes (Becton-Dickinson SST tube or equivalent), processed per manufacturer's instructions and stored at -70°C or below until transfer for analysis.

- PBMCs: Whole blood will be collected into K_2EDTA tubes and processed per manufacturer's instructions. Viable PBMCs will be stored in cryopreservation medium, at 5e6-1e7 per vial, in liquid nitrogen.

- Plasma samples: Whole blood will be collected in K_2EDTA tubes, kept at 4 degrees C and processed as per lab manual.
  Bronchial & Nasal Samples (Johns Hopkins only): as detailed in 10.3, samples will be stored in cryovials containing appropriate reagent and stored at -80◦C until processed as per lab manual.

10.5 Stool Samples

- **Stool must be collected by subjects using a kit that will be mailed or given to participants by study team. Upon collection, subjects will be asked to store specimen in a refrigerator or cool place until it can be brought with them to the clinic. Time points include:**

- **Once between Day -44 and -42 (prior to nivolumab +/- ipilimumab administration)**

- **Once between Day -16 to -14 (prior to nivolumab +/- ipilimumab administration)**
- **Every 3 months for the first year as feasible and after the first year every 6 months as feasible.**
- **The sociodemographic questionnaire must be completed with each stool collection time-point. An initial questionnaire must be completed with the first sample collection and a follow-up questionnaire should be completed for each subsequent time-point. If a patient**

does not provide a sample as scheduled, this will be documented and no questionnaire completed.

10.6 Methods of Analysis

10.6.1 Immunohistochemistry

Tumor and lymph node biopsies will be stained using commercially available and locally developed monoclonal antibodies. Analyses may include phosphorylated proteins of signaling pathways including but not limited to NF-kB, STAT3, RAS, MEK, and ERK; and phenotypes of infiltrating immune cell populations including but not limited to CD3, CD4, FoxP3, CD25, CD8, CD68, CD56, CD20, CD45RO and granzyme B. Peritumoral versus intratumoral infiltrates will be scored, since these staining patterns have been shown to correlate with clinical outcomes[84, 86-90]. Pathologists will assign an intratumoral and peritumoral immune cell infiltrate grade of (0) none, (1) rare lymphocytes (2) focal lymphohistocytic aggregates or (3) severe diffuse infiltration[91]. Pathologists will designate 3 representative fields to be evaluated by image analysis, which will allow for the data to be reported as a percentage of area with positive staining.

Immunohistochemical analysis of exploratory markers will focus on areas where the pathology co-investigators have established expertise, including but not limited to: the B7 family ligands PD-L1 (B7-H1), PD-L2 (B7-DC), B7-H3 and B7-H4, as well as inhibitory receptors on lymphocytes, including PD-1, 2B4, LAG-3, BTLA, and Tim-3; these cell surface molecules are candidates for therapeutic combinatorial antibody blockade. Expression of the ligands for Tim-3, BTLA and 2B4 (galectin 9, HVEM, and CD48, respectively) may also be evaluated as well as cytokine expression. These studies will provide a comprehensive view of cellular subsets and immune checkpoint molecule expression in tumors from untreated patients and how cellular subsets and key immune regulatory molecules are impacted intratumorally after treatment with anti-PD-1. Multiplex immunofluorescence may be performed in selected cases.

PD-L1 expression in FFPE specimens will be assessed with the DAKO 28-8 PharmDx Assary (+/- mAb 5H1 in selected cases)[4].

PD-L1 testing with the BMS assay and JHU assay will be prioritized above other exploratory testing of the tissue specimens.

10.6.2 Amplified In Situ Hybridization (ISH)

ISH will be performed on FFPE sections using the RNAscope method from Advanced Cell Diagnostics. Genes to be probed include specific cytokines, such as IFN-g, IL-17, IL-10, IL-22, TGF-b, IL-4, TNF-a and certain chemokines e.g. CXCR4. These studies will provide information on functional capacity of tumor infiltrating lymphocytes. Additionally, ISH

will be performed for selected molecules, such as LAG-3, that are also being assessed by IHC. This will provide cross-validation for the two techniques.

10.6.3 Laser Capture Microdissection (LCM) and RNA Analysis

Complimentary to the ISH, LCM followed by RNA Analysis may be performed on FFPE sections. LCM will be performed by trained pathologists. RNA analysis may consist of qRT-PCR for selected immune genes, some of which are going to be analyzed in parallel by ISH, and also by whole genome microarray, using the DASL system. These analyses will provide a broader gene expression profile for broadly defined areas of the tumor (ie infiltrating tumor rests vs peri-tumoral vs surrounding stroma) and will complement ISH and IHC analyses. In addition, fresh frozen tumor samples will be used for expression analyses by means of RNA sequencing.

10.6.4 Flow cytometric analysis of tumor and lymph nodes

Cryopreserved viable single cell suspensions will be thawed, and cells will be stained with specific monoclonal antibodies to assess coordinate expression of co-regulatory molecules by tumor infiltrating lymphocytes, draining lymph node cells and tumor cells. Multicolor flow cytometric analyses will be conducted. We will enumerate and characterize T cell subsets (e.g., CD4, CD8, CD25, HLA-DR, CD45RO, FoxP3, LAP, PD-1, PD-L1, PD-L2, LAG-3, ICOS, OX40, 41BB, central memory, effector memory), B cells (e.g., CD19, CD20, PD-1, PD-L2, ICOSL), dendritic cells and macrophages (e.g., CD68, CD83, CD1a, PD-L2, HLA-DR) and natural killer cells (CD56). Functional data and further demonstration of relevant T cell subsets will be gained from intracellular cytokine staining on T cells before and after non-specific CD3/28 activation (e.g., IFN-g, TNF-a, granzyme, IL-4, IL-10, and IL-17). The importance of these specific cytokines is that they mark distinct subsets of T cells with specific roles in pro- vs. anti-cancer immunity. In addition, blood samples will also be analyzed for the similar markers, and for cytokines by multiplex assays.

10.6.5 10.65.5 Bronchial and Nasal Epithelium analysis (Johns Hopkins only)

RNA sequencing including single-cell RNA-seq of bronchial and nasal epithelium will be performed. In addition, DNA and RNA will be extracted and gene-expression arrays will be analyzed to assess for both pre- and post-treatment differences, in addition to unique signatures that correspond with MPR. Pre-treatment gene array signatures will be associated with neoantingen burden and MANA-specific T-cell responses, in addition to other aspects of the tumor immune environment. Other genes and molecular pathways will be assessed via a multitude of a platforms potentially including DNA-

sequencing, proteomics, single-cell sequencing, droplet PCR, and neo-antigen prediction.

10.6.6 PBMC analysis

Assessments of coordinate expression of co-regulatory molecules by PBMCs will be performed using multicolor flow cytometric analyses. T cell subsets (including CD4, CD8, and Treg with CD25 and Foxp3) will be analyzed as well as co-stimulatory and co-inhibitory molecule expression and markers for T cell activation state (e.g., CD25, HLA-DR, CD45RO, LAP, PD-1, PD-L1, LAG-3, ICOS, OX40, 41BB, central memory, effector memory). B cells (CD19, CD20, PD-1, PD-L1, PD-L2, ICOSL), dendritic cells and macrophages (CD68, CD83, CD1a, PD-L1, PD-L2, 4-1BB, 4-1BBL, ICOSL, HLA-DR) and natural killer cells (CD56) will be enumerated and characterized. Myeloid derived suppressor cells (MDSCs) will be enumerated by staining for CD14, CD11b, and HLA-DR expression. Further cytokines produced by T cells, will be analyzed by intracellular cytokine staining and multiplex assay. In certain cases, tetramer staining for populations of antigen-specific T cell populations may be performed.

10.6.7 Pharmacodynamic assessment of nivolumab

Approximate quantitation of infused nivolumab bound to PD-1 receptors on the surface of T cells in the peripheral blood and within the resected tumor and lymph node specimens will be performed in Dr. Topalian's laboratory, according to published procedures[7]. This will provide information about tissue penetration of nivolumab, which has not been obtainable in prior studies.

10.6.8 Molecular pathway analysis

Genes and pathways that are significantly altered in post-therapy tumor tissues as compared to stage-matched untreated tumor tissues or pre-therapy tissues will be assessed by whole-genome analyses and confirmed by methods such as RNAseq or equivalent.

Whole blood collection will be performed for germline subtraction.

Serial circulating tumor DNA analyses will be performed and correlated with findings from analyses of the primary tumor and with response to treatment.

10..6.9 Serum analysis

Serum will be assessed for immunological factors which may include antibodies, cytokines and chemokines, as well as potentially for circulating tumor DNA.. This may include analysis of antibodies to angiopoietin-1/2, MIF, and VEGF-A with ELISAs, as

previously reported[93]. Patients that demonstrate high titer humoral reactions will then undergo detailed evaluation to isolate specific monoclonal antibodies.

10..6.10 Genomic and Mutation-associated neoantigen (MANA) Analyses

Genomic analyses will be performed by whole-exome sequencing in pre and post-treatment tumor tissue and matched normal tissue, to assess dynamics in the mutational landscape using described methods. A multi-dimensional neoantigen prediction algorithm that incorporates MHC binding affinity, epitope processing, self-similarity and gene expression to generate neoantigen candidates tailored to each individual's HLA haplotype will be used. The TCR repertoire will be assessed serially by means of TCR sequencing.

10..6.11 Liquid Biopsy Analysis

ctDNA dynamics will be assessed by targeted sequencing of serial plasma samples collected at the time intervals specified in the study timeline. For these analyses, a custom capture and sequencing approach called targeted error correction sequencing (TEC-Seq) will be used that allows for sensitive and specific detection of low abundance sequence alterations using next generation sequencing.

10.7 Leftover Samples

Any leftover study blood and tissue samples will be stored in the Laboratories of co-investigators for future research studies. These samples may be released for use in future studies after approval by the principal investigator and other regulatory bodies, as appropriate. Subjects will be asked to consent to the future use of samples in the consent document.

10.7.1 Additional Information

The study coordinator will keep a log (separate logs will be kept for the blood and tissue samples) that includes the study number, a specimen serial number, the patient's name, time point in therapy, and the date and time that the sample was drawn. The sample will be labeled with a serial number only. The laboratory technician will keep a log with the specimen number, conditions, processing and storage information.

The laboratory investigators will be blinded to the subject identifiers and clinical data while generating the research data; additionally, the reported results will not disclose any unique patient identifiers.

Note: The correlative sample collection schedules outlined above are based on an ideal subject. The sample schedule should be followed as closely as is realistically possible; however, the schedule may be modified due to problems such as scheduling delays or conflicts (e.g., clinic closure, poor weather conditions, vacations, etc.).

11. Adverse Events

11.1 General

This study will use the descriptions and grading scales found in the revised National Cancer Institute Common Terminology Criteria for Adverse Events (CTCAE) Version 4.0 for adverse event reporting that can be found at http://ctep.cancer.gov/reporting/ctc.html.

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

All adverse events experienced by subjects will be collected from the time of first dose of study medication, throughout the study and until the final assessment as outlined in the Study Calendar **(Section 8)**. Subjects continuing to experience toxicity after discontinuation of the study drug may be contacted for additional assessments until the toxicity has resolved or is deemed irreversible.

Any adverse event experienced during additional preoperative treatment or after the surgical procedure that the investigator feels is related to study treatment will be captured.

11.2 Definitions

11.2.1 Adverse Event (AE)

Defined as any new untoward medical occurrence or worsening of a pre-existing medical condition in a patient or clinical investigation subject administered an investigational (medicinal) product and that does not necessarily have a causal relationship with this treatment. An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding, for example), symptom, or disease temporally associated with the use of investigational product, whether or not considered related to the investigational product.

Adverse events can be spontaneously reported or elicited during open-ended questioning, examination, or evaluation of a subject. In order to prevent reporting bias, subjects should not be questioned regarding the specific occurrence of one or more AEs. Medical conditions/diseases present before starting study treatment are only considered adverse events if they worsen after starting study treatment (any procedures specified in the protocol). Adverse events occurring before starting study treatment but after signing the informed consent form will be recorded. Abnormal laboratory values or test results constitute adverse events only if they induce clinical signs or symptoms or require therapy.

11.2.2 Serious Adverse Event (SAE):

A serious AE (SAE) is any untoward medical occurrence that:

• results in death

• is life-threatening (defined as an event in which the subject was at risk of death at the time of the event; it does not refer to an event which hypothetically might have caused death if it were more severe)

• requires inpatient hospitalization or causes prolongation of existing hospitalization (see NOTE below)

• results in persistent or significant disability/incapacity

• is a congenital anomaly/birth defect

• is an important medical event (defined as a medical event(s) that may not be immediately life-threatening or result in death or hospitalization but, based upon appropriate medical and scientific judgment, may jeopardize the subject or may require intervention [i.e. medical, surgical] to prevent one of the other serious outcomes listed in the definition above.) Examples of such events include, but are not limited to, intensive treatment in an emergency room or at home for allergic bronchospasm; blood dyscrasias or convulsions that do not result in hospitalization.) Potential drug induced liver injury (DILI) is also considered an important medical event. (See **Section 7.3.4** for the definition of potential DLT.)

Suspected transmission of an infectious agent (ie, any organism, virus or infectious particle, pathogenic or non-pathogenic) via the study drug is an SAE.

Although pregnancy, overdose, cancer, and potential drug induced liver injury (DILI) are not always serious by regulatory definition, these events must be handled as SAEs.

The following hospitalizations are not considered SAEs for the purposes of this study:

- a visit to the emergency room or other hospital department lasting < 24 hours, that does not result in admission (unless considered "important medical event" or event life threatening)
- elective surgery, planned prior to signing consent
- admissions as per protocol for a planned medical/surgical procedure
- routine health assessment requiring admission for baseline/trending of health status (ie, routine colonoscopy)
- medical/surgical admission for purpose other than remedying ill health state and was planned prior to entry into the study. Appropriate documentation is required in these cases.
- admission encountered for another life circumstance that carries no bearing on health status and requires no medical/surgical intervention (ie, lack of housing, economic inadequacy, care-giver respite, family circumstances, administrative).

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

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

11.2.5 Relationship

The relationship of all adverse events and serious adverse events to study medication will be assessed by an investigator and assigned as follows:

Definitely: An adverse event which has a timely relationship to the administration of the investigational drug/agent, follows a known pattern of response, for which no alternative cause is present.

Probably: An adverse event, which has a timely relationship to the administration of the investigational drug/agent, follows a known pattern of response, but for which a potential alternative cause may be present.

Possibly: An adverse event, which has a timely relationship to the administration of the investigational drug/agent, follows no known pattern of response, but a potential alternative cause does not exist.

Unlikely: An adverse event which does not have a timely relationship to the administration of the investigational drug/agent, follows no known pattern of response, does not reappear or worsen after re-administration of the investigational drug/agent

(if applicable), and for which there is evidence that it is related to a cause other than the investigational drug/agent.

Unrelated: An adverse event, for which there is evidence that it is definitely related to a cause other than the investigational drug/agent. In general, there is no timely relationship to the administration of the investigational drug/agent, or if there is a timely relationship, the event does not follow a known pattern of response, and there is an alternative cause.

11.3 Serious Adverse Event Collection and Reporting

Following the subject's written consent to participate in the study, all SAEs, whether related or not related to study drug, must be collected, including those thought to be associated with protocol-specified procedures.

Criteria for dose modifications, delays, discontinuation, and resuming treatment are outlined in section 7.6.

General Management Algorithms for potential nivolumab-related toxicities are contained in Appendix D of this protocol and in the Investigators Brochure.

Management algorithms for chemotherapy-related adverse events for carboplatin and paclitaxel will follow standard of care at the treating institution.

All SAEs, must be collected after a study informed consent is signed that occur during the screening period and within 100 days of discontinuation of dosing for those subjects that receive study therapy (within 30 days of last visit for enrollment failure).The investigator should report any SAE occurring after these time periods that is believed to be related to study drug or protocol-specified procedure.

An SAE report should be completed for any event where doubt exists regarding its status of seriousness. If the investigator believes that an SAE is not related to study drug, but is potentially related to the conditions of the study (such as withdrawal of previous therapy, or a complication of a study procedure), the relationship should be specified in the narrative section of the SAE Report Form.

All SAEs, whether related or unrelated to nivolumab and all pregnancies must be reported to BMS (by the investigator or designee) and the Coordinating Center within 24 hours.

The principal investigator will notify the appropriate regulatory agencies of any serious adverse event due to any cause during the course of this investigation. These include the Johns Hopkins Cancer Center Data and Safety Monitoring Committee, and the Johns

Hopkins Medical Institutional Review Board (JHM-IRB) of The Johns Hopkins Medical Institutions. The required reporting time period is 3 days for fatal events, and 10 days for all other events.

For studies conducted under an Investigator IND, any event that is both serious and unexpected must be reported to the Food and Drug Administration (FDA) as soon as possible and **no later than 7 days** (for a death or life-threatening event) or **15 days** (for all other SAEs) **after the investigator's or institution's initial receipt of the information.** BMS will be provided with a simultaneous copy of all adverse events filed with the FDA. SAEs should be reported on MedWatch Form 3500A or similar form. It MUST include the institutional **AND** BMS study ID [per study Agreement]

MedWatch SAE forms should be sent to the FDA at:
MEDWATCH
5600 Fishers Lane
Rockville, MD 20852-9787
Fax: 1-800-FDA-0178 (1-800-332-0178)
http://www.accessdata.fda.gov/scripts/medwatch/

All SAEs should simultaneously be faxed or e-mailed to BMS at:
Global Pharmacovigilance & Epidemiology
Bristol-Myers Squibb Company
Fax Number: 609-818-3804
SAE Email Address: Worldwide.Safety@BMS.com

The study period during which adverse events will be reported is from the initiation of study procedures to the end of the study treatment follow-up, defined as 100 days following the last administration of nivolumab or ipilimumab treatment.
If only limited information is initially available, follow-up reports are required. (Note: Follow-up SAE reports should include the same investigator term(s) initially reported.) If an ongoing SAE changes in its intensity or relationship to study drug or if new information becomes available, a follow-up SAE report should be sent to BMS using the same procedure used for transmitting the initial SAE report.

In accordance with local regulations, BMS will notify investigators of all SAEs that are suspected (related to the investigational product) and unexpected (ie, not previously described in the Investigator Brochure). In the European Union (EU), an event meeting these criteria is termed a Suspected, Unexpected Serious Adverse Reaction (SUSAR).

Investigator notification of these events will be in the form of an expedited safety report (ESR).

SAEs must be recorded on the SAE Report Form. If only limited information is initially available, follow-up reports are required. (Note:
Follow-up SAE reports should include the same investigator term(s) initially reported.)
If an ongoing SAE changes in its intensity or relationship to study drug or if new information becomes available, a follow-up SAE report should be sent within 24 hours.
All SAEs should be followed to resolution or stabilization.

11.4 Non-serious Adverse Events

A non-serious adverse event is an AE not classified as serious.

11.4.1 Non-serious Adverse Event Collection and Reporting

The collection of non-serious AE information should begin at initiation of study drug.
Non-serious AE information should also be collected from the start of a lead-in period or other observational period intended to establish a baseline status for the subjects. All non-serious adverse events (not only those deemed to be treatment-related) should be collected continuously during the treatment period and for a minimum of 90 days following the last dose of study treatment.
Non-serious AEs should be followed to resolution or stabilization, or reported as SAEs if they become serious (see **Section 11.3**). Follow-up is also required for non-serious AEs that cause interruption or discontinuation of study drug, or those that are present at the end of study treatment as appropriate. All identified non-serious AEs must be recorded and described on the non-serious AE page of the CRF (paper or electronic).

11.5 Laboratory Test Abnormalities

Completion of supplemental CRFs may be requested for AEs and/or laboratory abnormalities that are reported/identified during the course of the study.
Serious AE CRF page or SAE Report Form (paper or electronic) as appropriate:

- Any laboratory test result that is clinically significant or meets the definition of an SAE
- Any laboratory abnormality that required the subject to have study drug discontinued
- Any laboratory abnormality that required the subject to receive specific corrective therapy.

It is expected that wherever possible, the clinical, rather than the laboratory term would be used by the reporting investigator (ie, anemia versus low hemoglobin value).

11.6 Pregnancy

If, following initiation of the investigational product, it is subsequently discovered that a study subject is pregnant or may have been pregnant at the time of investigational product exposure, including during at least 6 half-lives after product administration, the investigational product will be permanently discontinued in an appropriate manner. Protocol-required procedures for study discontinuation and follow-up must be performed on the subject unless contraindicated by pregnancy (ie, x-ray studies). Other appropriate pregnancy follow-up procedures should be considered if indicated.
The investigator must immediately notify the Johns Hopkins IRB of this event and complete and forward a Pregnancy Surveillance Form to BMS PVG within 24 hours and in accordance with SAE reporting procedures described in **Section 11.3**.

Follow-up information regarding the course of the pregnancy, including perinatal and neonatal outcome and, where applicable, offspring information must be reported on the Pregnancy Surveillance Form.
Any pregnancy that occurs in a female partner of a male study participant should be reported to the sponsor. Information on this pregnancy will be collected on the Pregnancy Surveillance Form.

11.7 Overdose

An overdose is defined as the accidental or intentional administration of any dose of a product that is considered both excessive and medically important. All occurrences of overdose must be reported as SAEs (see Section 11.3 for reporting details).

11.8 Other Safety Considerations

Any significant worsening noted during interim or final physical examinations, electrocardiograms, x-rays, and any other potential safety assessments, whether or not these procedures are required by the protocol, should also be recorded as a non-serious or serious AE, as appropriate, and reported accordingly.

12. Data and Safety Monitoring

12.1 Data Management

All information will be collected on study-specific case report forms by the study staff.

The completed forms will be forwarded for central review and inclusion in the study dataset with relevant source documentation as outlined in the case report forms. The data submission schedule is as follows:

At the time of registration:

- Registration Form
- Informed Consent Form (signed by the subject)
- Eligibility Checklist
- Source documents related to eligibility and randomization

Within 2 weeks after registration:

- Baseline study case report forms
- Pertinent source documents

Within 2 weeks after final dose of study medication:

- On study case report forms
- Pertinent source documents

The investigator will permit study-related monitoring, audits, and inspections by the IRB, government regulatory bodies, and University compliance and quality assurance groups of all study related documents. The investigator will ensure the capability for inspections of applicable study-related facilities (e.g. pharmacy, diagnostic laboratory, etc.).

The SKCCC Compliance Monitoring Program will provide external monitoring for JHU-affiliated sites in accordance with SKCCC DSMP (Version 6.0, 02/21/2019). The SMC Subcommittee will determine the level of patient safety risk and level/frequency of monitoring. Data monitoring of this protocol will occur on a regular basis. The protocol will be monitored internally (Johns Hopkins East Baltimore and Bayview Medical Center Campuses) at SKCCC by the Principal Investigator and externally by the SKCCC CRO in accordance with SKCCC guidelines.

The study team will follow the SKCCC Coordinating Center Operations Manual to verify the following items:

- Confirmation that the coordinating center PI has contact information for all centers
- Plan for review of each site's IRB approval documents and consent forms
- If federally funded research, confirmation that each participating site has on file an FWA with OHRP
- Method of assuring that all centers have the most current version of the protocol and amendments to the protocol will be communicated to all centers
- Plan for collection and management of data from all centers

- Process for reporting and evaluating protocol events and deviations from all centers

12.2 Meetings

Teleconferences of all investigators, research nurses and other study staff involved in the study will take place, starting once both sites have enrolled a subject. The following study team members involved with the conduct of the trial will be included as appropriate: study coordinators, data managers, research nurses, sub-investigators, collaborators (if applicable), and statistician.

During these meetings, matters related to the following will be discussed: enrollment rate relative to expectation, characteristics of participants, retention of participants, adherence to protocol (potential or real protocol violations), validity and integrity of the data, toxicities, acquisition of serum samples and transfer to lab, and progress of data for objectives.

12.3 Monitoring

Evaluation of safety will be monitored continuously through day 100 following the last dose of nivolumab. The evaluations will be conducted under the direction of Dr. Patrick Forde and the study statistician; additional information may be found in the statistical section.

13. Administrative Procedures

13.1 Protocol Amendments

Any changes to the protocol will be made in the form of an amendment and must be approved by the IRB before implementation. The Principal Investigator is responsible for the coordination and development of all protocol amendments.

13.2 Informed Consent

An investigator will explain to each subject the nature of the study, its purpose, procedures involved, expected duration, potential risks and benefits. Each subject will be informed that participation in the study is voluntary and that she may withdraw from the study at any time, and that withdrawal of consent will not affect her subsequent medical treatment. This informed consent will be given by means of a standard written statement and will be submitted for IRB approval prior to use. No patient will enter the study before her informed consent has been obtained. In accordance with the Health Information Portability and Accountability Act (HIPAA), the written informed consent document (or a separate document to be given in conjunction with the consent document) will include a subject authorization to release medical information to the

study sponsor and supporting agencies and/or allow these bodies, a regulatory authority, or Institutional Review Board access to subjects' medical information that includes all hospital records relevant to the study, including subjects' medical history.

13.3 Ethics and Good Clinical Practice

This study must be carried out in compliance with the protocol and Good Clinical Practice, as described in:

1. ICH Harmonized Tripartite Guidelines for Good Clinical Practice 1996.
2. US 21 Code of Federal Regulations dealing with clinical studies (including parts 50 and 56 concerning informed consent and IRB regulations).
3. Declaration of Helsinki, concerning medical research in humans (Recommendations Guiding Physicians in Biomedical Research Involving Human Subjects, Helsinki 1964, amended Tokyo 1975, Venice 1983, Hong Kong 1989, Somerset West 1996).

The investigator agrees to adhere to the instructions and procedures described in it and thereby to adhere to the principles of Good Clinical Practice that it conforms to.

13.4 Regulatory Authorities

13.4.1 Institutional Review Board

Information regarding study conduct and progress will be reported to the Institutional Review Board (IRB) per the current institutional standards of each participating center**.**

13.4.2 Food and Drug Administration (FDA)

13.5 Principal Investigator Responsibilities

The Protocol Chair is responsible for performing the following tasks:

- Coordinating, developing, submitting, and obtaining approval for the protocol as well as its subsequent amendments.
- Assuring that the correct version of the protocol is used.
- Taking responsibility for the overall conduct of the study and for monitoring the progress of the study.
- Reviewing and ensuring reporting of Serious Adverse Events (SAE).
- Reviewing data from all sites.

14. Statistical Considerations

Addition of two expansion cohorts:

This protocol is investigating the safety and feasibility of neoadjuvant immune checkpoint inhibition in subjects with resectable stage IB, II and IIIA NSCLC. The original arm of the protocol consisted of a six patient run-in phase followed by a 10 patient expansion cohort.

While encouraging pathological responses were seen with the original treatment plan, most patients had remaining tumor after two doses of nivolumab over one month of treatment. It is possible that six weeks of therapy and/or dual immune checkpoint inhibition is necessary to get maximal immune infiltration and tumor regression with neoadjuvant immune checkpoint inhibition. We, therefore, seek to expand the current 20-patient study to include two new treatment arms with a longer duration of treatment: a 15-patient arm treated with a six week course of nivolumab and a 15-patient arm treated with nivolumab and ipilimumab for six weeks. However, due to preliminary data from a phase III study of neoadjuvant immunotherapy in NSCLC in which the nivolumab + ipilimumab arm was discontinued prematurely, we have amended our protocol to close the nivolumab+ipilimumab (Arm A) prior to complete accrual.

Patients will initially be enrolled to Arm A of this protocol, this arm had intended to include 15 patients to receive nivolumab (on day -42, -28 and -14) and ipilimumab (on day -42 only) but as above will be closed early prior to complete accrual. Similar to the first portion of the study, there will be a six patient run-in for Arm A (nivolumab + ipilimumab) only to preliminarily assess safety. . The primary objective of this arm will be safety and feasibility of nivolumab + ipilimumab. Subsequently 15 patients will be enrolled to Arm B (nivolumab single agent on day -42, -28 and -14). A 6 patient safety run-in will not be performed for Arm B. The primary objective of this arm will be safety and feasibility of neoadjuvant nivolumab x 3 doses. Along with the original patient set, these two arms will provide information about the effect of a longer course of therapy on immune correlates, as well as the effect on pathologic regression of a combination anti-PD1 and anti-CTLA4 treatment approach and longer course of single agent anti-PD1.

Experience with nivolumab + ipilimumab (Arm A) as neoadjuvant immunotherapy in NSCLC at the time of the amendment closing this treatment arm (11/9/2018):

The safety run-in of this expansion arm (first six patients) was completed without any safety events, and all patients were able to proceed to surgery as planned. Three additional patients were enrolled. Two of these patients completed treatment without safety events, one proceeded to surgery as planned and one was found to have adrenal metastases following treatment prior to surgery, and did not undergo surgery. The ninth patient completed neoadjuvant therapy and proceeded to surgery, however this patient passed away in the postoperative period at MSKCC and the event was assessed as "possibly related" to pneumonitis from the neoadjuvant treatment and reported to regulatory authorities. Given this event and the discontinuation of the development of ipilimumab and nivolumab in early stage lung cancer, a decision was made to discontinue enrollment to this arm of the study.

Safety and feasibility were evaluable in nine patients. Two patients were not resected due to occult metastasis discovered at the time of surgery. Outcomes for subjects enrolled in Arm A were published in 2020 (Reuss et al. JITC 2020) [96].

Experience with nivolumab monotherapy (Arm B) at the time of the current amendment:

Accrual to Arm B is currently ongoing with no unexpected adverse events.

14.1 Study Design

Patients with operable stage I, II and IIIA non-small cell lung cancer (NSCLC) enrolled in arm A will receive nivolumab 3mg/kg IV for 3 doses (day -42, -28 and -14) with ipilimumab 1mg/kg IV for 1 dose on day -42 prior to surgical resection (between day -3 and day +10) while patients enrolled to arm B will receive single agent nivolumab 240mg IV for 3 doses on the same schedule. Patients in Arm C will receive three doses of nivolumab 360 mg IV, carboplatin AUC 6 IV, and paclitaxel 200 mg/m2 IV on prior to surgical resection. Patients will be observed for perioperative grade 3-4 adverse events through day 100 (or day 30 post surgery – whichever is longer) following the last dose of study drug(s). Safety and feasibility will be monitored continually throughout the study through biweekly meetings with investigators. A set of markers of immune reactivity measured in lung tumor resection specimens, draining lymph nodes, and peripheral blood will be evaluated. Feasibility in this study means the successful completion of preoperative treatment and proceeding to surgery without any extended treatment-related delays. Extended treatment-related delay is defined as >24 days from preplanned Day 0 in this context (>14 days from preplanned day 0 plus 10 extra days to allow for OR scheduling constraints etc.).

The primary endpoint statistical calculations will be based on the 9 treated patients in Arm A (nivolumab + ipilimumab) and subsequently 15 treated patients in Arm B (nivolumab x 3). The stopping rules for safety and feasibility will remain the same for Arm B.

Design and sample size Arm C:

The primary objective for this arm will be pathologic complete response (pCR), defined as no residual tumor cells on histolopathologic examination of resected tumor following neoadjuvant therapy. Sample size is based on an exact single-stage design, Hern (2001). Safety will be continuously monitored during the study.

A review of the literature was done to extract pCR data from clinical trials that could serve as references for this arm of the study. Fourteen trials with at least one relevant treatment arm were selected. The number of pCR responses observed out of the number of patients surgically evaluated from each study, table below, served as input for a random effects meta-analysis of these data. Assuming exchangeable pCR response rates, the predictive distribution based on the historical proportion of responders had mean pCR 8.9%, and median pCR 7.2% (with a 95% credible interval of 1.3% to 27.9%). For ease of use and interpretation, this predictive distribution can be approximated by a Beta density with matching mean and standard deviation, resulting in a Beta(1.9, 16.2) distribution. The mean of 8.9% will be our reference value (null) for pCR in Arm C and the distribution will be used informally as a summary of the historical information when interpreting the results of the study. Our alternative hypothesis is that pCR with the combination neoadjuvant chemo-immunotherapy will be 35% or higher. We will continue treating patients (up to a maximum of 20) until 15 have been treated and surgically evaluated for pCR. With this sample size, the hypothesis that the pCR is less than or equal to 8.9% could be evaluated with a type I error rate of less than 5% and the hypothesis that the pCR is greater than or equal to 35% could be evaluated with 82% power (type II error of 18%). Given the small number of patients in each arm and the importance of obtaining correlative data for all patients, there will no futility monitoring for this arm based on pCR.

Historical references for pCR

Reference	Number surgically evaluated	Number with pCR	% pCR
Pisters[1]	58	9	15.5 %
Pisters[2]	152	15	9.9 %
Roth[3]	20	0	0.0 %
Rosell[4]	27	1	3.7 %
Felip[5]	181	19	10.5 %
Gilligan[6]	230	8	3.5 %
Scagliotti[7]	110	5	4.5 %
Van Meerbeeck[8]	154	8	5.2 %
Nagai[9]	28	0	0.0 %
Betticher[10]	78	11	14.1 %
Jaklitsch[11]	63	0	0.0 %
Martini[12]	114	19	16.7 %
Depierre[13]	167	19	11.4 %
Westeel[14]	257	22	8.6 %

14.2 Objectives

14.2.1 Primary Objective

To investigate the safety and feasibility of neoadjuvant nivolumab +/- ipilimumab in subjects with resectable stage IB, II and IIIA NSCLC.

The primary objective of Arm C is to evaluate pCR, as measured by percent tumor regression, to neoadjuvant nivolumab with chemotherapy.

14.2.2 Secondary Objectives

i. To evaluate pathologic response to neoadjuvant nivolumab +/- ipilimumab in terms of percent tumor regression
ii. To evaluate the frequency of major pathologic response (MPR) to neoadjuvant treatment. MPR is defined as <10% remaining viable tumor in the resection specimen after neoadjuvant treatment
iii. To evaluate radiographic response to neoadjuvant therapy using RECIST 1.1
iv. Safety will be a secondary objective for Arm C

14.3 Primary Endpoint Definition

14.3.1 Primary Endpoints

Safety of nivolumab +/- ipilimumab administered preoperatively according to the planned schema in NSCLC.
Safety will be measured by:
• Frequency of drug related adverse events occurring up to 100 days after the last dose of study treatment or 30 days after surgery (whichever is longer).
• Frequency of serious adverse events occurring up to 100 days after the last dose of study treatment or 30 days after surgery (whichever is longer).
• Frequency of clinical laboratory test by worst toxicity grade using NCI CTC v4.0 (as assessed at the time intervals outlined in the study calendar in section 8, **Table 3**)

Feasibility of preoperative administration of nivolumab +/- ipilimumab in NSCLC.
Feasibility will be evaluated as the successful completion of preoperative treatment and proceeding to surgery without any extended treatment-related delays defined as >24 days from preplanned Day 0 in this context.

For Arm C, the primary endpoint will be pathologic complete response (pCR), defined as 0% residual tumor in the resection specimen on pathologic review. Secondary endpoints include safety adverse events, MPR, and radiographic response with RESIST 1.1.

14.4 Safety Endpoint

Safety stopping rule:

Nivolumab, 3 mg/kg IV, on days -28 and -14 prior to surgery has been tested for safety and feasibility in 19 patients enrolled in the first part of the study, all of these patients are in postoperative follow upAs of October 1, 2016, there were no delays to surgery, meaning this treatment plan was feasible for all enrolled patients. Eighteen of nineteen patients were able to receive both neoadjuvant doses. The one patient not receiving both doses of nivolumab developed fevers on day 7 with tumor cavitation possibly related to treatment (grade 3 SAE) and proceeded directly to surgery without the second dose of nivolumab. Surgery and recovery postoperatively was uncomplicated.

The primary DLTs of concern for safety monitoring in the new expansion cohorts will be grade 3-4 toxicities of the types listed in section 7.3.4. These include liver, GI, renal, pneumonitis and any other grade 3-4 toxicity that in the opinion of the investigator significantly interfered with the subjects' optimal perioperative management. They will be monitored continuously for fifteen patients in each arm through day 100 following the last dose of study treatment (or day 30 post surgery, whichever is longer).

For the first part of the study, we assumed that the risk of grade 3-4 toxicities in advanced NSCLC and other solid tumors is 25% and we used a Beta prior distribution with parameters 1 and 3. With this prior, there is 90% probability that this proportion is between 1.7% and 53.6%. The safety stopping rule for the new expansion cohorts will apply this prior distribution to the observed number of patients experiencing DLT and will compute the resulting probability of DLT. If the posterior probability of risk >.25, based on Bayes rule and the assumption implied by the prior, is 70% or higher the study will stop.

In the first six patients enrolled to Arm A there will be two modifications to the above stopping rule:

1. If the first patient on study experiences a DLT, we will not stop, but treat one additional patient before making a decision.
2. In the first six patients, if there has been one DLT and a second DLT is seen in the fifth or sixth patient, the study will be paused for an additional safety review and may or may not continue.

Table 1. Stopping rule for safety.

Stop if DLTs	2	3	4	5
and N patients	2-4	5-8	9-11	12-15

As an example, the rule will call for stopping the study if 4 patients out of the first 9 experiences grade 3-4 DLT. The following table shows the percent of the time that the stopping rule will stop the study under different hypothetical risks of toxicity, along with the average sample size (based on 5000 simulations). The third row of the table gives the percentage of simulated studies where a second toxicity in the fifth or sixth patient would have caused the study to be paused for a review and possibly stopped.

Table 2. Operating characteristics based on 5,000 simulations with 15 patients and safety stopping rule.

Simulated Risk of DLT	.10	.15	.20	.25	.30	.35	.40	.50
% of Time Study Stops	7.8%	19.4%	32.2%	48.0%	63.0%	75.7%	86.5%	96.3%
Expected Sample Size	14.2	13.2	12.1	10.7	9.3	8.0	6.6	4.8
% with additional safety review.	4.7%	6.7%	8.5%	8.2%	5.7%	4.0%	2.2%	0.7%

Safety Monitoring for Arm C:

The primary DLTs of concern for safety monitoring will be toxicities of the types listed in 7.6.1.2. They will be monitored continuously through day 100 following the last dose of study treatment (or day 30 post surgery, whichever is longer).

The safety stopping rule will pause the if the posterior probability of risk > 0.30, based on Bayes rule and the assumption implied by the prior, is 70% or higher. Based on previous studies of adjuvant therapies involving immunotherapy and chemo-immunotherapy, the frequency of grade 3-4 adverse has been fairly low: Forde (2018) 1/22 (5%) and Provencio (ASCO 2019) 6/46 (13%). There is potential for increased toxicity with the proposed combination however, and therefore the prior for this monitoring rule will be beta (1, 4). This means that our prior guess at the proportion of failures is 20.0%, and there is 90% probability that this proportion is between 1.3% and 52.7%. The stopping rule and operating characteristics are given in the tables below.

Arm C stopping rule for safety.

Stop if AEs	2	3	4	5	6	7
and N participants	2	3-5	6-8	9-11	12-14	15

Operating characteristics based on 5,000 simulations

Simulated Risk of DLT	.10	.15	.20	.25	.30	.35	.40	.50
% of Time Study Stops	2.0%	6.3%	12.6%	21.6%	34.3%	50.5%	62.2%	85.0%
Expected Sample Size	14.8	14.4	13.8	13.0	12.1	10.7	9.6	7.1

14.5 Feasibility Endpoint

14.5.1 Early stopping guideline for feasibility: The feasibility of neoadjuvant nivolumab+/-ipilimumab will be based on patients proceeding to surgery without extended treatment related delays. A treatment related delay will be considered "extended" if it is greater than 24 days following the initially planned surgery date. For feasibility, a toxicity of any grade, that in the judgment of the investigator or surgeon could adversely impact perioperative morbidity or mortality, should delay the planned operative date. We will use a probability-based decision rule for the study to decide if the probability successfully proceeding to surgery as planned is convincingly less than .90.

Previously we expected*, a priori*, the feasibility to be high and that 90% of patients would not have their surgery delayed. Based on results in the first arm of this study, where all 19 patients were feasible and proceeded to surgery without delay, we expect this will be true for the expansion cohorts as well. The monitoring rule for the expansion cohorts will therefore use an *a priori* optimistic Beta(9,1) prior distribution. This distribution corresponds to an assumption that 9 out of 10 patients will proceed to surgery as planned and 90% certainty that feasibility is between .715 and .994. This stopping rule will hold enrollment if, given the data, there is at least 90% probability that fewer than 90% of patients can continue to surgery without treatment related delays. The feasibility stopping rule calls for the study to be paused for a review if the number of patients successfully proceeding to surgery is too low. For example, if neither of the first 2 patients are able to proceed to surgery without a delay, the study should stop. If only 1 of the first 3 patients goes to surgery as planned, the study continues, but the study would be paused for a review if the fourth patient does not. If the feasibility stopping boundary is reached, we will reevaluate the clinical advantages of the treatment, pathological tumor response and prolonged PFS, against the risks and consider re-designing the regimen to allow more time between the last dose of study treatment and surgery to better manage potential

side effects. While an optimistic prior is used to define the stopping boundary, we will use a uniform prior in the analysis phase.

Table 3. Stopping Rule for Feasibility.

No. patients for whom the regimen is feasible	0	1	2	3	4	5	6	7
No. of patients	2	4	5	6	8	10	12	14

14.5.2 Operating characteristics: The operating characteristics of this feasibility rule have been calculated based on 5000 simulations. If the posterior certainty is 90% or higher that feasibility is less than 0.90, based on Bayes rule and the assumption of a Beta(9,1) prior, further study will be reconsidered. For data simulated with known probabilities of feasibility (θ), the second row in the table below shows the percent of time that the feasibility rule will determine that the underlying proportion of patients who can continue to surgery is below the benchmark of 90% and the study should be stopped early or after the 15th patient based on the feasibility rule. The fourth row in the table gives the additional percentage of studies for which the posterior probability of not being feasible at the end of the study is greater than the posterior probability being feasible. These studies would also be considered as having failed feasibility.

Table 4. Operating characteristics of feasibility rule based on 5,000 simulations.

True feasibility (θ)	0.30	0.40	0.50	0.60	0.70	0.75	0.80	0.85	0.90
% studies stopped	100%	99.8%	98.8%	92.4%	74.1%	58.3%	40.4%	22.0%	7.7%
Expected sample size	3.5	4.5	5.6	7.4	9.7	11.2	12.4	13.7	14.5
Additional studies with posterior probability infeasible >50% at final analysis	0.0%	0.2%	1.2%	7.1%	22.2%	33.7%	42.3%	44.7%	37.8%

14.5.3 Feasibility Sample Size and Accrual

The analysis plan for Arm A, which is being discontinued early will be descriptive based on nine patients. The following plan will now apply to Arm B of the study. The accrual rate for this study is expected to be 2-3 patients per month. Arm B will have enrolled fifteen patients if the study does not stop early. We will provide the posterior estimate and a 90% posterior credible interval. The final analysis will use a uniform reference prior for the proportion of patients for whom the neoadjuvant regimen is feasible in this setting. For example, if at least 11 patients were able to proceed directly to surgery after the regimen, then the posterior estimate will be 0.71, and the 90% credible interval will be (0.52, 0.87). Since this credible interval does not include 0.90, we will not consider neoadjuvant immunotherapy feasible in this setting. Using this criterion, at least 12 of 15 patients will have to have been able to go to surgery without problems (i.e., no issues

relating to feasibility) for us to consider the regimen feasible. If the neoadjuvant treatment is feasible for 12 of 15 patients, then the estimate of the chance the regimen is feasible will be 0.76 and the 90% credible interval will be (0.58, 0.91), which includes 0.90.

14.6 Statistical Analysis Plans

14.6.1 Analysis plan for safety

The proportion of DLTs will be reported with exact binomial 95% confidence intervals. All other adverse events will be similarly summarized by type and grade.

14.6.2 Analysis plan for feasibility

The following table shows the 90% credible intervals for the underlying probability of feasibility, based on different numbers of patients going to surgery without extensive delay, using a Beta (1,1) prior. The analysis plan is two-sided (5% in each tail), allowing for the full range of possible outcomes, while sample size is based on a one-sided consideration (10% in the upper tail). As an example of the final inference, if 14 out of 15 patients go to surgery without an extended delay, we will be fairly confident that feasibility is adequate. We will require that 12 out of 15 patients not have extended surgery delays for neoadjuvant immunotherapy to be considered for further study in this setting.

Number feasible out of 15, posterior estimate, and (90% Credible Interval)	**Decision**
7 out of 15 47.1 (27.9, 66.7)	Not Feasible
8 out of 15 53.0 (33.3, 72.1)	Not Feasible
9 out of 15 58.8 (39.1, 77.3)	Not Feasible
10 out of 15 64.7 (45.2, 82.2)	Not Feasible
11 out of 15 70.6 (51.6, 86.8)	Not Feasible
12 out of 15 76.5 (58.3, 91.0)	Feasible
13 out of 15 82.4 (65.6, 94.7)	Feasible
14 out of 15 88.2 (73.6, 97.7)	Feasible

15 out of 15 94.1 (86.6, 1.0)	Feasible

14.6.3 Analysis plan for Arm C

At least 4 patients will have to achieve pCR for the study to reject the null hypothesis that the proportion with pCR is less than or equal to 8.9%. The following table shows 90% confidence intervals based on possible numbers of pCRs out of 15 patients. For comparison, the analysis will also include a Bayesian interpretation. Based on the anticipated pCR for this combination of 35%, an optimistic prior is Beta(1.6, 3.0). This distribution corresponds to an assumption that 35% of patients will obtain pCR and 90% certainty that the pCR probability is between 6.4% and 71.9%. Combining this prior and the possible observed results, the table below also shows 90% credible intervals for the underlying probability of pCR. If nine out of 15 patients obtain pCR, we will be fairly confident that the probability of pCR is 35% or higher with the therapy.

Final inferences based on observing 4 or more pCR in 15 patients.

Observed pCR	90% Confidence Interval	90% Credible Interval
If 4 pCRs out of 15 pts:	(7.9, 45.4)	(13.5, 46.2)
If 5 pCRs out of 15 pts:	(13.3, 53.4)	(17.5, 51.8)
If 6 pCRs out of 15 pts:	(19.2, 60.8)	(21.8,57.1)
If 7 pCRs out of 15 pts:	(25.5, 67.9)	(26.2, 62.2)
If 8 pCRs out of 15 pts:	(32.1, 74.5)	(30.9, 67.2)
If 9 pCRs out of 15 pts:	(39.2, 80.8)	(35.7, 71.9)
If 10 pCRs out of 15 pts:	(46.6, 86.7)	(40.8, 76.5)
If 11 pCRs out of 15 pts:	(54.6, 92.1)	(46.1, 80.8)
If 12 pCRs out of 15 pts:	(63.0, 97.0)	(51.6, 84.9)
If 13 pCRs out of 15 pts:	(72.2, 100.0)	(57.3, 88.8)
If 14 pCRs out of 15 pts:	(82.7, 100.0)	(63.4, 92.3)
If 15 pCRs out of 15 pts:	NA	(73.8, 100.0)

14.7 Exploratory Immune Endpoints

14.7.1. Markers of response to anti-PD-1

Markers of immune reactivity will be prospectively measured in lung tumor resection specimens, draining lymph nodes (DLNs) and serial peripheral blood samples from stage IB/II/IIIA NSCLC patients who receive preoperative immune checkpoint inhibition. All samples will be analyzed using whole exome sequencing, RNA sequencing, TCR sequencing, multicolor flow cytometry, immunohistochemistry and in situ hybridization for candidate surrogate markers of immune response to anti-PD-1, depending on tissue availability. These analyses include assessment of frequency of coordinate expression of co-regulatory molecules **(Table 5)** by peripheral blood lymphocytes, tumor infiltrating lymphocytes, DLN cells and tumor cells. PD-L1, CD3, CD4, CD8, granzyme B, CD20, and CD56 staining will be performed on biopsy and resection specimen and frequency of expression tabulated. Analysis of circulating cell-free tumor DNA will be performed at serial timepoints. Additional molecules may be assessed as well.

Table 5 – Selected co-regulatory molecules to be tested

T Cell Subset	Principle Role
CD4, CD8	Interaction of TCR with APC
CD25	Forms receptor for IL-2, may suppress anti-tumor immunity
HLA-DR	Presentation of antigens for immune recognition
CD45RO	Expression may potentiate anti-tumor immunity
FoxP3	Essential for production of Tregs, may suppress anti-tumor immunity
LAP	Potential marker for activated Tregs
PD-1/PD-L1	Inhibit anti-tumor immunity, PD-1 is primary target of nivolumab
LAG-3	Inhibitory co-receptor may synergize with PD-1 to suppress anti-tumor immunity
ICOS	Stimulates T cell response to tumor
CTLA-4	Immune checkpoint inhibits anti-tumor immune response
OX40	Co-stimulatory, contributes to memory T cell response
4-1BB	Anti-4-1BB antibody may potentiate anti-tumor response

Abbreviations – CD, cluster differentiation; TCR, T-cell receptor; APC, antigen presenting cell; HLA-DR, human leukocyte antigen-D related; FoxP3, forkhead box P3; LAP, latency associated peptide; LAG-3, lymphocyte –activation gene 3; ICOS, Inducible T-cell costimulator; CTLA-4, cytotoxic T-lymphocyte antigen-4; PD-L1, Programmed death-ligand 1;

14.7.2 Analysis of exploratory immunologic endpoints

These exploratory analyses will be descriptive/graphical in nature, and are designed to generate new hypotheses to be tested in future clinical studies. When parameters of immune response are measured, continuous variables will be summarized with means and standard deviations. Dichotomous and categorical variables will be summarized using proportions with exact 95% confidence intervals and counts, respectively. These summaries will be computed for each treated patient at multiple time points before and

after nivolumab administration as indicated in the study schema. Plots will be used to show the changes in immune response over time both for each individual. For each patient, comparisons in the pre and post-nivolumab responses will be compared using paired t-tests, or Wilcoxon signed rank tests if data distribution is not normal, for continuous variables and McNemar's test for dichotomous or categorical variables. Associations between immune responses will be explored graphically (e.g. scatterplots, boxplots) and numerically (e.g. correlations, $\chi 2$ tests). Boxplot is for visualization purposes. The correlations of immune markers will be quantified by correlations coefficient. Similar comparisons will be performed between immunotherapy-treated patients, and control patients with resectable NSCLC who do not receive nivolumab or nivolumab and ipilimumab but are enrolled on a companion tissue collection protocol.

14.7.3 **Analysis of exploratory features of gut microbiota that correlate with clinical response.**

Integration of microbiome science into cancer therapeutics is a new field of study. There are as yet no prospective human studies to determine if the microbiome influences the response to cancer therapies. However, two recent mouse papers published in Science (insert PMID: 26541606 and PMID: 26541610) have suggested that cancer immunotherapy is impacted by the composition of the microbiome and that certain bacterial species can promote improved therapeutic responses to checkpoint inhibitor therapy. Thus, the goal of this sample collection is to facilitate exploratory, correlative analyses between the gut microbiome communities and responses to neoadjuvant checkpoint blockade therapy.

We will consider using 16S rRNA, shotgun metagenomics and/or RNA-seq for analyses. 16S rRNA sequencing data will be filtered for poor quality and contaminant/chimeric sequences, followed by taxonomic assignment using standard bioinformatic pipelines such as QIIME and Resphera Insight. To detect differentially abundant taxa between responders and non-responders, we will utilize the nonparametric Mann-Whitney test with correction for multiple hypothesis testing using the False Discovery Rate. To evaluate beta-diversity between responders and non-responders (i.e. shared total community composition), we will compute the UniFrac distance metric for all sample pairs, followed by principal coordinate analysis and significance testing with PERMANOVA. Functional inference of gene content from 16S rRNA data may also be performed using tools such as PICRUSt. While 16S rRNA identifies only bacterial sequences, metagenomics permits detection of viruses and fungi among others. Metagenomic analyses will be designed to remove human contaminant sequences followed by taxonomic assignment using Kraken and

Pathoscope. Functional characterization of metagenomic data will be performed using the HUMAnN tool. Additionally, RNA-seq analysis (meta-transcriptomics) enables characterization of actively transcribed microbial genes and RNA viruses, with the potential to explore the combined host:microbial interaction(s).

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Appendix A: TNM staging system for lung cancer (7th edition)

TNM staging system for lung cancer (7th edition)

Primary tumor (T)			
T1	Tumor ≤3 cm diameter, surrounded by lung or visceral pleura, without invasion more proximal than lobar bronchus		
T1a	Tumor ≤2 cm in diameter		
T1b	Tumor >2 cm but ≤3 cm in diameter		
T2	Tumor >3 cm but ≤7 cm, or tumor with any of the following features:		
	Involves main bronchus, ≥2 cm distal to carina		
	Invades visceral pleura		
	Associated with atelectasis or obstructive pneumonitis that extends to the hilar region but does not involve the entire lung		
T2a	Tumor >3 cm but ≤5 cm		
T2b	Tumor >5 cm but ≤7 cm		
T3	Tumor >7 cm or any of the following:		
	Directly invades any of the following: chest wall, diaphragm, phrenic nerve, mediastinal pleura, parietal pericardium, main bronchus <2 cm from carina (without involvement of carina)		
	Atelectasis or obstructive pneumonitis of the entire lung		
	Separate tumor nodules in the same lobe		
T4	Tumor of any size that invades the mediastinum, heart, great vessels, trachea, recurrent laryngeal nerve, esophagus, vertebral body, carina, or with separate tumor nodules in a different ipsilateral lobe		
Regional lymph nodes (N)			
N0	No regional lymph node metastases		
N1	Metastasis in ipsilateral peribronchial and/or ipsilateral hilar lymph nodes and intrapulmonary nodes, including involvement by direct extension		
N2	Metastasis in ipsilateral mediastinal and/or subcarinal lymph node(s)		
N3	Metastasis in contralateral mediastinal, contralateral hilar, ipsilateral or contralateral scalene, or supraclavicular lymph node(s)		
Distant metastasis (M)			
M0	No distant metastasis		
M1	Distant metastasis		
M1a	Separate tumor nodule(s) in a contralateral lobe; tumor with pleural nodules or malignant pleural or pericardial effusion		
M1b	Distant metastasis (in extrathoracic organs)		
Stage groupings			
Stage IA	T1a-T1b	N0	M0
Stage IB	T2a	N0	M0
Stage IIA	T1a,T1b,T2a	N1	M0
	T2b	N0	M0
Stage IIB	T2b	N1	M0
	T3	N0	M0
Stage IIIA	T1a,T1b,T2a,T2b	N2	M0
	T3	N1,N2	M0
	T4	N0,N1	M0
Stage IIIB	T4	N2	M0
	Any T	N3	M0
Stage IV	Any T	Any N	M1a or M1b

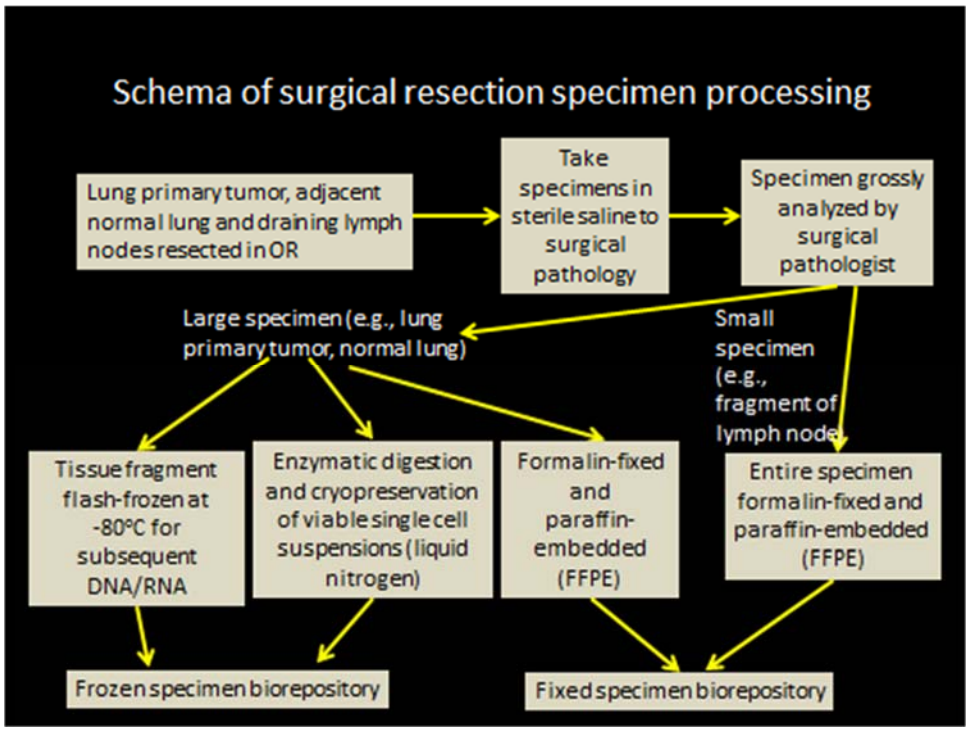
Adapted from: Goldstraw P, Crowley J, Chansky K, et al. The IASLC Lung Cancer Staging Project: Proposals for the revision of the TNM stage groups in the forthcoming (seventh) edition of the TNM classification of malignant tumours. J Thorac Oncol 2007; 2:706.

UpToDate

Appendix B: ECOG Performance Status Scale

Score	Definition
0	Asymptomatic
1	Symptomatic, fully ambulatory
2	Symptomatic, in bed less than 50% of day
3	Symptomatic, in bed more than 50% of day, but not bedridden
4	Bedridden

Appendix C: Guidelines for Tissue Banking Process



Note: Only tissue that is absolutely not needed for clinical diagnosis or staging should be collected for tissue banking**. If in doubt about this, do NOT submit specimens for banking.**

Banking of Frozen Tissue

1. Place a single tissue specimen flat in the plastic bag. A single tissue specimen's overall volume should be at least 1 cm3, and at most 3 – 4 cm3, with at least one dimension measuring 0.5 cm thick or less to facilitate quick freezing.

2. For a given case (patient), please collect sufficient **non-malignant and malignant** tissue. Tissue selected should be grossly viable, and grossly consistent with tumor or adjacent normal tissue (see #5 below for contraindications). Non-malignant (i.e. "adjacent normal") tissue should be collected at least **2 cm** from the primary tumor, subject to any limitations from the specimen's physical dimensions. Do not place tumor and non-malignant tissue in the same bag. For large tumors, do not place large pieces of tissue in a single bag. Rather, divide the tissue according to size guidelines in #1 above, and place each in an individual bag. Collect and separately identify both: 1) primary tumor and 2) metastatic lesions to lymph nodes or other tissues. Tissue will typically be taken by scalpel or dissection blade, though the use of 5 -7 mm skin punch biopsy tools could be considered in certain situations.

3. Immediately place the specimens for freezing in an isopentane or 2-methylbutane cryobath, or other effective liquid freezing agent. If no cryobath is available, then liquid N2 can be used as the freezing agent, in a properly insulated container and with sufficient safety precautions. The goal is to have

bankable tissue immersed in the bath **within 30 minutes of the OR's procurement** from the patient. If more cryobath space is needed, move **already frozen** tissue to a -80C freezer in order to make sufficient room. Make sure to check periodically for cryobath problems (e.g. not maintaining temperature, refrigerant level low), and call for appropriate maintenance as needed. **Do not freeze tissue by placing it fresh directly in the -80C freezer.**

4. On receipt by the tissue bank laboratory, the frozen tissue is embedded in OCT (Optimal Cutting Temperature medium), and a frozen section is stained with H&E and the section evaluated by the tissue bank pathologist for quality assurance (QA) purposes. A report on the histopathologic findings is filed or communicated as needed. The frozen section evaluation can also count for adjacent pieces of tissue if they were taken as a "mirror image" section to the surface cut for the frozen section.

5. General contraindications to tissue banking

DON'T bank tissue from these specimen types or situations:

- small tumors and other cases where all or most of the lesional tissue is needed for diagnosis
- surgical margins of resection specimens where tumor and benign areas cannot be clearly delineated grossly grossly visible areas of primarily necrosis, hemorrhage, or fat
- specimens which are known to have been delayed significantly more than 30 minutes past their procurement time in the OR
- tissue previously freeze-thawed, or frozen slowly (e.g. in the cryostat or -80 freezer)
- areas of deepest invasion, tumor/normal interface, tumor/capsule interface, extranodal extension of tumor, and other key landmarks needed for surgical pathology evaluation and/or tumor staging
- chemotherapy- or radiation-treated tumors
- diagnostic biopsies where most or all tissue must be submitted for pathology evaluation…most lymph node, GI, bone marrow, and liver biopsies fall in this category
- tissue clearly marked as intended for a special study such as immunofluorescence

Appendix D: MANAGEMENT ALGORITHMS

These general guidelines constitute guidance to the Investigator and may be supplemented by discussions with the Principal Investigator. The guidance applies to all immuno-oncology agents and regimens.

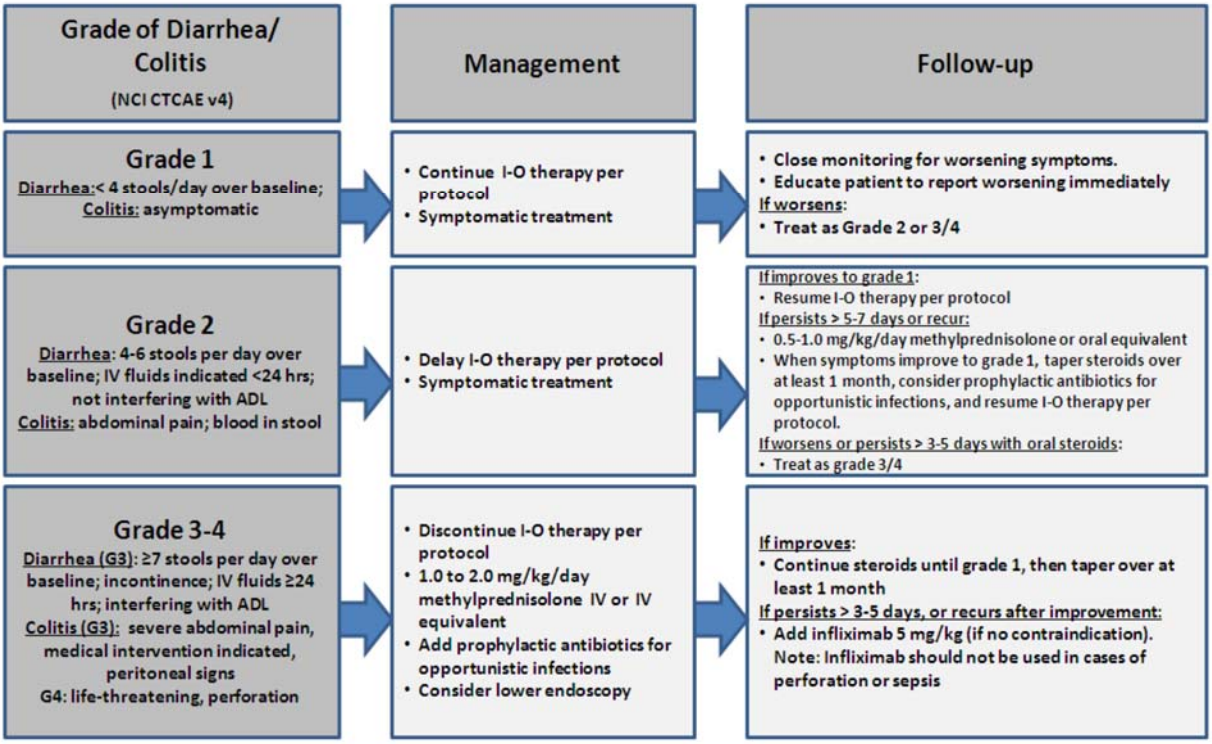
A general principle is that differential diagnoses should be diligently evaluated according to standard medical practice. Non-inflammatory etiologies should be considered and appropriately treated.

Corticosteroids are a primary therapy for immuno-oncology drug-related adverse events. The oral equivalent of the recommended IV doses may be considered for ambulatory patients with low-grade toxicity. The lower bioavailability of oral corticosteroids should be taken into account when switching to the equivalent dose of oral corticosteroids.

Consultation with a medical or surgical specialist, especially prior to an invasive diagnostic or therapeutic procedure, is recommended.
The frequency and severity of the related adverse events covered by these algorithms will depend on the immuno-oncology agent or regimen being used.

GI Adverse Event Management Algorithm

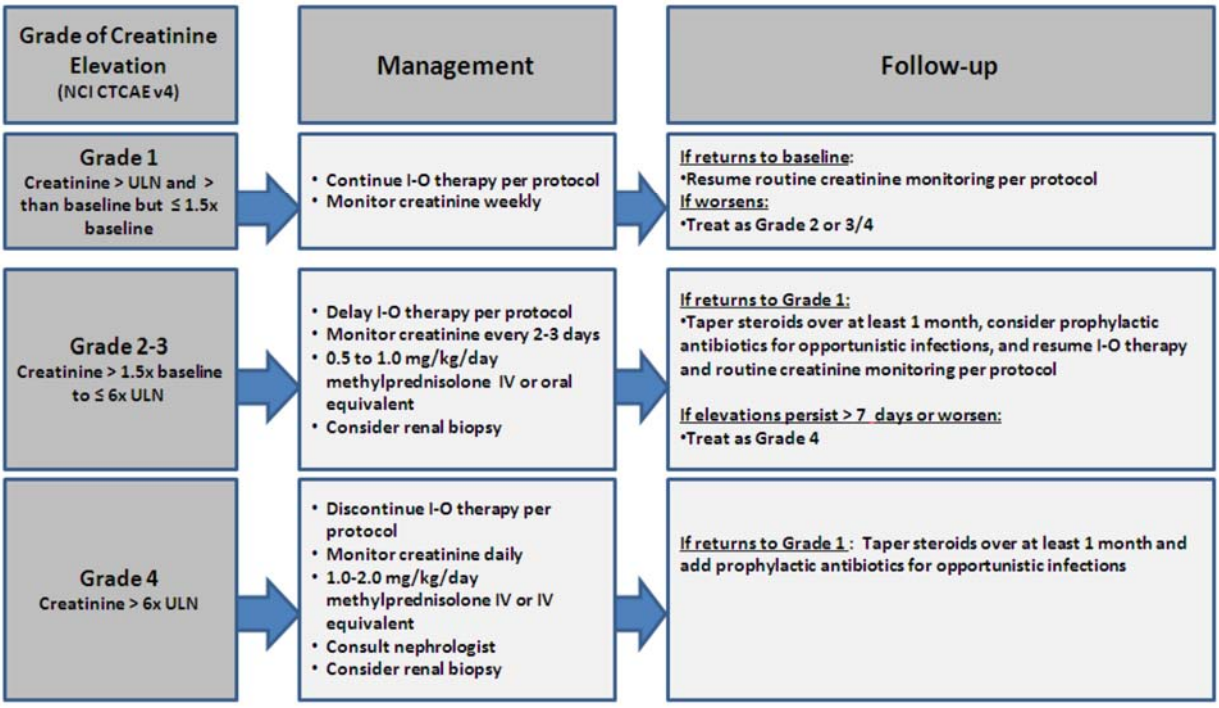
Rule out non-inflammatory causes. If non-inflammatory cause is identified, treat accordingly and continue I-O therapy. Opiates/narcotics may mask symptoms of perforation. Infliximab should not be used in cases of perforation or sepsis.



Patients on IV steroids may be switched to an equivalent dose of oral corticosteroids (e.g. prednisone) at start of tapering or earlier, once sustained clinical improvement is observed. Lower bioavailability of oral corticosteroids should be taken into account when switching to the equivalent dose of oral corticosteroids.

Renal Adverse Event Management Algorithm

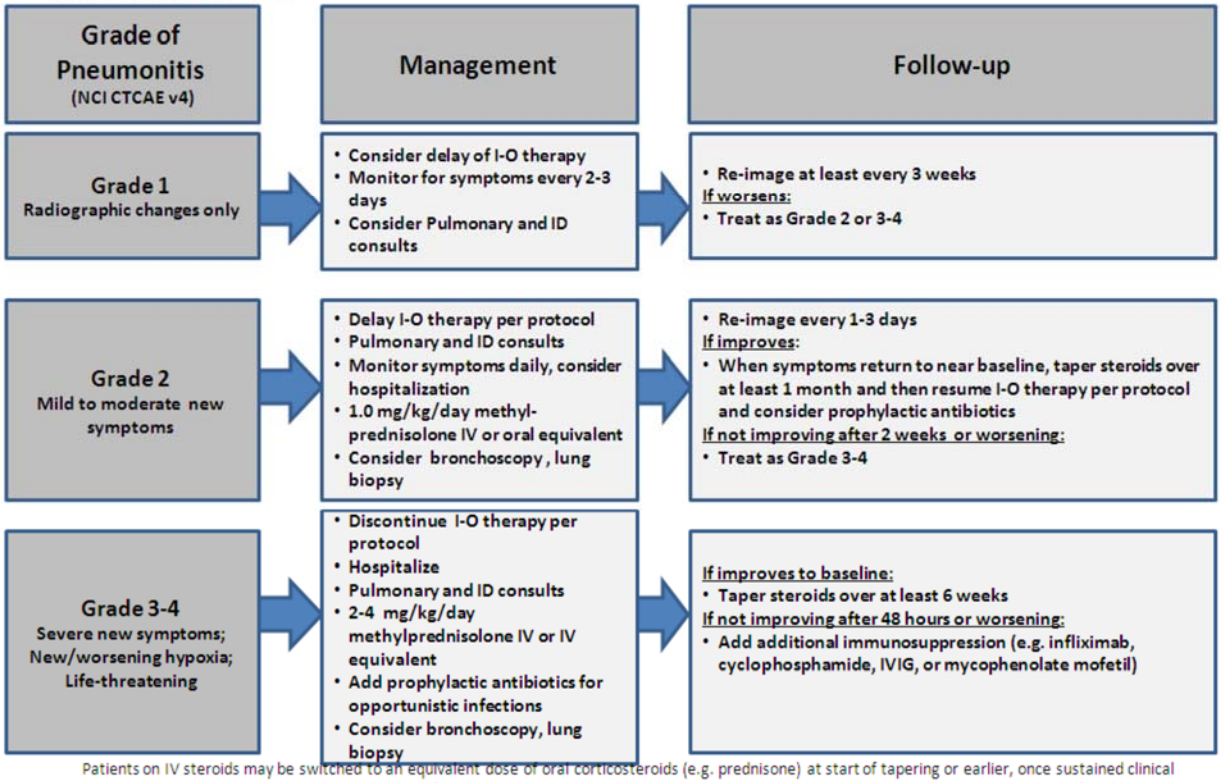
Rule out non-inflammatory causes. If non-inflammatory cause, treat accordingly and continue I-O therapy



Patients on IV steroids may be switched to an equivalent dose of oral corticosteroids (e.g. prednisone) at start of tapering or earlier, once sustained clinical improvement is observed. Lower bioavailability of oral corticosteroids should be taken into account when switching to the equivalent dose of oral corticosteroids.

Pulmonary Adverse Event Management Algorithm

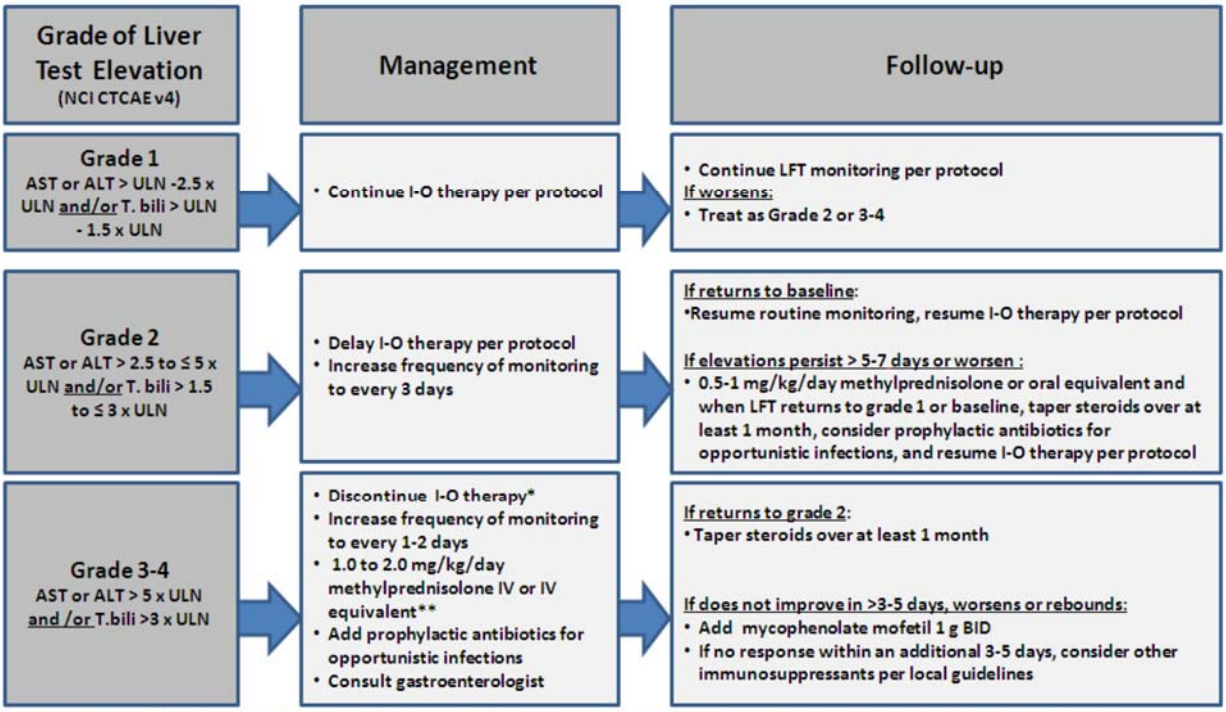
Rule out non-inflammatory causes. If non-inflammatory cause, treat accordingly and continue I-O therapy. Evaluate with imaging and pulmonary consultation.



Patients on IV steroids may be switched to an equivalent dose of oral corticosteroids (e.g. prednisone) at start of tapering or earlier, once sustained clinical improvement is observed. Lower bioavailability of oral corticosteroids should be taken into account when switching to the equivalent dose of oral corticosteroids.

Hepatic Adverse Event Management Algorithm

Rule out non-inflammatory causes. If non-inflammatory cause, treat accordingly and continue I-O therapy. Consider imaging for obstruction.



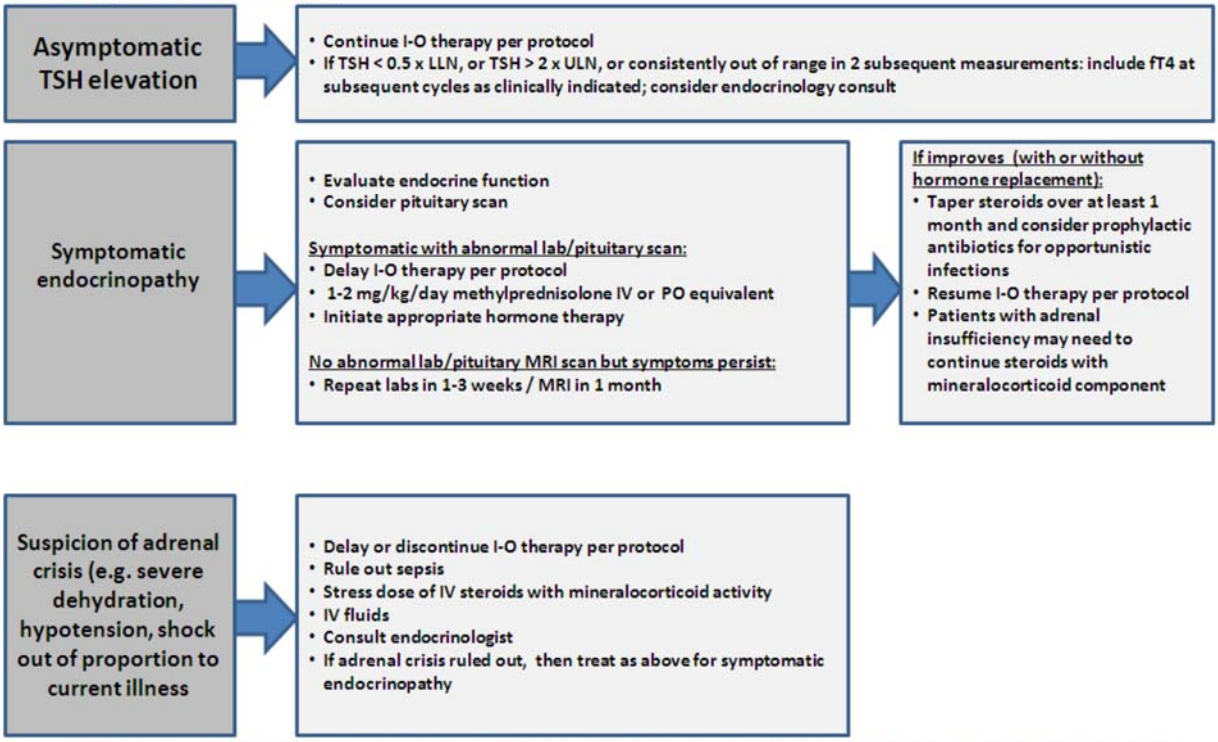
Patients on IV steroids may be switched to an equivalent dose of oral corticosteroids (e.g. prednisone) at start of tapering or earlier, once sustained clinical improvement is observed. Lower bioavailability of oral corticosteroids should be taken into account when switching to the equivalent dose of oral corticosteroids.

*I-O therapy may be delayed rather than discontinued if AST/ALT ≤ 8 x ULN and T.bili ≤ 5 x ULN.

**The recommended starting dose for grade 4 hepatitis is 2 mg/kg/day methylprednisolone IV.

Endocrinopathy Management Algorithm

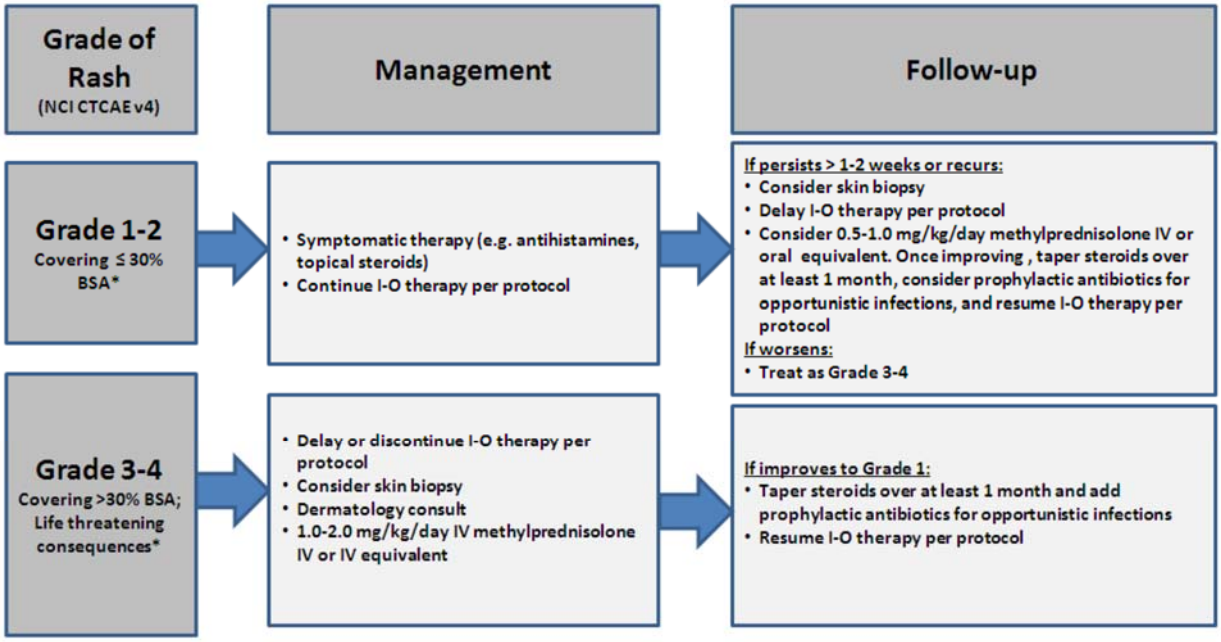
Rule out non-inflammatory causes. If non-inflammatory cause, treat accordingly and continue I-O therapy. Consider visual field testing, endocrinology consultation, and imaging.



Patients on IV steroids may be switched to an equivalent dose of oral corticosteroids (e.g. prednisone) at start of tapering or earlier, once sustained clinical improvement is observed. Lower bioavailability of oral corticosteroids should be taken into account when switching to the equivalent dose of oral corticosteroids.

Skin Adverse Event Management Algorithm

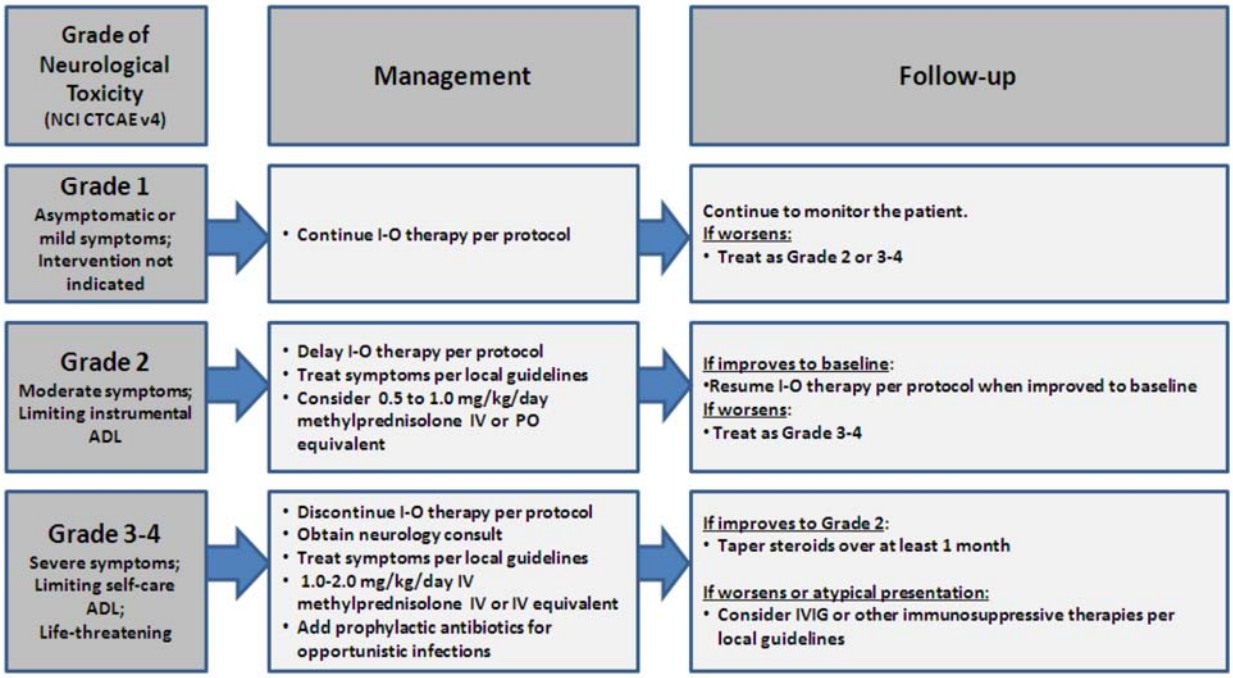
Rule out non-inflammatory causes. If non-inflammatory cause, treat accordingly and continue I-O therapy.



Patients on IV steroids may be switched to an equivalent dose of oral corticosteroids (e.g. prednisone) at start of tapering or earlier, once sustained clinical improvement is observed. Lower bioavailability of oral corticosteroids should be taken into account when switching to the equivalent dose of oral corticosteroids.
*Refer to NCI CTCAE v4 for term-specific grading criteria.

Neurological Adverse Event Management Algorithm

Rule out non-inflammatory causes. If non-inflammatory cause, treat accordingly and continue I-O therapy.



Patients on IV steroids may be switched to an equivalent dose of oral corticosteroids (e.g. prednisone) at start of tapering or earlier, once sustained clinical improvement is observed. Lower bioavailability of oral corticosteroids should be taken into account when switching to the equivalent dose of oral corticosteroids.

APPENDIX E: Sociodemographic Charactiersitics Questionnaire

Date: Month_____ / Day _____ / Year_____
Study ID Number: _____

INITIAL SOCIODEMOGRAPHIC CHARACTERISTICS QUESTIONNAIRE

FAMILY QUESTIONS

Both genetics and environment could be risk factors for the development of cancer. For this reason, it is important to determine your biological relationship with your family.

5. Are you adopted? □ Yes □ No
6. How many of each of the following family members do you have?
Brothers: _____Sisters: _____Sons: _____ Daughters: _____

SMOKING QUESTIONS

7. Do you smoke cigarettes? ☐ **Yes** ☐ **No, never** ☐ **Not currently but I have in the past**

8. At what age did you start smoking? _____

9. When did you quit smoking cigarettes? _____

10. How many total years have you or did you regulary smoke cigarettes? _____ _____

11. During the time you usually smoked regularly, how many cigarettes do or did you usually smoke per day? _____

12. Do you smoke cigars? □ Yes □ No, never □ Not currently but I have in the Past

13. At what age did you start smoking cigars? _____

14. When did you quit smoking cigars? _____

15. How many years in total did you regularly smoke cigars? _____

16. Do you use smokeless tobacco or other nicotine products? (i.e. chewing tobacco, snuff, e-cigarette, nicotine patch or gum) ☐ **Yes** ☐ **No**

If yes, please indicate type(s):

_____

17. Were you exposed to asbestos, that you know of? □ Yes □ No, never

18. Were you exposed to any other potential harmful exposures to your lung? □ Yes □ No, never

ALCOHOL QUESTIONS

19. Have you ever drunk alcoholic beverages, such as beer, wine, or liquor regularly, that is at least once a month? ☐ Yes ☐ No

20. At what age did you start drinking alcoholic beverages regularly, i.e. at least once a month?

_____years of age

21. Before the age of 40, how many drinks of beer (12 oz.), wine (5oz.), or liquor (1 oz.) did you usually drink per week?

More than one per week. Please indicate number_____
Less than one per week_____
Never drank before age 40 _____

22. After the age of 40, how many drinks of beer (12 oz.), wine (5 oz.), or liquor (1 oz.) did you usually drink per week?

More than one per week. Please indicate number _____
Less than one per week _____
Never drank after age 40 _____
Currently aged less than 40 years _____

MEDICAL QUESTIONS

23. Have you taken antibiotics in the last 3 months? ☐ Yes ☐ No
A list of antibiotics is attached for you to refer to. If no, skip to question 25.

24. If you know the name of the antibiotic(s), please write it here.

_____
_____

25. Have you had a bronchoscopy in the last 3 months? ☐ Yes ☐ No

26. Have you taken oral corticosteroids in the last 2 weeks? ☐ Yes ☐ No
(Oral corticosteroids examples: prednisone, dexamethasone, methlyprednisone, hydrocortisone)

27. If you answered Yes to (26), please write the name and dose here and when you took these.

_____

_____

28. Have you taken inhaled corticosteroids in the last 2 weeks? ☐ Yes ☐ No
(Inhaled corticosteroids examples: budesonide, fluticasone, beclomethasone, ciclesonide)

29. If you answered Yes to (28), please write the name and dose here and when you took these.

_____

_____

30.Do you have sleep apnea? ☐ Yes ☐ No

31.Do you have reflux disease? ☐ Yes ☐ No

32.Do you have any other chronic lung conditions? ☐ Yes ☐ No

33. If you answered Yes to (32), please write the name of the condition here

_____

_____

DENTAL HEALTH

34. About how often do you visit a dentist?

- ☐ Less than every 6 months.
- ☐ Between every 6-12 months.
- ☐ Greater than every 12 months.
- ☐ I have never been to a dentist.

35. Overall, how would you rate the health of your teeth and gums?

☐ Excellent ☐ Good ☐ Fair ☐ Poor

36. Have you ever had treatment for gum disease such as scaling and root planing, sometimes called deep cleaning?

☐ Yes ☐ No

37. Have you ever had any teeth become loose on their own, without an injury? (Not including baby teeth). ☐ Yes ☐ No

DIET QUESTIONS

38. Do you eat meat? Meat is defined as beef, chicken, pork, lamb or venison:

- **A. I do not eat meat.** ☐
- **B. I have eaten meat in the last year.** ☐
- **C. I have eaten meat one or more times a month during the last year.** ☐
- **D. I have eaten meat one or more times a week during the last year.** ☐

39. Do you eat Fish? Fish is defined as all fish and shellfish:

- **A. I do not eat fish.** ☐
- **B. I have eaten fish in the last year.** ☐
- **C. I have eaten fish one or more times a month during the last year.** ☐
- **D. I have eaten fish one or more times a week during the last year** ☐

40. Do you eat Eggs?

- **A. I do not eat eggs** ☐
- **B. I have eaten eggs in the last year** ☐
- **C. I have eaten eggs one or more times a month during the last year** ☐
- **D. I have eaten eggs one or more times a week during the last year** ☐

41. Do you eat Cheese? (This includes fresh, soft, aged or cottage cheese as well as sour cream):

- **A. I do not eat cheese** ☐
- **B. I have eaten cheese in the last year** ☐
- **C. I have eaten cheese one or more times a month during the last year** ☐
- **D. I have eaten cheese one or more times a week during the last year** ☐

42. Do you drink Milk? Milk is defined as milk from a cow, goat or sheep (not soy, coconut or almond milk, for example). If you put milk on your cereal, you should not answer (A).

- **A. I do not drink milk** ☐
- **B. I drank milk in the last year** ☐
- **C. I drank milk one or more times a month during the last year** ☐
- **D. I drank milk one or more times a week during the last year** ☐

43. Do you eat yogurt?

- **A. I do not eat yogurt** ☐
- **B. I have eaten yogurt in the last year** ☐
- **C. I have eaten yogurt one or more times a month during the last year** ☐
- **D. I have eaten yogurt one or more times a week during the last year** ☐

44. Do you take probiotics (live bacteria supplement)? ☐ Yes ☐ No

If yes (question 44) and if you know the name of the probiotic product(s), please write here:

_____

_____

45. Do you take vitamin supplements? ☐ **Yes** ☐ **No**

If yes (question 45) and if you know the name of the vitamin supplement(s), please write it/them here:

_____

_____

WORK AND PHYSICAL ACTIVITY

46. What is your current employment status?

☐ Employed/self-employed ☐ Unemployed ☐ Retired ☐ Disabled

47. How would you categorize your physical activity on the job?

☐ Mostly sedentary or light activity (e.g. mostly sitting, standing, lifting light objects of less than 3
kilos).

☐ Mostly medium activity (e.g. much walking, climbing stairs).

☐ Mostly intense activity (e.g. heavy construction work).

☐ Unemployed/retired/disabled

48. What type of exercise (physical activity) do you do regularly (at least 3 times per week)?

☐ Mostly moderate activity (slow walking, gardening, golfing etc.)

☐ Mostly vigorous activity (running, swimming, bicycling, football etc.)

☐ I do not exercise regularly

49. At age 20, what type of exercise did you do regularly (at least 3 times per week)?

☐ Mostly moderate activity (slow walking, gardening, golfing etc.)

☐ Mostly vigorous activity (running, swimming, bicycling, football etc.)

☐ I did not exercise regularly

APPENDIX F: Follow-Up Sociodemographic Charactiersitics Questionnaire

FOLLOW-UP SOCIODEMOGRAPHIC CHARACTERISTICS QUESTIONNAIRE

Date: Month_____ / Day _____ / Year_____

Study ID Number: _____

Part I: Smoking Questions

1. Are you currently smoking?

☐ **Yes**

☐ **No**

If so, how many cigarettes/day? _____

Part II: Alcohol Questions

1. Are you currently consuming alcoholic beverages such as wine, beer or liquor regularly?

☐ **Yes**

☐ **No**

If yes, please indicate pattern:

More than 1 per week. _____ If checked, please indicate number_____

Less than 1 per week_____

Part III: Medical Questions

1. Have you taken antibiotics since completing the last questionnaire?

☐ **Yes**

☐ **No (Skip to question 3)**

A list of antibiotics is attached for you to refer to.

2. If you know the name of the antibiotic(s), please write it here. _____

_____

3. Have you had a bronchoscopy since completing the last questionnaire?

☐ **Yes**

☐ **No**

4. Have you taken oral corticosteroids since completing the last questionnaire?

☐ **Yes**

☐ **No (Skip to question 6)**

(Oral corticosteroids examples: prednisone, dexamethasone, methlyprednisone, hydrocortisone)

5. If you answered Yes to (4), please write the name and dose and when you took these.

_____

6. Have you taken inhaled corticosteroids since completing the last questionnaire?

☐ **Yes**

☐ **No (Skip to Part IV)**

(Inhaled corticosteroids examples: budesonide, fluticasone, beclomethasone, ciclesonide)

7. If you answered Yes to (6), please write the name and dose and when you took these.

_____

Part IV: Diet Questions

1. Have you changed your diet since completing the last questionnaire?

☐ **Yes**

☐ **No**

If yes, please answer questions 2-9 below.

2. Do you eat meat? Meat is defined as beef, chicken, pork, lamb or venison:

A. I do not eat meat. □

B. I have eaten meat in the last year. □

C. I have eaten meat one or more times a month during the last year. □

D. I have eaten meat one or more times a week during the last year. □

3. Do you eat Fish? Fish is defined as all fish and shellfish:

A. I do not eat fish. □

B. I have eaten fish in the last year. □

C. I have eaten fish one or more times a month during the last year. □

D. I have eaten fish one or more times a week during the last year □

4. Do you eat Eggs?

A. I do not eat eggs □

B. I have eaten eggs in the last year □

C. I have eaten eggs one or more times a month during the last year □

D. I have eaten eggs one or more times a week during the last year □

5. Do you eat Cheese? (This includes fresh, soft, aged or cottage cheese as well as sour cream):

A. I do not eat cheese □

B. I have eaten cheese in the last year □

C. I have eaten cheese one or more times a month during the last year □

D. I have eaten cheese one or more times a week during the last year □

6. Do you drink Milk? Milk is defined as milk from a cow, goat or sheep (not soy, coconut or almond milk, for example). If you put milk on your cereal, you should not answer (A).

A. I do not drink milk □

B. I drank milk in the last year □

C. I drank milk one or more times a month during the last year □

D. I drank milk one or more times a week during the last year □

7. Do you eat yogurt?

A. I do not eat yogurt □

B. I have eaten yogurt in the last year □

C. I have eaten yogurt one or more times a month during the last year □

D. I have eaten yogurt one or more times a week during the last year □

8. Do you take probiotics (live bacteria supplement)?

☐ **Yes**

☐ **No**

If yes (question 8) and if you know the name of the probiotic product(s), please write it/them here:

_____

9. Do you take vitamin supplements?

☐ **Yes**

☐ **No**

If yes (question 9) and if you know the name of the vitamin supplement(s), please write it/them here:

_____