TITLE: Induction IRX-2 immunotherapy to promote immunologic priming and enhanced response to neoadjuvant pembrolizumab + chemotherapy in triple negative breast cancer (TNBC)

IND NUMBER: 146229

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PROTOCOL VERSION/DATE: Version 28-Jul-2021

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1.0 TRIAL SUMMARY

Abbreviated Title	Induction immunotherapy to promote immunologic priming and enhanced response to neoadjuvant pembrolizumab + chemotherapy in triple negative breast cancer (TNBC)
Sponsor Product Identifiers	MK-3475 (Pembrolizumab) IRX-2
Trial Phase	II
Clinical Indication	Neoadjuvant treatment for stage II-III TNBC
Trial Type	Interventional
Type of control	Pembrolizumab on a background of chemotherapy
Route of administration	Intravenous (pembrolizumab, cyclophosphamide, paclitaxel, doxorubicin), subcutaneous (IRX-2)
Trial Blinding	Open-label
Treatment Groups	There are two phases of therapy and randomization to treatment versus control: • Induction Phase o Control Arm: Pembrolizumab o Arm A: Cyclophosphamide + IRX-2 + Pembrolizumab o Subsequent arms to be added in protocol amendment • Neoadjuvant Phase o Control Arm: Pembrolizumab + Chemotherapy o Arm A: Pembrolizumab + Chemotherapy with IRX-2 re-induction o Subsequent arms to be added in protocol amendment <u>Induction phase:</u> Pembrolizumab: 200 mg fixed dose, intravenously (IV) single dose on day 1 of the induction phase [All arms] IRX-2: 2mL subcutaneous (2 x 1mL subcutaneous injection) daily for 10 days spanning days 4-21 during induction phase [Arm A] Cyclophosphamide: 300 mg/m^2, IV single dose on day 1 of the induction phase [Arm A] <u>Neoadjuvant Phase:</u> Pembrolizumab: 200mg fixed dose, intravenously (IV) Q3W of cycles 1-4 of the T regimen (treatment 1) and Q3W of cycles 1-4 of the AC regimen [All arms] Paclitaxel: 80 mg/m^2, IV, weekly, on Days 1, 8, 15 of Cycles 1-4 of the paclitaxel regimen (Treatment 1) [All arms] Followed by:

	IRX-2: 2mL subcutaneous (2 x 1mL subcutaneous injection) daily for 10 days over any 14 day period following completion of Treatment 1 and to be completed prior to advancement to Treatment 2 [Arm A] Doxorubicin: 60 mg/m^2, IV, Q3W, on Day 1 of Cycles 1-4 of the doxorubicin + cyclophosphamide (AC) regimen (Treatment 2) [All arms] Cyclophosphamide: 600 mg/m^2, IV, Q3W, on Day 1 of Cycles 1-4 of the AC regimen (Treatment 2) [All arms] Note: each cycle = 21 days
Number of trial subjects	Estimated 30 subjects will be enrolled (additional subjects pending subsequent expansions).
Estimated duration of trial	Approximately 2 years experimental arm and Arm A, from the time the first subject signs the informed consent until the last subject's last study-related visit.
Duration of Participation	Each subject will participate in the trial for approximately 34-36 weeks from the time the subject signs the Informed Consent Form (ICF) through completion of study treatment. After a screening phase of ~28 days, each subject will be receiving study treatment based on the randomization schedule for approximately 30 weeks (induction phase plus 8 cycles neoadjuvant phase followed by definitive surgery 3-6 weeks after conclusion of the last cycle of the neoadjuvant treatment.)

Randomization Ratio	1:1 experimental:control for arm A

A list of abbreviations used in this document can be found in Section 10.3.

2.0 TRIAL DESIGN

2.1 Trial Design

This is a Phase II, randomized, open-label trial to evaluate the clinical and immunologic activity of pembrolizumab plus chemotherapy when combined with various immunotherapy induction regimens as neoadjuvant therapy for triple negative breast cancer (TNBC).

A commonly used standard neoadjuvant regimen for TNBC is a weekly taxane (e.g., paclitaxel 80mg/m^2) for 12 weeks followed by an anthracycline (e.g., doxorubicin 60 mg/m^2 plus cyclophosphamide at 600 mg/m^2 [AC]) every 3 weeks (Q3W) for 4 cycles.

The chemotherapy regimen included in this study is built upon the aforementioned regimen. Pembrolizumab in combination with this regimen will be studied as part of a multi-arm study that randomizes subjects to receive:

- Control Arm: (Pembro + ACT): Pembrolizumab induction (single-dose 200mg IV), followed by pembrolizumab Q3W + paclitaxel (T) weekly x 4 cycles, followed by pembrolizumab + doxorubicin + cyclophosphamide (AC) Q3W x 4 cycles as neoadjuvant therapy prior to surgery.
- Arm A: (Pembro + IRX-2 + ACT): Pembrolizumab (single-dose 200mg IV) cyclophosphamide (single-dose 300 mg/m2 IV) + IRX-2 induction (1mL SQ x 2 daily, x 10 days), followed by pembrolizumab Q3W + paclitaxel (T) weekly x 4 cycles, followed

by IRX-2 re-induction (1mL SQ x 2 daily, x 10 days), followed by pembrolizumab + doxorubicin + cyclophosphamide (AC) Q3W x 4 cycles as neoadjuvant therapy prior to surgery.

- Subsequent induction therapy arms are to be included in protocol amendments.

Note: 1 cycle = 21 days

In the neoadjuvant phase, the dose and schedule of pembrolizumab will be fixed at 200 mg Q3W; and a commonly used AC dose/schedule will be used, i.e., doxorubicin 60 mg/m^2 Q3W plus cyclophosphamide 600 mg/m^2 Q3W. The dose level of paclitaxel is 80 mg/m^2 weekly.

Tumor Biopsy for Translational Research: Subjects with locally advanced TNBC are required to submit tissue from the diagnostic biopsy, or undergo a dedicated core needle biopsy during the screening period. Additionally, subjects will be undergo research core needle biopsy at the end of the induction treatment phase and prior to commencing the neoadjuvant treatment phase. Tumor tissue samples will also be collected at definitive surgery for subjects who have not achieved a pCR. All tumor tissue samples collected at different time points during the study will be submitted to Providence Cancer Institute for translational research as described in more detail in the laboratory manual.

Definitive surgery: Definitive surgery such as breast conservation surgery (BCS) or mastectomy with or without axillary lymph node dissection will be performed as part of the local standard of care approximately 3-6 weeks following the completion or early discontinuation of the treatments in the Neoadjuvant Treatment Phase. A thorough evaluation of breast cancer status, pathological staging per the current American Joint Committee of Cancer (AJCC) Edition 8 Breast Cancer Staging criteria and assessment of surgical margins will be performed by the local pathologist on all the tissues removed during the surgery.

Post-surgical therapy: Subjects may receive adjuvant radiation therapy and systemic therapy at the discretion of the treating provider.

Safety: Study treatment will continue until completion of study treatment, disease progression in the neoadjuvant phase or until recurrence (local or distance) after surgery, unacceptable adverse event(s), intercurrent illness that prevents further administration of study treatment, Investigator's decision to withdraw the subject from study treatment, pregnancy of the subject, noncompliance with study treatment or procedure requirements, consent withdrawal, becoming lost-to-follow-up, death, or administrative reasons that require cessation of treatment. Adverse events will be monitored throughout the trial and graded according to the National Cancer Institute (NCI) Common Terminology Criteria for Adverse Events (CTCAE) Version 5.0.

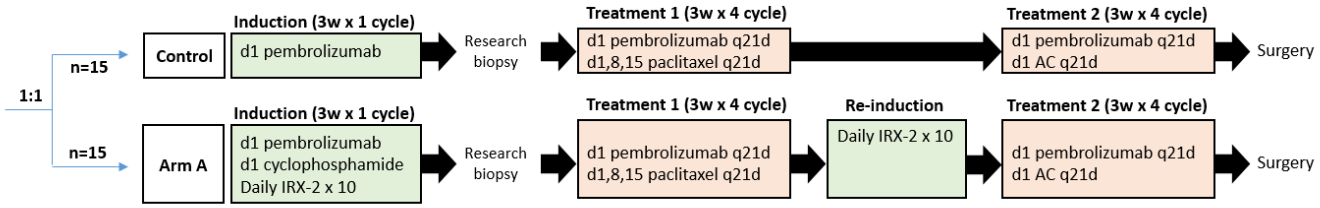
Specific procedures to be performed during the trial, as well as their prescribed times and associated visit windows, are outlined in the Trial Flow Chart - Section 6.0. Details of each procedure are provided in Section 7.0 – Trial Procedures.

The study will be conducted in conformance with Good Clinical Practices (GCPs).

2.2 Trial Diagram

The trial diagram is depicted in Figure 2.2-1.

Figure 2.2-1: Trial diagram (Control and Arm A)



AC refers to doxorubicin plus cyclophosphamide. Details of treatments are listed in subsequent sections. Following completion of accrual of Arm A (n=15) and control arm (n=15), the trial may either terminate, or may continue to enroll additional arms at n=15, each randomized 1:1 against control (n=5), with additional agents as specified in subsequent protocol amendments. See section 5.0 for additional treatment management guidelines.

3.0 OBJECTIVE(S) & HYPOTHESIS(ES)

In male and female subjects at least 18 years of age with newly diagnosed, locally advanced TNBC:

3.1 Primary Objective & Hypothesis

1) **Objective:** To evaluate the rate of pCR of pembrolizumab plus chemotherapy plus various induction regimens using the definition of ypT0/Tis ypN0 (i.e., no invasive residual in breast or nodes; noninvasive breast residuals allowed) as assessed by the local pathologist at the time of definitive surgery in subjects with locally advanced TNBC.

   Hypothesis: Administration of induction immunotherapy enhances activity of pembrolizumab plus chemotherapy, as measured by the rate of pCR using the definition of ypT0/Tis ypN0 as assessed by the local pathologist at the time of definitive surgery in subjects with locally advanced TNBC.

The study is considered to have met its primary objective if an induction regimen (example: Arm A: IRX-2) is found to have pCR rates similar or superior to historical controls of pembrolizumab plus carboplatin-containing chemotherapy obtained from the phase III KEYNOTE-522 clinical trial.

3.2 Secondary Objective(s)

1) **Objective**: To evaluate the rate of response using the residual cancer burden (RCB) index in subjects with locally advanced TNBC tumors treated with pembrolizumab plus chemotherapy plus various induction immunotherapy regimens.
2) **Objective:** To determine the safety and tolerability of pembrolizumab in combination with neoadjuvant chemotherapy and various induction immunotherapy regimens in locally advanced TNBC subjects.

3) **Objective:** To evaluate stromal TIL quantity[1] changes of therapy, comparing baseline diagnostic biopsy to post-induction research biopsy core tissue specimen.

3.3 Exploratory Objectives

1) **Objective**: To characterize TIL phenotype and activation status at baseline and following induction immunotherapy and/or pembrolizumab by various assays such as multispectral immunofluorescence (mIHC), and/or deep sequencing (TCRseq and/or RNAseq/Nanostring).
2) **Objective:** To evaluate for dynamic tumoral/immune cell PD-L1 expression following induction therapies (using assays such as mIHC, transcriptome, and/or PD-L1 assay).
3) **Objective:** To identify molecular (genomic, metabolic and/or proteomic) biomarkers that may be indicative of clinical response/resistance, safety, pharmacodynamic activity, and/or the mechanism of action of pembrolizumab+chemotherapy plus various induction immunotherapy regimens.

4.0 BACKGROUND & RATIONALE

4.1 Background

Refer to the Investigator's Brochure (IB)/approved labeling for detailed background information on MK-3475 and IRX-2.

4.1.1 Pharmaceutical and Therapeutic Background

4.1.1.1 TNBC Background

Breast cancer is the most commonly diagnosed malignancy and the second leading cause of cancer death in women. In the United States, the estimated number of new cases and death from breast cancer in 2018 is approximately 268,670 and 41,400, respectively.[2] Triple-negative breast cancer, or TNBC, which is phenotypically defined by lack of estrogen receptor (ER) and progesterone receptor expression, and the absence of human epidermal growth factor receptor-2 (HER2) overexpression and/or amplification, accounts for approximately 15-20% of all breast cancers.[3] Compared to other breast cancer subtypes, TNBC is associated with poor overall survival and high risk of disease recurrence relative to other tumor types. [3,4]

4.1.1.2 Standard-of-care therapy for TNBC

Neoadjuvant chemotherapy has emerged as the standard-of-care treatment option for locally advanced (i.e. stage II-III) TNBC in the United States. Systemic therapy is given prior to definitive surgery, which may be beneficial relative to an adjuvant approach because it allows for assessment of disease response by pathology review. Anthracycline/taxane-based regimens have been considered an important and standard part of treatment strategy for patients with locally advanced TNBC for both tumor control and improving the curability rate [5].

The poor long-term outcome in TNBC was found to be driven by those who did not achieve pCR after neoadjuvant chemotherapy. Patients who achieved pCR demonstrated sustained

clinical benefit regardless of breast cancer subtypes [6]. Recently, a large pooled analysis demonstrated strong association of pCR, when defined as no tumor in both breast and lymph nodes (ypT0 ypN0 or ypT0/is yp N0) following neoadjuvant therapy for breast cancer, with improved long-term benefit as measured by EFS and OS. Furthermore, this association was found to be strongest in patients with TNBC [7].

Because of the poorer survival for patients that did not achieve pCR after neoadjuvant chemotherapy, there is great interest in examining whether additional therapy after surgery will improve the RFS for this group of patients. The CREATE-X study demonstrated significant improvement in both EFS and OS for patients with HER2-negative breast cancer, positive lymph node and non-pCR, receiving capecitabine compared to controls [8]. In previous studies, the GeparTrio and GeparQuattro, the rate of pCR did not improve with the addition of capecitabine [9,10]. However, while previous adjuvant studies such as the GEICAM/2003-10, FINXX and/or CBCSG10 did not show a statistically significant improvement in RFS and/or OS with the addition of capecitabine in all patients, there may be a benefit in subsets of patients with TNBC, albeit with increased toxicities [11]. Thus, confirmation studies are needed. Meanwhile, current standard of care for patients who do not obtain pCR include observation, adjuvant capecitabine, or clinical trial participation.

The findings that the pCR is correlated with survival have led to increased efforts in identifying new drugs and drug combinations that can deliver higher pCR in TNBC.

Addition of a taxane to anthracycline-based regimen as adjuvant chemotherapy has been shown to improve both DFS and OS in locally-advanced breast cancer. Currently, pCR rate for standard regimen with paclitaxel followed by an anthracycline and cyclophosphamide is about 30% [12]. Because patients without pCR have significantly worse outcomes, efforts are underway to find novel combinations to improve the pCR.

4.1.1.3 Targeting PD-1 Immune Checkpoints for Cancer Treatment

It is widely accepted that cancer cells carry tumor-specific or tumor-associated antigens and therefore are immunogenic and subject to immune surveillance of the human body. However, cancer cells can often escape immune system's surveillance and control via various mechanisms and progress into clinically evident disease, a process called cancer immunoediting.[13] The ability of human cancer to evade the destruction of the immune system has recently been recognized as an emerging hallmark of cancer.

In the adaptive immune system, cytotoxic T-lymphocytes cells (CTLs, also called CD8+ or effector T cells) can recognize foreign antigens presented on the surface of antigen presenting cells (APC) via T cell receptor (TCR) and become activated executing the cell killing function. TCR-mediated T cell activations are tightly controlled by co-stimulatory and co-inhibitory signals or pathways that are triggered by the interactions between T cell surface receptors and their ligands. These inhibitory pathways, also called immune checkpoints, are crucial for maintaining self-tolerance and minimizing collateral tissue damage in the event of immune response to pathogens. Cancer can exploit immune checkpoint pathways as one of the key mechanisms to avoid being detected and destroyed. Therefore, restoration of endogenous anti-cancer immunity by immune checkpoint blockade has become an attractive strategy of cancer immunotherapy.

Among many of the agents in clinical development that target immune checkpoint pathways, those that target pathways controlled by programmed cell death protein 1 (PD-1) and cytotoxic T-lymphocyte-associated antigen-4 (CTLA-4) are the most advanced and have shown unprecedented clinical anticancer activities and durable responses across multiple solid tumors. Immune checkpoint inhibiting agents that have been approved by the US Food and Drug Administration (FDA) include ipilimumab, a full human anti-CTLA-4 monoclonal antibody (mAb), pembrolizumab (MK-3475), a humanized mAb targeting PD-1, and nivolumab, a full human mAb targeting PD-1 (see details in Yervoy® US label, Keytruda® US Label, and Opdivo® US label).

PD-1 is a member of the extended CD28/CTLA-4 family of T cell regulators. It is a transmembrane receptor including an extracellular domain that resembles the immunoglobulin variable region, a transmembrane region, and an intracellular tail that contains separate potential phosphorylation sites for signaling. Binding of PD-1 to its ligands PD-L1 (also named B7-H1) and/or programmed death – ligand 2 (PD-L2) (also named B7-DC) will trigger downstream signaling inside T cells leading to decreased cytokine production such as IL-2, inhibition of cell proliferation, reduced T cell effector function and survival. Unlike CTLA-4 which modulates the early phase of activation of naïve or memory T cells, PD-1 is expressed on antigen-experienced T cells in the peripheral tissues and therefore regulates the effector phase of the T-cell activity.

The PD-1 receptor-ligand interaction is a major pathway hijacked by tumors to suppress immune control. The normal function of PD-1, expressed on the cell surface of activated T cells under healthy conditions, is to down-modulate unwanted or excessive immune responses, including autoimmune reactions. PD-1 (encoded by the gene Pdcd1) is an immunoglobulin (IgG) superfamily member related to CD28 and cytotoxic CTLA-4, which has been shown to negatively regulate antigen receptor signaling upon engagement of its ligands (PD-L1 and/or PD-L2). The structures of murine PD-1 alone, and in complex with its ligands, were first resolved (Ref: 0422GD, 0422GF), and more recently the nuclear magnetic resonance–based (NMR-based) structure of the human PD-1 extracellular region and analyses of its interactions with its ligands were also reported. PD-1 and family members are type I transmembrane glycoproteins containing an IgG Variable-type (V-type) domain responsible for ligand binding and a cytoplasmic tail which is responsible for the binding of signaling molecules. The cytoplasmic tail of PD-1 contains 2 tyrosine-based signaling motifs, an immunoreceptor tyrosine-based inhibition motif (ITIM) and an immunoreceptor tyrosine-based switch motif (ITSM). Following T cell stimulation, PD-1 recruits the tyrosine phosphatases Src homology phosphatase (SHP)-1 and SHP-2 to the ITSM motif within its cytoplasmic tail, leading to the dephosphorylation of effector molecules, such as CD3 zeta (CD3ζ), protein kinase C-theta (PKCθ), and zeta-chain-associated protein kinase 70 kDa (ZAP70), which are involved in the CD3 T cell signaling cascade. The mechanism by which PD-1 down-modulates T cell responses is similar to, but distinct from, that of CTLA-4. PD-1 was shown to be expressed on activated lymphocytes, including peripheral CD4+ and CD8+ T cells, B cells, T regs and natural killer cells (NKCs). Expression has also been shown during thymic development on CD4-CD8–double negative T cells as well as subsets of macrophages and dendritic cells. The ligands for PD-1 (PD-L1 and PD-L2) are constitutively expressed or can be induced in a variety of cell types. PD-L1 is expressed at low levels on various nonhematopoietic tissues, most notably on vascular endothelium, whereas PD-L2 protein is only detectably expressed on

antigen-presenting cells found in lymphoid tissue or chronic inflammatory environments. Both ligands are type I transmembrane receptors containing both IgV- and gC-like domains in the extracellular region and short cytoplasmic regions with no known signaling motifs. Binding of either PD-1 ligand to PD-1 inhibits T cell activation triggered through the T cell receptor. PD-L2 is thought to control immune T cell activation in lymphoid organs, whereas PD-L1 serves to dampen unwarranted T cell function in peripheral tissues. Although healthy organs express little (if any) PD-L1, a variety of cancers were demonstrated to express abundant levels of this T cell inhibitor, which, via its interaction with the PD-1 receptor on tumor-specific T cells, plays a critical role in immune evasion by tumors. As a consequence, the PD-1/PD-L1 pathway is an attractive target for therapeutic intervention in cancer.

4.1.1.4 Targeting PD-1 Immune Checkpoints for TNBC

Several studies have demonstrated that the presence of tumor-infiltrating T-lymphocytes (TILs) correlated with better prognosis in TNBC, independent of systemic therapy.[14] In addition, unsupervised gene expression profiling of TNBCs has identified a gene signature enriched for cytotoxic CD8+ T cell genes and natural killer cell (NKC) activity, which is predictive of good clinical outcome. These findings suggest an active role of acquired immunity in concurring TNBC.[15]

PD-L1, which is not detected in normal breast tissue, has been reported to be expressed in about half of all breast cancers, particularly in hormone-receptor-negative, high grade and proliferative tumors.[16] The presence of Treg cells, tumor PD-L1 expression, and PD-1–positive TILs has been associated with high histologic grade, ER negativity, and prominent tumor lymphocytic infiltration. Recently, it is reported that PD-L1 messenger ribonucleic acid (mRNA) is expressed in nearly 60% of breast tumors, independently of hormone receptor status, and is positively correlated with PD-L1 protein expression and increased TILs. Another study mining The Cancer Genome Atlas ribonucleic acid (RNA) sequencing data showed that PD-L1 gene expression is significantly higher in TNBCs compared to non-TNBCs, and is associated with Phosphatase and Tensin Homolog (PTEN) loss. This evidence demonstrates that TNBCs are characterized by PD-L1 positivity and presence of TILs, and thus suggest that PD-1 immune checkpoint inhibition is a therapeutic strategy worthy of further investigation for the treatment of this aggressive breast cancer subtype.

4.1.2 Induction Immunotherapy approaches to enhance response to chemotherapy and immune checkpoint therapy

Despite the above associations, TNBC remains a heterogeneous disease, with only a subset of tumors expressing PD-L1 and exhibiting dense baseline TIL infiltrate, suggesting that the addition of immune checkpoint antibodies to chemotherapy may not be an effective strategy for all locally advanced TNBCs. Complementary immunotherapeutic approaches are being explored in combination with anti-PD-1/L1 to facilitate anti-tumor immune responses. Examples include therapies that alter the microenvironment or cytokine milieu, or therapies that induce immunogenic cell death and/or antigen presentation.

Recently, the phase II TONIC trial evaluated an induction approach whereby metastatic TNBCs were treated for two weeks with various therapies (radiotherapy, low-dose cyclophosphamide, low-dose doxorubicin, low-dose platinum) preceding anti-PD-1. In this

study, clinical response appeared to be potentiated with the addition of induction low-dose doxorubicin and/or cisplatin.[17]

The overarching goal in this protocol is to sequentially evaluate whether various candidate induction strategies may be used to enhance response to immune checkpoint therapy plus chemotherapy in locally advanced TNBC.

4.1.2.1 IRX-2 cytokine therapy as an induction therapy preceding anti-PD-1

IRX-2-is a cell-derived biologic with multiple active cytokine components produced under pharmaceutical standards as discussed in more detail in the IB. Briefly, human leukocytes ("buffy coats") pooled from multiple donors are stimulated with phytohemagglutinin and ciprofloxacin. Subsequently, the phytohemagglutinin, ciprofloxacin and all cellular elements are removed or significantly reduced, and the cell-free supernatant is filter sterilized, nanofiltered to clear viral particles, vialed, and frozen as IRX-2. IL-2 is the major cytokine in IRX-2, followed by IFN-γ, TNF-α, and interleukin 1 beta (IL-1β). These cytokines when studied individually enhance cell-mediated immunity via several different mechanisms discussed below and in the IRX-2 IB.

Recent *in vitro* studies have elucidated several potential mechanisms of action of IRX-2, and these various mechanisms of action need not be exclusive. IRX-2 treatment of human monocyte-derived dendritic cells results in changes consistent with the development of mature activated dendritic cells. Specifically, IRX-2 increased the percentage of cells expressing CD83 and CCR7, markers for dendritic cell maturation and migration and increased the expression of multiple markers that are critical mediators of T cell activation [18]. Similar results were obtained in a later study in cells obtained from patients with HNSCC[19]. Also, in an in vitro study of peripheral blood mononuclear cells obtained from patients with HNSSC, IRX-2 up-regulated cytotoxicity of NK cells and did so more effectively than IL-2[20].

IRX-2 can also protect T cells from activation induced cell death by reversing microvesicle induced inhibition of the PI3K/Akt pathway and correcting the imbalance of pro- versus anti-apoptotic proteins induced by tumor-derived microvesicles[21,22]. IRX-2 was superior to recombinant IL-7 and IL-15 in protecting T cells from tumor-induced apoptosis. The presence of IRX-2 in a tumor microenvironment model promoted the induction and expansion of IFN-γ+T-bet+ Teff and significantly decreased the induction of inducible IL-10+TGF-β+ Treg. The responsible mechanism involved IFN-γ+-driven T cell polarization towards Teff and suppression of Treg differentiation[20]. In the Phase 2a study in subjects with HNSSC (described below in Section 1.3.1), IRX-2 mediated reductions in circulating B and NKT cell numbers, suggesting redistribution of these cells to tissues[23]. A decrease in naïve T cells was also noted, suggesting their upregulation to memory T cells, while unchanged numbers of Tregs (suppressor T cells) after IRX-2 therapy indicated that IRX-2 does not expand this compartment, potentially benefiting anti-tumor immune responses.

IRX-2 has been shown to induce enhanced T cell responses when administered with tumor antigen vaccines, raising the possibility that IRX-2 treatment in subjects with HNSCC enhances endogenous antigen-specific T cell responses to the tumor.[24]

4.1.2.1.1 Delivery of IRX-2 via breast lymphatics

The route of administration of IRX-2 takes advantage of the normal pathways of lymph node activation. Normally, lymphatics drain from an area of disease, such as a tumor bed, and antigens and other factors associated with disease migrate in the lymphatics to the regional nodes. By presenting the cytokine-containing biologic in the area of the tumor-draining lymph nodes rather than systemically, there is an opportunity to mobilize antigen presenting cells and enhance dendritic cell function as well as directly activate T cells to proliferate and become cytotoxic lymphocytes. Additionally, subcutaneous administration has been less toxic since the systemic cytokine drug concentration is much lower.

Periareolar injection of radioactive colloid is an acceptable and clinically utilized technique for localizing sentinel lymph nodes in breast cancer[25] and therefore it is assumed that periareolar injection of IRX-2 leads to update of the IRX-2 product into the draining lymph nodes of the tumor. Peri-areolar injections have been shown to be safe and feasible, with pharmacodynamic activity, in a pilot/phase Ib study in early stage breast cancer, which is described later in detail.[26]

4.1.2.1.2 Components of IRX-2 Induction regimen

The IRX-2 induction regimen being investigated in this trial includes single-dose cyclophosphamide (300mg/m^2 IV on day 1), followed by 10 peri-areolar subcutaneous injections of the IRX-2 cytokine product (2 x 1mL product daily).

The IRX-2 regimen includes cyclophosphamide, which is thought to mediate T-regulatory cell depletion and immunogenic cell death. Evidence indicates that cyclophosphamide inhibits Treg number and/or function[27]. Thus many clinical trials that involve immunotherapy or attempt to stimulate immune response to tumor antigens have employed low dose cyclophosphamide (300 mg/m2) as a component of the treatment regimen. This immunomodulatory dose is half of the typical anti-cancer dose included in the AC regimen, and is intended to enhance the development of cell-mediated immunity by providing contra-suppression of tumor-associated immune suppression (to reduce the number and function of regulatory T cells, i.e. Treg). Pathways of local immune tolerance, escape mechanisms active within the tumor microenvironment and superimposed potent systemic mechanisms of immune tolerance have been reviewed and are discussed in more detail in the Investigator's Brochure (Section 3.4.1). The use of cytotoxic chemotherapy in doses and schedules designed to abrogate specific mechanisms of immune tolerance in order to release the full potential of an antitumor immune response is discussed. Specifically, cyclophosphamide may be used to prime the immune system by promoting the differentiation of CD4+ T helper cells and by abrogating the suppressive influence of CD4+CD25+ T regulatory (Treg) cells. In the absence of Treg influence, high-avidity CD8+ T cells are recruited to an antigen-specific immune response. Cyclophosphamide also facilitates the establishment of memory CD8+ T cells. Thus inclusion of cyclophosphamide in combination with other immune-modulatory agents is supported by both pre-clinical and clinical data as reviewed in more detail by Emens (2005) and in the Investigator's Brochure (Section 3.4.1).

In previous trials, IRX-2 was co-administered with oral indomethacin and oral vitamin supplementation, based upon preclinical rationale the nonselective COX-1/COX-2 inhibition may results in reversal of immunosuppression induced by prostaglandin. However, because a

number of subjects with breast cancer experienced adverse gastrointestinal effects related to the indomethacin requiring discontinuation of this component of the regimen, subsequent clinical studies of IRX-2 will not include oral indomethacin or supplements.

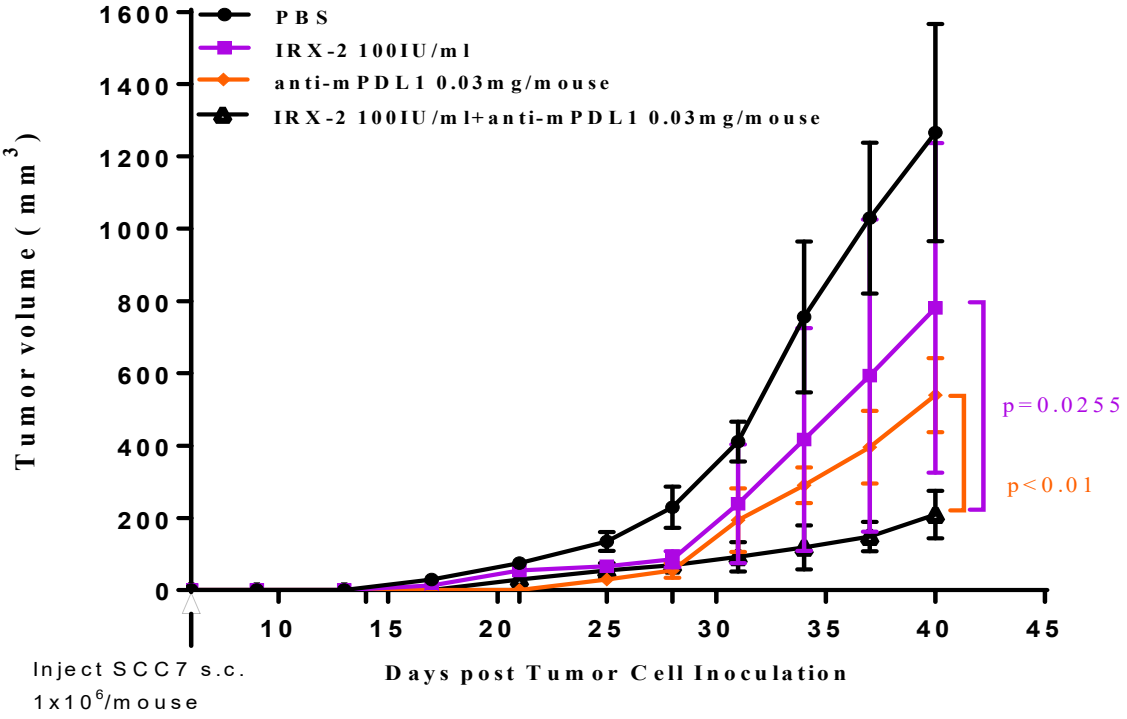
4.1.2.1.3 Preclinical rationale to combine IRX-2 with anti-PD1

Recently it has been shown that the degree of lymphocyte infiltration is an important prognostic factor for treatment with anti-PD-1 monoclonal antibodies [28]. Furthermore, the degree of tumoral and/or immune cell PD-L1 expression may predict clinical benefit in the setting of anti-PD-1/L1 therapy. These two data provide a compelling rationale for consideration of IRX-2 as an induction agent. In a phase Ib trial in early stage breast cancer, IRX-2 induction therapy was shown to increase stromal TIL count and to increase mRNA transcripts of PD-L1.[26]. In a Phase 2 trial, IRX-2 was shown to increase T cell infiltration into the tumor and this correlated with overall survival in HNSCC[29]. We have found increases in PD-L1 in 4 of 7 Head and neck patients treated with neoadjuvant IRX-2 regimen as documented by Perkin-Elmer multiplex IHC platform. In addition similar findings were documented by NanoString for both PD-L1 and CTLA-4 on 3 of 7 patients. Together with increases in lymphocytic infiltration seen in 21 of 25 patients[29] by standard IHC, a known important predictive parameter for response to many checkpoint inhibitors, there is a strong rationale for the combination.

IRX-2 has also been shown to induce enhanced T cell responses when administered with tumor antigen vaccines, raising the possibility that IRX-2 treatment in patients with HNSCC enhances endogenous antigen-specific T-cell responses to the tumor[24].

Recently at the University of Michigan using a SCC7 model of squamous cell head and neck cancer, the combination of anti-PD-L1 plus IRX-2 resulted in synergistic slowing in tumor outgrowth relative to control or monotherapy with either IRX-2 or anti-PD-L1. In this model, SCC7 tumors were injected orthotopically (1×10^6 tumor cells injected into SC flanks), followed by IRX-2 100IU/mL at day 14 (weekly x 3) and/or anti-PD-L1 (0.03mg twice weekly x 3 weeks). Mice were observed before tumor sacrifice for analysis of lung/primary tumor/spleen and PBMCs. As shown in figure 4.1.2.1.3-1, combination therapy was associated with significant slowing of tumor outgrowth relative to control (PBS) or either monotherapy arm (unpublished, data shown below).

Figure 4.1.2.1.3-1: synergistic efficacy of IRX-2 plus anti-PD-L1 in a SCC7 orthotopic model



4.1.2.1.4 Rationale to include IRX-2 re-induction following Treatment 1 and preceding Treatment 2

No clinical or preclinical data are available to ascertain the optimal duration of IRX-2 therapy in the context of anti-PD-1 and/or neoadjuvant chemotherapy in breast cancer. However, IRX-2 is known to facilitate antigen presentation and recruit immune cells. Because Treatment 2 constitutes a new chemotherapy type (doxorubicin and cyclophosphamide) with a unique potential for immunologic priming, in arm A a re-induction period of 10 days of IRX-2 injections will be administered following completion of Treatment 1 (paclitaxel/pembrolizumab) and Treatment 2 (AC/pembrolizumab). IRX-2 will be given at least 1 day following completion of Treatment 1 (i.e. after cycle 4 day 15) to mitigate potential concerns of toxicity with concurrent therapy.

4.1.3 Summary of Clinical Activities: Pembrolizumab

Pembrolizumab is a potent and highly selective humanized mAb of the IgG4/kappa isotype designed to directly block the interaction between PD-1 and its ligands, PD-L1 and PD-L2. Details regarding preclinical, clinical pharmacology, and clinical efficacy and safety studies can be found in pembrolizumab clinical IB, the US label, and the Summary of Product Characteristics (SmPC).

Pembrolizumab has demonstrated robust, substantial, and clinically-meaningful benefit in the treatment of a number of solid tumors, based on RECIST 1.1 and immune-related RECIST (irRECIST) recommendations. Pembrolizumab has been generally well tolerated, as expected based on preclinical findings and data from other anti-PD-1 monoclonal antibodies. Pharmacokinetics were as expected, based on pembrolizumab being an IgG mAb and based on preclinical data, which support dosing once every 2 or 3 weeks.

4.1.3.1 Summary of Clinical Data Supporting Pembrolizumab Use for Treatment of Metastatic TNBC

4.1.3.1.1 KEYNOTE-012 (KN012)

In Study KN012, a cohort of 32 female patients with metastatic TNBC, with PD-L1 positivity (defined as PD-L1 expression in ≥1% tumor cells or in stroma, using a prototype assay and the 22C3 antibody) was enrolled and received pembrolizumab 10 mg/kg Q2W dose. Subjects with a median age of 51.9 years (range: 29-72 years) and PD-L1 (+) metastatic TNBC (mTNBC) were enrolled in the study. The currently available prevalence of PD-L1 positivity in mTNBC is 58%, as determined by this study KN012. Most of these patients had received and progressed on multiple lines of therapy for advanced disease (median number of prior treatments for metastatic disease was 3). Based on a data cutoff of 06-Nov-2014, 5 (15.6%) patients experienced at least one drug-related SAE; each of 4 patients experienced one of the following: Grade 3 anemia, headache, aseptic meningitis or pyrexia, and a fifth patient experienced Grade 5 disseminated intravascular coagulation (DIC) with thrombocytopenia and decreased blood fibrinogen. Of the 27 patients with centrally confirmed measurable disease, 1 (3.7%) patient had a complete response (CR), 4 patients (14.8%) had a confirmed partial response (PR), 25.9% had stable disease (SD), and 44.4% had progressive disease (PD), based on RECIST 1.1 as assessed by the central imaging vendor. At this cutoff, the median duration of response had not been reached (range: 15 to 40+ weeks), and 3 patients (1 CR; 2 PR) were still on treatment after at least 11 months. Given that the current systemic treatments had little effect in this setting, this result looks very promising.

4.1.3.1.2 Additional Ongoing Clinical Studies with Pembrolizumab in breast cancer

Two clinical studies are currently investigating the efficacy of single agent pembrolizumab as later line of treatment for mTNBC, namely KEYNOTE-086 (KN086), KEYNOTE-119 (KN119), and 2 clinical studies are investigating the efficacy of combination of pembrolizumab with chemotherapy (KEYNOTE-355 [KN355] and KEYNOTE-173 [KN173]).

- KN086 (NCT02447003): A Phase II Clinical Trial of Pembrolizumab (MK-3475) as Monotherapy for Metastatic Triple Negative Breast Cancer (mTNBC) – (KEYNOTE-086)
- KN119 (NCT02555657): A Randomized, Open-Label, Phase III Clinical Trial of Single Agent Pembrolizumab vs Single Agent Chemotherapy per Physician's Choice for Metastatic Triple Negative Breast Cancer (mTNBC) – (KEYNOTE-119)

- KN173 (NCT02622074): A Phase 1b Study to Evaluate safety and clinical activity of Pembrolizumab (MK-3475) in combination with Chemotherapy as Neoadjuvant Treatment for Triple Negative Breast Cancer (TNBC) - (KEYNOTE-173)
- KN355 (NCT02819518): A Randomized, Double-Blind, Phase III Study of Pembrolizumab (MK-3475) plus Chemotherapy vs Placebo plus Chemotherapy for Previously Untreated Locally Recurrent Inoperable or Metastatic Triple Negative Breast Cancer – (KEYNOTE-355)

A number of clinical trials are ongoing that evaluate pembrolizumab in the neoadjuvant or adjuvant setting, including the randomized comparison of neoadjuvant chemotherapy with/without pembrolizumab for triple negative breast cancer (KEYNOTE-522), as well as a randomized assessment of adjuvant pembrolizumab versus placebo in triple negative breast cancers not experiencing pathologic complete response (SWOG 1418).

Ongoing clinical studies are also being conducted in melanoma, NSCLC, head and neck cancer, breast cancer, gastric cancer, colorectal cancer, a number of other advanced solid tumor indications, and hematologic malignancies. For further details, please refer to the IB.

4.1.4 Summary of Clinical Activities: IRX-2

4.1.4.1 Phase I clinical trial

IRX-2 2004-B was a multicenter, Phase 1 trial in subjects with advanced stage HSNCC who had progressed after surgery and/or radiation therapy that was designed to evaluate the clinical and laboratory safety and tolerability of the IRX-2 regimen. Results of the study are presented in the IB and have been published. The reported toxicities were acceptable overall and did not preclude proceeding to additional trials.

4.1.4.2 Phase 2 trial in HNSCC

IRX-2 2005 A was a multi-center trial entitled "A Phase 2, Open-label Trial of the Safety and Biological Effect of Pre-operative Subcutaneous IRX-2 (with Cyclophosphamide, Indomethacin, and Zinc) in Subjects with Resectable Cancer of the Head and Neck." Results of the study are presented in the IB and have been published[20,29-31] and are also summarized here.

The study objectives were to determine the safety of the IRX-2 regimen when used as neoadjuvant (preoperative) therapy and to evaluate clinical, pathological, and radiographic tumor response and disease-free survival (DFS) and overall survival (OS).

Changes in tumor size between baseline and immediately pre-operative, i.e. after the IRX-2 regimen, measurements were evaluable in 23 subjects. Based on measurement of the longest single tumor diameter, tumor growth or shrinkage was as follows: 4 subjects had a decrease in tumor size, 16 subjects had no significant change, and 3 subjects had an increase in tumor size.[30] Review of the radiologic findings in relation to the pathology finding in the resection specimens, however, established that lymph node or tumor enlargement due to reactive hyperplasia could not be distinguished from enlargement related to tumor growth.

One subject underwent a fluorodeoxyglucose positron emission tomography (FDG-PET) CT scan at baseline and at completion of the IRX-2 regimen. Elevated glycolytic activity was

observed in 2 lymph nodes and in the primary tumor on the baseline PET scan. At the completion of the IRX-2 regimen, there was a 75% decrease in glycolytic activity in these lesions.

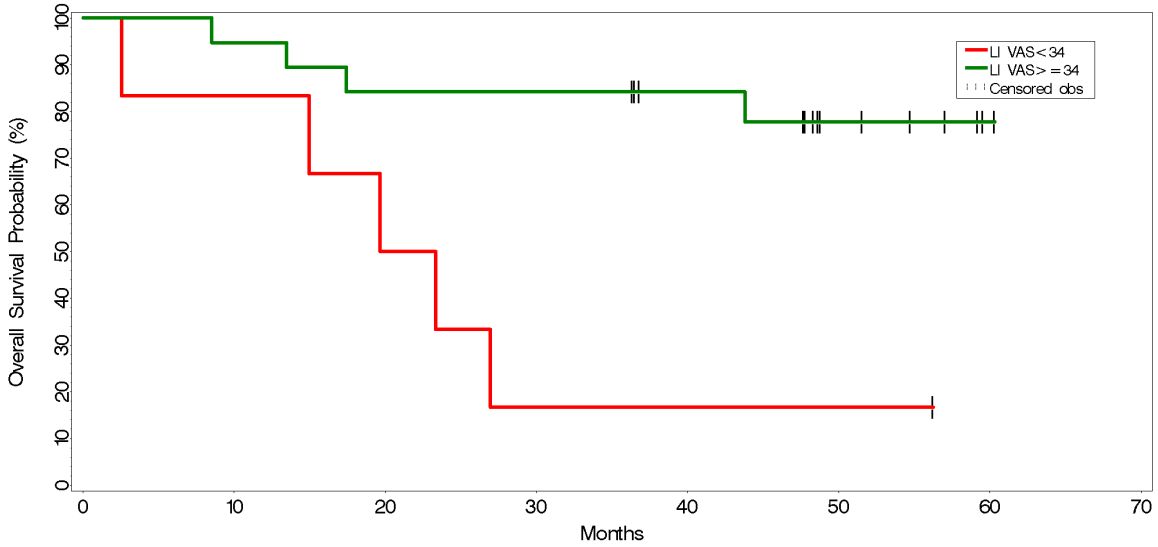
After 5 years of follow-up, median DFS and OS had not been reached. These results for both DFS and OS appeared to be slightly superior to those observed in a comparable group of 81 historical controls, treated at the University of Michigan and matched for baseline characteristics.

The IRX-2 regimen was tolerated with minimal toxicity. Compliance was excellent; all subjects completed the regimen and there were no unplanned delays in surgery as a result of the immunotherapy regimen. There were no reports from investigators of any unanticipated or unusual delays or difficulties in performing the planned resections or reconstructions or during the postoperative recovery.

The most common adverse events (AEs) were headache (30%), injection site pain (22%), nausea 22%), constipation (15%), dizziness (15%), fatigue (11%), aspiration pneumonia (11%), anemia (11%) and myalgia (7%). All were Grade 1-2 except for the aspiration pneumonias (one Grade 3, one Grade 4) and all resolved without sequelae. There were only minor (Grade 1) alterations in post-treatment laboratory values. Eight serious adverse events (SAEs) in 7 subjects were reported during treatment and the 30-day post-operative period: aspiration pneumonia (n = 3), respiratory tract infection, asthma exacerbation, wound infection, neck abscess and alcohol withdrawal (n = 1 each); only the postoperative wound infection was considered related to the study treatment. Note that many of these AEs, including e.g. aspiration pneumonia, wound infections, alcohol withdrawal, are common in HNSCC subjects during their pre- and post-operative course. During treatment, several subjects noted decreased pain or improved swallowing and no significant progressive symptoms were noted.

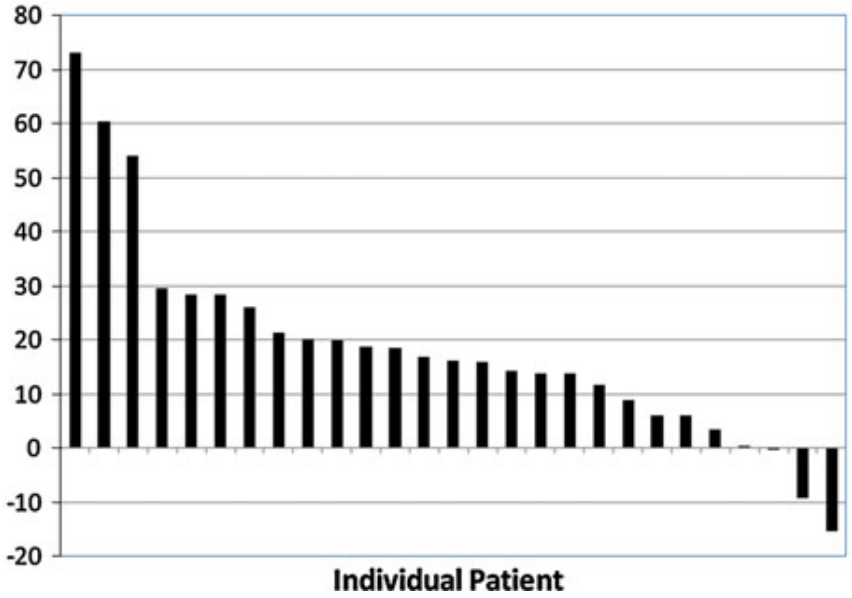
Pretreatment tumor biopsies and the tumor surgical specimens from 25 subjects were characterized for lymphocyte infiltration, necrosis and fibrosis using both hematoxylin and eosin stains and immunohistochemistry[29]. Kaplan-Meier estimates of overall survival are displayed in Figure 1 as a function of high and low lymphocyte infiltration in the surgical specimens after the IRX-2 immunotherapy. Eighteen subjects were in the better survival group and 7 were in the inferior survival group; the survival curves are significantly different ($p < 0.05$).

Figure 4.1.4.2-1: Overall Survival by High and Low Lymphocyte Infiltration in the Surgical Resection Specimen (Study IRX-2 2005-A)



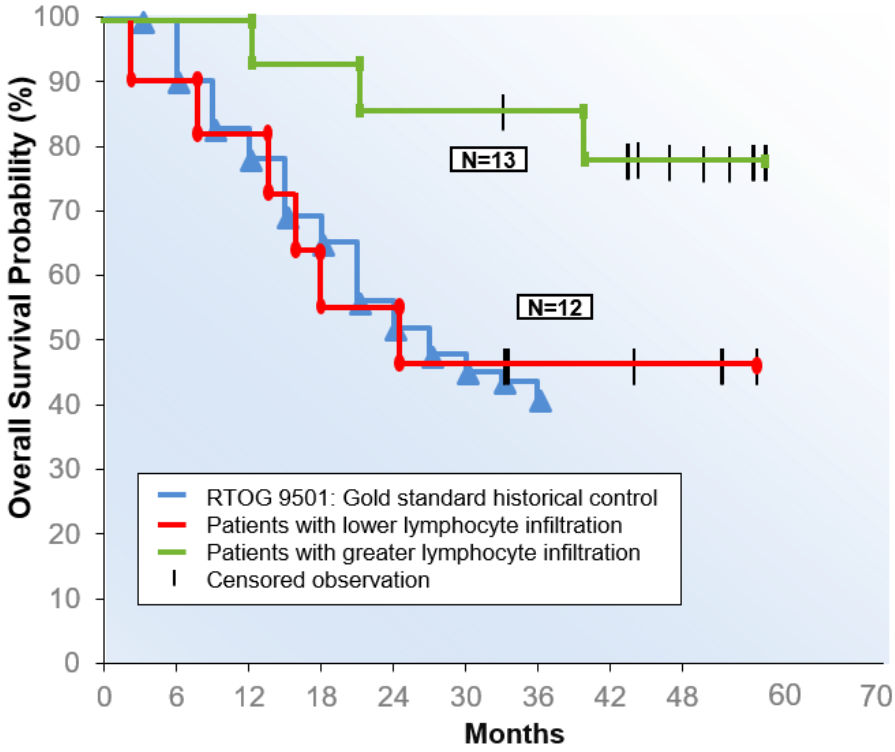
When lymphocyte infiltration in the pretreatment biopsies was compared to that in the resected surgical specimen, increases in lymphocyte infiltration were seen as shown in Figure 2 (change in mean lymphocyte infiltration from the biopsy to the surgical specimen is shown on the y-axis).

Figure 4.1.4.2-2: Lymphocyte Infiltration (Study IRX-2 2005-A)



In addition, subjects in whom the greatest increase in tumor lymphocyte infiltration from biopsy to surgery (n=14) was observed had a trend toward superior survival compared to subjects in whom no or more limited change was observed (n=11) as shown in Figure 3 (p = 0.10) (Brooklyn Immunotherapeutics, unpublished observations).

Figure 4.1.4.2-3: Overall Survival vs. Change in Lymphocyte Infiltration (Study IRX-2 2005-A)



Peripheral blood lymphocyte subsets also were monitored pre- and post-treatment with the IRX-2 regimen to evaluate changes induced by the IRX-2 regimen (summarized in Section 4.1.4.2 above and in the IB). The IRX-2 regimen-mediated reductions in B and NKT cell numbers in the blood suggested a redistribution of these cells to tissues while the unchanged numbers of Tregs after IRX-2 therapy indicated that IRX-2 does not expand this compartment, potentially benefiting anti-tumor immune responses.

4.1.4.3 Phase Ib Trial of IRX2 in early stage breast cancer

A phase Ib trial evaluating safety and feasibility of IRX2 in stage I-III (any histology) breast cancer has completed accrual. Preliminary results were presented at SITC 2017 and ACSO 2018.[26] The primary outcome of the study was to evaluate the safety/tolerability of IRX2 during a pre-surgical window-of-opportunity in patients for whom surgical lumpectomy/mastectomy was planned. In this study, all 16 subjects (n=16/16, 100%) tolerated

the therapy without dose interruption or reduction of the IRX-2 product. No unexpected surgical complications or delays were observed.

Pharmacodynamic assessment of the tumor bed and PBMCs was conducted. All patients (n=16/16) completed and tolerated the regimen with no surgical delays or treatment-attributed grade III/IV toxicities. Common adverse events (occurring in >15% subjects) attributed to IRX-2 injections were: injection site reaction (grade 1, n=8/16), bruising (grade 1, n=7/16), and pain (grade 1, n=3/16). Common adverse events attributed to low-dose cyclophosphamide were: fatigue (grade 1, n=5/16) and nausea (grade 1/2, n=3/16). Treatment was associated with an increase in sTIL score (Wilcoxon signed-rank p=.04), with 4/10 sTIL-low tumors (0-10% score) re-categorized to sTIL-moderate (11-50% score). Increases in PD-L1 RNA expression were observed (Wilcoxon signed-rank p=.04) in 12/16 tumors (median 57% increase, range: -53% to 185% increase), as well as increases in Nanostring cell signatures (natural killer cell and Th1 signatures). In blood, increases in CD4 and CD8 effector T-cell activation (ICOS, HLA-DR, and CD38) and T-reg depletion were observed.

4.1.5 Rationale for the Trial and Selected Subject Population

It is well known that TNBC has the worst prognosis and is the most difficult to treat among the breast cancer subtypes. Recent clinical trials data support the possibility that pembrolizumab may increase pathologic complete response rates when combined with standard-of-care neoadjuvant paclitaxel, doxorubicin, and cyclophosphamide.[32]. These findings are being confirmed in a large placebo-controlled study evaluating neoadjuvant paclitaxel, carboplatin, doxorubicin, and cyclophosphamide, +/- pembrolizumab versus placebo (Merck KEYNOTE-522).

Anticipating that the addition of pembrolizumab improves outcome in KEYNOTE-522, one future direction will be to incorporate induction immunotherapy approaches to enhance immune response. If proven, such an approach may allow for de-escalation of the chemotherapy backbone, thereby reducing toxicity and morbidity associated with curative-intent therapy in this disease.

Because the addition of carboplatin is not a universally accepted standard treatment for locally advanced TNBC (owing to disparate data on long term survival outcomes), and because the addition of carboplatin substantially increases the likelihood of hematologic toxicities and other toxicities, the rationale of this trial is to evaluate the efficacy and tolerability of various immunotherapy/chemotherapy combination approaches that omit carboplatin. The trial is designed to offer preliminary point estimates of pCR rates, which could be compared to contemporary trials data from the I-SPY-2 and KEYNOTE-522 trial. Furthermore, randomization to pembrolizumab-only induction arm will afford an opportunity to conduct a comparative analysis of immunologic activity, as measured by serial blood and tissue biopsy following induction immunotherapy.

Arm 2 of the trial will evaluate whether the addition of IRX-2 to pembrolizumab is associated with favorable pCR rates and favorable immunologic activity. Subsequent induction agents will be studied in future arms of the study. Arm 1 (control) will enroll in an ongoing fashion).

4.1.6 Rationale for Dose Selection/Regimen/Modification

4.1.6.1 Rationale for Testing Pembrolizumab in Combination with the Selected TNBC Neoadjuvant Regimen

The rationale for testing pembrolizumab in combination with the selected chemotherapy regimens is as follows. Pembrolizumab functions as an immune checkpoint blockade by targeting PD-1, which helps to restore the endogenous anti-cancer immunity. Pembrolizumab has shown significant clinical anti-cancer activity across multiple tumor types including melanoma, NSCLC, head and neck cancer, bladder cancer and has gained FDA approval for treating advanced melanoma, head and neck, and NCSLC. Preliminary data have also shown promising clinical activity of pembrolizumab in metastatic TNBC patients who have failed multiple prior treatments. Therefore, further testing of pembrolizumab in both the metastatic and early stage such as a neoadjuvant and/or adjuvant setting is warranted.

Pembrolizumab relies on a functional immune system to exert its anti-tumor effect. Theoretically, an even greater tumor cell reduction might be achieved by enhancing the antigen presentation via administration of pembrolizumab in combination with standard cytotoxic chemotherapy, provided that the immune suppression by some of these agents (e.g., cyclophosphamide) do not significantly compromise the anti-tumor effect of pembrolizumab. Optimal supportive care may alleviate some of these potential negative impacts.

4.1.6.1.1 Rationale for evaluating combination immunotherapy with a carboplatin-sparing neoadjuvant chemotherapy background regimen

Across 2 randomized trials, adding carboplatin in combination with weekly paclitaxel at 80 mg/m^2 versus paclitaxel alone followed by the standard anthracycline/cyclophosphamide combination has shown increased pCR rates as neoadjuvant treatment for TNBC via 2 randomized trials using either weekly carboplatin at AUC 2 (the Phase II GeparSixto trial), (Ref: 047FC8) or carboplatin at AUC 6 Q3W (the Phase III CALGB 40603 trial). Due to toxicity, in the GeparSixto trial, the dose of carboplatin was reduced to AUC 1.5. A meta-analysis by Petrelli et al to compare TNBC patients who received carboplatin vs. those who did not receive carboplatin in the neoadjuvant setting, showed the risk of not having a pCR for those without carboplatin was 1.45 (95% CI, 1.25-1.68, $p<0.0001$) compared to those who have received carboplatin. Based upon the potential for improved pCR rates, the phase III KEYNOTE-522 study is evaluating whether the addition of pembrolizumab may improve the reponse to a neoadjuvant regimen containing paclitaxel, carboplatin, doxorubicin, and cyclophosphamide.

However, despite trials showing improved pCR with carboplatin, there are still limited prospective data confirming that the addition of carboplatin improves long term survival or recurrence free survival. Furthermore, hematologic toxicities are significantly higher with the addition of carboplatin. For example, in the CALGB 40603 study the carboplatin AUC 6 Q3W plus weekly paclitaxel 80 mg/m^2 arm showed statistically significant increase in Grade 3/4 neutropenia (56% vs. 22%) and Grade 3/4 thrombocytopenia (20% vs. 4%) compared to paclitaxel alone arm.

Because of the lack of long-term survival data and because of the increased toxicities, it may be of interest to evaluate combination immunotherapy approaches with a carboplatin-sparing chemotherapy backbone. This study will estimate the pCR rate of a chemotherapy backbone regimen of weekly paclitaxel, doxorubicin, and cyclophosphamide, but omitting carboplatin and adding a secondary immunotherapy agent. The goal is to achieve pCR rates that are similar to carboplatin-containing chemo-immunotherapy, but without the added toxicities of carboplatin.

Refer to the Investigator's Brochure for additional preclinical and clinical data.

4.1.6.2 Rationale of Pembrolizumab Dose

The optimal duration of pembrolizumab has not been tested, and therefore for this trial, pembrolizumab will only be given during the neoadjuvant phase and will not be continued beyond surgery. If during the trial pembrolizumab is demonstrated to improve long-term outcomes when administered beyond surgery, the protocol may be amended to allow for extended pembrolizumab dosing.

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

- Clinical data from 8 randomized studies demonstrating flat dose- and exposure-efficacy relationships from 2 mg/kg Q3W to 10 mg/kg every 2 weeks (Q2W)

- Clinical data showing meaningful improvement in benefit-risk including OS at 200 mg Q3W across multiple indications, and

- Pharmacology data showing full target saturation in both systemic circulation (inferred from pharmacokinetic [PK] data) and tumor (inferred from physiologically based pharmacokinetic [PBPK] analysis) at 200 mg Q3W

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

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

Finally, population PK analysis of pembrolizumab, which characterized the influence of body weight and other subject covariates on exposure, has shown that the fixed-dosing provides similar control of PK variability as weight based dosing, with considerable overlap in the distribution of exposures from the 200 mg Q3W fixed dose and 2 mg/kg Q3W dose. Supported by these PK characteristics, and given that fixed-dose has advantages of reduced dosing complexity and reduced potential of dosing errors, the 200 mg Q3W fixed-dose was selected for evaluation across all pembrolizumab protocols.

4.1.6.1 Rationale for pembrolizumab dosing and schedule during the induction period

The pembrolizumab dose during induction period will be 200mg IV on day 1, similar to the neoadjuvant period. Subjects will advance to the neoadjuvant phase after at least 2 weeks following pembrolizumab dosing, but no more than 26 days. Pembrolizumab has been studied and demonstrated to be safe at doses up to 10mg/kg q2wk.

4.1.6.2 Rationale for IRX-2 dosing, delivery method, and schedule

The IRX-2 dosing schedule and regimen are directly informed by a preceding phase Ib study in ESBC that demonstrated safety/tolerability of the regimen, as well as an associated expansion of TILs and PD-L1 expression within the tumor[26]. Cyclophosphamide will be given at 300mg IV on day 1, followed by the IRX-2 biologic administered on days 4-21 (daily on any 10 days, 2 x 1mL daily).

A re-induction period of IRX-2 will occur following completion of treatment 1 (paclitaxel/pembrolizumab) and preceding initiation of treatment 2 (AC/pembrolizumab). This will consist of only IRX-2 biologic (daily on any 10 days, 2 x 1mL daily). The rationale is that pre-treatment with IRX-2 may enhance the microenvironment of the residual tumor and draining lymph nodes, and facilitate priming associated with the initiation of AC chemotherapy.

4.1.6.3 Rationale for Dose Interval for Neoadjuvant therapy

The standard neoadjuvant treatment is typically 4 cycles of taxane (12 × weekly dosing) followed by 4 cycles of anthracycline/cyclophosphamide combination (e.g., doxorubicin 60 mg/m^2 + cyclophosphamide 600 mg/m^2 Q3W). This trial will test the new combination using the same dosing interval and schedule, with the inclusion of an induction immunotherapy Rationale for Endpoints

4.1.6.4 Efficacy Endpoint

pCR is considered a surrogate measure of long-term survival in patients with TNBC, and is considered by the FDA as a viable surrogate for event free survival following the FDA

guidance on "Pathological Complete Response in Neoadjuvant Treatment of High-Risk Early-Stage Breast Cancer". Patients who achieved pCR have demonstrated sustained clinical benefit regardless of breast cancer subtypes. Recently, a large pooled analysis demonstrated strong association of pCR, when defined as no tumor in both breast and lymph nodes (ypT0 ypN0 or ypT0/is yp N0) following neoadjuvant therapy for breast cancer, with improved long-term benefit as measured by event-free survival and overall survival.[7].

Following these data, the study will use pCR to preliminary evaluate whether various combination therapies are meritorious for further study in a larger trial.

4.1.6.5 Safety Endpoints

Safety parameters such as incidence of AE/SAEs (including fatal SAEs), immune-related AEs (irAEs) and laboratory abnormalities, rates of dose interruption and discontinuation due to AEs, and ECI are important endpoints for safety and tolerability evaluations.

4.1.6.6 Exploratory Biomarker Research

Cancer immunotherapies represent an important and novel class of anti-tumor agents. However, the mechanism of action of these exciting new therapies is not completely understood and much remains to be learned regarding how best to leverage these new drugs in treating patients. Thus, to aid future patients, it is important to investigate the determinants of response or resistance to cancer immunotherapy as well as determinants of adverse events in the course of our clinical trials. These efforts will identify novel predictive/pharmacodynamic biomarkers and generate information that will better guide single-agent and combination therapy with immuno-oncology drugs. To identify novel biomarkers, we will collect biospecimens (blood components, tumor material, etc.) to support analyses of cellular components (e.g., protein, deoxyribonucleic acid [DNA], RNA, metabolites) and other circulating molecules. Investigations may include but are not limited to:

Germline (blood) Genetic Analyses (e.g., SNP Analyses, Whole Exome Sequencing, Whole Genome Sequencing)

This research will evaluate whether genetic variation within a clinical trial population correlates with response to the treatment(s) under evaluation. If genetic variation is found to predict efficacy or adverse events, the data might inform optimal use of therapies in the patient population. Furthermore, it is important to evaluate germline DNA variation across the genome in order to interpret tumor-specific DNA mutations. Finally, microsatellite instability (MSI) may be evaluated as this is an important biomarker for some cancers (i.e., colorectal cancer).

Genetic (DNA) Analyses from Tumor

The application of new technologies, such as next generation sequencing, has provided scientists the opportunity to identify tumor-specific DNA changes (i.e., mutations, methylation status, microsatellite instability). Key molecular changes of interest to immune-oncology drug development include the mutational burden of tumors and the clonality of T-cells in the tumor microenvironment. Increased mutational burden (sometimes referred to as a "hyper-mutated" state) may generate neo-antigen presentation in the tumor microenvironment. To conduct this type of research, it is important to identify tumor-specific mutations that occur across all genes in the tumor genome. Thus, genome wide approaches may be used for this effort. Note that

in order to understand tumor-specific mutations; it is necessary to compare the tumor genome with the germline genome. Microsatellite instability (MSI) may also be evaluated as this is an important biomarker for some cancers (i.e., colorectal cancer).

Tumor and Blood RNA Analyses

Both genome-wide and targeted messenger RNA (mRNA) expression profiling and sequencing in tumor tissue and in blood may be performed to define gene signatures that correlate to clinical response to treatment with pembrolizumab or other immunotherapies. Pembrolizumab induces a response in tumors that likely reflects an inflamed/ immune phenotype. Specific immune-related gene sets (such as those capturing interferon-gamma transcriptional pathways) may be evaluated and new signatures may be identified. Individual genes related to the immune system may also be evaluated (e.g., IL-10). MicroRNA profiling may also be pursued.

Proteomics and Immunohistochemistry using Blood or Tumor

Tumor and blood samples from this study may undergo proteomic analyses (e.g., PD-L1 immunohistochemistry [IHC]). PD-L1 protein level in tumor sections, assessed by IHC, has been shown to correlate with response to pembrolizumab in patients with NSCLC, and an IVD device has been developed for use with pembrolizumab in NSCLC. Preliminary data indicate that this association may also be true in additional cancer types (i.e., TNBC, H&N, and gastric). Additional tumor or blood-derived proteins may also correlate with response to pembrolizumab. Therefore, tumor tissue may be subjected to proteomic analyses using a variety of platforms that could include but are not limited to immunoassays, liquid chromatography/mass spectrometry. This approach could identify novel protein biomarkers that could aid in patient selection for pembrolizumab (MK-3475) therapy.

Anti-drug Antibodies (ADA)

A portion of scheduled research sera obtained before and during therapy from patients in the IRX treatment arm will be stored according to manufacturer's instructions in case it becomes necessary to perform immunogenicity testing for anti-drug antibodies (ADA).

Other Blood Derived Biomarkers

In addition to expression on the tumor tissue, PD-L1 and other tumor derived proteins can be shed from tumor and released into the blood. Assays such as enzyme-linked immunoassay measure such proteins in serum. Correlation of expression with response to pembrolizumab therapy may identify new approaches for predictive biomarkers in blood, representing a major advance from today's reliance on assessing tumor biomarkers. This research would serve to develop such assays for future clinical use.

Translational Research

The tumor microenvironments before treatment and after the combinations will be characterized, and this may include the presence and changes of TILs, immune-related mRNA expression signatures, and PD-L1 expression. In addition, tumor genetic profiling such as genetic testing for mutational burden based on tumor samples collected at Screening will be performed. Additional translational research may include T cell clonality, neoantigen expression, presence and changes in circulating tumor markers such as circulating tumor DNA (ctDNA), and serum microRNA (miRNA) and protein changes at Screening and following

treatment. Correlation of clinical response (pCR and ORR) to tumor/ circulating markers at Screening and after treatments may be evaluated.

4.1.6.7 Future Biomedical Research

The investigator will conduct Future Biomedical Research on specimens consented for future biomedical research during this clinical trial. This research may include genetic analyses (DNA), gene expression profiling (RNA), proteomics, metabolomics (serum, plasma) and/or the measurement of other analytes, depending on which specimens are consented for future biomedical research.

4.2 Benefit/Risk

Relative to standard-of-care, subjects in clinical trials generally cannot expect to receive direct benefit from treatment during participation, as clinical trials are designed to provide information about the safety and effectiveness of an investigational medicine. Additional details regarding specific benefits and risks for subjects participating in this clinical trial may be found in the accompanying IB and Informed Consent documents.

5.0 METHODOLOGY

5.1 Entry Criteria

5.1.1 Subject Inclusion Criteria

In order to be eligible for participation in this trial, the subject must:

1. Be willing and able to provide written informed consent for the trial. The subject may also provide consent for Future Biomedical Research. However, the subject may participate in the main trial without participating in Future Biomedical Research.
2. Be a male or female subject ≥18 years of age on day of signing informed consent.
3. Have locally confirmed TNBC, as defined by the most recent ASCO/CAP guidelines.
4. Have previously untreated locally advanced non-metastatic (M0) TNBC defined as the following combined primary tumor (T) and regional lymph node (N) staging per the current AJCC Version 8 staging criteria for breast cancer staging criteria as assessed by the investigator based on radiological and/or clinical assessment:
    - T1c, N1-N2
    - T2, N0-N2
    - T3, N0-N2
    - T4a-d, N0-N2

    Note: bilateral tumors (i.e., synchronous cancers in both breasts) and/or multi-focal (ie, 2, separate lesions in the same quadrant)/multi-centric (ie, 2 separate lesions in different quadrants) tumors are allowed, as well as inflammatory breast cancer, and the tumor with the most advanced T stage should be used to assess the eligibility.

5. Provide a core needle biopsy consisting of at least 1 separate tumor-bearing cores from the primary tumor at screening for translational research (archival is acceptable if sufficient tumor is available; slides are acceptable if at least 15 are available). Also agree to on study biopsy post induction therapy and prior to initiating combination pembrolizumab and paclitaxel.
6. Have Eastern Cooperative Oncology Group (ECOG) performance status of 0 or 1 assessed within 14 days of treatment initiation.
7. Demonstrate adequate organ function as defined in Table 5.1.1-1. All screening labs should be performed within 14 days of treatment initiation.

Table 5.1.1-1 Adequate Organ Function Laboratory Values

Organ System	Laboratory Value
Hematological	
Absolute neutrophil count (ANC)	≥1,500 cells/μL without granulocyte colony-stimulating factor (G-CSF) support within 2 weeks prior to the first dose of study treatment
Platelet count	≥100,000/μL without transfusion within 2 weeks prior to the first dose of study treatment
Hemoglobin	≥9 g/dL or ≥5.6 mmol/L without transfusion or EPO dependency
Renal	
Serum creatinine **OR** Calculated creatinine clearance (CrCl) (calculated per institutional standard)	≤1.5 X upper limit of normal (ULN) **OR** ≥50 mL/min
Hepatic	
Total bilirubin	≤1.5 X ULN **OR** Direct bilirubin ≤ULN for subjects with total bilirubin levels >1.5 X ULN
Aspartate aminotransferase [AST (SGOT)] and alanine aminotransferase [ALT (SGPT)]	≤2.5 X ULN
Albumin	≥3.0 g/dL
Lactate dehydrogenase (LDH)	<2.5 X ULN
Coagulation	
International Normalized Ratio (INR) or prothrombin time (PT)	≤1.5 X ULN unless subject is receiving anticoagulant therapy as long as PT or aPTT/PTT is within therapeutic range of intended use of anticoagulants
Activated partial thromboplastin time (aPTT) or partial thromboplastin time (PTT)	≤1.5 X ULN unless subject is receiving anticoagulant therapy as long as PT or aPTT/PTT is within therapeutic range of intended use of anticoagulants

8. Have left ventricular ejection fraction (LVEF) of ≥50% or ≥ institution lower limit of normal (LLN) as assessed by echocardiogram (ECHO) or multigated acquisition (MUGA) scan performed at screening.

9. Males and female subjects of childbearing potential (Section 5.7.2 – Contraception) must be willing to use an adequate method of contraception as outlined in Section 5.7.2 – Contraception, for the course of the study through 12 months after the last dose of study medication for subjects who have received cyclophosphamide, and 6 months after the last dose of study medication for subjects who did not.

   Note: Abstinence is acceptable if this is the usual lifestyle and preferred contraception for the subject.

10. Female subject of childbearing potential must have a negative urine or serum pregnancy test within 72 hours prior to receiving the first dose of study medication. If the urine test is positive or borderline a serum pregnancy test will be required.

5.1.2 Subject Exclusion Criteria

The subject must be excluded from participating in the trial if the subject:

1. Has a history of invasive malignancy ≤5 years prior to signing informed consent except for adequately treated basal cell or squamous cell skin cancer or in situ cervical cancer.
2. Has received prior chemotherapy, targeted therapy, and radiation therapy within the past 12 months.
3. Has received prior therapy with an anti-PD-1, anti-PD-L1, or anti-PD-L2 agent or with an agent directed to another co-inhibitory T-cell receptor (e.g., CTLA-4, OX-40, CD137) or has previously participated in MK-3475 clinical trials.
4. Is currently participating in or has participated in an interventional clinical trial with an investigational compound or device within 4 weeks of the first dose of treatment in this current trial.

   Note: subject should be excluded if he/she received an investigational agent with anti-cancer or anti-proliferative intent within the last 12 months.

5. Has received a live vaccine within 30 days of the first dose of study treatment.

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

6. Has an active autoimmune disease that has required systemic treatment in past 2 years (i.e., with use of disease modifying agents, corticosteroids or immunosuppressive drugs). Replacement therapy (e.g., thyroxine, insulin, or physiologic corticosteroid replacement therapy for adrenal or pituitary insufficiency) is not considered a form of systemic treatment.
7. Has a diagnosis of immunodeficiency or is receiving systemic steroid therapy or any other form of immunosuppressive therapy within 7 days prior to the first dose of trial treatment.
8. Has a known history of Human Immunodeficiency Virus (HIV) (HIV 1/2 antibodies).
9. Has known active Hepatitis B (e.g., HBsAg reactive) or Hepatitis C (e.g., HCV RNA [qualitative] is detected).

10. Has a history of (non-infectious) pneumonitis that required steroids or current pneumonitis.
11. Has an active infection requiring systemic therapy.
12. Has significant cardiovascular disease, such as:
    - History of myocardial infarction, acute coronary syndrome or coronary angioplasty/stenting/bypass grafting within the last 6 months
    - Congestive heart failure (CHF) New York Heart Association (NYHA) Class II-IV or history of CHF NYHA class III or IV
13. Has a history or current evidence of any condition, therapy, lab abnormality or other circumstance that might expose the subject to risk by participating in the trial, confound the results of the trial, or interfere with the subject's participation for the full duration of the trial in the opinion of the Investigator.
14. Has known psychiatric or substance abuse disorders that would interfere with cooperation with the requirements of the trial in the opinion of the Investigator.
15. Is pregnant or breastfeeding, or expecting to conceive children within the projected duration of the trial, starting with the screening visit through 12 months after the last dose of trial treatment for subjects who have received cyclophosphamide, and for 6 months after the last dose of study medication for subjects who have not.
16. Has a known hypersensitivity to the components of the study therapy or its analogs.
17. Has a known history of active TB (Bacillus Tuberculosis).
18. Allergy to ciprofloxacin (or other quinolones).
19. History of allogeneic stem cell and/or solid organ transplant.

5.2 Trial Treatment(s)

The treatments to be used in this trial are outlined in the below tables:

Table 5.2-1: Trial treatments: Control Arm

Drug	Dose/ Potency	Dose Frequency	Route of Administration	Dosing Time of each 3-week cycle	Use
Pembrolizumab (MK-3475)	200 mg	Q3W	IV infusion	Day 1 in the Induction Phase; Day 1 of Cycles in the Neoadjuvant Phases (8 cycles)	Experimental
Paclitaxel	80 mg/m^2	Weekly	IV Infusion	Days 1, 8, 15 of Cycles 1-4 of Treatment 1	Standard of care
Doxorubicin	60 mg/m^2	Q3W	IV Injection	Day 1 of Cycles 1-4 of Treatment 2	Standard of care

Drug	Dose/ Potency	Dose Frequency	Route of Administration	Dosing Time of each 3-week cycle	Use
Cyclophosphamide	600 mg/m^2	Q3W	IV Infusion	Day 1 of Cycles 1-4 of Treatment 2	Standard of care
Abbreviations: IV = intravenous; Q3W = every 3 weeks					

Nab-paclitaxel may be substituted for paclitaxel due to medical necessity (e.g. hypersensitivity reaction) per local standard practice. If substituted, administer nab-paclitaxel at 125 mg/m2 IV over 30 minutes (±10 minutes) weekly for total of 12 weeks including paclitaxel doses that were previously administered before substitution). Premedication is not generally necessary prior to nab-paclitaxel, although may be administered per standard practice or in patients with prior mild to moderate hypersensitivity reactions.

Table 5.2-2 Trial Treatments: Arm A

Drug	Dose/ Potency	Dose Frequency	Route of Administration	Dosing Time of each 3-week cycle	Use
Pembrolizumab (MK-3475)	200 mg	Q3W	IV infusion	Day 1 in the Induction Phase; Day 1 of Cycles in the Neoadjuvant Phases (8 cycles)	Experimental
IRX-2	2mL (230 units of IL-2)	Daily x 10 days	Two 1mL subcutaneous periareolar injections	Any 10 days over days 4-21 of the induction Phase, and daily for 10 days over any 14 day period following completion of Treatment 1 and to be completed prior to advancement to Treatment 2.	Experimental
Paclitaxel	80 mg/m^2	Weekly	IV Infusion	Days 1, 8, 15 of Cycles 1-4 of Treatment 1	Standard of care
Doxorubicin	60 mg/m^2	Q3W	IV Injection	Day 1 of Cycles 1-4 of Treatment 2	Standard of care
Cyclophosphamide	300mg/m^2 600 mg/m^2	Once Q3W	IV Infusion IV Infusion	Day 1 (Induction Phase) Day 1 of Cycles 1-4 of Treatment 2	Experimental Standard of care
Abbreviations: IV = intravenous; Q3W = every 3 weeks					

Nab-paclitaxel may be substituted for paclitaxel due to medical necessity (e.g. hypersensitivity reaction) per local standard practice. If substituted, administer nab-paclitaxel at 125 mg/m2 IV over 30 minutes (±10 minutes) weekly for total of 12 weeks including paclitaxel doses that were previously administered before substitution). Premedication is not generally necessary prior to nab-paclitaxel, although may be administered per standard practice or in patients with prior mild to moderate hypersensitivity reactions.

Trial Treatment should begin within 3 business days of randomization.

Pembrolizumab will be provided by Merck. IRX-2 will be provided by Brooklyn Immunotherapeutics, or locally by the trial site.

Pembrolizumab Storage

Pembrolizumab (MK-3475) Powder for Solution for Infusion vials should be stored at refrigerated conditions 2 – 8 °C (36 - 46 ºF). Prior to reconstitution, the vial of lyophilized powder can be out of refrigeration (temperatures at or below 25°C (77°F)) for up to 24 hours.

Pembrolizumab (MK-3475) solutions may be stored at room temperature for a cumulative time of up to 6 hours. The 6 hour countdown begins when the vial is pierced, and includes room temperature storage of reconstituted drug product solution in vials, room temperature storage of admixture solutions in the IV bags and the duration of infusion. (Please note this 6 hour timeframe is to provide a microbial control strategy. The microbial clock only starts when the product stopper is pierced and not when the vial is removed from the refrigerator.)

In addition, reconstituted vials and/or IV bags may be stored under refrigeration at 2 °C to 8 °C (36 °F to 46 °F), total cumulative storage time at room temperature and refrigeration should not exceed 24 hours.

If refrigerated, allow the vials and/or IV bags to come to room temperature prior to use.

IRX-2 Storage

Each subcutaneous injection of IRX-2 comes in 1.0 ml frozen vials from the manufacturer. IRX-2 must be stored frozen at a range of −15◦C to –50◦ C until the date on the expiry label. Vials should be thawed at room temperature prior to administration. In some cases, it may take as long as 60 minutes for small precipitates in the vial to go into solution; this is acceptable. Please refer to the Investigator's Brochure for detailed information on IRX-2 preparation and administration.

The vials MAY NOT be returned to the freezer or refrigerator once placed at room temperature. The prepared drug for injection must be used within 24 hours of removing the vial from frozen storage.

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

5.2.1 Dose Modification for Pembrolizumab

AEs associated with pembrolizumab exposure may represent an immunologic etiology. These immune-related AEs (irAEs) may occur shortly after the first dose or several months after the last dose of pembrolizumab treatment and may affect more than one body system simultaneously. Therefore, early recognition and initiation of treatment is critical to reduce complications. Based on existing clinical trial data, most irAEs were reversible and could be managed with interruptions of pembrolizumab, administration of corticosteroids and/or other supportive care. For suspected irAEs, ensure adequate evaluation to confirm etiology or exclude other causes. Additional procedures or tests such as bronchoscopy, endoscopy, skin biopsy may be included as part of the evaluation. Based on the severity of irAEs, withhold or permanently discontinue pembrolizumab and administer corticosteroids. Dose modification and toxicity management guidelines for irAEs associated with pembrolizumab are provided in the below tables.

Table 5.2.1-1 Dose Modification and Toxicity Management Guidelines for Immune-related AEs Associated with Pembrolizumab

General instructions:

1. Corticosteroid taper should be initiated upon AE improving to Grade 1 or less and continue to taper over at least 4 weeks.
2. For situations where pembrolizumab has been withheld, pembrolizumab can be resumed after AE has been reduced to Grade 1 or 0 and corticosteroid has been tapered. Pembrolizumab should be permanently discontinued if AE does not resolve within 12 weeks of last dose or corticosteroids cannot be reduced to ≤10 mg prednisone or equivalent per day within 12 weeks.
3. For severe and life-threatening irAEs, IV corticosteroid should be initiated first followed by oral steroid. Other immunosuppressive treatment should be initiated if irAEs cannot be controlled by corticosteroids.

Immune-related AEs	**Toxicity grade or conditions (CTCAEv5.0)**	**Action taken to pembrolizumab**	**irAE management with corticosteroid and/or other therapies**	**Monitor and follow-up**
Pneumonitis	Grade 2	Withhold	• Administer corticosteroids (initial dose of 1-2 mg/kg prednisone or equivalent) followed by taper	• Monitor participants for signs and symptoms of pneumonitis • Evaluate participants with suspected pneumonitis with radiographic imaging and initiate corticosteroid treatment • Add prophylactic antibiotics for opportunistic infections
	Grade 3 or 4, or recurrent Grade 2	Permanently discontinue		
Diarrhea / Colitis	Grade 2 or 3	Withhold	• Administer corticosteroids (initial dose of 1-2 mg/kg prednisone or equivalent) followed by taper	• Monitor participants for signs and symptoms of enterocolitis (ie, diarrhea, abdominal pain, blood or mucus in stool with or without fever) and of bowel perforation (ie, peritoneal signs and ileus).

	Grade 4	Permanently discontinue		• Participants with ≥ Grade 2 diarrhea suspecting colitis should consider GI consultation and performing endoscopy to rule out colitis. • Participants with diarrhea/colitis should be advised to drink liberal quantities of clear fluids. If sufficient oral fluid intake is not feasible, fluid and electrolytes should be substituted via IV infusion.
AST / ALT elevation or Increased bilirubin	Grade 2	Withhold	• Administer corticosteroids (initial dose of 0.5- 1 mg/kg prednisone or equivalent) followed by taper	• Monitor with liver function tests (consider weekly or more frequently until liver enzyme value returned to baseline or is stable
	Grade 3 or 4	Permanently discontinue	• Administer corticosteroids (initial dose of 1-2 mg/kg prednisone or equivalent) followed by taper	
Type 1 diabetes mellitus (T1DM) or Hyperglycemia	Newly onset T1DM or Grade 3 or 4 hyperglycemia associated with evidence of β-cell failure	Withhold	• Initiate insulin replacement therapy for participants with T1DM • Administer anti-hyperglycemic in participants with hyperglycemia	• Monitor participants for hyperglycemia or other signs and symptoms of diabetes.
Hypophysitis	Grade 2	Withhold	• Administer corticosteroids and initiate hormonal replacements as clinically indicated.	• Monitor for signs and symptoms of hypophysitis (including hypopituitarism and adrenal insufficiency)
	Grade 3 or 4	Withhold or permanently discontinue[1]		
Hyperthyroidism	Grade 2	Continue		

	Grade 3 or 4	Withhold or permanently discontinue[1]	• Treat with non-selective beta-blockers (eg, propranolol) or thionamides as appropriate	• Monitor for signs and symptoms of thyroid disorders.
Hypothyroidism	Grade 2-4	Continue	• Initiate thyroid replacement hormones (eg, levothyroxine or liothyroinine) per standard of care	• Monitor for signs and symptoms of thyroid disorders.
Nephritis and Renal dysfunction	Grade 2	Withhold	• Administer corticosteroids (prednisone 1-2 mg/kg or equivalent) followed by taper.	• Monitor changes of renal function
	Grade 3 or 4	Permanently discontinue		
Myocarditis	Grade 1 or 2	Withhold	• Based on severity of AE administer corticosteroids	• Ensure adequate evaluation to confirm etiology and/or exclude other causes
	Grade 3 or 4	Permanently discontinue		
All other immune-related AEs	Intolerable/ persistent Grade 2	Withhold	• Based on type and severity of AE administer corticosteroids	• Ensure adequate evaluation to confirm etiology and/or exclude other causes
	Grade 3	Withhold or discontinue based on the type of event. Events that require discontinuation include and not limited to: Gullain-Barre Syndrome, encephalitis		
	Grade 4 or recurrent Grade 3	Permanently discontinue		

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

NOTE:

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

Dose Modification and Toxicity Management of Infusion Reactions Related to Pembrolizumab

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

Table 5.2.1-2 Pembrolizumab Infusion Reaction Dose modification and Treatment Guidelines

NCI CTCAE v5.0 Grade	Treatment	Premedication at Subsequent Dosing
Grade 1 Mild reaction; infusion interruption not indicated; intervention not indicated	Increase monitoring of vital signs as medically indicated until the participant is deemed medically stable in the opinion of the investigator.	None
Grade 2 Requires therapy or infusion interruption but responds promptly to symptomatic treatment (e.g., antihistamines, NSAIDs, narcotics, IV fluids); prophylactic medications indicated for ≤24 hrs	**Stop Infusion.** Additional appropriate medical therapy may include but is not limited to: IV fluids Antihistamines NSAIDs Acetaminophen Narcotics Increase monitoring of vital signs as medically indicated until the participant is deemed medically stable in the opinion of the investigator. If symptoms resolve within 1 hour of stopping drug infusion, the infusion may be restarted at 50% of the original infusion rate (e.g. from 100 mL/hr to 50 mL/hr). Otherwise dosing will be held until symptoms resolve and the participant should be premedicated for the next scheduled dose. **Participants who develop Grade 2 toxicity despite adequate premedication should be permanently discontinued from further study drug treatment**	Participant may be premedicated 1.5h (± 30 minutes) prior to infusion of pembrolizumab with: Diphenhydramine 50 mg po (or equivalent dose of antihistamine). Acetaminophen 500-1000 mg po (or equivalent dose of analgesic).

Grades 3 or 4 Grade 3: Prolonged (i.e., not rapidly responsive to symptomatic medication and/or brief interruption of infusion); recurrence of symptoms following initial improvement; hospitalization indicated for other clinical sequelae (e.g., renal impairment, pulmonary infiltrates) Grade 4: Life-threatening; pressor or ventilatory support indicated	**Stop Infusion.** Additional appropriate medical therapy may include but is not limited to: Epinephrine** IV fluids Antihistamines NSAIDs Acetaminophen Narcotics Oxygen Pressors Corticosteroids Increase monitoring of vital signs as medically indicated until the participant is deemed medically stable in the opinion of the investigator. Hospitalization may be indicated. **In cases of anaphylaxis, epinephrine should be used immediately. **Participant is permanently discontinued from further study drug treatment.**	No subsequent dosing

Appropriate resuscitation equipment should be available at the bedside and a physician readily available during the period of drug administration.

For further information, please refer to the Common Terminology Criteria for Adverse Events v5.0 (CTCAE) at http://ctep.cancer.gov

Other Allowed Dose Interruptions for Pembrolizumab

Pembrolizumab may be interrupted for situations other than treatment-related AEs such as medical/surgical events or logistical reasons not related to study therapy. Subjects should be placed back on study therapy within 6 weeks of the scheduled interruption, unless otherwise discussed with the Sponsor. The reason for interruption should be documented in the subject's study record.

5.2.2 Dose Modifications for IRX-2

Based upon the relative safety of IRX-2 in the phase Ib pilot study in breast cancer, as well as in a number of preceding phase I/II clinical trials in head/neck cancer (as well as in a number of ongoing trials in other indications), dose reductions or discontinuations are not expected to be necessary.

Because dose reductions have not been previously studied, no dose reductions of IRX-2 will be allowed on this protocol.

IRX-2 doses may be delayed or discontinued at any time if unanticipated and intolerable toxicities emerge. Subjects requiring a delay of more than 7 days due to toxicity should discontinue IRX-2 permanently and advance to biopsy and the neoadjuvant phase of treatment, and should not receive re-induction IRX-2. Subjects who require IRX-2 discontinuation for any reason should advance to biopsy and the neoadjuvant phase of treatment, and should not receive re-induction IRX-2.

5.2.3 Dose Modifications for Chemotherapy Agents

Suggested dose modifications and guidelines for growth factor support for paclitaxel are detailed in table 5.2.3-1 and for doxorubicin and cyclophosphamide table 5.2.3-2

Of note, primary prophylaxis with growth factor support is allowed with chemotherapy. Local guidelines related to treatment holds and dose reduction for chemotherapy should be followed, if different than the recommendations outlined in table 5.2.3-1 and -2.

Table 5.2.3-1: Dose Modification Guideline for Paclitaxel

Toxicities	Grade or actual value	Paclitaxel
Hematological		
Neutropenia	≥1000/mm^3 Grade 2/Grade 1	No change to paclitaxel • For ANC ≤1500/mm^3, prophylactic myeloid growth factors (filgrastim) may be used at the discretion of the investigator, o Should not be given on the same day as chemotherapy. o Pegfilgrastim may not be used with paclitaxel due to its weekly dosing schedule
	<1000/mm^3 Grade 3/Grade 4	Hold paclitaxel until ANC ≥1000/mm^3. Prophylactic G-CSF may be used between days 2–6 at discretion of the investigator. Pegfilgrastim may not be used with paclitaxel due to its weekly dosing schedule. Resume paclitaxel based on timing of recovery: • ≤1 week: No change to paclitaxel • >1 but <3 weeks: Dose-reduce paclitaxel to 70 mg/m^2 for all subsequent cycles. • ≥3 weeks: Stop paclitaxel (see general instruction below)
Febrile neutropenia	ANC ≤1000/mm^3, fever ≥38.5°C Grade 3 and Grade 4	Hold paclitaxel until resolved (ANC >1000/mm^3, fever <38.5°C, and resolution of any signs of infection). Prophylactic G-CSF may be used between days 2–6 at discretion of the investigator. Pegfilgrastim may not be used with paclitaxel due to its weekly dosing schedule. Resume paclitaxel according to number of episodes: • First episode: no change to paclitaxel. Consider adding prophylactic GCSF for subsequent cycles. • Second episode: Reduce paclitaxel to 70 mg/ for all subsequent doses. • Third episode: Discontinue paclitaxel (see general instruction below)
Thrombocytopenia	75-<100,000/mm^3 Grade 1	Hold paclitaxel until ≥100,000/mm^3, resume treatment based on timing of recovery: • ≤1 week — no change to paclitaxel. • >1 but <3 weeks — Reduce paclitaxel to 70 mg/m^2 for all subsequent doses. • ≥3 weeks: Discontinue paclitaxel (see general instruction below)
	<75,000/mm^3 ≥Grade 2	Hold paclitaxel until ≥100,000/mm^3. • Reduce paclitaxel to 70 mg/m^2 for all subsequent doses. • Stop paclitaxel if held for ≥3 weeks in a row, (see general instruction below)
Anemia	All grades	No change to paclitaxel • Iron studies should be done and iron should be replaced as indicated. • Red blood cell transfusions can be given at the investigator's discretion.
Nausea/Vomiting	Grade 1 or 2	No change to paclitaxel
	≥Grade 3	Hold paclitaxel until resolved to ≤Grade 1. • Resume paclitaxel at previous dose with modification of premedication

		• Second episode ≥Grade 3 despite with maximum supportive care, reduce paclitaxel to 70 mg/m^2 for all subsequent cycles
Mucositis/Stomatitis	Grade 1 or 2	No change to paclitaxel
	≥Grade 3	Hold paclitaxel until resolved to ≤Grade 1. • Resume paclitaxel at previous dose with modification of premedication • Second episode ≥Grade 3 despite with maximum supportive care, reduce paclitaxel to 70 mg/m^2 for all subsequent cycles
Neurotoxicity	Grade 1–2	No change to paclitaxel
	Grade 3	Hold paclitaxel until neuropathy improves to ≤Grade 2. • Resume paclitaxel dose reduced to 70 mg/m^2 for all subsequent doses. • Discontinue paclitaxel if held for ≥3 weeks in a row, (see general instruction below)
	Grade 4	• Discontinue paclitaxel if held for ≥3 weeks in a row, (see general instruction below)
Hepatic	Grade 1	• No change to paclitaxel
	≥Grade 2 or 3	• Bilirubin fractionation should be performed if total bilirubin >1.5xULN. Dose may continue if isolated bilirubinemia is mostly indirect such as in subject with Gilbert • Hold paclitaxel until resolve to Grade 1 and resume the dose at previous level • Discontinue paclitaxel if held for ≥3 weeks in a row, (see general instruction below)
	Grade 4	• Discontinue paclitaxel (see general instruction below) Note all concurrent ALT/AST >3×ULN and Total bilirubin >2×ULN should be discontinued and evaluated for potential Hy's law
Anaphylaxis /hypersensitivity	Mild	• Complete paclitaxel infusion, observe until symptom resolved
	Moderate	• Stop infusion and treat per standard practice • Resume infusion at half of the infusion speed if symptom resolve • Stop if symptom recurs
	Severe	• Stop infusion immediately and discontinue treatment (see general instruction below)
Other significant toxicities excluding fatigue, alopecia and leukopenia at discretion of the investigators	Grade 2	• Hold paclitaxel until resolve to ≤Grade 1 • Resume at the previous dose and increase supportive care measure, if available
	≥Grade 3	• Hold paclitaxel, and discuss with sponsor medical monitor for further instructions • If ≥Grade 3 toxicity recurs upon rechallenge, discontinue treatment permanently

Table 5.2.3-2: Dose Modification Guideline for Doxorubicin and Cyclophosphamide (AC)

Toxicities	Grade or actual value	Doxorubicin and cyclophosphamide
Hematological		
Neutropenia	≥1000/mm^3 (Grade 2/Grade 1)	No change to AC • For ANC ≤1500/mm^3, prophylactic myeloid growth factors (filgrastim or pegfilgrastim) may be used at the discretion of the investigator.
	<1000/mm^3 Grade 3/Grade 4	Hold AC until ANC ≥1000/mm^3. • Prophylactic myeloid growth factors (filgrastim or pegfilgrastim) may be used at the discretion of the investigator Resume AC based on timing of recovery: • <1 week: No change to AC • ≥1 but <3 weeks: Reduce AC by 20% for all subsequent cycles. • ≥3 weeks: discontinue AC (see general instruction below)
Febrile neutropenia	ANC ≤1000/mm^3, fever ≥38.5°C Grade 3 and Grade 4	Hold AC until resolved (ANC >1000/mm^3, fever <38.5°C, and resolution of any signs of infection • Prophylactic myeloid growth factors (filgrastim or pegfilgrastim) may be used at the discretion of the investigator. Resume AC according to number of episodes: • First episode: no change to AC. • Second episode: Reduce AC by 20% for all subsequent cycles • Third episode: Discontinue AC (see general instruction below)
Thrombocytopenia	75–<100,000/mm^3 Grade 1	Hold AC until ≥100,000/mm^3, resume AC based on timing of recovery: • ≤1 week — no change to AC. • >1 but <3 weeks — Reduce AC by 20% for all subsequent cycles • ≥3 weeks: Discontinue AC (see general instruction below)
	<75,000/mm^3 ≥Grade 2	Hold AC until ≥100,000/mm^3. • Reduce AC or EC by 20% for all subsequent cycles • Discontinue AC if held for ≥3 weeks in a row, (see general instruction below)
Anemia	All grades	No change to AC • Iron studies should be done and iron should be replaced as indicated. • Red blood cell transfusions can be given at the investigator's discretion.
Nausea/Vomiting	Grade 1 or 2	No change to AC
	≥Grade 3	Hold AC until resolved to ≤Grade 1. • Resume AC at previous dose with modification of premedication • Second episode ≥Grade 3 despite with maximum supportive care, reduce AC by 20% for all subsequent cycles
Mucositis/Stomatitis	Grade 1 or 2	No change to AC
	≥Grade 3	Hold AC until resolved to ≤Grade 1.

		• Resume AC at previous dose with modification of premedication • Second episode ≥Grade 3 despite with maximum supportive care, reduce AC by 20% for all subsequent cycles
Hepatic	Grade 1	• No change to AC
	≥Grade 2 or 3	• Hold AC until resolve to Grade 1 and resume the dose at previous level • Discontinue AC if held for ≥3 weeks in a row, (see general instruction below)
	Grade 4	• Discontinue AC (see general instruction below) Note all concurrent ALT/AST >3×ULN and Total bilirubin >2×ULN should be discontinued and evaluated for potential Hy's law
Cardiac toxicity	Grade 1 or 2	• No change to AC
	≥Grade 3	• Discontinue doxorubicin (see general instruction below)
Anaphylaxis/ hypersensitivity	Mild	• Complete AC infusion, observe until symptom resolved
	Moderate	• Stop infusion and treat per standard practice • Resume infusion at half of the infusion speed if symptom resolve • Stop if symptom recurs
	Severe	• Stop infusion immediately and discontinue treatment (see general instruction below)
Other significant toxicities excluding fatigue, alopecia and leukopenia at discretion of the investigators	Grade 2	• Hold AC until resolve to ≤Grade 1 • Resume at the previous dose and increase supportive care measure, if available
	≥Grade 3	• Hold AC and discuss with sponsor medical monitor for further instructions • If ≥Grade 3 toxicity recurs upon rechallenge, discontinue treatment permanently

Instruction on Discontinuation of a Component or Entire Regimen

During the first part of the combination therapy, if 1 or more than 1 component must be discontinued or delayed due to toxicity, the investigator can select 1 of the following options at his/her own discretion for the subject (table 5.2.3-1 and -2).

- If paclitaxel is discontinued due to toxicity related to paclitaxel,
    - Option 1 Stop the first part of the neoadjuvant chemotherapy (pembrolizumab + paclitaxel); start and complete the AC regimen as planned per protocol, then followed by surgery..
    - Option 2 Nab-paclitaxel may be substituted for paclitaxel due to medical necessity (e.g. hypersensitivity reaction) per local standard practice. If substituted, administer nab-paclitaxel at 125 mg/m2 IV over 30 minutes (±10 minutes) weekly for total of 12 weeks including paclitaxel doses that were previously administered before substitution). Premedication is not generally necessary prior to nab-paclitaxel, although may be administered per standard practice or in patients with prior mild to moderate hypersensitivity reactions.

  - Option 3 In cases nab-paclitaxel may not be immediately feasible (insurance auth), option 1 may be followed, with subsequent completion of taxane after receipt of AC.
  - If taxane doses are held or skipped, if feasible the full course of taxane therapy should be completed thereafter.
  - In arm A, IRX2 reinduction is intended to be administered following treatment 1 and preceding treatment 2. However, in the above instances, re-induction may be held/omitted or delayed at the discretion of the PI

- If only pembrolizumab is discontinued due to pembrolizumab toxicity,
  - Continue with paclitaxel regimen alone as planned per protocol, then start and complete the AC regimen as planned per protocol, then followed by surgery.
  - IRX-2 re-induction may be administered if thought to be safe after review of scenario with principal investigator
- If only IRX-2 is discontinued due to IRX-2 toxicity
  - Continue with other therapies per protocol, then followed by surgery

During the second part of the combination therapy, if one or more than one component should be discontinued, the investigator can select 1 of the following options at his/her own discretion for the subject:

- If doxorubicin and/or cyclophosphamide are discontinued due to doxorubicin and/or cyclophosphamide toxicity,
  - Discontinue all study treatment including pembrolizumab and proceed with surgery.
- If only pembrolizumab is discontinued due to pembrolizumab toxicity,
  - Continue doxorubicin and cyclophosphamide for the remaining cycles as planned per protocol, and followed by surgery.

Subjects who are discontinued from the study treatment and continue with another neoadjuvant treatment prior to the definitive surgery will be evaluable for pCR, but will be considered non-pCR. In these cases, pathology response at the time of definitive surgery will be evaluated and reported for exploratory/descriptive purposes.

Instruction on Making up Missed Doses:

If a dose delay does not require discontinuation of chemotherapy, subjects may resume treatment with the next scheduled dose in the regimen and continue on treatment to complete the full number of cycles per protocol.

5.2.4 Timing of Dose Administration

On each trial treatment dosing day, trial treatments should be administered after all procedures/ assessments have been completed as listed in the Section 6 – Trial Flow Chart.

Refer to the product label for detailed instructions on preparation and administration precautions on combination chemotherapy agents included in the trial: paclitaxel, doxorubicin, and cyclophosphamide.

5.2.4.1 Pembrolizumab

For the induction phase, pembrolizumab is administered on day 1 of therapy. For the neoadjuvant phase, pembrolizumab will be administered on Cycle 1 Day 1 (with a window of +5 days) as the first trial treatment; for Cycles 1-4 of the Neoadjuvant Treatment Phase (Treatment 1) every 3 weeks (± 2 days); and for Cycles 1-4 of the Neoadjuvant Treatment Phase (Treatment 2) every 3 weeks (+/- 3 days. Trial treatments should be administered in accordance with the schedules provided in Section 6 – Trial Flow Chart.

On Day 1 of each Cycle, a fixed dose of 200 mg pembrolizumab will be administered as a 30 minute IV infusion. Sites should make every effort to target infusion timing to be as close to 30 minutes as possible. However, given the variability of infusion pumps from site to site, a window of -5 minutes and +10 minutes is permitted (i.e., infusion time is 30 minutes: -5 min/+10 min).

When pembrolizumab is administered on the same day with chemotherapy agents, pembrolizumab should be administered prior to chemotherapy agents.

5.2.4.2 IRX-2

IRX-2 should be administered as two subcutaneous injections of 1 mL each, in the periareolar region of the affected breast. Each injection should be given subcutaneously approximately 1 cm from the outer margin of the areola. A 4-6 mm, 28-30 gauge needle is recommended; injections should consistently enter the fat just underneath the skin. The first injection should be administered on the same axis as the tumor location as identified by physical exam, mammography or ultrasound. The second injection on the same day should be rotated 90 degrees clockwise to the first injection. On the second day of IRX-2 injections, the first injection should be again administered on the same axis as the tumor location, but the second injection should be rotated 90 degrees counter-clockwise to the first injection, i.e. to maximize the likelihood of drug delivery to the lymphatics, the location of the second injection each day should be alternated between clockwise and counter-clockwise from the primary injection site. Each injection site each day should be gently massaged for one minute to promote entry into lymphatics.

5.2.4.3 Paclitaxel

Paclitaxel, at a dose level of 80 mg/m^2, will be administered on Days 1, 8, and 15 during Cycles 1-4 of the paclitaxel regimen (Treatment 1) as IV infusion as instructed per product label.

Paclitaxel should be administered after pembrolizumab. Additional premedication should be administered as per standard practice. It is preferable that dexamethasone be tapered and discontinued by cycle 2 if no infusion reactions are incurred.

5.2.4.4 Cyclophosphamide

5.2.4.4.1 Induction phase

Cyclophosphamide will be administered at a dose level of 300mg/m^2 intravenously on day 1 [arm A only], as instructed per product label. The relatively low dose of cyclophosphamide has low to moderate emetic risk per NCCN Guidelines. Use of corticosteroids as an antiemetic is neither required nor permitted because of its potential immunosuppressive effect. An antiemetic regimen of lorazepam and a serotonin (5-HT3) antagonist is recommended.

5.2.4.4.2 Neoadjuvant phase

Cyclophosphamide at a dose level of 600 mg/m^2 will be administered intravenously as instructed per product label on Day 1 of Cycles 1-4 of the AC regimen (Treatment 2) following the administration of pembrolizumab. Additional premedication should be administered as per standard practice.

5.2.4.5 Doxorubicin

Doxorubicin at a dose level of 60 mg/m^2 should be administered IV push on Day 1 of Cycles 1-4 of the AC regimen (Treatment 2) as instructed per product label following the administration of pembrolizumab. Additional premedication should be administered as per standard practice. Doxorubicin should be avoided for subjects who had previous exposure to doxorubicin of more than 200 mg/m^2.

5.3 Randomization or Treatment Allocation

Treatment allocation/randomization will occur centrally at Providence Cancer Institute, details on randomization are in statistical methods.

5.4 Stratification

Patients will not be stratified.

5.5 Concomitant Medications/Vaccinations (Allowed & Prohibited)

Medications or vaccinations specifically prohibited in the exclusion criteria are not allowed during the ongoing trial. If there is a clinical indication for any medication or vaccination specifically prohibited during the trial, discontinuation from trial therapy or vaccination may be required. The investigator should discuss any questions regarding this with the Coordinating Center Principal Investigator. The final decision on any supportive therapy or vaccination rests with the investigator and/or the subject's primary physician. However, the decision to continue the subject on trial therapy or vaccination schedule requires the mutual agreement of the treating investigator, the Coordinating Center Principal Investigator and the subject.

5.5.1 Acceptable Concomitant Medications

All treatments that the investigator considers necessary for a subject's welfare may be administered at the discretion of the investigator in keeping with the community standards of medical care. All concomitant medication will be recorded on the case report form (CRF) including all prescription, over-the-counter (OTC), herbal supplements, and IV medications and fluids. If changes occur during the trial period, documentation of drug dosage, frequency, route, and date may also be included on the CRF. Note: the use of GnRH therapy (eg, goserelin acetate [Zolodex®]) for ovarian preservation and bisphosphonates or rank ligand inhibitors to prevent osteopenia or osteoporosis is allowed during chemotherapy.

Supportive care is permitted for managing drug-related toxicities. See guidelines in Section 5.6 –Rescue Medications & Supportive Care for more details.

All prior medications received within 30 days before the screening visit, and all new concomitant medications given from the screening visit through the safety follow-up visit should be recorded. After the Adjuvant Phase safety follow-up visit, record all medications administered for the treatment of SAEs and ECIs as defined in Section 9.1 - Common Terminology Criteria for Adverse Events (CTCAE) and Section 9.2 - Definitions.

5.5.2 Prohibited Concomitant Medications

Subjects are prohibited from receiving the following therapies from the time of screening until completion of all study treatments (subsequent off study treatments may be administered prior to surgery if a biopsy is performed and there is histologic confirmation of residual disease following all treatments on this study):

- Immunotherapy not specified in this protocol
- Chemotherapy not specified in this protocol
- Investigational agents not specified in this protocol
- Radiation therapy. (Post-operative radiation therapy is acceptable according to the standard of care, as applicable.)
- Live vaccines within 30 days prior to the first dose of trial treatment and while participating in the trial. Examples of live vaccines include, but are not limited to, the following: measles, mumps, rubella, varicella, zoster, yellow fever, intranasal influenza, rabies, BCG, and typhoid vaccine.

  Note: Seasonal influenza vaccines for injection are generally inactivated flu vaccines and are allowed; however intranasal influenza vaccines (e.g., Flu-Mist®) are live attenuated vaccines, and are not allowed.
- Glucocorticoids for any purpose other than to modulate symptoms from an irAE of suspected immunologic etiology or for use as a pre-medication for chemotherapeutic agents specified in the protocol.
    - *Note: Inhaled steroids are allowed for management of asthma.*
    - *Note: Use of prophylactic corticosteroids to avoid allergic reactions (e.g., to IV contrast dye) is permitted.*

Subjects who are discontinued from the study treatment and continue with another neoadjuvant treatment prior to the definitive surgery will be considered treatment failures and will not be evaluable for pCR. Subjects may receive other medications that the investigator deems to be medically necessary. If there is a clinical indication for any medication or vaccination specifically prohibited during the trial, discontinuation from trial therapy may be required.

The Exclusion Criteria describes other prior medications prohibited during study treatment.

Site staff should refer to the local product label for permitted and prohibited medications, as well as, drug interactions for each chemotherapy agent used as trial treatment.

5.6 Rescue Medications & Supportive Care

5.6.1 Supportive Care Guidelines for Pembrolizumab

Subjects should receive appropriate supportive care measures as deemed necessary by the treating investigator. Suggested supportive care measures for the management of adverse events with potential immunologic etiology are outlined along with the dose modification guidelines in Section 5.2.1. Where appropriate, these guidelines include the use of oral or IV treatment with corticosteroids as well as additional anti-inflammatory agents if symptoms do not improve with administration of corticosteroids. Note that several courses of steroid tapering may be necessary as symptoms may worsen when the steroid dose is decreased. For each disorder, attempts should be made to rule out other causes such as metastatic disease or bacterial or viral infection, which might require additional supportive care. The treatment guidelines are intended to be applied when the investigator determines the events to be related to pembrolizumab.

Note: If after the evaluation of the event, it is determined not to be related to pembrolizumab, the Investigator does not need to follow the treatment guidance. Refer to Section 5.2.1 for guidelines regarding dose modification and supportive care.

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

5.6.2 Supportive Care Guidelines for Chemotherapy Agents

Instructions regarding supportive care for the chemotherapeutic agents administered in this study can be found in the local product label for each agent. Infusion reactions and injection site reactions will be managed by the investigators according to the local product labels.

5.7 Diet/Activity/Other Considerations

5.7.1 Diet

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

5.7.2 Contraception

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

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

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

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

OR

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

OR

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

Female and male subjects of reproductive potential must agree to avoid becoming pregnant (or getting their partner pregnant) while receiving study drug and for 12 months after the last dose of study drug for subjects who have received cyclophosphamide, and for 6 months after the last dose of study medication for subjects who have not, by complying with one of the following:

1. practice abstinence[†] from heterosexual activity;

OR

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

Acceptable methods of contraception are[‡]:

1. Single method (one of the following is acceptable):
    - intrauterine device (IUD)
    - vasectomy of a female subject's male partner
    - contraceptive rod implanted into the skin
2. Combination method (requires use of two of the following):
    - diaphragm with spermicide (cannot be used in conjunction with cervical cap/spermicide)
    - cervical cap with spermicide (nulliparous women only)
    - contraceptive sponge (nulliparous women only)
    - male condom or female condom (cannot be used together)
    - hormonal contraceptive: oral contraceptive pill (estrogen/progestin pill or progestin-only pill), contraceptive skin patch, vaginal contraceptive ring, or subcutaneous contraceptive injection

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

Subjects should be informed that taking the study medication may involve unknown risks to the fetus (unborn baby) if pregnancy were to occur during the study. In order to participate in the study subjects of childbearing potential must adhere to the contraception requirement (described above) from the day of study medication initiation (or 14 days prior to the initiation of study medication for oral contraception) throughout the study period up to 12 months after the last dose of trial therapy for subjects who have received cyclophosphamide, and for 6 months after the last dose of study medication for subjects who have not. If there is any question that a subject of childbearing potential will not reliably comply with the requirements for contraception, that subject should not be entered into the study.

5.7.3 Use in Pregnancy

If a subject inadvertently becomes pregnant while on treatment with pembrolizumab, the subject will immediately be removed from study treatment.

5.7.4 Use in Nursing Women

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

5.8 Subject Withdrawal/Discontinuation Criteria

5.8.1 Discontinuation of Treatment

Discontinuation of treatment does not represent withdrawal from the trial.

As certain data on clinical events beyond treatment discontinuation may be important to the study, they must be collected through the subject's last scheduled follow-up, even if the subject has discontinued treatment. Therefore, all subjects who discontinue trial treatment prior to completion of the treatment will still continue to participate in the trial as specified in Section 6.0 - Trial Flow Chart and Section 7.1.4.1.

Subjects may discontinue treatment at any time for any reason or be dropped from treatment at the discretion of the investigator should any untoward effect occur. In addition, a subject may be discontinued from treatment by the investigator if treatment is inappropriate, the trial plan is violated, or for administrative and/or other safety reasons. Specific details regarding procedures to be performed at treatment discontinuation are provided in Section 7.1.4 – Other Procedures.

A subject must be discontinued from treatment, but continue to be monitored in the trial for any of the following reasons:

- Unacceptable adverse experiences as described in Section 9.1 - Common Terminology Criteria for Adverse Events (CTCAE) and Section 9.2 - Definitions.

- The subject or subject's legally acceptable representative requests to discontinue treatment.
- Intercurrent illness that prevents further administration of treatment
- Investigator's decision to withdraw the subject from study treatment due to disease progression or other reasons.
- The subject has a confirmed positive serum pregnancy test
- Noncompliance with trial treatment or procedure requirements

The Early Discontinuation and Safety Follow-up visit procedures are listed in Section 6 – Trial Flow Chart and Section 7.1.5 – Visit Requirements. Following completion of treatment, each subject will be followed for 30 days for any adverse events (SAEs will be collected for 90 days after completion of treatment or 30 days following completion of treatment if the subject initiates new anticancer therapy, whichever is earlier, as described in Section 9.3 –Adverse Event Reporting).

Discontinuation from treatment is "permanent." Once a subject is discontinued, he/she shall not be allowed to restart treatment.

5.8.2 Withdrawal from the Trial

A subject must be withdrawn from the trial if the subject or subject's legally acceptable representative withdraws consent from the trial.

If a subject withdraws from the trial, they will no longer receive treatment or be followed at scheduled protocol visits.

Specific details regarding procedures to be performed at the time of withdrawal from the trial including the procedures to be performed should a subject repeatedly fail to return for scheduled visits and/or if the study site is unable to contact the subject, as well as specific details regarding withdrawal from Future Biomedical Research are outlined in Section 7.1.4 – Other Procedures.

5.9 Subject Replacement Strategy

Subjects will be replaced in the following scenarios:

- Discontinuation from trial treatment or withdraws from the trial without receipt of at least a partial dose of therapy. In the case that a subject withdraws prior to receipt of at least a partial dose, the subject will not be evaluable and will be replaced.
- Additional subjects may be randomized to ensure that at least n=15 viable paired tumor specimens are available for experimental arms, and n=10 viable paired tumor specimens are available for control arm.

5.10 Beginning and End of the Trial

The overall trial begins when the first subject signs the informed consent form. The overall trial ends when the last subject completes the last study-related visit, withdraws from the trial or is lost to follow-up (i.e. the subject is unable to be contacted by the investigator).

5.11 Clinical Criteria for Early Trial Termination

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

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

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

Further recruitment in the study or at (a) particular study site(s) may be stopped due to insufficient compliance with the protocol, GCP, and/or other applicable regulatory requirements or procedure-related problems or if the number of discontinuations for administrative reasons is too high.

6.0 TRIAL FLOW CHART

Table 6.0-1: Control Arm Flow Chart

Trial Period:	**Screening Phase**	**Induction Phase**			**Neoadjuvant Treatment Phase (a)**									**30 Day Safety Follow-Up (b)**	**Definitive Surgery**	**Early Discon Visit (c)**	**Follow-up**
					Treatment 1					**Treatment 2**							
					(Paclitaxel)					**(AC)**							
Treatment Cycle:	Screening	D1	D8 +/- 2d	D15	C1D1 (21 days +/- 5d of D1 induction)	C1 D8	C1 D15	C2-C4 D1	C2-C4 D8, D15	C1D1	C2D1	C3D1	C4D1				Q3mos for 3 years, then Q6mos for 5 years from definitive surgery.
Scheduling Window (Days)	-28 to 0					±2	±2	±2	±2	±3	±3	±3	±3	±7	Surgery		±30
Informed Consent (d)	X																
Inclusion/Exclusion Criteria	X																
Demographics and Medical History	X																
Prior and Concomitant Medication Review (e)	X				X			X		X	X	X	X	X		X	
Treatment allocation/randomization	X (w)																
Clinical/Pathological tumor staging	X														X		
Pembrolizumab		X			X			X		X	X	X	X				
Paclitaxel					X	X	X	X	X								
Doxorubicin										X	X	X	X				
Cyclophosphamide										X	X	X	X				

Trial Period:	**Screening Phase**	**Induction Phase**			**Neoadjuvant Treatment Phase (a)**									**30 Day Safety Follow-Up (b)**	**Definitive Surgery**	**Early Discon Visit (c)**	**Follow-up**
					Treatment 1					**Treatment 2**							
					(Paclitaxel)					**(AC)**							
Treatment Cycle:	Screening	D1	D8 +/- 2d	D15	C1D1 (21 days +/- 5d of D1 induction)	C1	C1	C2-C4 D1	C2-C4 D8, D15	C1D1	C2D1	C3D1	C4D1				Q3mos for 3 years, then Q6mos for 5 years from definitive surgery.
						D8	D15										
Review Adverse Events (f)	X	X			X	X	X	X	X	X	X	X	X	X		X	
12-Lead ECG (Locally performed)	X									X (g)				X		X (x)	
MUGA or ECHO for LVEF Assessment	X									X (g)				X		X (x)	
Full Physical Examination	X																
Directed Physical Examination					X			X		X	X	X	X	X		X	
Vital Signs, Height and Weight (h)	X				X	X	X	X	X	X	X	X	X	X		X	
ECOG Performance Status (i)	X				X			X		X	X	X	X	X		X	
Pregnancy Test – Urine or Serum β-HCG (j)	X																
Blood for menopausal status (if applicable) (k)	X																
PT/INR and aPTT/ PTT (l,n)	X													X		X	
CBC with Differential (m,n,o,q)	X	X (q)	X (q)		X	X	X	X	X	X	X	X	X	X		X	
Chemistry Panel (m,n,o)	X				X	X	X	X	X	X	X	X	X	X		X	

Trial Period:	**Screening Phase**	**Induction Phase**			**Neoadjuvant Treatment Phase (a)**									**30 Day Safety Follow-Up (b)**	**Definitive Surgery**	**Early Discon Visit (c)**	**Follow-up**
					Treatment 1					**Treatment 2**							
					(Paclitaxel)					**(AC)**							
Treatment Cycle:	Screening	D1	D8 +/- 2d	D15	C1D1 (21 days +/- 5d of D1 induction)	C1 D8	C1 D15	C2-C4 D1	C2-C4 D8, D15	C1D1	C2D1	C3D1	C4D1				Q3mos for 3 years, then Q6mos for 5 years from definitive surgery.
Urinalysis (m,n,o)	X							X		X			X	X		X	
T3, FT4 and TSH (m,n,o)	X									X				X		X	
Cortisol (m,n,u)	X																
Troponin (m,n,v)	X																
LDH (m,n)	X													X		X	
Assessment of disease progression (p)								X		X	X	X	X	X		X	X
Blood for Biomarker Analysis (q)		X (q)	X(q)		X (q)			X (q)		X (q)	X (q)			X (q)		X (q)	
pCR assessment															X		
Core tumor tissue biopsy for translational research (s)	X (s)			X(s)										* (s)	X (r,t)		
Definitive surgery															X (r,t)		
Survival																	X

AC = doxorubicin + cyclophosphamide; CBC = Complete Blood Count; Discon = discontinuation; DNA = deoxyribonucleic acid; ECG = electrocardiogram; ECHO = Echocardiogram; ECOG = Eastern Cooperative Oncology Group; PRO = Patient Reported Outcomes; ; FFPE = formalin-fixed paraffin-embedded; FT4=Free thyroxine 4; HCG = human chorionic gonadotropin; LDH = Lactate Dehydrogenase; LVEF = Left ventricular ejection fraction; MUGA = Multigated Acquisition; NCI = National Cancer Institute; pCR = Pathological Complete Response; PT/INR = Prothrombin Time/International Normalized Ratio; aPTT/PTT= Activated Prothrombin Time/Partial thromboplastin time; RNA = ribonucleic acid; T3 = Triiodothyronine; TNBC = Triple-negative Breast Cancer; TSH = Thyroid stimulating hormone.

Trial Period:	**Screening Phase**	**Induction Phase**			**Neoadjuvant Treatment Phase (a)**									**30 Day Safety Follow-Up (b)**	**Definitive Surgery**	**Early Discon Visit (c)**	**Follow-up**
					Treatment 1					**Treatment 2**							
					(Paclitaxel)					**(AC)**							
Treatment Cycle:	Screening	D1	D8 +/- 2d	D15	C1D1 (21 days +/- 5d of D1 induction)	C1	C1	C2-C4 D1	C2-C4 D8, D15	C1D1	C2D1	C3D1	C4D1				Q3mos for 3 years, then Q6mos for 5 years from definitive surgery.
						D8	D15										

a. In general, assessments/procedures are performed on Day 1 of each cycle prior to dosing of any study treatment (or prior to weekly dosing of paclitaxel) unless otherwise specified. Each treatment cycle is 3 weeks (21 days). If the treatment is delayed, all procedures should be performed based on the new dosing schedule.

b. The 30-Day Safety Follow-Up visit should be performed within 30 days +/- 7 days after last dose of chemotherapy. If surgery is scheduled to occur less than 30 days after the end of Neoadjuvant Treatment Phase, the Safety Follow-up Visit should occur before surgery. If patient is to receive another treatment for their breast cancer prior to surgery and following end of study treatment, the 30 Day Safety Follow-up should be performed prior to initiation of new treatment. If an Early Discontinuation Visit occurs, then every attempt should be made to perform a 30-Day Safety Follow-Up visit (30 days ± 7 days).

c. The Early Discontinuation Visit should be conducted if subject discontinues all protocol-specified treatment after Treatment 1 Cycle 1 through 30 Day Safety Follow-up.

d. Written consent must be obtained prior to performing any protocol-specified procedures. If the signature falls outside of the 28 day screening window, the consent form does not need to be resigned. Results of a test performed prior to the subject signing consent as part of routine clinical management are acceptable in lieu of a screening test, if performed within the specified time frame (e.g., within 28 days prior to the first dose of trial treatment).

e. Prior medications – Record all medications taken within 30 days prior to the screening visit. Concomitant medications – Enter new medications started during the screening period through the Safety Follow-up Visit or Early Discontinuation, whichever is earlier. Record all medications taken for AEs

f. AEs and laboratory safety measurements will be graded per NCI CTCAE Version 5.0. All AEs, whether gradable by CTCAE or not, will also be evaluated for seriousness.

g. After Treatment 1 Cycle 4, and prior to dosing the subject with AC, the 12-lead ECG and MUGA/ECHO must be performed to ensure adequate cardiac function. If taxane aborted early due to toxicity, the 12-lead EKG and MUGA/ECHO are not required prior to starting AC and should be obtained at Investigator discretion.

h. Vital signs to include temperature, pulse, respiratory rate and blood pressure. Height will be measured at screening only; weight will be measured at baseline and preceding each dose of chemotherapy/pembrolizumab. Vital signs will be collected during treatment cycle.

<table>
<tr><th rowspan="3">Trial Period:</th><th rowspan="3">Screening Phase</th><th rowspan="3" colspan="3">Induction Phase</th><th colspan="10">Neoadjuvant Treatment Phase (a)</th><th rowspan="3">30 Day Safety Follow-Up (b)</th><th rowspan="3">Definitive Surgery</th><th rowspan="3">Early Discon Visit (c)</th><th rowspan="3">Follow-up</th></tr>
<tr><th colspan="6">Treatment 1</th><th colspan="4">Treatment 2</th></tr>
<tr><th colspan="6">(Paclitaxel)</th><th colspan="4">(AC)</th></tr>
<tr><td rowspan="2">Treatment Cycle:</td><td rowspan="2">Screening</td><td rowspan="2">D1</td><td>D8 +/- 2d</td><td rowspan="2">D15</td><td rowspan="2">C1D1 (21 days +/- 5d of D1 induction)</td><td>C1</td><td>C1</td><td rowspan="2">C2-C4 D1</td><td rowspan="2">C2-C4 D8, D15</td><td rowspan="2">C1D1</td><td rowspan="2">C2D1</td><td rowspan="2">C3D1</td><td rowspan="2">C4D1</td><td rowspan="2"></td><td rowspan="2"></td><td rowspan="2"></td><td rowspan="2">Q3mos for 3 years, then Q6mos for 5 years from definitive surgery.</td></tr>
<tr><td></td><td>D8</td><td>D15</td></tr>
<tr><td colspan="20">i. ECOG performance status at Screening to be performed within 14 days prior to of the first dose of trial treatment. ECOG performance status will also be performed prior to the start of every treatment cycle, at 30-Day follow-up visits, and at the Early Discontinuation visit.</td></tr>
<tr><td colspan="20">j. For women of childbearing potential, a serum or urine pregnancy test should be performed within 72 hours prior to first dose of trial treatment. If urine pregnancy results are positive or cannot be confirmed as negative, a serum pregnancy test performed by the local study site laboratory will be required. Monthly pregnancy testing should be conducted as per local regulations where applicable.</td></tr>
<tr><td colspan="20">k. Blood for menopausal status may be required for certain subjects as described in Section 7.1.3.1 – Menopausal Status.</td></tr>
<tr><td colspan="20">l. Coagulation factors (PT/INR and aPTT/PTT) should be tested within 14 days of treatment initiation and at the time points specified. Additional testing to be conducted as clinically indicated for subjects taking anticoagulant therapy.</td></tr>
<tr><td colspan="20">m. After Treatment 1 Cycle 1, pre-dose lab samples can be collected up to 72 hours prior to the scheduled time point.</td></tr>
<tr><td colspan="20">n. Screening laboratory samples will be collected within 14 days prior to study treatment initiation.</td></tr>
<tr><td colspan="20">o. Unresolved abnormal labs that are drug-related AEs should be followed until resolution. Labs do not need to be repeated after the end of treatment if labs are within normal range.</td></tr>
<tr><td colspan="20">p. Assessment includes (per local or institutional guidelines): disease progression that precludes definitive surgery, local or distant recurrence, development of a second primary malignancy, or death.</td></tr>
</table>

Trial Period:	**Screening Phase**	**Induction Phase**			**Neoadjuvant Treatment Phase (a)**									**30 Day Safety Follow-Up (b)**	**Definitive Surgery**	**Early Discon Visit (c)**	**Follow-up**
					Treatment 1					**Treatment 2**							
					(Paclitaxel)					**(AC)**							
Treatment Cycle:	Screening	D1	D8 +/- 2d	D15	C1D1 (21 days +/- 5d of D1 induction)	C1	C1	C2-C4 D1	C2-C4 D8, D15	C1D1	C2D1	C3D1	C4D1				Q3mos for 3 years, then Q6mos for 5 years from definitive surgery.
						D8	D15										
q. Blood for exploratory biomarkers (Three 10cc green top tube, one 5cc cytochex tube, and one SST tube; CPT tube acceptable as alternative to green top if the green top is not feasible) will be collected on Induction day 1, day 8 +/- 2 days, at pre-dose on Day 1 of Cycles 1 and 2 of Treatment 1 (paclitaxel), Day 1 of Cycles 1 and 2 of Treatment 2 (AC), and at Early Discontinuation or safety follow-up visit (within 30 days +/- 7 days of last chemo) . A CBC must be collected on all days that research blood is collected. See Laboratory Manual. Any leftover samples from the blood studies will be stored for future biomedical research.																
r. If feasible, a tumor tissue sample will also be collected at definitive surgery for subjects who have not achieved a pathological complete response (pCR)																	
s. During screening, formalin-fixed paraffin-embedded (FFPE) tumor tissue samples or slides obtained at subject's initial diagnosis maybe submitted and used to satisfy the baseline requirement if sufficient tissue for analysis is available). Induction Phase biopsy to be collected within 7 days prior to starting Treatment 1. *In certain cases, a post-treatment biopsy will be conducted per the treating physician if clinically indicated to confirm the presence of residual disease burden, and to guide subsequent therapy in the neoadjuvant setting. In this case, remaining tissue may be used for research purposes following confirmation of residual disease.																	
t. Detailed pathological staging of all tumor tissue collected during definitive surgery for determination of pCR.																	
u. Cortisol to be determined in the morning.																	
v. Troponin to be measured at screening and then as clinically indicated. w. Induction Day 1 must occur within 3 business days of randomization x. ECG and ECHO/MUGA need not be repeated at early discontinuation if performed within previous 12 weeks																	

Table 6.0-2: Arm A (IRX-2) Flow Chart

Trial Period:	**Screening Phase**	**Induction Phase**			**Neoadjuvant Treatment Phase (a)**										**30 Day Safety Follow-Up (b)**	**Definitive Surgery**	**Early Discon Visit (c)**	**Follow-up**
					Treatment 1 (Paclitaxel)					**IRX-2 Reinduction**	**Treatment 2 (AC)**							
Treatment Cycle:	Screening	D1	Any 10 days between day 4-21	Within 7 days of last IRX-2	C1D1 (21 days +/- 5d of D1 induction)	C1 D8	C1 D15	C2-C4 D1	C2-C4 D8, D15	Any 10 days over 14 day window	C1 D1	C2 D1	C3 D1	C4 D1				Q3mos for 3 years, then Q6mos for 5 years from definitive surgery.
Scheduling Window (Days)	-28 to 0					±2	±2	±2	±2		±3	±3	±3	±3	±7	Surgery		±30
Informed Consent (d)	X																	
Inclusion/Exclusion Criteria	X																	
Demographics and Medical History	X																	
Prior and Concomitant Medication Review (e)	X				X			X			X	X	X	X	X		X	
Treatment allocation/randomization	X(x)																	
Clinical/Pathological tumor staging	X															X		
Pembrolizumab		X			X			X			X	X	X	X				
Paclitaxel					X	X	X	X	X									
Doxorubicin											X	X	X	X				
Cyclophosphamide											X	X	X	X				
Induction Cyclophosphamide		X																
IRX-2			X							X (w)								

Trial Period:	**Screening Phase**	**Induction Phase**			**Neoadjuvant Treatment Phase (a)**										**30 Day Safety Follow-Up (b)**	**Definitive Surgery**	**Early Discon Visit (c)**	**Follow-up**
					Treatment 1 (Paclitaxel)					**IRX-2 Reinduction**	**Treatment 2 (AC)**							
Treatment Cycle:	Screening	D1	Any 10 days between day 4-21	Within 7 days of last IRX-2	C1D1 (21 days +/- 5d of D1 induction)	C1 D8	C1 D15	C2-C4 D1	C2-C4 D8, D15	Any 10 days over 14 day window	C1 D1	C2 D1	C3 D1	C4 D1				Q3mos for 3 years, then Q6mos for 5 years from definitive surgery.
Review Adverse Events (f)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X		X	
12-Lead ECG (Locally performed)	X										X (g)				X		X (y)	
MUGA or ECHO for LVEF Assessment	X										X (g)				X		X (y)	
Full Physical Examination	X																	
Directed Physical Examination					X			X			X	X	X	X	X		X	
Vital Signs, Height and Weight (h)	X				X	X	X	X	X		X	X	X	X	X		X	
ECOG Performance Status (i)	X				X			X			X	X	X	X	X		X	
Pregnancy Test – Urine or Serum β-HCG (j)	X																	
Blood for menopausal status (if applicable) (k)	X																	
PT/INR and aPTT/ PTT (l,n)	X														X		X	
CBC with Differential (m,n,o,q)	X	X (q)	X(q)		X	X	X	X	X	X(q)	X	X	X	X	X		X	
Chemistry Panel (m,n,o)	X				X	X	X	X	X		X	X	X	X	X		X	
Urinalysis (m,n,o)	X							X			X			X	X		X	

Trial Period:	**Screening Phase**	**Induction Phase**			**Neoadjuvant Treatment Phase (a)**										**30 Day Safety Follow-Up (b)**	**Definitive Surgery**	**Early Discon Visit (c)**	**Follow-up**
					Treatment 1 (Paclitaxel)					**IRX-2 Reinduction**	**Treatment 2 (AC)**							
Treatment Cycle:	Screening	D1	Any 10 days between day 4-21	Within 7 days of last IRX-2	C1D1 (21 days +/- 5d of D1 induction)	C1 D8	C1 D15	C2-C4 D1	C2-C4 D8, D15	Any 10 days over 14 day window	C1 D1	C2 D1	C3 D1	C4 D1				Q3mos for 3 years, then Q6mos for 5 years from definitive surgery.
T3, FT4 and TSH (m,n,o)	X										X				X		X	
Cortisol (m,n,u)	X																	
Troponin (m,n,v)	X																	
LDH m,n)	X														X		X	
Assessment of disease progression (p)								X			X	X	X	X	X		X	X
Blood for Biomarker Analysis (q)		X (q)	X (q)		X (q)			X (q)		X(q)	X (q)	X (q)			X (q)		X (q)	
pCR assessment																X		
Core tumor tissue biopsy for translational research (s)	X (s)			X											* (s)	X (r,t)		
Definitive surgery																X (r,t)		
Survival																		X

AC = doxorubicin + cyclophosphamide; CBC = Complete Blood Count; Discon = discontinuation; DNA = deoxyribonucleic acid; ECG = electrocardiogram; ECHO = Echocardiogram; ECOG = Eastern Cooperative Oncology Group; PRO = Patient Reported Outcomes; ; FFPE = formalin-fixed paraffin-embedded; FT4=Free thyroxine 4; HCG = human chorionic gonadotropin; LDH = Lactate Dehydrogenase; LVEF = Left ventricular ejection fraction; MUGA = Multigated Acquisition; NCI = National Cancer Institute; pCR = Pathological Complete Response; PT/INR = Prothrombin Time/International Normalized Ratio; aPTT/PTT= Activated Prothrombin Time/Partial thromboplastin time; RNA = ribonucleic acid; T3 = Triiodothyronine; TNBC = Triple-negative Breast Cancer; TSH = Thyroid stimulating hormone.

Trial Period:	**Screening Phase**	**Induction Phase**			**Neoadjuvant Treatment Phase (a)**										**30 Day Safety Follow-Up (b)**	**Definitive Surgery**	**Early Discon Visit (c)**	**Follow-up**
					Treatment 1 (Paclitaxel)					**IRX-2 Reinduction**	**Treatment 2 (AC)**							
Treatment Cycle:	Screening	D1	Any 10 days between day 4-21	Within 7 days of last IRX-2	C1D1 (21 days +/- 5d of D1 induction)	C1	C1	C2-C4	C2-C4	Any 10 days over 14 day window	C1	C2	C3	C4				Q3mos for 3 years, then Q6mos for 5 years from definitive surgery.
						D8	D15	D1	D8, D15		D1	D1	D1	D1				
a. In general, assessments/procedures are performed on Day 1 of each cycle prior to dosing of any study treatment (or prior to weekly dosing of paclitaxel) unless otherwise specified. Each treatment cycle is 3 weeks (21 days). If the treatment is delayed, all procedures should be performed based on the new dosing schedule.																		
b. The 30-Day Safety Follow-Up visit should be performed within 30 days +/- 7 days after last dose of chemotherapy. If surgery is scheduled to occur less than 30 days after the end of Neoadjuvant Treatment Phase, the Safety Follow-up Visit should occur before surgery. If patient is to receive another treatment for their breast cancer prior to surgery and following end of study treatment, the 30 Day Safety Follow-up should be performed prior to initiation of new treatment. If an Early Discontinuation Visit occurs, then every attempt should be made to perform a 30-Day Safety Follow-Up visit (30 days ± 7 days).																		
c. The Early Discontinuation Visit should be conducted if subject discontinues all protocol-specified treatment after Treatment 1 Cycle 1 through 30 Day Safety Follow-up..																		
d. Written consent must be obtained prior to performing any protocol-specified procedures. If the signature falls outside of the 28 day screening window, the consent form does not need to be resigned. Results of a test performed prior to the subject signing consent as part of routine clinical management are acceptable in lieu of a screening test, if performed within the specified time frame (e.g., within 28 days prior to the first dose of trial treatment).																		
e. Prior medications – Record all medications taken within 30 days prior to the screening visit. Concomitant medications – Enter new medications started during the screening period through the Safety Follow-up Visit or Early Discontinuation, whichever is earlier. Record all medications taken for AEs																		
f. AEs and laboratory safety measurements will be graded per NCI CTCAE Version 5.0. All AEs, whether gradable by CTCAE or not, will also be evaluated for seriousness.																		
g. After Treatment 1 Cycle 4, and prior to dosing the subject with AC, the 12-lead ECG and MUGA/ECHO must be performed to ensure adequate cardiac function. If taxane aborted early due to toxicity, the 12-lead EKG and MUGA/ECHO are not required prior to starting AC and should be obtained at Investigator discretion.																		
h. Vital signs to include temperature, pulse, respiratory rate and blood pressure. Height will be measured at screening only; weight will be measured at baseline and at each cycle. Vital signs will be collected during treatment cycle.																		

Trial Period:	**Screening Phase**	**Induction Phase**			**Neoadjuvant Treatment Phase (a)**										**30 Day Safety Follow-Up (b)**	**Definitive Surgery**	**Early Discon Visit (c)**	**Follow-up**
					Treatment 1 (Paclitaxel)					**IRX-2 Reinduction**	**Treatment 2 (AC)**							
Treatment Cycle:	Screening	D1	Any 10 days between day 4-21	Within 7 days of last IRX-2	C1D1 (21 days +/- 5d of D1 induction)	C1	C1	C2-C4	C2-C4	Any 10 days over 14 day window	C1	C2	C3	C4				Q3mos for 3 years, then Q6mos for 5 years from definitive surgery.
						D8	D15	D1	D8, D15		D1	D1	D1	D1				
i. ECOG performance status at Screening to be performed within 14 days prior to of the first dose of trial treatment. ECOG performance status will also be performed prior to the start of every treatment cycle, at 30-Day follow-up visits, and at the Early Discontinuation visit.																		
j. For women of childbearing potential, a serum or urine pregnancy test should be performed within 72 hours prior to first dose of trial treatment. If urine pregnancy results are positive or cannot be confirmed as negative, a serum pregnancy test performed by the local study site laboratory will be required. Monthly pregnancy testing should be conducted as per local regulations where applicable.																		
k. Blood for menopausal status may be required for certain subjects as described in Section 7.1.3.1 – Menopausal Status.																		
l. Coagulation factors (PT/INR and aPTT/PTT) should be tested within 14 days of treatment initiation and at the time points specified. Additional testing to be conducted as clinically indicated for subjects taking anticoagulant therapy.																		
m. After Treatment 1 Cycle 1, pre-dose lab samples can be collected up to 72 hours prior to the scheduled time point.																		
n. Screening laboratory samples will be collected within 14 days prior to study treatment initiation.																		
o. Unresolved abnormal labs that are drug-related AEs should be followed until resolution. Labs do not need to be repeated after the end of treatment if labs are within normal range.																		
p. Assessment includes (per local or institutional guidelines): disease progression that precludes definitive surgery, local or distant recurrence, development of a second primary malignancy, or death.																		

Trial Period:	**Screening Phase**	**Induction Phase**			**Neoadjuvant Treatment Phase (a)**										**30 Day Safety Follow-Up (b)**	**Definitive Surgery**	**Early Discon Visit (c)**	**Follow-up**
					Treatment 1 (Paclitaxel)					**IRX-2 Reinduction**	**Treatment 2 (AC)**							
Treatment Cycle:	Screening	D1	Any 10 days between day 4-21	Within 7 days of last IRX-2	C1D1 (21 days +/- 5d of D1 induction)	C1	C1	C2-C4	C2-C4	Any 10 days over 14 day window	C1	C2	C3	C4				Q3mos for 3 years, then Q6mos for 5 years from definitive surgery.
						D8	D15	D1	D8, D15		D1	D1	D1	D1				
q. Blood for exploratory biomarkers (Three 10cc green top tube, one 5cc cytochex tube, and one SST tube; CPT tube acceptable as alternative to green top if the green top is not feasible) will be collected on Induction day 1 (before cyclophosphamide), at pre-dose on day 8 +/- 2 days, at pre-dose on Day 1 of Cycles 1 and 2 of Treatment 1 (paclitaxel), Day 1 pre-dose of IRX-2 re-induction, Day 1 pre-dose of Cycles 1 and 2 of Treatment 2 (AC), and at Early Discontinuation or safety follow-up (30 day from last chemo +/- 7 days). Serum obtained at each of these time points will be stored to allow for anti-drug antibody (ADA) testing if necessary. A CBC must be collected on all days that research bloods are collected. See Laboratory Manual. Any leftover samples from the blood studies will be stored for future biomedical research.																	
r. If feasible, a tumor tissue sample will also be collected at definitive surgery for subjects who have not achieved a pathological complete response (pCR)																		
s. During screening, formalin-fixed paraffin-embedded (FFPE) tumor tissue samples or slides obtained at subject's initial diagnosis maybe submitted and used to satisfy the baseline requirement if sufficient tissue for analysis is available). *In certain cases, a post-treatment biopsy will be conducted per the treating physician if clinically indicated to confirm the presence of residual disease burden, and to guide subsequent therapy in the neoadjuvant setting. In this case, remaining tissue may be used for research purposes following confirmation of residual disease.																		
t. Detailed pathological staging of all tumor tissue collected during definitive surgery for determination of pCR.																		
u. Cortisol to be determined in the morning.																		
v. Troponin to be measured at screening and then as clinically indicated.																		
w. IRX-2 re-induction consists of IRX-2 injections, may commence as early as 1 day after last dose of treatment 1 (1 day after c4d15 paclitaxel/pembrolizumab), and should be completed prior to commencing treatment 2. Please refer to Section 5.0 for guidance in setting of early termination of taxane. x. Induction day 1 must occur within 3 business days of randomization. y. ECG and ECHO/MUGA need not be repeated at early discontinuation if performed within previous 12 weeks																		

7.0 TRIAL PROCEDURES

7.1 Trial Procedures

The Trial Flow Chart - Section 6.0 summarizes the trial procedures to be performed at each visit. Individual trial procedures are described in detail below. It may be necessary to perform these procedures at unscheduled time points if deemed clinically necessary by the investigator.

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

7.1.1 Administrative Procedures

7.1.1.1 Informed Consent

Consent must be documented by the subject's dated signature or by the subject's legally acceptable representative's dated signature on a consent form along with the dated signature of the person conducting the consent discussion. A copy of the signed and dated consent form should be given to the subject before participation in the trial.

7.1.1.2 Inclusion/Exclusion Criteria

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

7.1.1.3 Medical History

A medical history will be obtained by the investigator or qualified designee. Medical history will include all active conditions, and any condition diagnosed within the prior 10 years that are considered to be clinically significant by the Investigator. Any autoimmune disorders, regardless of onset date, should be recorded.

7.1.1.3.1 Disease Details

Details regarding subject's TNBC diagnosis and status at baseline must be thoroughly evaluated by the investigator or qualified designee and recorded.

7.1.1.4 Prior and Concomitant Medications Review

7.1.1.4.1 Prior Medications

The investigator or qualified designee will review prior medication use, including any protocol-specified washout requirement, and record prior medication taken by the subject within 30 days prior to screening visit.

7.1.1.4.2 Concomitant Medications

The investigator or qualified designee will record all concomitant medication, if any, taken by the subject within 30 days before the first dose of trial treatment through the Adjuvant Treatment Phase Safety Follow-up Visit. All medications related to reportable AEs, SAEs and ECIs, including AEs and SAEs following the Adjuvant Treatment Phase, 30-day Safety Follow-up Visit, and Early Discontinuation visit should be recorded.

7.1.1.5 Research Participant Registration

To register a patient, email the Data Management Office of the Providence Cancer Institute at OREACRIRandomization@providence.org with the following information:

- Investigator's name
- Patient's initials and date of birth
- Eligibility Verification
- Date of first dose.

Patients must meet all of the eligibility requirements and undergo all pre-study procedures. If a patient enrolls in the study, but does not receive study therapy, the patient's enrollment may be canceled. Reasons for cancellation will be documented in writing. Any patient whose enrollment was canceled before receiving study therapy will be replaced.

Assignment of Study Numbers:

Study Numbers will be assigned at enrollment based on order of enrollment. For example:

- MFH is the 4th patient enrolled to study. Study Number PMI – 04
- B-A is 9th patient enrolled to study. Study Number PMI - 09

All case report forms, study reports, and laboratory samples for research tests, including immune parameters or pharmacokinetics, will be labeled with the full patient study number.

7.1.1.6 Assignment of Treatment/Randomization Number

All eligible subjects will be randomly allocated and will receive a treatment/randomization number. The treatment/randomization number identifies the subject for all procedures occurring after treatment allocation/randomization. Once a treatment/randomization number is assigned to a subject, it can never be re-assigned to another subject. A single subject cannot be assigned more than 1 treatment/randomization number. Study treatment should begin on the day of randomization or, at most, within 3 days post randomization. Randomization will occur via the Data Management Office of the Providence Cancer Institute.

7.1.1.7 Trial Compliance (Medication)

Interruptions from the protocol specified treatment for greater than 6 weeks between doses require consultation with the principal investigator.

Administration of trial medication will be overseen by the investigator and/or trial staff. The total volume of IRX-2, pembrolizumab alone and/or combination product infused will be

compared to the total volume prepared to determine compliance with each dose of pembrolizumab and/or combination product administered.

The instructions for preparing and administering pembrolizumab will be provided in the Pharmacy Manual.

Refer to the product label for instructions on preparation and administration precautions on combination chemotherapy agents included in the trial: paclitaxel; doxorubicin and cyclophosphamide.

7.1.2 Clinical Procedures/Assessments

7.1.2.1 Adverse Event Monitoring

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

7.1.2.2 12-Lead Electrocardiogram

A 12-lead electrocardiogram (ECG) will be performed according to the study calendar.

7.1.2.3 Echocardiography or Multigated Acquisition Scan

An ECHO or MUGA scan will be required at screening for subjects with locally advanced TNBC to determine study eligibility. The assessment method will be at the investigator's discretion and per the local standard of care. Additional assessments will be performed according to the study calendar and as clinically necessary.

7.1.2.4 Physical Examination

7.1.2.4.1 Full Physical Examination

The investigator or qualified clinical designee will perform a complete physical examination during the screening period. Clinically significant abnormal findings should be recorded as medical history. Post randomization, physical examinations should be performed at the discretion of the physician according to the subject's signs and symptoms.

7.1.2.4.2 Directed Physical Examination

The investigator or qualified clinical designee will perform directed physical examinations to assess subject's TNBC status according to the time points as specified in the study calendar. New clinically significant abnormal findings should be recorded as AEs.

7.1.2.5 Vital Signs

The Investigator or qualified clinical designee will take vital signs according to the study calendar. Height will be measured at screening only. Vital signs should be taken prior to treatment administration.

7.1.2.6 Eastern Cooperative Oncology Group Performance Status

The investigator or qualified clinical designee will assess ECOG performance according to the study calendar.

7.1.2.7 Tumor Tissue Biopsy and Sample Collection

In accordance with the study inclusion criteria, subjects with locally advanced TNBC are required to have a core needle biopsy consisting of at least 1 tumor cores, utilizing multiple passes (fine needle aspirate not adequate) performed during or preceding the Screening Period for research. Diagnostic biopsy FFPE material is sufficient if at least 1 core (or at least 15 slides if not possible) is available to be submitted. Tumor tissue samples will also be collected for both Control Arm and Arm A (IRX) patients during induction phase prior to initiation of Treatment 1 as per Arm specific Flow Chart calendar. Ideally at least 3 cores will be collected during study biopsies. Tumor tissue samples will also be collected from definitive surgery tissue for subjects who have not achieved a pCR. Detailed instructions for tissue collection, processing and shipment are provided in the Laboratory Manual.

7.1.2.8 Imaging Disease Assessment

Subjects must have evidence of M0 disease based on the assessments from their initial diagnosis. In the event of suspected regional or distant metastasis during Screening, subjects should be thoroughly evaluated as clinically indicated; and those with metastatic disease should be excluded. Imaging (e.g., CT, MRI, Bone Scan) will be performed at the discretion of the investigator, as per the local institution's standard of care.

7.1.2.9 Definitive Surgery

Approximately 3-6 weeks following completion of the Neoadjuvant Treatment Phase or Early Discontinuation, subjects will undergo definitive surgery per local standard of care. Details regarding date of surgery, type of surgery, tumor resectability etc. will be recorded. Pathological staging per the current AJCC 8th edition staging criteria and assessment of surgical margins will be performed by the local pathologist on all the tissues removed during the surgery. For subjects who did not achieve a pCR, tumor tissue samples should be collected and submitted for translational research.

In cases were there is evidence of clinical or radiographic residual disease at the completion of study therapy: if the investigating provider wishes to provide additional neoadjuvant therapy, the subject may undergo clinical biopsy to confirm non-pCR, and this may serve as a reasonable substitute to definitive surgery for ascertaining the primary endpoint. In this case, tissue from both the biopsy, and ultimately the resection specimen, should be obtained for research analysis.

7.1.3 Laboratory Procedures/Assessments

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

Table 7.1.3-1 Laboratory Tests

Hematology	Chemistry	Urinalysis	Other
Hematocrit	Albumin	Blood	Serum β-human Chorionic Gonadotropin (β-hCG)[a]
Hemoglobin	Alkaline Phosphatase	Glucose	PT (INR)[b]
Platelet Count	Alanine Aminotransferase (ALT)	Protein	aPTT/PTT[b]
White Blood Cell - WBC (total and differential)	Aspartate Aminotransferase (AST)	Specific Gravity	Total Triiodothyronine (T3)[c]
Red Blood Cell Count	Carbon Dioxide (CO_2 or Bicarbonate)[d]	Microscopic exam, if abnormal results are noted	Free Thyroxine (FT4)
Absolute Neutrophil Count	Calcium	Urine Pregnancy Test[a]	Thyroid Stimulating Hormone (TSH)
Absolute Lymphocyte Count	Chloride		FSH (+/-) Estradiol if clinically indicated to establish menopausal status[e]
	Creatinine or Creatinine clearance (CrCl)		
	Glucose		
	Lactate Dehydrogenase (LDH) [i]		
	Phosphorus		
	Potassium		
	Sodium		
	Total Bilirubin		
	Direct Bilirubin, if Total Bilirubin is elevated above the upper limit of normal[i]		
	Total Protein		
	Blood Urea Nitrogen or Urea[g]		
	Uric Acid[i]		
	Cortisol[h]		
	Troponin[i]		

a. Perform on women of childbearing potential only. Urine pregnancy test is preferred. If the urine test is positive or cannot be confirmed as negative, a serum pregnancy test will be required. The serum or urine pregnancy test should be performed within 72 hours prior to first dose of trial treatment
b. Coagulation factors (PT/INR and aPTT/PTT) should be tested as part of the screening procedures and at the time points specified in the Trial Flow Chart. Additional testing to be conducted as clinically indicated for subjects taking anticoagulant therapy.
c. Total T3 is preferred; if not available free T3 may be tested.
d. If considered standard of care in your region. If these tests are not done as part of standard of care in your region then these tests do not need to be performed.
e. Blood for menopausal status is only required for some subjects
g. Blood urea nitrogen is preferred; if not available urea may be tested.
h. Cortisol is to be determined in the morning at screening.
i. Troponin, LDH, Uric acid, direct bilirubin (if total bilirubin above ULN) measured at screening and then as clinically indicated.

7.1.3.1 Menopausal Status

The menopausal status (pre- or post-menopausal) for women with locally advanced TNBC must be documented. Laboratory studies to assess menopausal status should be obtained per local standard of care when indicated.

7.1.3.2 Blood Collections Samples for Exploratory Biomarker Analyses

Detailed instructions for sample collection, processing and shipment are provided in the Laboratory Manual. The following specimens are to be retained as part of Future Biomedical Research: leftover DNA for future research, leftover tumor tissue, leftover RNA, leftover plasma and serum from exploratory biomarker studies

7.1.4 Other Procedures

7.1.4.1 Withdrawal/Discontinuation

Subjects who discontinue treatment prior to completion of the treatment regimen should be encouraged to continue to be followed for all remaining study visits. When a subject discontinues/withdraws from participation in the trial, all applicable activities scheduled for the Early Discontinuation visit should be performed at the time of discontinuation. Any adverse events which are present at the time of discontinuation/withdrawal should be followed. Subjects may withdraw consent at any time by contacting the principal investigator for the main trial.

7.1.5 Visit Requirements

7.1.5.1 Screening

Approximately 28 days prior to treatment allocation/randomization, potential subjects will be evaluated to determine that they fulfill the entry requirements. Results of a test performed prior to the subject signing consent as part of routine clinical management are acceptable in lieu of a screening test if performed within the specified time frame.

Screening procedures are to be completed within 28 days prior to treatment allocation/randomization except for the following:

- Laboratory tests and ECOG performance status are to be performed within 14 days prior to treatment allocation/randomization.
- For women of reproductive potential, a urine and/or serum pregnancy test will be performed within 72 hours prior to receiving the first dose of study medication.

7.1.5.2 Treatment Cycles

Visit timing requirements during the treatment period are delineated in section 6 study calendar.

7.1.5.3 Definitive Surgery

Definitive surgery is to be conducted per standard of care.

7.1.5.4 Post-Treatment Visits

7.1.5.4.1 Early Discontinuation Visit

The Early Discontinuation Visit should be conducted if subject discontinues all protocol-specified treatment after Treatment 1 Cycle 1 through the end of treatment. If the Discontinuation Visit occurs 30 days from the last dose of study treatment, at the time of the mandatory Safety Follow-Up Visit, procedures listed in the Early Discontinuation should be performed.

7.1.5.4.2 Safety Follow-up Visits

Mandatory Safety Follow-up Visits should be conducted according to the above treatment calendar.

7.1.5.4.3 Unscheduled Visit

Subjects who experience a toxicity that requires discontinuation of all components (i.e., pembrolizumab + paclitaxel) should be observed for recovery prior to initiation of the next treatment phase (i.e. treatment 1, reinduction, or treatment 2). Results from assessments performed during the Unscheduled Visit are acceptable in lieu of repeating assessments during the next treatment phase if performed within 3 days prior.

7.1.5.4.4 Long Term Follow-up

Subjects will be followed per standard-of-care periodically in routine clinical visits by their treating physician. During these visits, and via telephone calls if necessary, subjects will be assessed for disease status/recurrence every three months for 3 years, then every six months for 5 years from definitive surgery.

7.2 TRIAL GOVERNANCE AND OVERSIGHT

7.2.1 Monitoring/Oversight Plan

Oversight of patient safety will include review of adverse events as well as study progress and outcomes. Patient updates, outcomes, and recruitment and retention of patients will be reviewed on a regular basis by the PI, research nurse, and data coordinator. Deviations will be reviewed during oversight meetings and/or through internal reporting procedures. Aggregate protocol deviations are monitored for trends to be reviewed by the Providence Cancer Institute Clinic and Research Quality Committee. In addition, a “first patient review” is conducted and documented for all clinical trials. This review includes treatment administration, deviations, and SAEs to ensure any compliance and/or safety issues are addressed prior to the enrollment of additional patients.

Study monitoring activities (Quality Control Reviews) are performed by clinical research staff members who have completed specialized training in study monitoring procedures and human subjects' protections. Individuals who perform study monitoring activities do not report to Principal Investigators or research scientists and may not monitor studies for which they have direct responsibility. Results of study monitoring activities will be reported to applicable study personnel, Clinical Trials Management and Quality Assurance. Study monitoring activities are conducted regularly and include (but are not limited to) review and verification of the following:

- Eligibility
- Informed Consent process
- Adherence to protocol treatment plan
- Case Report Forms (CRFs)
- Source Documentation
- Adverse Events
- Regulatory

Quality Assurance

Quality Assurance (QA) personnel will perform monitoring activities on a routine basis following the monitoring plan. If necessary, QA will determine follow-up actions to resolve significant findings. QA has the authority to request immediate corrective action if significant patient safety issues are identified. QA will track and trend results from routine monitoring activities as well as associated corrective and preventive actions. If necessary, a QA summary report will be provided to the IRB at the time of continuing review. QA personnel do not have a direct reporting relationship to the principal investigator and are not responsible for enrollment or coordination of care for study participants.

8.0 STATISTICAL ANALYSIS PLAN

This section outlines the statistical analysis strategy and procedures for the study. If, after the study has begun, changes are made to primary and/or key secondary hypotheses, or the statistical methods related to those hypotheses, then the protocol will be amended (consistent with ICH Guideline E-9).

8.1 Statistical Analysis Plan Summary

This is a signal-seeking, phase II trial to evaluate multiple novel immunotherapy agents in combination with pembrolizumab and chemotherapy in a neo-adjuvant setting. Patients will be randomized to either a concurrent control arm (pembro and chemotherapy) or an experimental arm (a novel immunotherapy agent in combination with pembro and chemotherapy). Each novel immunotherapeutic agent will be evaluated sequentially. A concurrent control arm will be used to compare exploratory molecular endpoints. A table below shows the number of patients in each arm and concurrent control arm.

Table 8.1-1: Sample Sizes

Control Arm (Pembro + Chemo)	Arm A (IRX2 + Control)	Subsequent arms (pending protocol amendment)
15	15	
5		15

A rationale of the study design is to allow a rapid identification of a novel neo-adjuvant immunotherapy with a promising signal. Upon the identification, such an arm can be expanded within the same protocol (via protocol amendment) or designed as a separate, larger phase II trial. Immunotherapy agents in subsequent arms will be determined and added to the protocol in the future as a new agent becomes available for the trial. Because experimental arms use a novel combination, we will implement a continual toxicity monitoring and conduct real-time toxicity monitoring to allow suspension of the trial in the presence of unacceptable toxicity.

Primary and Secondary Endpoints: The primary endpoint is pathologic complete response (pCR) by AJCC 8th edition definition (ypT0/is ypN0) in each arm. The secondary endpoints are safety and tolerability profile, residual cancer burden (RCB) score, and increase in tumor infiltrating lymphocyte by H&E. Exploratory endpoints are TIL quantity, phenotype and activation status at baseline and following neo-adjuvant treatment by multispectral immunofluorescence and deep sequencing (TCRseq and RNAseq), and dynamic tumoral/immune cell PD-L1 expression.

Sample Size and Accrual: The merit of the experimental regimen will be based upon a combined assessment of clinical activity, tolerability, and biomarkers activity. Therefore, sample sizes are not determined based upon a formalized power calculation. A sample size of n=15 for each experimental arm would allow a meaningful biomarkers comparative assessment, as well as a meaningful preliminary estimation of pCR rate, according to the below table:

Table 8.1-2: exact pCR confidence intervals at various response levels

Observed pCR rate	90% CI	With 80% certainty, the pCR rate is at least:
n=15/15 (100%)	81.8-100%	89.8%
n=14/15 (93.3%)	72.1-99.7%	81.3%
n=13/15 (86.6%)	63.7-97.6%	73.6%
n=12/15 (80%)	56.0-94.3%	66.3%
n=11/15 (73.3%)	48.9-90.3%	59.2%
n=10/15 (66.6%)	42.3-85.8%	52.4%

The observed clinical activity of the experimental regimen will be estimated as above and interpreted in the context of historical control data from the ISPY2 trial (60% predicted pCR) and a contemporary randomized trial (Merck KEYNOTE-522) which is evaluating pembrolizumab plus neoadjuvant carboplatin-containing chemotherapy for the same patient population. For example, a sample size of 15 would provide 83% power to discriminate a pCR

rate of 60% (the expected pCR rate in the control arm from the I-SPY2 trial) vs. 85% based on a one-sample binomial test with a one-sided exact significance level of 9%.

Because this is a small, signal-seeking study, regimens that achieve pCR rates numerically higher than the historical control, even if not statistically significant based upon this threshold, may still be considered meritorious for further study if robust pharmacodynamic changes are observed in comparison with the control arm.

Assuming an annual accrual rate of 20 patients each year, each experimental arm will take 1-1.5 years to complete the accrual.

Randomization: A randomization list will be generated prior to the start of the trial using a permuted block randomization with a block size of 3 per Arm (6).

Analysis Populations: An intent to treat analysis set includes all patients who consent to enroll in a trial and are randomized to either a control or experimental arm, regardless of actual exposure to the neo-adjuvant treatment. An intent to treat analysis set is used to report accruals and demographic characteristics of trial participants. A safety analysis set includes patients who are exposed to at least one dose of the experimental treatment. Safety endpoints will be evaluated using the safety analysis set. An evaluable analysis set includes those with a specific endpoint available, e.g., pCR evaluable patients includes all patients with pCR outcome is available. An analysis set used for each endpoint will be defined and stated clearly in the results reporting.

Statistical Analysis: Because each arm is sequentially tested, the analysis will be conducted each arm separately as soon as the data for each arm become available. A control arm may be analyzed multiple times during the trial depending on the needs for the concurrent control data. For the primary endpoint, a point and 90% exact binomial confidence interval of pCR will be provided for each arm. For safety and tolerability, all serious adverse events (SAE) will be summarized according to major organ categories of the NCI CTCAE v5.0. The incidence and rate of SAE will be presented for each arm. For residual cancer burden (RCB) score and increase in tumor infiltrating lymphocyte by H&E, descriptive statistics (mean, median, range, and standard deviation) will be provided for each arm. Wherever possible, data visualization techniques (e.g., histogram, box plot and waterfall plot) will be used to display the data for exploratory data analyses.

Continuous Toxicity Monitoring: A dose-limiting toxicity (DLT) is defined as any Grade 3 or higher toxicity defined by NCI CTCAE v5.0. We derived sequential boundaries based on the method proposed by Ivanova et al.[33], which has 90% probability of stopping when the underlying DLT probability is 33%. Specifically, the trial will be suspended for further safety review if the number of patients experiencing DLT is bn or greater among n patients.

Table 8.1-3 Continuous Toxicity Monitoring

Number of Patients, n	3	6	9	12	15
Boundary, b_n	1	2	3	4	5

Missing Values: Data will be analyzed using all available data, and missing values will not be imputed. A sensitivity analysis may be performed to evaluate robustness of the results in cases where there are substantial amount of missing data. Every efforts will be made to minimize missing data, especially for the primary and secondary endpoints.

8.2 Analysis Endpoints

Efficacy and safety endpoints that will be evaluated are listed below.

8.2.1 Efficacy Endpoints

Primary

Pathological Complete Response (pCR) Rate (ypT0/Tis ypN0)

pCR rate (ypT0/Tis ypN0) is defined as the proportion of subjects without residual invasive cancer on hematoxylin and eosin evaluation of the complete resected breast specimen and all sampled regional lymph nodes following completion of neoadjuvant systemic therapy per the current AJCC staging criteria assessed by the local pathologist at the time of definitive surgery.

Subjects who don't receive any study medication will be replaced. Subjects who are discontinued from the study treatment and continue with other neoadjuvant treatment not specified by the study prior to definitive surgery will be classified as not having a pCR (non-responders) in the efficacy analyses, regardless of the results obtained from the surgery.

Subjects who are discontinued from study treatment due to the reasons that preclude surgery are considered non-responders.

Subjects without pCR data due to any reason will be counted as non-responders.

Secondary

The secondary endpoints are safety and tolerability profile (point estimates and 90% confidence intervals of toxicity incidence, assessed by CTCAE v5.0, residual cancer burden (RCB) score, and increase in tumor infiltrating lymphocyte by H&E (comparing diagnostic biopsy to post-induction biopsy. Tumor infiltrating lymphocytes will be assessed by the San Antonio stromal TIL criteria[1]. The rationale for this endpoint is that therapy-associated tumor lymphocyte infiltration may be a surrogate of immune activation/stimulation. The stromal TIL criteria is validated to be both predictive and prognostic in breast cancer, with reasonable intra-observer reproducibility. Scores will be reported as an average of two blinded pathologist reads.

8.2.2 Other Exploratory Endpoints

Immune-based biomarkers, owing to their exploratory nature, will be primarily reported descriptively. If data permit, it may be possible to formally test whether each of the experimental interventions is associated with a change in a specific biomarker relative to the control arm using testing methods appropriate for the nature of the data type for each biomarker. Such statistical testing will be reserved for biomarkers that have significant

rationale founded upon published pre-clinical and preceding clinical data. To account for multiple-testing bias, such tests will be adjusted for multiple comparisons using a method such as the Bonferroni correction.

9.0 REGULATORY AND REPORTING REQUIREMENTS

9.1 Common Terminology Criteria for Adverse Events (CTCAE)

This study will utilize the Common Terminology Criteria for Adverse Events (CTCAE) active version (v5.0). The CTCAE active version can be accessed from the CTEP home page (http://ctep.cancer.gov).

9.2 Definitions

Adverse event:

Any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medical treatment or procedure regardless of whether it is considered related to the medical treatment or procedure (attribution of unrelated, unlikely, possible, probable, or definite). Medical conditions or diseases present before starting the investigational drug will be considered as treatment-related AEs if they worsen after starting study treatment.

Serious adverse event:

Any adverse drug experience occurring at any dose that results in any of the following outcomes: Death, a life-threatening adverse drug experience, inpatient hospitalization or prolongation of existing hospitalization, a persistent or significant disability/incapacity, or a congenital anomaly/birth defect. Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered a serious adverse drug experience when, based upon appropriate medical judgment, they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition. For this trial, SAEs will be captured from the start of study treatment until completion of adjuvant treatment.

Unexpected adverse event:

Any adverse drug experience, the specificity or severity of which is not consistent with the current investigator brochure; or, if an investigator brochure is not required or available, the specificity or severity of which is not consistent with the risk information described in the general investigational plan or elsewhere in the current application, as amended.

Disability:

A substantial disruption of a person's ability to conduct normal life functions.

Life-threatening adverse drug experience:

Any adverse drug experience that places the patient or subject, in the view of the investigator, at immediate risk of death from the reaction as it occurred, i.e., it does not include a reaction that, had it occurred in a more severe form, might have caused death.

Unanticipated Problem:

An unanticipated problem is an adverse event that is (i) unexpected; (ii) serious; and (iii) felt by the investigator to be possibly, probably, or definitely related to the research intervention. Only adverse events that meet this definition need be reported to the IRB.

For more information on the definition of an unanticipated problem and reporting requirement, consult current PH&S AE Guidelines on the IRB website (http://phsnet.phsor.org/institutional_review_board).

9.3 Adverse Event Reporting

Adverse events will be reported from the time of the first dose through 90 days following cessation of treatment, or the initiation of new anti-cancer therapy, whichever is earlier.

Any serious adverse event, or follow up to a serious adverse event, that occurs to any subject from the time of the first dose through 90 days following cessation of treatment, or the initiation of new anti-cancer therapy, whichever is earlier, whether or not related to Merck/Brooklyn Immunotherapeutic product, must be reported within 24 hours to the Sponsor (via email at OREACRISAEreporting@providence.org) and within 2 working days to Merck Global Safety (Attn: Worldwide Product Safety; FAX 215-661-6229) and Brooklyn ITX (via email SAE@linical.accelovance.com, and safety@brooklynitx.com) via the MedWatch 3500 reporting form.

IRB:

An unanticipated event that is serious and definitely or probably caused by the study treatment (drugs or device) will be reported to the IRB of record in accordance with their guidelines and within their timelines.

FDA Reporting:

The Principal Investigator is responsible for determining whether or not the suspected adverse reaction meets the criteria for expedited reporting in accordance with Federal Regulations (21 CFR §312.32). A suspected adverse reaction must be both serious and unexpected in order to meet the reporting requirements.

An adverse event or suspected adverse reaction is considered “unexpected” if it is not listed in the investigator brochure or is not listed at the specificity or severity that has been observed;

or, if an investigator brochure is not required or available, is not consistent with the risk information described in the general investigational plan or elsewhere in the current application, as amended.

An adverse event or suspected adverse reaction is considered "serious" if, in the view of either the investigator, it results in any of the following outcomes: Death, a life-threatening adverse event, inpatient hospitalization or prolongation of existing hospitalization, a persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions, or a congenital anomaly/birth defect. Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered serious when, based upon appropriate medical judgment, they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition.

The timeline for submitting an IND safety report to FDA is no later than 15 calendar days after the Investigator determines that the suspected adverse reaction qualifies for reporting (21 CFR 312.32(c)(1)).

Any unexpected fatal or life-threatening suspected adverse reaction will be reported to FDA no later than 7 calendar days after the Investigator's initial receipt of the information (21 CFR 312.32(c)(2)).

Any relevant additional information that pertains to a previously submitted IND safety report will be submitted to FDA as a Follow-up IND Safety Report without delay, as soon as the information is available (21 CFR 312.32(d)(2)).

9.4 Continuing Review and Final Reports

An annual progress report (continuing review) will be submitted to the IRB for the duration of the study. A final report to the IRB will be submitted at the summation of the study. Safety and efficacy reports will be submitted to Merck and Brooklyn Immunotherapeutics regularly once enrollment commences.

9.5 Protocol Amendments

Once the protocol has been approved by the IRB, any changes to the protocol must be documented in the form of an amendment. The amendment must be approved by IRB prior to implementation. If it becomes necessary to alter the protocol to eliminate an immediate hazard to patients, an amendment may be implemented prior to IRB approval. In this circumstance, however, the Investigator must then notify the IRB in writing within five (5) working days after implementation from the IRB will be forwarded to the FDA.

9.6 Record Retention

The Principal Investigator is required to maintain adequate records of the disposition of the drug, including dates, quantity, and use by subjects, as well as written records of the disposition of the drug when the study ends.

The Principal Investigator is required to prepare and maintain adequate and accurate case histories that record all observations and other data pertinent to the investigation on each individual administered the investigational drug or employed as a control in the investigation. Case histories include the case report forms and supporting data including, for example, signed and dated consent forms and medical records including, for example, progress notes of the physician, the individual's hospital chart(s), and the nurses' notes. The case history for each individual shall document that informed consent was obtained prior to participation in the study.

Study documentation includes all CRFs, data correction forms or queries, source documents, Sponsor-Investigator correspondence, monitoring logs/letters, and regulatory documents (e.g., protocol and amendments, IRB correspondence and approval, signed patient consent forms). Source documents include all recordings of observations or notations of clinical activities and all reports and records necessary for the evaluation and reconstruction of the clinical research study.

In accordance with FDA regulations, the investigator shall retain records for a period of 2 years following the date a marketing application is approved for the drug for the indication for which it is being investigated; or, if no application is to be filed or if the application is not approved for such indication, until 2 years after the investigation is discontinued and FDA is notified.

9.7 Data Management

The Principal Investigator and/or his/her designee, will prepare and maintain adequate and accurate participant case histories with observations and data pertinent to the study. Study specific Case Report Forms (CRFs) will document safety and treatment outcomes for safety monitoring and data analysis. All study data will be entered into Velos eResearch via eCRFs. The Clinical Research Associate (CRA) will complete the CRFs as soon as possible upon completion of the study visit; the Investigator will review and approve the completed CRFs.

The information collected on CRFs shall be identical to that appearing in original source documents. Source documents will be found in the patient’s electronic medical records. All printed source documentation should be kept in separate research folders for each patient.

10.0 LIST OF REFERENCES

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APPENDICES

10.1 ECOG Performance Status Scale

Grade	Description
0	Fully active, able to carry on all pre-disease performance without restriction
1	Restricted in physically strenuous activity but ambulatory and able to carry out work of a light or sedentary nature, e.g., light house work, office work
2	Ambulatory and capable of all self-care but unable to carry out any work activities; up and about more than 50% of waking hours
3	Capable of only limited self-care; confined to bed or chair more than 50% of waking hours
4	Completely disabled; cannot carry on any self-care; totally confined to bed or chair
5	Dead
Oken M, Creech R, Tormey D, et al. Toxicity and response criteria of the Eastern Cooperative Oncology Group. *Am J Clin Oncol.* 1982;5:649-655. http://ecog-acrin.org/resources/ecog-performance-status	

10.2 Common Terminology Criteria for Adverse Events V5.0 (CTCAE)

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

10.3 Abbreviations

Abbreviation/ Term	Definition
AC	Doxorubicin + Cyclophosphamide
ACT	Doxorubicin + Cyclophosphamide + Paclitaxel
ADA	Anti-Drug Antibodies
AE	Adverse Event
AJCC	American Joint Committee on Cancer
ALT	Alanine Aminotransferase
ASaT	All Subjects as Treated
ASCO	American Society of Clinical Oncology
AST	Aspartate Aminotransferase
aPTT	Activated partial thromboplastin time
AUC	Area under the curve
β-hCG	β-human Chorionic Gonadotropin
BCS	Breast Conservation Surgery
BRCA	Breast Cancer 1
CAP	College of American Pathologists
Cb	Carboplatin
CbK	Carboplatin + Pembrolizumab
CBC	Complete Blood Count
CD8+	Cluster of Differentiation 8 positive
CHF	Congestive Heart Failure
CI	Confidence Interval
cN+	Palpable and/or sonographically suspicious lymph nodes
cN0	Node-negative disease;
CR	Complete Response
CRF	Case Report Form
CSR	Clinical Study Report
CT	Computed Tomography
CTCs	Circulating Tumor Cells
CTCAE	Common Toxicity Criteria for Adverse Events
ctDNA	circulating tumor DNA
CTLA-4	Cytotoxic T-lymphocyte-associated antigen-4
CTL	Circulating T Lymphocytes
DIC	Disseminated intravascular coagulation
DMC	Data Monitoring Committee
DNA	Deoxyribonucleic acid
EC	Epirubicin + cyclophosphamide
ECG	Electrocardiogram
ECHO	Echocardiogram
ECI	Events of Clinical Interest
ECOG	Eastern Cooperative Oncology Group
EDC	Electronic Data Capture
eCRF	Electronic Case Report Form
EOC	Executive Oversight Committee
EFS	Event-free Survival
EGFR	Epidermal Growth Factor Receptor
EORTC	European Organisation for Research and Treatment of Cancer
EORTC QLQ-BR23	European Organisation for Research and Treatment of Cancer Breast Cancer–Specific Quality of Life Questionnaire

Abbreviation/ Term	Definition
EORTC QLQ-C30	European Organisation for Research and Treatment of Cancer Quality of Life Questionnaire Core 30
ePRO	Electronic Patient Reported Outcomes
EQ-5D™	EuroQol-5 Dimension Questionnaire
ER	Estrogen Receptor
ERC	Ethics Review Committee
FA	Final Analysis
FBR	Future Biomedical Research
FDA	Food and Drug Administration
FDAAA	Food and Drug Administration Amendments Act
FFPE	Formalin-fixed Paraffin Embedded
FTV	Functional Tumor Volume
GCP	Good Clinical Practice
HBsAg	Hepatitis B surface Antigen
HCV	Hepatitis C Virus
HER2	Human Epidermal Growth Factor Receptor 2
HIV	Human Immunodeficiency Virus
HNSCC	Head and neck squamous cell cancer
HR	Hazard Ratio
IA	Interim Analysis
IB	Investigator's Brochure
ICF	Informed Consent Form
ICH	International Conference on Harmonization
IgG	Immunoglobulin
IHC	Immunohistochemistry
INR	International Normalized Ratio
irAEs	Immune-related Adverse Events
IRB	Institutional Review Board
ITIM	Immunoreceptor tyrosine-based inhibition motif
ITSM	Immunoreceptor tyrosine-based switch motif
IV	Intravenous
IVRS	Interactive Voice Response System
IWRS	Integrated Web Response System
K	Pembrolizumab
KAC	Pembrolizumab + doxorubicin + cyclophosphamide
KCb	Pembrolizumab + carboplatin
KEC	Pembrolizumab + epirubicin + cyclophosphamide
KN	KEYNOTE
KX	Pembrolizumab + paclitaxel
LMP	Last Menstrual Period
LVEF	Left ventricular ejection fraction
M0	non-metastatic
mAb	Monoclonal antibody
MAH	Marketing Authorisation Holder
miRNA	microRNA
MRI	Magnetic Resonance Imaging
mRNA	Messenger Ribonucleic Acid
mTNBC	Metastatic Triple-negative Breast Cancer
MUGA	Multigated Acquisition
N	Node
NACT	Neoadjuvant chemotherapy

Abbreviation/ Term	Definition
NCI	National Cancer Institute
NKC	Natural Killer Cell
NMR	Nuclear Magnetic Resonance
NSCLC	Non-Small Cell Lung Cancer
NYHA	New York Heart Association
ORR	Objective Response Rate
OS	Overall Survival
OTC	Over-the-counter
pCR	Pathological Complete Response
PD	Progressive Disease
PD-1	Programmed cell death protein 1
PD-L1	Programmed Death - Ligand 1
PD-L2	Programmed Death - Ligand 2
PIN	Personal Identification Number
PK	Pharmacokinetic
PR	Partial Response
PT	Prothrombin Time
PTT	Partial thromboplastin time
PTEN	Phosphatase and Tensin Homolog
Q2W	Every 2 Weeks
Q3W	Every 3 Weeks
QoL	Quality of Life
RCB	Residual Cancer Burden
RECIST	Response Evaluation Criteria in Solid Tumors
RNA	Ribonucleic Acid
SAC	Scientific Advisory Committee
SAE	Serious Adverse Events
SAP	Statistical Analysis Plan
SD	Stable Disease
SHP	Src homology phosphatase
SOP	Standard Operating Procedures
sSAP	Supplemental statistical analysis plan
SmPC	Summary of Product Characteristics
T	Tumor
T3	Total triiodothyronine
T4	Free thyroxine
TCR	T Cell Receptor
TILs	Tumor-Infiltrating T Lymphocytes
TNBC	Triple-negative Breast Cancer
TSH	Thyroid Stimulating Hormone
ULN	Upper Limit of Normal
WBC	White Blood Cell
X	Paclitaxel
yCN0	Node-negative disease after chemotherapy

11.0 SIGNATURES

I agree to conduct this clinical trial in accordance with the design outlined in this protocol and to abide by all provisions of this protocol (including other manuals and documents referenced from this protocol). I agree to conduct the trial in accordance with generally accepted standards of Good Clinical Practice. I also agree to report all information or data in accordance with the protocol and, in particular, I agree to report any serious adverse events as defined in Section 7.0 – TRIAL PROCEDURES (Adverse Event Monitoring). I also agree to handle all clinical supplies provided by the Sponsor and collect and handle all clinical specimens in accordance with the protocol. I understand that information that identifies me will be used and disclosed as described in the protocol, and that such information may be transferred to countries that do not have laws protecting such information. Since the information in this protocol and the referenced Investigator's Brochure is confidential, I understand that its disclosure to any third parties, other than those involved in approval, supervision, or conduct of the trial is prohibited. I will ensure that the necessary precautions are taken to protect such information from loss, inadvertent disclosure or access by third parties.

Coordinating Site Lead Principal Investigator

PRINTED NAME	David Page, MD
TITLE	Assistant Member, Earle A. Chiles Research Institute, Providence Portland Medical Center
SIGNATURE	
DATE SIGNED	

Participating Site Principal Investigator

PRINTED NAME	
TITLE	
SIGNATURE	
DATE SIGNED	