Phase Ib/II Study of Neoadjuvant Pembrolizumab (MK-3475) with Gemcitabine-Cisplatin (cisplatin eligible) or Gemcitabine (cisplatin-ineligible) in Subjects with T2-4aN0M0 Urothelial Cancer: HCRN GU14-188

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I confirm I have read this protocol, I understand it, and I will work according to this protocol and to the ethical principles stated in the latest version of the Declaration of Helsinki, the applicable guidelines for good clinical practices, or the applicable laws and regulations of the country of the study site for which I am responsible, whichever provides the greater protection of the individual. I will accept the monitor's overseeing of the study. I will promptly submit the protocol to applicable ethical review board(s).

_____ _____
Signature of Site Investigator Date

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Site Investigator Name (printed)

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Site Investigator Title

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Name of Facility

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Location of Facility (City and State)

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SYNOPSIS

TITLE	Phase Ib/II Study of Neoadjuvant Pembrolizumab with Gemcitabine-Cisplatin (cisplatin-eligible) or Gemcitabine (cisplatin-ineligible) in subjects with T2-4aN0M0 Urothelial Cancer: HCRN GU14-188
SHORT TITLE	Study of neoadjuvant pembrolizumab in combination with gemcitabine based therapy in cis-eligible or -ineligible subjects with urothelial cancer
PHASE	Phase Ib/II
OBJECTIVES	Primary Objective: • Phase Ib: safety of pembrolizumab in combination with gemcitabine-cisplatin • Phase II: rate of pathologic muscle invasive response (PaIR), *i.e.* ypT0, Tis, Ta, T1 ypN0 at radical cystectomy. Secondary Objectives: • Determine the 18month-relapse free survival (RFS) and overall survival (OS) • To evaluate the safety and tolerability of pembrolizumab in combination with gemcitabine or gemcitabine-cisplatin. • Determine the radical cystectomy (RC) rate in subjects who are cisplatin–eligible and –ineligible Exploratory Objectives: • Assess antitumor activity by PD-L1 immunohistochemistry (IHC) grade and correlate with 18mo-RFS for possible predictive or prognostic biomarker development. • To assess prognostic and predictive associations between pre and post-treatment changes in peripheral blood mononuclear cells (PBMCs), lymphocyte subsets, with 18 month-RFS. • To correlate responses to bladder cancer subtype of basal or luminal (and possibly p-53 like, though methodology for determining this phenotype in FFPE tissue is in progress)[1] • To correlate pathologic response to prior bacillus Calmette-Guerin (BCG) exposure.
STUDY DESIGN	Each subject will participate in the trial for up to 2 years from the time the subject signs the Informed Consent Form (ICF) through the final contact. After a screening of up to 28 days, cohort I cisplatin eligible subjects will begin phase Ib of the protocol to assess safety of the combination. Treatment is Days 1 and 8 of each 3-week dosing cycle. Treatment will continue for 4 cycles of gemcitabine-cisplatin (GC). The experimental intervention, pembrolizumab, will be given every 3 weeks, starting C1D8, for 5

	doses. Once the maximum tolerated dose (MTD) is determined in Phase Ib, cohort I will be treated in the same manner in Phase II with a primary endpoint of efficacy. Once Phase II opens, it will also start accruing cisplatin ineligible (though cystectomy eligible), subjects on "cohort II" with treatment of weekly gemcitabine (G) x3, on and every 28 day cycle for 3 cycles. Pembrolizumab is the same as in cohort I: starts on C1D8 and continues every 3 weeks for 5 doses. Subjects will then have surgery to remove their primary tumor within 2-7 weeks after their last dose of neoadjuvant therapy. The primary Phase II endpoint of PaIR (ypT0, Tis, Ta, T1 ypN0) will be determined. Subjects will then be followed for an additional 18 months with restaging scans every 12 weeks.
ESTIMATED NUMBER OF SUBJECTS	Approximately 81 subjects will be enrolled
PARTICIPATING CENTERS	Up to 8 centers in the HCRN
KEY ELIGIBILITY CRITERIA	**Subject Inclusion Criteria** 1. Have histologically confirmed muscle invasive disease of the urinary bladder, renal pelvis, or ureters. For subjects who have tumors limited to the upper tract including renal pelvis or ureters, muscle invasive disease does not need to be pathologically proven, and a CT urogram must be performed (MRI is not acceptable to meet this criterion). To be eligible, subjects with upper tract tumors of the renal pelvis and ureter(s) must meet a high risk assessment defined as: tumor ≥1cm *and/or* hydronephrosis *and/or* high grade pathology *and/or* multifocal disease, where a radical NU approach to treat localized disease is warranted.[2] 2. Histology must be urothelial carcinoma or urothelial carcinoma with mixed histology. 3. Clinical stage cT2-4aN0M0. See exclusion criteria for acceptable N0 determination / lymph node size. 4. Have surgical intent for radical cystectomy (RC) or nephroureterectomy (NU) must be documented by treating urologist. 5. Have an archived tumor block available to submit unstained slides for PD-L1 expression, basal and luminal subtype analysis- MANDATORY. For subjects without this, a biopsy prior to starting therapy is strongly encouraged. (See Study

	Procedures Manual for collection, labeling and shipping instructions.) 6. Demonstrate adequate organ function as defined in 7. Table 2, all screening labs should be performed within 28 days of study registration. **Cohort I – Cisplatin eligible** In addition to the inclusion criteria listed above, Cohort I subjects must satisfy **all** of the following criteria: • Glomerular filtration rate (GFR) or creatinine clearance (Ccr) ≥ 50 mL/min. (24 hour urine preferred). The cisplatin dose will be split over two days for values between 50-59 mL/min • ECOG PS 0, 1 (and not 2) • Hearing impaired ≤ grade 1 (may or may not be enrolled in a monitoring program) • Peripheral neuropathy ≤ grade 1 **Cohort II – Cisplatin *Ineligible*** In addition to the inclusion criteria listed above, Cohort II subjects must also meet any **one** of the following criteria: • GFR or Ccr: 30-49 (24 hour urine preferred). OR • ECOG PS 2 OR • Hearing impaired ≥ grade 2 as assessed by treating physician (may or may not be enrolled in a monitoring program). OR • Peripheral neuropathy of Grade 2-4 See full exclusion criteria for further details **Subject Exclusion Criteria** The subject must be excluded from participating in the trial if the subject: 1. Is not a surgical candidate. 2. Has abdomino-pelvic short axis lymph node of ≥15mm without biopsy. See full exclusion criteria for further details. 3. Has a diagnosis of immunodeficiency or received systemic steroid therapy of a prednisone equivalent of greater than 10mg within 7 days prior to study registration. Subjects on steroids for physiologic replacement due to a non-cancer related cause would not be excluded. 4. Has had a prior monoclonal antibody within 28 days prior to study registration or who has not recovered (i.e., ≤ Grade 1 or at baseline) from adverse events due to agents administered more than 28 days earlier. 5. Has a known additional malignancy that is progressing or requires active treatment ≤ 48 months of study registration.

	Exceptions include basal cell carcinoma of the skin, squamous cell carcinoma of the skin, or in situ cervical cancer that has undergone potentially curative therapy. See full exclusion criteria for further details. 6. Has active New York Heart Association (NYHA) Stage III/IV heart failure. See full exclusion criteria for further details. 7. Has known active central nervous system (CNS) metastases and/or carcinomatous meningitis. 8. Has an active autoimmune disease requiring systemic treatment within the past 3 months or a documented history of clinically severe autoimmune disease, or a syndrome that requires systemic steroids or immunosuppressive agents. See full exclusion criteria for further details. 9. Has evidence of interstitial lung disease 10. Has a history of (non-infectious) pneumonitis that required steroids, or currently has pneumonitis. 11. Has an active infection requiring systemic therapy. 12. Has received prior therapy with an anti-PD-1, anti-PD-L1, anti-PD-L2, anti-CD137, or anti-Cytotoxic T-lymphocyte-associated antigen-4 (CTLA-4) antibody (including ipilimumab or any other antibody or drug specifically targeting T-cell co-stimulation or checkpoint pathways). 13. Has a known history of Human Immunodeficiency Virus (HIV) (HIV 1/2 antibodies). 14. Has known active Hepatitis B (e.g., HBsAg reactive) or Hepatitis C (e.g., HCV RNA [qualitative] is detected). 15. Has received a live vaccine within 30 days prior to study registration. See full exclusion criteria for further details.
STATISTICAL CONSIDERATIONS	**Phase Ib**: This is a 3+3 design in the cisplatin-eligible group only. Two pembrolizumab dose levels of 200mg, and 120mg will be investigated with gemcitabine/cisplatin. The MTD will be the highest Pembrolizumab dose level in combination with gemcitabine/cisplatin at which no more than 1 of 6 subjects experience a dose limiting toxicity (DLT) related to pembrolizumab (and not of gemcitabine or cisplatin). Once the MTD is established, this portion of the study will close and the Phase II will open to cohort I and II at the recommended phase II dose (RP2D). Including a ~4% subject drop out, the maximal sample size is 14. **Phase II**: Cohort I and II subjects will be treated at RP2D from the phase 1b portion in independent Simon two-stage designs. For cohort I, the null hypothesis that the true pathologic muscle invasive response rate (PaIR) is 23% will be tested against a one-sided alternative. In the first stage, 15 subjects will be accrued. If

	there are 4 or fewer responses in these 15 subjects, cohort I will be stopped. Otherwise, 17 additional subjects will be accrued for a total of 32. The null hypothesis will be rejected if 12 or more responses are observed in 32 subjects. This yields a type I error rate of 4% and power of 86% when the true response rate is 48%. For cohort II, the null hypothesis that the true PaIR rate is 18% will be tested against a one-sided alternative. In the first stage, 19 subjects will be accrued. If there are 4 or less responses in these 19 subjects, the cohort will be stopped. Otherwise, 16 additional subjects will be accrued for a total of 35. The null hypothesis will be rejected if 11 or more responses are observed in 35 subjects. This yields a type I error rate of 4% and power of 86% when the true response rate is 40%.
ESTIMATED ENROLLMENT PERIOD	Estimated 16 months
ESTIMATED STUDY DURATION	Estimated 36 months

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SCHEMA

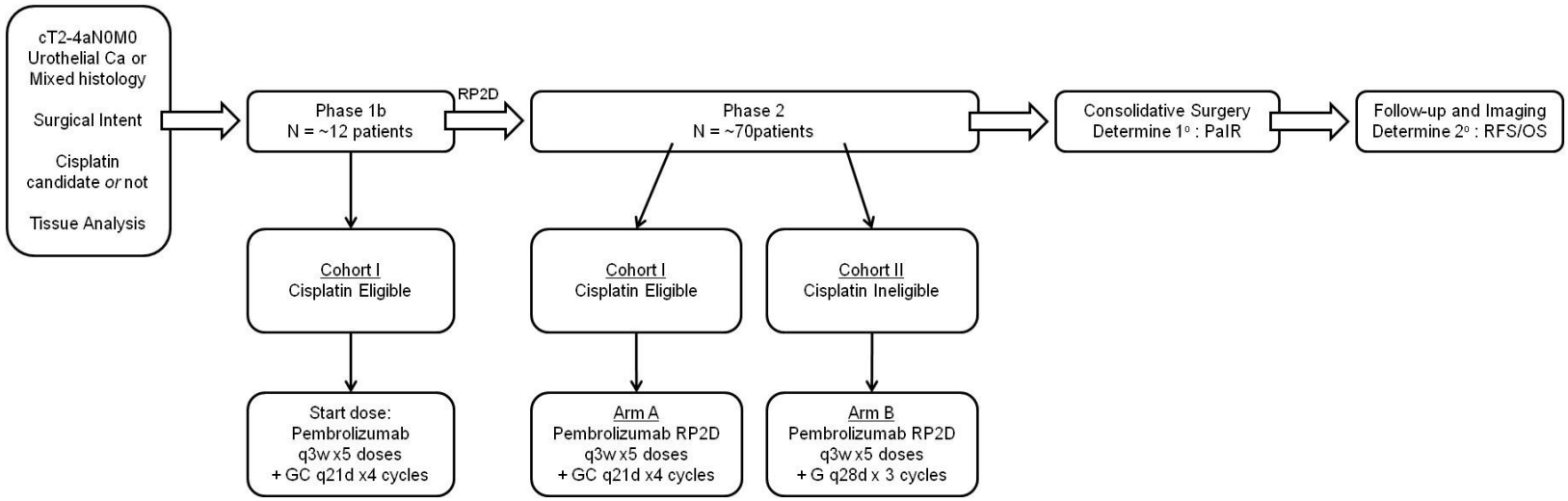


Figure 1. Schema of HCRN GU14-188 trial: neoadjuvant pembrolizumab in combination with gemcitabine based therapy in cisplatin-eligible or -ineligible subjects with urothelial cancer. RP2D, recommended phase II dose; GC, gemcitabine-cisplatin; G, gemcitabine; PaIR, pathologic muscle invasive response or p≤T1; RFS, relapse free survival; OS, overall survival

1. BACKGROUND AND RATIONALE

1.1. Background

1.1.1. Pharmaceutical and Therapeutic Background

The importance of intact immune surveillance in controlling outgrowth of neoplastic transformation has been known for decades[3]. Accumulating evidence shows a correlation between tumor-infiltrating lymphocytes (TILs) in cancer tissue and favorable prognosis in various malignancies[4-8]. In particular, the presence of $CD8^+$ T-cells and the ratio of $CD8^+$ effector T-cells / FoxP3+ regulatory T-cells seems to correlate with improved prognosis and long-term survival in many solid tumors.

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 Ig superfamily member related to CD28 and CTLA-4 which has been shown to negatively regulate antigen receptor signaling upon engagement of its ligands (PD-L1 and/or PD-L2) [9,10]. The structure of murine PD-1 has been resolved[11]. PD-1 and family members are type I transmembrane glycoproteins containing an Ig 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 SHP-1 and SHP-2 to the ITSM motif within its cytoplasmic tail, leading to the dephosphorylation of effector molecules such as CD3ζ, PKCθ and ZAP70 which are involved in the CD3 T-cell signaling cascade[9,12-14]. The mechanism by which PD-1 down modulates T-cell responses is similar to, but distinct from that of CTLA-4 as both molecules regulate an overlapping set of signaling proteins[15,16]. 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[17,18]. Expression has also been shown during thymic development on $CD4^-CD8^-$ (double negative) T-cells as well as subsets of macrophages and dendritic cells[19]. The ligands for PD-1 (PD-L1 and PD-L2) are constitutively expressed or can be induced in a variety of cell types, including non-hematopoietic tissues as well as in various tumors[15,20-22]. Both ligands are type I transmembrane receptors containing both IgV- and IgC-like domains in the extracellular region and contain 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-L1 is expressed at low levels on various non-hematopoietic tissues, most notably on vascular endothelium, whereas PD-L2 protein is only detectably expressed on antigen-presenting cells found in lymphoid tissue or chronic inflammatory environments. PD-L2 is thought to control immune T-cell activation in lymphoid organs, whereas PD-L1 serves to dampen unwarranted T-cell function in peripheral tissues[15]. Although healthy organs express little (if any) PD-L1, a variety of cancers were demonstrated to express abundant levels of this T-cell inhibitor. PD-1 has been suggested to regulate tumor-specific T-cell expansion in subjects with melanoma (MEL)[23]. This suggests that the PD-1/PD-L1 pathway plays a critical role in tumor immune evasion and should be considered as an attractive target for therapeutic intervention.

Pembrolizumab (previously known as MK-3475, or SCH 900475) is a potent and highly selective humanized monoclonal antibody (mAb) of the IgG4/kappa isotype designed to directly block the interaction between PD-1 and its ligands, PD-L1 and PD-L2. It received accelerated FDA approval for second line treatment of metastatic melanoma in September 2014.

1.1.2.Preclinical and Clinical Trial Data

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

1.2. Rationale

1.2.1. Rationale for the Trial and Subject Population

Bladder cancer is the second most common genitourinary (GU) malignancy in the US and the most common histology is urothelial carcinoma (UC). Distant recurrences occur in 20-50% of patients compared to 5-15% locoregional recurrences, and once metastases occur, the 5-year OS is 5.4%. Neoadjuvant chemotherapy followed by radical cystectomy (RC) with pelvic lymph node dissection is the current optimal treatment approach for Stage II or III disease based on level 1 evidence. Patients that achieve a pathologic muscle invasive response (PaIR) – defined as p≤T1 (and N0M0) disease at RC – have the best chance for cure.[24]

Neoadjuvant therapy for stage II or III muscle invasive urothelial cancer includes methotrexate, vinblastine, doxorubicin, cisplatin (MVAC)[22,23] or Gemcitabine-Cisplatin (GC) as extrapolated from data in the metastatic setting[24-26] and one prospective trial[27],[24,25] or Gemcitabine-Cisplatin (GC) as extrapolated from data in the metastatic setting[26-28] and one prospective trial[29]. Though these regimens remain the standard 1st line option, the survival benefit is limited for non-PaIR patients[30]. Despite many attempts at improving this outcome with single agents, and some pathway targeting agents; no agents have been proven to improve the modest outcomes of the GC and MVAC regimens. Bladder cancer has a high mutation burden[31] that can confer preserved signaling of driver pathways despite cytotoxic chemotherapy or targeted agents. A strategy that incorporates immune checkpoint blockade can potentially improve both recognition and targeted destruction of these neo-antigen expressing cells.

Bladder cancer is an immunogenic cancer that has been shown to be infiltrated by T regulatory cells (Tregs), express high levels of inhibitory cytokines, high prevalence of B7-H1 (PD-L1) expression[32], has survival outcomes that correlate with PD-L1 expression[33], and notably, respond to a broad lymphocytic inflammatory response to bacillus calmette-guerin (BCG) and BCG-interferon. Pembrolizumab blocks the ligation of PD-1 with PD-L1 or PD-L2, a key co-regulator interaction that can shift the balance of the immune system towards an anti-tumor response.

The motivating rationale for this combination trial is therefore based on 1) improved release of tumor antigen, specifically, urothelial cancer's high neo-antigen burden, using cytotoxic chemotherapy and 2) breaking immune tolerance and allowing renewed host patient immune surveillance for antigenic processing and immune mediated tumor cell death. Gemcitabine is cytotoxic to bladder cancer and can thereby cause tumor antigen release, reduces MDSCs to enhance immune surveillance[47], and also prevents PD-1 dependent $CD4^+$ T cell tolerization[50], which make it an attractive choice to study in combination with pembrolizumab. The hypothesis being tested in this trial is that the anti-PD-1 antibody, pembrolizumab, plus a gemcitabine

backbone (alone or with cisplatin) is a well tolerated neoadjuvant combination that can lead to a high rate of deep and durable responses in UC.

1.2.2. Rationale for inclusion of cisplatin-ineligible subjects

Pembrolizumab is well tolerated with a different side effect profile than MVAC or gemcitabine/cisplatin (GC)[34] and is therefore an attractive compound to also evaluate in the cisplatin-unfit population[35]. UC is unfortunately a disease of the elderly and due to age-associated and disease-associated comorbidities, over 50% of patients are unfit for cisplatin or doxorubicin[35-38] _ENREF_32. Common reasons to avoid doxorubicin and/or cisplatin include renal dysfunction, performance status of 2, grade 2 or greater hearing loss, neuropathy, or heart failure. Though nearly half of patients older than 70 years old have a Ccr of <50 mL/min _ENREF_27and may not be represented well in clinical trials[25,39], retrospective data in bladder cancer patients does show that cisplatin is tolerated similarly despite age, so long as they are otherwise cisplatin eligible[40]. Substituting carboplatin for cisplatin in the neoadjuvant setting has been evaluated in 3 prospective phase II trials: M-CAVI (methotrexate, carboplatin, vinblastine), and 2 trials of combination carboplatin-paclitaxel-gemcitabine (GCT). The M-CAVI regimen treated 47 T2-4N0 bladder cancer patients and found pT0 rates were 26.5% and disease specific survival of 42% at 2years[41]. In a phase II trial of 68 patients with adequate renal function and cT2-4N0-1 disease, those who received GCT had a pT0 rate of 22% of the intent to treat population. Unfortunately 79% had grade 3-4 hematologic toxicity[42]. SWOG conducted a phase II trial of neoadjuvant GCT followed by cystoscopic surveillance or immediate RC for cT0 status after therapy[43]. Of 74 patients, cT0 was achieved in 34 (46%), - and of these 34, only 10 went on to have RC. Of these 10, 6 (60%) actually had persistent cancer in their RC specimen despite cT0 status. In summary, while these 3 trials did replace cisplatin with carboplatin, they did not actually target or include the cisplatin-ineligible patients and failed to show sufficient safety or efficacy with these combinations.

Neoadjuvant carboplatin/gemcitabine (CaG) was evaluated in a single institution retrospective cohort of cisplatin-ineligible bladder cancer patients with cT2-T4 and/or node positive disease and were compared to a contemporary group who received a cisplatin combination regimen. Of the 41 patients included in the CaG cohort, 26.8% had crossed over from up to 3 cycles of a prior cisplatin-containing regimen to then finish neoadjuvant with the CaG regimen. No survival advantage was noted.

RC alone has been reviewed in a retrospective cohort of ~1000 patients and found a pathologic complete response (pCR) rate of 6.3%. A prospective evaluation of single modality RC pCR rate was determined to be 15% in the comparator arm of the Phase III SWOG 8710 trial[24,44]. These patients had prior diagnostic (and occasionally maximal) transurethral resection of bladder tumor (TURBT) to yield a pCR rate of ~15% at RC. It's important, however, to understand this rate should only be viewed within the context of being a comparator group against which a difference with neoadjuvant therapy was assessed within the pre-specified controls of the clinical trial. A pathologic CR rate for RC-only would require a clinical trial that controls for extent of TURBT prior to RC, and reproducible pathologic specimen sectioning (compulsive, detailed sectioning of whole-mounts may reveal Tis). Thus, given the available data, a large retrospective cohort may actually be a more relevant pCR rate for RC only groups.

Single agent gemcitabine has activity in invasive urothelial cancer. It has been studied in one phase I trial, 2 Phase II trials of previously treated patients, and 2 trials in previously untreated patients in the advanced or metastatic setting. The phase I trial of 15 patients had one CR, and 3 PR[45]. The two phase II trials of previously treated patients demonstrated a response rate of 28 and 50%, respectively[46,47]. Two Phase II trials of previously untreated patients found an ORR of 28%- with three CRs in patients with liver metastases, and an ORR of 24.3%, respectively[47,48]. All trials used a weekly gemcitabine times three on a 4-week cycle. The single agent gemcitabine administration regimen for this trial of cohort II- cisplatin-ineligible patients was based on this experience.

In summary, there is a need for better outcomes in bladder cancer patients. The most critical need includes those patients that are ineligible to receive cisplatin where current guideline recommendations are for RC or clinical trial[37].

1.2.3. Rationale for combining pembrolizumab with gemcitabine and/or cisplatin:

Currently, non-metastatic UC is optimally treated with neoadjuvant cisplatin based therapy: gemcitabine-cisplatin (GC); or methotrexate-vinblastine-doxorubicin-cisplatin (MVAC), which have well known 'macro' effects on the immune system such as neutropenia and lymphopenia; however, their effects on tumor-specific immune responses are becoming better appreciated. Their ability to augment immune effector and suppressor cells and cytokine milieu can be taken advantage of to beneficially modulate pembrolizumab immune checkpoint therapy **(Table 1).**

Active Bladder Agent	**Immune effects**
Cisplatin	Class I HLA expression Inhibits STAT6 expression of PD-L2 (B7-DC) Enhance Fas/ICAM-1 expression for Ag specific CTL killing
Gemcitabine	Increased Ag presentation Inhibit and reduce myeloid derived suppressor cells Enhance Fas expression CD40 based T cell stimulation Prevent PD-1 dependent $CD4^+$ T-cell tolerization

Table 1: Immune modulating effects of cytotoxic chemotherapy for bladder cancer. [49-52] ENREF_43 DC, dendritic cell; HLA, human leukocyte antigen; Ag, antigen; CTL, cytotoxic T lymphocyte; MHC, major histocompatibility complex

There is preclinical biology as well as clinical support for the immunomodulating effects of bladder-specific neoadjuvant therapies including cisplatin, gemcitabine, vinblastine, doxorubicin, and methotrexate.

Cisplatin can stimulate antigen presentation, increase HLA expression, and improve factors for cytotoxic T lymphocyte kill. It also inhibits STAT6 expression of PD-L2 which has unknown effects on PD-1 inhibition and tumor surveillance, though likely does not impede it[50,51].

Gemcitabine has been shown *in vivo* and in clinical trials to favorably augment tumor specific $CD4^+$ and $CD8^+$ ratios and to induce responses through effects it also has on the B-lymphocytes.[53,54] _ENREF_8 Gemcitabine, reduces myeloid-derived suppressor cells (MDSCs) to enhance immune surveillance[49] and also prevents PD-1 dependent $CD4^+$ T cell tolerization[52]. Preclinical data with Gemcitabine in combination with pembrolizumab shows synergistic tumor inhibition compared to monotherapy in a variety of syngeneic mouse tumor models (MK-3475) IB ed.6 rev10Jan2014).

Several ongoing early phase trials combining immune checkpoint inhibitors with platinum doublet regimens in non-small cell lung cancer (NSCLC) and SCLC have had manageable toxicities (clinicaltrails.gov ID: NCT02039674, NCT01285609, NCT01454102)[55,56] _ENREF_43. In an interim report of the Phase I multi-arm trial of an anti-PD-1 inhibitor in combination with 3 different platinum doublet regimens for NSCLC, the GC combination arm showed good tolerability without any treatment discontinuations and grade 3-4 toxicity limited to 6% anemia[55].

Based on the SWOG neoadjuvant MVAC trial and recent neoadjuvant phase II trials of dose dense/accelerated MVAC[24,57,58] a rationale for potential beneficial immunomodulation by MVAC was explored. There are preclinical data that suggest vinblastine and cisplatin could support a pembrolizumab directed anti-tumor response. Methotrexate and doxorubicin, however, are more likely to have counterproductive effects when used in combination with pembrolizumab. Doxorubicin has been shown to induce expansion of monocytic myeloid suppressor cells which may interfere with anti-PD-1 therapy[52]. While methotrexate (MTX) exhibits some beneficial immune anti-tumor properties (such as DC-mediated antigen presentation), methotrexate also notably has anti-inflammatory effects that are mediated thru its primary mechanism of action of dihydrofolate reductase (DHFR) inhibition. As an anti-cancer agent, MTX's competitive inhibition of DHFR leads to depletion of reduced folates which are critical for nucleotide synthesis and cell survival. This same mechanism also benefits autoimmune and rheumatologic conditions by suppressing the inflammatory cascade. DHFR inhibition by MTX leads to *adenosine accumulation*, an important signaling molecule for inhibiting the inflammatory cascade via receptor A2a on T cells[59]. (The anti-inflammatory mechanisms of MTX may be mediated by additional non-adenosine pathways and are beyond the scope of this protocol.) Of specific relevance, is that adenosine is essential for T_{reg} mediated immune suppression[60] and acts via stimulating adenosine receptors on T effector cells which *increases expression* of negative co-stimulatory molecules such as PD-1 and CTLA-4[61,62] _ENREF_60. Pembrolizumab combination with MTX could require dose escalation or alternative scheduling regimens of pembrolizumab to overcome MTX's affects on PD-1 and, therefore, a MVAC/ddMVAC combination arm is not pursued as an intervention in this protocol.

Timing of cytotoxic chemotherapy in combination with immune checkpoint inhibitors is an important consideration for overall efficacy as well as toxicities. Often, steroids are given as supportive care with cisplatin-based regimens as an anti-emetic for up to 4 days after the cisplatin dose. Here we choose to time the delivery of the PD-1 inhibitor at day 8, to avoid overlap with dexamethasone and just before immune reconstitution from maximal myelosuppression, and, as important, the cisplatin dosing option for subjects with GFR 50-59 is to be split over day 1 and 2[63-66]. This is also a critical time as T-cell subsets emerge from

myelosuppression and can have a favorable anti-tumor milieu. Additionally, while this is a 'sensitive' period for hematopoiesis, and hematopoietic precursors do express PD-L1, there are no reports or preclinical data to suggest PD-1 signaling is essential to normal hematopoiesis and therefore would not predict dose-limiting cytopenia from this timing and combination.

1.2.4. Rationale for Timing of Consolidative Surgery

Timing of RC after neoadjuvant therapy is an important consideration for this multimodality intervention. Delivery of cystectomy within 10 weeks from the last dose of a combination neoadjuvant chemotherapy regimen of 9-12 weeks in duration was found to not compromise survival in a retrospective cohort of 153 patients[67]. This HCRN GU14-188 trial delivers experimental pembrolizumab therapy 3 weeks after the last dose of gemcitabine (G) or gemcitabine-cisplatin (GC), and therefore requires delivery of RC (or nephroureterectomy, NU) within 2 to 7 weeks after this dose.

1.2.5. Rationale for Dose Selection/Regimen/Modification

An open-label Phase I trial (Protocol 001) is currently being conducted to evaluate the safety and clinical activity of single agent pembrolizumab. The dose escalation portion of this trial evaluated three dose levels, 1 mg/kg, 3 mg/kg, and 10 mg/kg, administered every 2 weeks (Q2W) in subjects with advanced solid tumors. All three dose levels were well tolerated and no dose-limiting toxicities were observed. This first in human study of pembrolizumab showed evidence of target engagement and objective evidence of tumor size reduction at all dose levels (1 mg/kg, 3 mg/kg and 10 mg/kg Q2W). No MTD has been identified to date with 10 mg/kg Q2W as the highest dose tested in PN001. Recent data from other clinical studies within the pembrolizumab program has shown that a lower dose of pembrolizumab and a less frequent schedule may be sufficient for target engagement and clinical activity.

Pharmacokinetic (PK) data analysis of pembrolizumab administered Q2W and Q3W showed slow systemic clearance, limited volume of distribution, and a long half-life (refer to IB). Pharmacodynamic (PD) data (IL-2 release assay) suggested that peripheral target engagement is durable (>21 days). These early PK and PD data provide scientific rationale for tests that compared Q2W and Q3W dosing schedules.

A population pharmacokinetic analysis has been performed using serum concentration time data from 476 patients. Within the resulting population PK model, clearance and volume parameters of pembrolizumab were found to be dependent on body weight. The relationship between clearance and body weight, with an allometric exponent of 0.59, is within the range observed for other antibodies and would support both body weight normalized dosing or a fixed dose across all body weights. Pembrolizumab has been found to have a wide therapeutic range based on the melanoma indication. The differences in exposure for a 200 mg fixed dose regimen relative to a 2 mg/kg Q3W body weight based regimen are anticipated to remain well within the established exposure margins of 0.5 – 5.0 for pembrolizumab in the melanoma indication. The exposure margins are based on the notion of similar efficacy and safety in melanoma at 10 mg/kg Q3W vs. the proposed dose regimen of 2 mg/kg Q3W (i.e. 5-fold higher dose and exposure). The population PK evaluation revealed that there was no significant impact of tumor burden on exposure. In addition, exposure was similar between the NSCLC and melanoma indications. Therefore, there are no anticipated changes in exposure between different indication settings.

1.2.6. Combination Risk Assessment

Treatment with GC is a standard of care option for eligible subjects with T2-T4aN0M0 urothelial cancer. Myelosuppression/cytopenias is a dose limiting toxicity of gemcitabine; and nephrotoxicity and nausea are the most common and/or serious adverse events for cisplatin. The most common adverse event (AE) identified (~10%) to date for pembrolizumab are fatigue, nausea, and less commonly, flu-like symptoms and are often grade 1-2. Phase I trials did not reach a MTD as there were very few (<3%) reports of grade 3-4 AE. Most of these events occurred at the 10 m/kg dosing, prior to the PK and clinical efficacy data suggested a flat dose of 200 mg (this flat dose is ~5-fold lower than the 10 mg/kg dose for an average size adult, see section 1.2.5) to be the ideal monotherapy recommended phase II dose (RP2D). Hematologic toxicity has not been noted. As pembrolizumab toxicities are infrequent and mild (and were determined at a dose nearly 4-5-fold higher than the current RP2D monotherapy dose), there is very little expectation for overlapping toxicities.

1.2.7. Rationale for Endpoints

1.2.7.1. Efficacy Endpoints

The primary efficacy endpoint is pathologic muscle invasive response (PaIR), or ≤ypT1N0, rate at RC/NU with lymph node dissection. It has been shown to correlate with survival in subjects treated with neoadjuvant therapy and is the ideal immediate datapoint collected in this neoadjuvant trial. The pathologic staging will be assessed by the study site pathologist and compared to the initial clinical TNM (cTNM, Tumor Node Metastasis) at diagnosis (recorded at study entry) for primary efficacy endpoint analysis. CT scan (or MRI per treating physician) restaging will be obtained within 21 days prior to planned RC/NU and an additional cTNM stage will be determined by the site investigator. Recognizing that the primary efficacy endpoint is dependent on consistent cTNM staging, site investigators shall refer to section 9 (Table 9a) for determining cTNM stage at diagnosis (recorded at study entry) and again pre-surgery. Investigators may also refer to AJCC staging manual, 7th edition (2009) or the NCCN guidelines page ST-2 as an additional reference when considering how to distinguish cT2 (muscle invasion) from cT3 (bladder wall thickening) from cT4 (fixed mass on exam under anesthesia or extra-vesicle extension to perivesicle fat).[68,69]

Radiographic (CT or MRI) RECIST 1.1 (Section 9.7) will be used for evaluation of disease response over time.

1.2.7.2. Safety Endpoints

The safety primary endpoint in this study is the incidence of non-gemcitabine/cisplatin related DLTs observed in the DLT evaluation period. Adverse events and laboratory test values observed in this study are also safety endpoints. In addition to general laboratory tests, immune laboratory tests will be evaluated considering the mode of action of pembrolizumab.

1.2.7.3. Biomarker Research

PD-L1 is a ligand for PD-1 and pembrolizumab attempts to disrupt the interaction of two proteins. High expression of PD-L1 on tumor cells (and to a lesser extent of PD-L2) has been found to correlate with poor prognosis and survival in various cancer types, including UC, RCC, pancreatic carcinoma, hepatocellular carcinoma, ovarian carcinoma, and NSCLC. The

correlation between baseline PD-L1 expression levels and tumor response will be evaluated as an exploratory objective. PD-L1 expression levels will be measured in archival tumors or biopsy samples determined to be evaluable by central lab. Pre-treatment tissue samples for this analysis are mandatory, and when not available a biopsy should be considered if sufficient neoplastic tissue is thought to still be available.

a) PD-L1: correlate with response to PD-1 inhibition
   Source: FFPE- Mandatory submission of unstained slides from an archival biopsy at diagnosis or pretreatment biopsy. When not available, a biopsy should be considered if sufficient neoplastic tissue is thought to still be available.
   Metric: PD-L1 IHC staining 0-3/3
b) Basal or Luminal subtypes[1] – correlate with response to chemotherapy.
   Source: FFPE- Mandatory submission of unstained slides from an archival biopsy at diagnosis or pretreatment biopsy.
   Metrics: IHC: CD 44, CD20, CD 5/6

Selected Sites where possible will participate in additional tissue acquisition for analysis:

c) FACS of PBMCs
   Source: PBMCs- optional submission of cryopreserved PBMCs for analysis collected at pre-treatment: cycle 1 day 1, cycle 1 day 8, cycle 1 day 11-15 (preferably closer to day 11).
   Metrics: CD3, CD4, CD8, CD25, Foxp3, NK cell, and MDSC markers
d) Tissue associated lymphocytes for 'in silica' Immunoscore
   Source: optional submission of 2 to 3 fresh punch (≥2mm diameter) biopsies of luminal surface of the cystectomy specimen obtained within 60 minutes of cystectomy. Fresh frozen samples to be sent overnight.
   Metrics: Lymphocyte subset analysis using Affymetrix gene array.

2. STUDY OBJECTIVES & HYPOTHESES

2.1. Primary Objective & Hypothesis

Determine the safety and efficacy of neoadjuvant pembrolizumab (MK-3475) combination with gemcitabine-cisplatin or gemcitabine in subjects with cT2-T4aN0M0 urothelial cancer (UC). As pembrolizumab has been well tolerated and without overlapping toxicity, a short Phase Ib study will be done to ensure tolerability in the Gemcitabine/Cisplatin cohort ("cohort I"), which will then move to a phase II where the primary endpoint of pathologic muscle invasion response (PaIR, ≤ypT1N0) will be measured. PaIR is defined as the absence of residual muscle invasive cancer in the surgical specimen.

- The PaIR will be determined from standard-of-care RC or NU lymph node dissection and assessed by the site investigator and his/her pathologist at their respective study site.

This phase II study will follow a two-stage Simon design to test the null hypothesis in each cohort of cisplatin-eligible and cisplatin-ineligible subjects.

Phase Ib: Safety of pembrolizumab in combination with gemcitabine-cisplatin

Phase II: Rate of pathologic muscle invasive response (PaIR), *i.e.* ypT0, Tis, Ta, T1 ypN0 at RC with pelvic lymph node dissection.or NU with lymph node dissection.

Hypothesis: Pembrolizumab activity can be modulated by a gemcitabine containing regimen and will have efficacy in bladder cancer subjects in the neoadjuvant setting when compared to historical controls.

2.2. Secondary Objectives & Hypotheses

- Determine the pT0 rate, 18mo-relapse free survival (RFS) and overall survival (OS)
- Evaluate the safety and tolerability of pembrolizumab in combination with gemcitabine or gemcitabine-cisplatin.
- Determine the radical cystectomy (RC) rate in subjects who are cisplatin–eligible and –ineligible

Hypothesis/Rationale: Multimodality therapy is necessary to attain the best chances at long term overall survival for this population of subjects with urothelial cancer. Reasons for not proceeding to RC or NU may include both cancer and non-cancer related issues.

2.3. Exploratory Objectives

- Assess antitumor activity by PD-L1 immunohistochemistry (IHC) grade and correlate with 18mo-RFS for possible predictive or prognostic biomarker development.
- Assess prognostic and predictive associations between pre and post-treatment changes in peripheral blood mononuclear cells (PBMCs), and tissue -associated lymphocyte subsets, with 18 month-RFS.
- Correlate responses to bladder cancer subtype of basal or luminal (and possibly p-53 like, though methodology for determining this phenotype in FFPE tissue is in progress)[1]3.0
- Correlate pathologic response to prior bacillus Calmette-Guerin (BCG) exposure.

3 ELIGIBLITY CRITERIA

3.1 Diagnosis/Condition for Entry into the Trial

Subjects with cT2-4aN0M0 urothelial cancer over the age of 18 whom a urologist has assessed and documented to be an appropriate surgical candidate (within 40 days of trial registration) with intent for consolidative surgical management. Histology may be urothelial carcinoma (transitional cell carcinoma) *or* urothelial carcinoma with additional histologic features (such as squamous, adenocarcinoma, sarcomatoid, small cell, etc). Upper tract disease with plan for nephroureterectomy (NU) and lymph node dissection is also acceptable.

3.2. 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.

2. ≥ 18 years of age on day of signing informed consent.

3. ECOG PS ≤ 2; please see section 3.4 for specific details regarding ECOG PS for each cohort

4. Have histologically confirmed muscle invasive disease of the urinary bladder. For subjects who have tumors limited to the upper tract including renal pelvis or ureters, muscle invasive disease does not need to be pathologically proven, and a CT urogram must be performed (MRI is not acceptable to meet this criterion). To be eligible, subjects with upper tract tumors of the renal pelvis and ureter(s) must meet a high risk assessment defined as: tumor ≥ 1cm *and/or* hydronephrosis *and/or* high grade pathology *and/or* multifocal disease, where a radical NU approach to treat localized disease is warranted.[2]

5. Histology must be urothelial carcinoma (transitional cell carcinoma) *or* urothelial carcinoma with mixed histology/features.

6. Clinical stage cT2-4aN0M0. Please see exclusion criteria for acceptable N0 determination/lymph node size.

8. Have a surgical evaluation that documents the plan for multimodality therapy with a consolidative radical cystectomy or nephroureterectomy.
NOTE on surgical intent: Criteria for acceptable surgical risk are not defined and per treating urologist. *Minimum* guidance on surgical intent includes subjects who do not have significant cardiovascular disease such as NHYA class III or IV heart failure, unstable arrhythmias or angina, active CAD, and/or EF<25%. Specific diagnostic testing to determine surgical intent is not required and per treating urologist or oncologist discretion.

9. Having an archived tumor block available to submit 11 unstained slides for PD-L1 expression, basal and luminal subtype analysis is MANDATORY for subjects with bladder cancer (and optional for those with tumors limited to the upper tract if sufficient tissue is not available). If slides are not available, a biopsy is strongly encouraged to obtain tissue for submission (See Study Procedures Manual for collection, labeling and shipping instructions.)

10. Subjects on full dose anticoagulants must be on a stable regimen of warfarin or low molecular weight heparin (LMWH) for at least two weeks.

11. Demonstrate adequate organ function as defined in Table 2: All screening labs should be performed within 28 days of study registration.

Table 2: Adequate Organ Function Laboratory Values

System	Laboratory Value
Hematological	
Absolute neutrophil count (ANC)	≥1,500 /mcL
Absolute lymphocyte count	≥350 mcL
Platelets	≥100,000 / mcL
Hemoglobin	≥9 g/dL or ≥5.6 mmol/L
Renal	
Measured or calculated[a] creatinine clearance	≥30 mL/min
Hepatic	
Serum total bilirubin	≤ 1.25 X ULN **OR** ≤ 2.5 xULN for subjects with Gilbert's disease
AST (SGOT) and ALT (SGPT)	≤ 2 X ULN

System	Laboratory Value
Coagulation	
International Normalized Ratio (INR) or Prothrombin Time (PT)	≤1.5 X ULN unless subject is receiving anticoagulant therapy as long as PT or PTT is within therapeutic range of intended use of anticoagulants
Activated Partial Thromboplastin Time (aPTT)	≤1.5 X ULN unless subject is receiving anticoagulant therapy and as long as PT or PTT is within therapeutic range of intended use of anticoagulants

[a]Creatinine clearance is calculated per institutional standard formula.

12. Female subjects of childbearing potential must have a negative urine or serum pregnancy test within 72 hours prior to study registration. If the urine test is positive or cannot be confirmed as negative, a serum pregnancy test is required.

13. Female subjects of childbearing potential must be willing to adequate contraception, be surgically sterile, or abstain from heterosexual intercourse for the course of the study and through 120 days after the last dose of study medication. **NOTE:** Subjects of childbearing potential are those who have not been surgically sterilized or have not been free from menses for > 1 year.

14. Male subjects must agree to use a barrier method of male contraception starting with the first dose of study therapy and through 120 days after the last dose of study therapy.

3.3. Subject Exclusion Criteria

Subjects may not have any of the following:

1. A non-surgical approach recommended by the treating urologist due to any reason.
   a) Criteria for surgical intent are not defined and, rather, suitability is determined and documented by the subject's treating urologist. *Minimum guidance* on surgical intent includes subjects who do not have significant cardiovascular disease such as NHYA class III or IV heart failure, unstable arrhythmias or angina, active CAD, and/or EF<25%. Specific cardiopulmonary diagnostic testing to determine surgical intent is not required and per treating urologist or oncologist discretion.

2. Has abdomino-pelvic short axis lymph node of ≥ 15mm without biopsy. **NOTE**: A subject with a staging biopsy proving a non-neoplastic process/N0 *will* meet inclusion.

3. Subjects with disease that is limited to the upper tract urothelial cancer and is considered low risk defined as: unifocal disease *and* tumor size <1cm, *and* low grade cytology, *and* without an invasive aspect on CT-urography.[2]

4. Is currently participating in or has participated in a study of an investigational agent or using an investigational device within 28 days prior to study registration.

5. Has a diagnosis of immunodeficiency or received systemic steroid therapy of a prednisone equivalent of greater than 10mg within 7 days prior to study registration. Subjects on steroids for physiologic replacement due to a non-cancer related cause would not be excluded.

6. Has had a prior monoclonal antibody ≤ 28 days prior to study registration or who has not recovered (i.e., ≤ Grade 1 or at baseline) from adverse events due to agents administered more than 28 days earlier.

7. Has had prior chemotherapy, targeted small molecule therapy, or radiation therapy for urothelial carcinoma.

8. Has a known additional malignancy that is progressing or required treatment ≤ 48 months of study registration. Exceptions include basal cell carcinoma of the skin, squamous cell carcinoma of the skin, *in situ* cervical cancer that has undergone potentially curative therapy, stable (as defined by PSA change, checked within 30 days) and untreated very low-risk or low-risk prostate cancer defined by current NCCN guidelines.

9. Has known active central nervous system (CNS) metastases and/or carcinomatous meningitis. Brain imaging is not required and per discretion of treating physician.

10. Has an active autoimmune disease requiring systemic treatment within the past 3 months or a documented history of clinically severe autoimmune disease, or a syndrome that requires systemic steroids or immunosuppressive agents. **NOTE**: Subjects with vitiligo or resolved childhood asthma/atopy would be an exception. Subjects that require intermittent use of bronchodilators or local steroid injections would not be excluded from the study. Subjects with hypothyroidism stable on hormone replacement or Sjogren's syndrome will not be excluded from the study.

11. Has evidence of interstitial lung disease

12. Has a history of (non-infectious) pneumonitis that required steroids, or currently has pneumonitis.

13. Has an active infection requiring systemic therapy.

14. Has a history or current evidence of any condition, therapy, or laboratory abnormality that might confound the results of the trial, interfere with the subject's participation for the full duration of the trial, or is not in the best interest of the subject to participate, in the opinion of the site investigator.

15. Has known psychiatric or substance abuse disorders that would interfere with cooperation with the requirements of the trial.

16. Is pregnant or breastfeeding, or expecting to conceive or father children within the projected duration of the trial, starting with the pre-screening or screening visit through 120 days after the last dose of trial treatment.
17. Has received therapy with hematopoietic growth factor such as G-CSF or GM-CSF in the 14 days prior to registration.

18. Has received prior therapy with an anti-PD-1, anti-PD-L1, anti-PD-L2, anti-CD137, or anti-Cytotoxic T-lymphocyte-associated antigen-4 (CTLA-4) antibody (including ipilimumab or any other antibody or drug specifically targeting T-cell co-stimulation or checkpoint pathways).

19. Has a known history of Human Immunodeficiency Virus (HIV) (HIV 1/2 antibodies).

20. Has known active Hepatitis B (e.g., HBsAg reactive) or Hepatitis C (e.g., HCV RNA [qualitative] is detected).

21. Has received a live vaccine within 30 days prior to the first dose of trial treatment. **NOTE**: Examples of live vaccines include, but are not limited to, the following: measles, mumps, rubella, chicken pox, yellow fever, rabies, BCG, and typhoid (oral) vaccines. Seasonal influenza vaccines for injection are generally killed virus vaccines and are allowed; however intranasal influenza vaccines (e.g. Flu-Mist®) are live attenuated vaccines, and are not allowed.

3.4 Cohort Assignment

3.4.1 Cohort I – Cisplatin *eligible*

In addition to the inclusion criteria listed above in Sections 3.1 and 3.2, Cohort I subjects must satisfy all of the following criteria:

- GFR or Ccr ≥ 50 mL/min (24 hour urine preferred). The cisplatin dose will be split over two days for values between 50-59 mL/min. See section 5.5.1 and 5.5.4.
- ECOG PS 0, 1 (and not 2)
- Hearing impaired ≤ Grade 1 (may or may not be enrolled in a monitoring program)
- Peripheral neuropathy ≤ Grade 1.

3.4.2 Cohort II – Cisplatin *Ineligible*

In addition to the inclusion criteria listed above in Sections 3.1 and 3.2, Cohort II subjects must also be documented to have surgical intent for radical cystectomy (RC) or nephroureterectomy (NU) and also meet any one or more of the following criteria:

- GFR or Ccr: 30-49 (24 hour urine preferred). **OR**
- ECOG PS 2 **OR**
- Hearing impaired ≥ Grade 2 as assessed by treating physician (may or may not be enrolled in a monitoring program) **OR**
- Peripheral neuropathy Grade 2-4

4. SUBJECT REGISTRATION

All subjects must be registered through the Hoosier Cancer Research Network (HCRN) electronic data capture (EDC) system. A subject will be considered registered when they are given an "on study" date in the EDC system. Subjects must be registered prior to starting protocol therapy and begin therapy **within five business days** of registration.

5. TREATMENT PLAN

5.1. Overview

This is a two-part trial with a one-arm Phase Ib portion followed by a two-arm Phase II portion. The study treatment is stratified into two cohorts based on cisplatin eligibility:

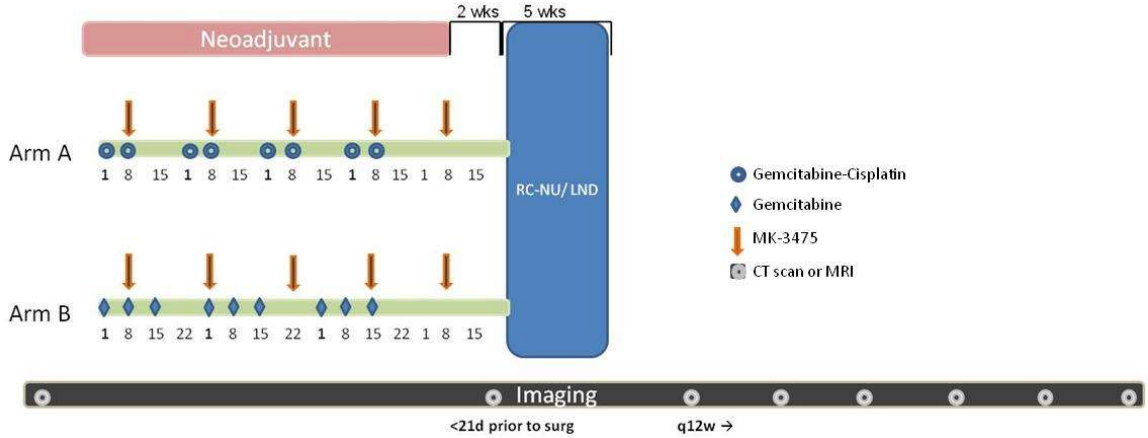
Phase Ib Cycle = 21 days:

- Cohort I only (receive treatment in Phase Ib and II): cisplatin eligible subjects receive gemcitabine/cisplatin every 3 weeks, repeated for 4 cycles. *Subjects with Ccr of 50-59 mL/min must follow split dosing of cisplatin over two days* (see section 5.5.1 and 5.5.4). Pembrolizumab will be given every 3 weeks for 5 doses. The starting dose will be 200 mg (Dose level 0). **NOTE:** the last dose of pembrolizumab falls on what would be day 8 of a 5th 'chemo' cycle, however gemcitabine/cisplatin is NOT GIVEN.

Phase II:

- Arm A; Cohort I; Cycle = 21 days: (receive treatment in Phase Ib and II): cisplatin eligible subjects receive gemcitabine/cisplatin every 3 weeks, repeated for 4 cycles. *Subjects with Ccr of 50-59 mL/min must follow split dosing of cisplatin over two days* (see Table 3). Pembrolizumab is given every 3 weeks for 5 doses starting with Cycle 1 Day 8 (C1D8). **NOTE:** the last dose of pembrolizumab falls on what would be day 8 of a 5th 'chemo' cycle, however gemcitabine/cisplatin is NOT GIVEN (figure 2).
- Arm B; Cohort II; Cycle = 28 days: (receive treatment in Phase II only): cisplatin ineligible subjects receive gemcitabine every week (weekly) for three consecutive weeks, repeated every 28 days for 3 cycles. Pembrolizumab is given every 3 weeks for 5 doses starting with Cycle 1 Day 8 (C1D8). **NOTE:** due to the timing of gemcitabine cycles every 4 weeks, and every three week dosing of pembrolizumab, there are two doses of pembrolizumab given during cycle 2: days 1 and 22. Additionally, the last dose of pembrolizumab falls on what would be day 8 of a 4th 'chemo' cycle; however gemcitabine is NOT GIVEN (figure 2).

Figure 2. Phase II Cisplatin Eligible Treatment Schema



5.2 Phase Ib: Dose Finding for pembrolizumab in cisplatin-eligible subjects

Cycle = 21 days

Cohort	Dose Level	MK-3475[1]	Gemcitabine	Cisplatin[2]	
				CLCr ≥60 mL/min	CLCr 50-59 mL/min
I	0	200mg IV q 3week	1000 mg/m^2 IV D1,8 q21d x4	70 mg/m^2 IV D1 q21d x4	35 mg/m^2 IV D1, 2 or D1,8 q21d x4
I	-1	120mg IV q 3week			

1: The last dose of pembrolizumab falls on what would be Cycle 5 Day 8, however gemcitabine-cisplatin is NOT GIVEN.
2: See section 5.5.4 regarding use of split-dose cisplatin. A Day 1 and 2 regimen is preferred over Day 1 and 8 though per treating physician discretion. Subjects with Ccr of ≥ 60 mL/min will receive cisplatin on Day 1 only.

5.2.1 Dose Finding Guidelines

A given dose cohort may be expanded up to a total of 12 subjects if further evaluation of the frequency of a given toxicity is warranted, based upon the observed safety profile in the 6 subjects already recruited in the cohort, and where the incidence of the confirmed DLT does not exceed 33%.

Number of subjects with DLT in dose-level cohort	Action
0 of 3 subjects	proceed to cohort expansion of an additional 3 subjects
1 of 3 subjects	accrue 3 additional subjects at current dose level for a total of 6 subjects
≤1 of 6 subjects	Proceed to Phase II
2 or more subjects in a dosing cohort (up to 6)	MTD has been exceeded. Reduce dose level and re-evaluate.

5.2.2 DLT Definition

The pembrolizumab DLT is defined as a drug-related AE occurring in the first 4 weeks of treatment of pembrolizumab (C1D8 – C2D14) and meeting one of the criteria listed below. Ascribing a DLT to a respective drug in this combination with gemcitabine-cisplatin requires attention to known DLTs and AEs. As myelosuppression is the recognized DLT of gemcitabine and an AE of cisplatin, hematologic toxicity during the Phase Ib should primarily follow the dose modification tables for gemcitabine and cisplatin in Section 6.0.

If any of these known AEs do occur, the recommended phase II dose (RP2D) will be discussed amongst HCRN, Merck, and the sponsor-investigator prior to finalizing the dose and treatment schedule:

- Grade 4 febrile neutropenia with vasopressor requirements >12 hours.
- Grade 4 non-hematologic toxicity (see exceptions below)
- Grade 3 clinically significant non-hematological drug-related AEs that cannot be managed with adequate supportive therapy within 14 days of the onset of the event (see Table 5)
- Grade 4 acute pancreatitis: non-alcoholic and non-cholelithiasis related in a subject without prior history suggesting a drug induced etiology.
- Any grade ≥ 3 toxicity occurring beyond the 6 week window of treatment which is considered dose-limiting in the judgment of the site investigator and sponsor-investigator.

The following will <u>not</u> be considered DLTs:

- Grade 4 neutropenia or thrombocytopenia that causes a treatment delay of ≤ 14 days.
- Acute kidney injury of Grade 3 or 4 that improves to ≤Grade 2 within 14 days following appropriate management.
- ALT or AST ≥ 8x ULN (but <grade 4) without bilirubin elevation (defined as total bilirubin ≤ 2.0xULN, or ≤ 3.0 x ULN in a subject with Gilbert's syndrome, or direct bilirubin ≤ 35%) that improves to ≤ Grade 2 within 14 days following appropriate management
- Grade 4 nausea, vomiting, or diarrhea
- Grade 3 febrile neutropenia and grade 4 that has stabilized (off vasopressor support) within 12 hours
- Grade 3 electrolyte abnormalities that are corrected within 72 hours and without clinical sequelae.
- Grade 3 endocrinopathy that is adequately controlled by hormonal replacement.
- Grade 3 AE of tumor flare (defined as local pain/spasms, irritation, hematuria)
- Grade 3 AE of urinary tract obstruction (such as due to tumor flare causing hydronephrosis) that resolves to ≤ Grade 2 within 96 hours (with or without intervention).
- Grade 3 infusion reaction which resolves within 6 hours to ≤ Grade 1.

5.3 Phase II: Cisplatin-Eligible and Cisplatin-Ineligible Subjects

Table 3: Cohort I; Arm A treatment: Gemcitabine/Cisplatin; Cycle = 21 days

Drug	Dose	Route	Day of Administration	Repeated Number
Gemcitabine	1000 mg/m^2	IV	Day 1, 8	every 3 weeks x 4 cycles
Cisplatin[1]	70 mg/m^2	IV	Day 1	every 3 weeks x 4 cycles
	35 mg/m^2	IV	Day 1,2[1]	every 3 weeks x 4 cycles
Pembrolizumab[2]	RP2D mg	IV	Day 8	every 3 weeks x 5 doses

[1]:Subjects with Ccr of 50-59 mL/min (determined each cycle) must follow split dosing of cisplatin over Days 1 and 2 (preferred) *or* Days 1 and 8. Subjects with Ccr of ≥ 60 mL/min will receive cisplatin on Day 1 only.
[2]: The last dose of pembrolizumab falls on what would be Cycle 5 Day 8, however, gemcitabine-cisplatin is NOT GIVEN.

Table 4: Cohort II, Arm B (and Cohort I-p) Treatment; Cycle = 28 days

Drug	Dose	Route	Day of Administration	Repeated Number
Gemcitabine	1000 mg/m^2	IV	Day 1, 8, 15	every **4 weeks** x 3 cycles
Pembrolizumab[1]	RP2D mg	IV	Cycle 1 Day 8 Cycle 2 Day 1 Cycle 2 Day 22 Cycle 3 Day 15 "Cycle 4" Day 8	every 3 weeks x 5 doses

[1]The last dose of pembrolizumab falls on what would be Cycle 4 Day 8, however, gemcitabine is NOT GIVEN.

5.3.1 Pre-medications

Dexamethasone should be kept to a minimum and used only during cisplatin use and not when gemcitabine is given without cisplatin. Pre-medications should follow the guidelines below, however, the site investigator may select alternative regimens (such as olanzipine containing, or other) for specific subjects that require more tailored antiemetic therapy. Pembrolizumab does not need to be given with pre-medications, though non-corticosteroid premeds can be introduced depending on overall subject tolerance on an as needed basis.

The following table is adapted from the Antiemesis Guidelines of the National Comprehensive Cancer Network (NCCN) v2.2014. Additional details can be found on page AE-4 for Gemcitabine administration, and on page AE-2 for cisplatin.

Chemotherapy	Premed Day	Premeds	Suggested Home Regimen
Gemcitabine [without concomitant cisplatin]	Day 1 or 8	Avoid Dexamethasone 5-HT3 antagonist: *eg.* Ondansetron 12-16mg IV or po and/or Prochlorperazine 10mg IV or po and/or NK-1 antagonist: Fosaprepitant IV or po	Consider Proton Pump Inhibitor Non-glucocorticoid PRN regimen
Cisplatin	Day 1 or 2 (or 8)	Dexamethasone 12mg IV and NK-1 antagonist: aprepitant po or fosaprepitant IV and 5-HT3 antagonist: *eg.* Ondansetron 16mg IV and/or Lorazepam IV, po, or sublingual	Consider Proton Pump Inhibitor Dexamethasone 8mg po days 2-4 AND aprepitant 80mg po days 2-3) OR Dexamethasone 8mg po once day 2 then 8mg po bid days 3 and 4 (with fosaprepitant IV po on Day1) and 5-HT3 antagonist: *eg.* Ondansetron 8mg q8 prn and/or Lorazepam 0.5-2mg po or sublingual q4-6hrs days 1-4
Pembrolizumab	q3weeks	Avoid Dexamethasone No pre-meds are necessary, however, may consider 5-HT3 antagonist	Consider Proton Pump Inhibitor Non-glucocorticoid PRN regimen

5.4 Study Drug Administration

Trial treatment will be administered after all procedures/assessments have been completed as detailed in the Study Schedule of Events (Section 8). A treatment window of within ± 3 days of each cycle is allowed due to administrative reasons provided the subject meets all treatment parameters. All trial treatments will be administered on an outpatient basis.

Dosing interruptions are permitted in the case of medical/surgical events or logistical reasons not related to study therapy (e.g., elective surgery, unrelated medical events, patient vacation, and/or holidays). Subjects should be placed back on study therapy within 2 weeks of the scheduled interruption, unless otherwise discussed with the sponsor-investigator. The reason for interruption should be documented in the subject's study record.

When applicable, gemcitabine-cisplatin or gemcitabine should be administered prior to the pembrolizumab experimental therapy.

Arm A: Cohort I treatment with gemcitabine and cisplatin will continue for a maximum of 4 cycles (cycle = 21days).

Arm B: Cohort II treatment with gemcitabine weekly will continue for a maximum of 3 cycles (cycle = 28 days).

5.4.1. Pembrolizumab Administration

Pembrolizumab will be administered as a 30 minute IV infusion (treatment cycle intervals may be increased due to toxicity as described in Section 6.0). 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).

The Pharmacy Manual contains specific instructions for pembrolizumab dose calculation, reconstitution, preparation of the infusion fluid, and administration.

5.4.2. Gemcitabine Administration

Gemcitabine administration is per institutional guidelines. Where possible, on-time cycles and full doses (gemcitabine dose intensity) are important for optimal pathologic outcome in this curative-intent setting. Site investigators are STRONGLY ENCOURAGED to use filgrastim to minimize gemcitabine dose reductions in the setting of neutropenia (with *or without* febrile neutropenia). An example would be to dose filgrastim on days 4-6 of a treatment cycle to minimize a chance for a D8 dose hold or reduction. PEG-filgrastim may be considered on D9 of a cohort I subject, or filgrastim during the neutropenic phase. Subjects who experience febrile neutropenia should receive filgrastim or PEG-filgrastim (see section 6.3.4).

5.4.3. Cisplatin Administration

Cisplatin administration is per institutional guidelines.

5.5. Dose Calculations

5.5.1. Renal Function Determination

Estimation of glomerular filtration rate (GFR) may be made using a 24 hour urine or the treating institution's standard for calculation of creatinine clearance (Ccr). A modified Cockcroft-Gault equation is suggested for calculation:

Male:

$$\text{Ccr (mL/min)} = \frac{(140 - a \quad in\ years) \cdot (actual\ weight\ in\ kg)}{72 \cdot (serum\ creatinine)}$$

Female:

$$\text{Ccr (mL/min)} = 0.85 \cdot \frac{(140 - age\ in\ years) \cdot (actual\ weigh \quad in\ kg)}{72 \cdot (serum\ creatinine)}$$

A Ccr from properly collected timed (18-24 hours) urine usually supersedes a calculated value for determination of cisplatin eligibility and dosing. Accuracy of timed urine collection, however, is dependent on patient compliance and proper collection instructions should be confirmed with the patient. Improper collection methods disqualify use of the timed collection results and a calculated Ccr value should be used. Results from a measured nuclear GFR study may also be used at the discretion of the site investigator.

5.5.2. Pembrolizumab

The starting dose amount required to prepare the pembrolizumab infusion solution is a fixed dose of 200 mg (or 120mg, or RP2D) and does not require calculation. It is given IV every 21 days for 5 cycles.

5.5.3. Gemcitabine

Cohort I: Cisplatin-eligible subjects: Gemcitabine 1000 mg/m^2 IV on Days 1 and 8, every 21 days for 4 cycles.

Cohort II: Cisplatin-ineligible subjects: Gemcitabine 1000 mg/m^2 IV on Days 1, 8, 15, every 28 days for 3 cycles.

Site investigators are strongly encouraged to use Filgrastim to minimize gemcitabine dose holds or reductions (see section 6.3.4).

5.5.4. Cisplatin and Split Dose Cisplatin

Subjects with creatinine clearance ≥ 60 mL/min will receive cisplatin at a dose of 70 mg/m^2 IV per institutional guidelines on Day 1 every 21 days for 4 cycles. At least 1L of normal saline will be given IV for hydration prior to cisplatin. Subjects may receive additional IV hydration, mannitol, electrolytes, or furosemide according to institutional practice.

Split-dose cisplatin regimens and schedules for bladder cancer have not yet been evaluated in a randomized controlled trial (and currently being evaluated in a Phase II, NCT02030574), however is considered for many physicians to be a conventional practice in the appropriate setting. In this trial, **at the beginning of any cycle, subjects whose creatinine clearance is ≥50 and <60 mL/min will be treated with cisplatin 35 mg/m^2 IV on Day 1 and Day 2 of the cycle.** This Day 1 and 2 splitting is considered acceptable conventional care and is strongly preferred over the alternative Day 1 and 8 approach in this setting. Though day 1 and 8 dosing may be a more convenient schedule, the day 1 and 2 splitting limits dexamethasone exposure to the first 4 days of a cycle and will allow for washout prior to dosing pembrolizumab. Additionally, Day 1 and 2 dosing likely preserves the theoretic gemcitabine-platinum anti-neoplastic synergy. For extenuating scheduling difficulties (such as subject commutes from over 90 minutes and unwilling), treating physician should document this and use discretion to allow for a Day 1 and 8 split. In cases of Day 1 and 8 cisplatin splitting, the subject should remain on a Day 1 and 8 schedule and not revert to Day 1 and 2 at any point. Subjects may receive additional IV hydration, mannitol, electrolytes, or furosemide according to institutional practice. At least 1 L of normal saline will be given IV for hydration prior to cisplatin.

Day 2 or Day 8 renal labs are not required prior to redosing on the split-dose regimen, and per treating physician discretion.

If at the beginning of a subsequent cycle, creatinine clearance recovers to ≥ 60 mL/min, subjects may resume full cisplatin on Day 1 of the cycle (with any applicable dose modifications).

For prophylaxis of cisplatin-induced emesis, a 5-HT3 antagonist and a corticosteroid course of ≤4 days are recommended per NCCN guidelines. NK1 receptor antagonists such as aprepitant are also encouraged.

5.5.5. Missed Dose

Missed doses of gemcitabine-cisplatin or gemcitabine are not made up. If Day 1 treatment cannot be administered, initiation of all protocol treatment for that cycle should be delayed. If the dose of gemcitabine-cisplatin or gemcitabine is held/omitted in the middle of a cycle, the pembrolizumab should be given per the toxicity criteria outlined in Section 6.0. If gemcitabine-cisplatin or gemcitabine is delayed from the beginning of a cycle, pembrolizumab should also be delayed and given during the cycle with gemcitabine-cisplatin or gemcitabine on resumption.

Any delays will be recorded in the appropriate electronic case report form(s). If needed, a site investigator may consult with the sponsor-investigator by contacting the HCRN Project Manager.

If a subject discontinues gemcitabine-cisplatin or gemcitabine treatment due to toxicity, the subject may elect to receive single-agent pembrolizumab every 3 weeks per the protocol schedule for a total of 5 doses, or may discontinue all study treatment and choose second-line treatment or consolidative surgery.

5.6 Concomitant Medications/Vaccinations (allowed & prohibited)

Medications or vaccinations specifically prohibited in the exclusion criteria are not allowed during the ongoing trial. If there is a clinical indication for one of these or other medications or vaccinations specifically prohibited during the trial, discontinuation from trial therapy or vaccination may be required. The site investigator should discuss any questions regarding this with the sponsor-investigator by contacting the HCRN project manager. The final decision on any supportive therapy or vaccination rests with the site 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 site investigator and the subject. Please contact the HCRN project manager in such cases.

5.6.1 Acceptable Concomitant Medications

All treatments that the site investigator considers necessary for a subject's welfare may be administered at the discretion of the site 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. All concomitant medications received within 28 days before the first dose of trial treatment and 30 days after the last dose of trial treatment should be recorded.

5.6.2 Prohibited Concomitant Medications

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

- Anti-cancer systemic chemotherapy or biological therapy
- Immunotherapy not specified in this protocol
- Chemotherapy not specified in this protocol
- Investigational agents other than pembrolizumab
- Radiation therapy
- Live vaccines within 30 days prior to study registration and while participating in the trial. Examples of live vaccines include, but are not limited to, the following: measles, mumps, rubella, chicken pox, yellow fever, rabies, BCG, and typhoid (oral) vaccine. Seasonal influenza vaccines for injection are generally killed virus vaccines and are allowed; however intranasal influenza vaccines (e.g. Flu-Mist®) are live attenuated vaccines, and are not allowed.
- Glucocorticoids for delayed emesis prophylaxis of cisplatin (no longer than 4 day course) or to modulate symptoms from an event of clinical interest of suspected immunologic etiology are allowed. Otherwise, a total of prednisone or prednisone equivalent of 10mg per day or less is allowed. Above a prednisone equivalent of 10mg a day of systemic

glucocorticoids are not allowed. The use of physiologic doses of corticosteroids (for specific non-cancer related conditions) may be approved after consultation with the sponsor-investigator. Please contact the HCRN project manager.

- OTC herbals, botanicals, and high dose anti-oxidant vitamins/supplements should be kept to an absolute minimum and are discouraged. While there is no known interacting harm nor benefit, the potential for harm while receiving an immunologic agent such as pembrolizumab is possible and therefore discouraged.
- Continuous steroids (premeds of ≤ 5 days as a cisplatin delayed antiemetic regimen is acceptable) meet exclusionary criteria, and NSAIDS- other than aspirin for cardiovascular risk - should be minimized.
- Any additional medication prohibited in the Exclusion Criteria.

Subjects who, in the assessment by the site investigator, require the use of any of the aforementioned treatments for clinical management should be removed from the trial. Subjects may receive other medications that the site investigator deems to be medically necessary.

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

5.7 Supportive Care

5.7.1 Diet

Subjects should maintain a normal diet unless modifications are required to manage an AE

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. Non-pregnant, non-breast-feeding women may be enrolled if they are willing to use adequate birth control or are considered highly unlikely to conceive. Highly unlikely to conceive is defined as 1) surgically sterilized, or 2) postmenopausal (a woman who is ≥45 years of age and has not had menses for greater than 1 year will be considered postmenopausal), or 3) not heterosexually active for the duration of the study. The two birth control methods can be either two barrier methods or a barrier method plus a hormonal method to prevent pregnancy. Subjects should start using birth control from study Visit 1 throughout the study period up to 120 days after the last dose of study therapy. If there is any question that a subject will not reliably comply with the requirements for contraception, that subject should not be entered into the study.

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

5.7.3 Use in Pregnancy

If a subject inadvertently becomes pregnant while on treatment with pembrolizumab, the subject will immediately be removed from the study. The site will contact the subject at least monthly

and document the subject's status until the pregnancy has been completed or terminated. The outcome of the pregnancy will be reported to HCRN without delay and within 24 hours if the outcome is a serious adverse experience (e.g., death, abortion, congenital anomaly, or other disabling or life-threatening complication to the mother or newborn). The site investigator will make every effort to obtain permission to follow the outcome of the pregnancy and report the condition of the fetus or newborn to HCRN. If a male subject impregnates his female partner the study personnel at the site must be informed immediately and the pregnancy reported to HCRN and followed as described above and in the Adverse Event section.

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 Adverse Event Grading and Dose Levels

The NCI Common Terminology Criteria for Adverse Events (CTCAE) Version 4.0 will be used to grade adverse events. Subjects will be evaluated for adverse events (all grades), serious adverse events, and adverse events requiring study drug interruption or discontinuation at each study visit during the treatment portion of the study.

Subjects enrolled in this study will be evaluated clinically and with standard laboratory tests before and at regular intervals during their participation in this study as specified in the Schedule of Events.

5.8.1 Dose Levels for Gemcitabine

Subjects who require more than two permanent dose reductions will be removed from gemcitabine treatment.

Cohort	Dose Level	Gemcitabine
I, II	0	1000 mg/m^2
I, II	-1	750 mg/m^2
I, II	-2	500 mg/m^2

5.8.2 Dose Levels for Cisplatin

Cohort	Dose Level	Cisplatin	
		ClCr ≥ 60 mL/min	ClCr ≥ 50 and <60 mL/min[1]
I	0	70 mg/m^2 on Day 1	35 mg/m^2 on Days 1, 2
I	-1	50 mg/m^2 on Day 1	25 mg/m^2 on Days 1, 2
I	-2	36 mg/m^2 on Day 1	18 mg/m^2 on Days 1, 2

[1]Day 1 and 2 splitting is considered acceptable conventional care and is encouraged over the alternative Day 1 and 8 approach. See section 5.5.4.

5.9 Post Neoadjuvant Restaging and Consolidative Surgery

5.9.1 Post chemotherapy Restaging Evaluation

Subjects should undergo a post neoadjuvant restaging evaluation per conventional care. This may be an extensive evaluation that includes cystoscopy[68,69], however, must at the very least be a CT of the abdomen-pelvis within 21 days of the planned surgery. MRI is also acceptable per institution practice or treating physician discretion. A clinical TNM staging prior to surgery should be assigned and documented. Imaging that suggests progression or new lesions, however, should be further evaluated with a biopsy prior to making a decision on definitive surgical consolidation. If progression is a concern, pseudo-progression due to tumor lymphocyte infiltration should be ruled out with a biopsy, especially if the area of concern involves abdomino-pelvic draining lymph nodes. See irRC criteria (Table 9b) as well. Progression or pseudo-progression at post neoadjuvant chemotherapy that impacts decision making for definitive surgery is considered an event of clinical interest and should be discussed with the HCRN project manager/sponsor-investigator.

5.9.2 Timing of Consolidative Surgery

To be evaluable for the primary endpoint analysis, subjects should undergo consolidative surgery with radical cystectomy and bilateral lymph node dissection or nephroureterectomy and lymph node dissection once subject is appropriately stabilized after neoadjuvant chemotherapy. Timing should fall no sooner than: 2 weeks after the last dose of pembrolizumab, or 3 weeks after the last dose of cytoxic chemotherapy, whichever is later. Completing definitive cancer management in a timely manner is important. While both neoadjuvant therapy and surgery with lymph node dissection can provide optimal control of the cancer, there may be situations that arise where delays due to toxicity of neoadjuvant therapy could lead to poor overall outcome if surgery is excessively delayed. Please see section 6.0 for criteria where treatment should stop and focus should turn to toxicity management and optimizing the patient for surgical consideration.

Consolidative surgery should be performed no later than 7 weeks after the last dose of pembrolizumab, or whenever safely possible after recovery from toxicity. It's encouraged for the treating physician to have frequent communication with the subject's urologist about course of neoadjuvant therapy, relevant toxicity, and timing of consolidative surgery.

Surgical approach and diversion is per discretion of the treating urologist and subject and may be open, laproscopic, or robotic.

6. DOSE MODIFICATIONS AND TOXICITY MANAGEMENT

Treatment labs, including timed urine, should be obtained no more than 3 days prior to initiating a new cycle. It's encouraged to obtain the most current labs possible prior to making a dosing modification. If multiple values are available, the determining value will be the most current available or per site investigator discretion. *Dose timing and dose intensity are important paradigms of curative intent therapy and filgrastim administration should be considered to avoid dose delays or reductions when possible (see section 6.3.4).*

6.1 Pembrolizumab Dose Modifications and Toxicity Management

The pembrolizumab dose remains the same during the Phase II portion, and only may change during phase Ib. In general, dose modifications for hematologic toxicity will follow chemotherapy-driven management guidelines. While there may be hematologic parameter related delays for pembrolizumab, the future dose level and interval does not change. It is possible, however, to have a pembrolizumab attributable hematologic toxicity, including immune related events where a work up is warranted. If suspected, the treating physician is encouraged to work up a suspected immune related hematologic toxicity and discuss a treatment plan with the sponsor-investigator. A total of five pembrolizumab doses should be given, and may require additional dosing beyond chemotherapy in the case of delays. Permanent discontinuation should be considered for any severe or life-threatening event.

When toxicities are *assessed to be most likely associated with pembrolizumab,* Table 5 should be followed.

In general, if toxicity attributed to pembrolizumab does not resolve to Grade 0-1 within 12 weeks after last infusion, pembrolizumab treatment should be discontinued and may discuss with the sponsor-investigator for continuation on trial. With site investigator and sponsor-investigator agreement, subjects with a laboratory adverse event still at Grade 2 after 12 weeks may continue treatment in the trial only if asymptomatic and controlled. *There are specific criteria to follow for renal, hepatic, and hematologic toxicity discussed in the following sections.*

Subjects who experience a recurrence of the same severe or life-threatening event at the same grade or greater with re-challenge of pembrolizumab should be discontinued from pembrolizumab treatment.

Table 5: Pembrolizumab dose modification guidelines for drug-related adverse events.

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 (CTCAEv4.0)	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). • Participants with ≥ Grade 2 diarrhea suspecting colitis should consider GI consultation and performing endoscopy to rule out colitis.
	Grade 4	Permanently discontinue		

				• 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	• Treat with non-selective beta-blockers (eg, propranolol) or thionamides as appropriate	• Monitor for signs and symptoms of thyroid disorders.
	Grade 3 or 4	Withhold or permanently discontinue[1]		
Hypothyroidism	Grade 2-4	Continue	• Initiate thyroid replacement hormones (eg, levothyroxine or liothyroinine) per standard of care	• Monitor for signs and symptoms of thyroid disorders.
	Grade 2	Withhold		

Nephritis and Renal dysfunction	Grade 3 or 4	Permanently discontinue	• Administer corticosteroids (prednisone 1-2 mg/kg or equivalent) followed by taper.	• Monitor changes of renal function
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).

6.2 Hematologic Toxicity; Cohort I; Gemcitabine, Cisplatin and Pembrolizumab

Gemcitabine and cisplatin doses may be modified separately based on individual toxicity according to the rules outlined below. There is no dose reduction below level -2 for gemcitabine or cisplatin. If dose reduction below level -2 is required, gemcitabine and/or cisplatin should be discontinued**.**

6.2.1 Cohort I; Initiation of a new cycle with Gemcitabine and Cisplatin

For Day 1 of each cycle, subjects must have an ANC ≥ 1,500 AND platelets ≥ 100,000 before receiving treatment with gemcitabine-cisplatin or pembrolizumab. Subjects that do not meet these values on Day 1 will have their cycle start delayed and any pembrolizumab doses during that cycle will be similarly delayed. Resume the gemcitabine-cisplatin new cycle and pembrolizumab dosing once the ANC improves to ≥1,500 AND platelets ≥100,000.

If treatment is/was delayed:

- For up to 1 week, resume treatment at the previous doses of gemcitabine-cisplatin. After the delay, pembrolizumab dosing and interval should resume to stay "in-sequence" with gemcitabine-cisplatin cycle.
- For more than 1 week and less than 4 weeks, reduce gemcitabine and cisplatin by one dose level for this and all subsequent cycles. Once the cycle is resumed, pembrolizumab dosing and its' interval should resume to stay "in-sequence" with gemcitabine-cisplatin cycle. Do not adjust pembrolizumab dose or cycle length.
- If a second treatment delay of more than one week and less than 4 weeks occurs, reduce gemcitabine and cisplatin an additional dose level. Pembrolizumab dosing should restart on the planned cycle day (for Cohort I, this is generally Day8). Do not adjust pembrolizumab dose or cycle length.
- Greater than 4 weeks due to hematologic toxicity, discontinue all therapy and initiate workup to optimize the subject for surgical consideration.

Workup of cytopenia etiology beyond bone marrow suppression is encouraged at any time, and per treating physician judgment.

6.2.2 Cohort I; Arm A; Criteria for Gemcitabine and Cisplatin Day 8 treatment

A complete blood count with differential must be drawn the day of treatment. Dose timing and dose intensity are important paradigms of curative intent therapy and filgrastim administration should be considered to avoid dose delays or reductions when possible (see section 6.3.4).

For ANC 500-999 or platelets 50,000-74,999, decrease gemcitabine by one dose level for this and all subsequent doses. For subjects receiving split dose cisplatin during this cycle, decrease cisplatin by one dose level for this and all subsequent doses (effective with current dose if on a Day 1 and Day 8 regimen, or with the next cycle of a Day 1 and Day 2 regimen, see section 5.5.4). Do not adjust pembrolizumab dose or cycle length.

For ANC < 500 or platelets < 50,000, skip gemcitabine and decrease gemcitabine by one dose level for all subsequent doses. For subjects on split dose cisplatin during this cycle, also skip cisplatin and decrease cisplatin by one dose level for this all subsequent doses (effective with current dose if on a Day 1 and Day 8 regimen, or with the next cycle of a Day 1 and Day 2 regimen, (see section 5.5.4).

For *second* occurrences of Day 8 ANC <500 and platelets <50,000:

- Modifications should follow the gemcitabine and cisplatin dose reductions as outlined above.

Day 8: Cohort I; Arm A; Treatment Toxicity Guidelines for Dose/Interval Changes:

ANC Day 8	Platelets Day 8	Gemcitabine[1] Day 8	Split Dose Cisplatin Day 8[2]	Current Cycle Pembrolizumab Treatment Guidelines based on Day 8[3]
≥ 1000 AND	≥ 100,000	No dose modification	No dose modification	Treat on schedule. No modification.
≥ 1000 AND	75,000 – 99,999	No dose modification	No dose modification	Treat on schedule. No modification.
500-999 or	50,000 – 74,999	Decrease by 1 dose level for this dose and all subsequent doses.	Decrease by 1 dose level for this dose and all subsequent doses.	Treat on schedule. No modification.
< 500 or	< 50,000	Omit gemcitabine and reduce by 1 dose level for subsequent doses. This dose reduction is permanent	Omit cisplatin and reduce by 1 dose level for subsequent doses. This dose reduction is permanent	Treat on schedule. No modification.
< 500 AND	< 50,000	Omit gemcitabine and reduce by 1 dose level for subsequent doses. This dose reduction *is* permanent	Omit cisplatin and reduce by 1 dose level for subsequent doses. This dose reduction is permanent	Treat on schedule. No modification.

1: Gemcitabine dose-levels reductions of-1 or -2 (see section 5.8.1) take effect with the next cycle. This is relevant when considering pembrolizumab interval modification and dosing during the severe cytopenia scenarios that distinguish subjects having had gemcitabine dose reduced to -1, or -2.
2: Only applies when split regimen cisplatin is given on Day 1 and 8; Days 1&2 is the preferred schedule and there are no labs or dose modifications necessary on Day 2.
3: For the purposes of dose modifications, hematologic toxicity will not be attributed to pembrolizumab. If suspected due to pembrolizumab, consider a workup and discuss with the sponsor investigator.

6.3 Cohort II (and Cohort I-p); Arm B; Gemcitabine and Pembrolizumab

Gemcitabine doses may be modified separately based on individual toxicity according to the rules outlined below. There is no dose reduction below level -2 for gemcitabine. If dose reduction below level -2 is required, gemcitabine should be discontinued**.**

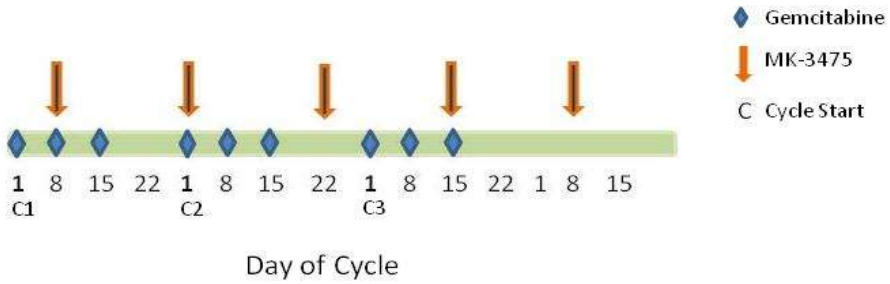


Figure 3. Cohort II (and Cohort I-p); Arm B: Arm B treatment includes subjects on Cohort II and certain cases of Cohort I-p. Pembrolizumab treatment interval is every 3 weekswith a total of 5 doses.Consolidative surgery and follow up not shown. See also Figure 2 and Table 4.

6.3.1 Cohort II (and Cohort I-p); Arm B; New Cycle Initiation Criteria for Gemcitabine and Pembrolizumab

For Day 1 of each Arm B cycle, subjects must have an ANC ≥ 1,500 AND platelets ≥ 100,000 before receiving treatment with gemcitabine or pembrolizumab. Resume the gemcitabine new cycle and pembrolizumab dosing once the ANC improves to ≥ 1,500 AND platelets ≥ 100,000. Dose timing and dose intensity are important paradigms of curative intent therapy and filgrastim administration should be considered to avoid dose delays or reductions when possible (see section 6.3.4).

If treatment is/was delayed:

- For up to 1 week, resume treatment at the previous doses of gemcitabine. Pembrolizumab should be similarly delayed and restarted on the planned day "within" the gemcitabine cycle as shown in Figure 3 or Table 4**.** For pembrolizumab dosing within the cycle, see guidelines below. Do not adjust pembrolizumab dose or cycle length.
- For more than 1 week and less than 4 weeks, reduce gemcitabine by one dose level for this and all subsequent cycles. Once the cycle is resumed, pembrolizumab dosing and its interval should be similarly delayed and restarted on the planned day "within" the gemcitabine cycle (see Figure 3 or Table 4 for pembrolizumab dosing within the cycle). Do not adjust pembrolizumab dose or cycle length.
- If a second treatment delay of more than 1 week and less than 4 weeks occurs, reduce gemcitabine an additional dose level. Pembrolizumab dosing should restart on the planned cycle day (see Figure 3 or Table 4 for pembrolizumab dosing within the cycle). Do not adjust pembrolizumab dose or cycle length..
- Greater than 4 weeks due to hematologic toxicity, discontinue all therapy and initiate workup to optimize the subject for surgical consideration.

Workup of cytopenia etiology beyond bone marrow suppression is encouraged at any time, and

per treating physician judgment.

6.3.2 Cohort II (and Cohort I-p); Arm B; Criteria for Gemcitabine and Pembrolizumab Day 8 & 15 & 22

A complete blood count with differential must be drawn on the days of treatment.
On Days 8, 15, and [where/when applicable] 22 of each cycle the dose of gemcitabine and pembrolizumab may be modified per the tables below. The modifications for pembrolizumab doses that fall on Day 22 of a cycle are the same as the Day 15 guidelines; gemcitabine is never dosed on Day 22. If subjects had a non-permanent gemcitabine dose reduction on day 15 and their ANC > 1500 and platelets are > 100,000 by day 1 of the next cycle, they will return to the dose prior to the non-permanent reduction. Dose timing and dose intensity are important paradigms of curative intent therapy and filgrastim administration should be considered to avoid dose delays or reductions when possible (see section 6.3.4). See Tables below.

For *second* occurrences of Day 8, 15, or 22 ANC <500 and platelets <50,000:

- Modifications should follow the gemcitabine dose reductions as outlined in the toxicity tables below.

Do not adjust pembrolizumab dose or cycle length.

Cohort II (and Cohort I-p); Arm B; Day 8 Toxicity Guidelines for Dose/Interval Changes

ANC Day 8	Platelets Day 8	Gemcitabine Day 8	Is Pembrolizumab Treatment Due? Day 8
≥ 1000 AND	≥ 100,000	No dose modification	Treat on schedule. No modification.
500-999 or	50,000 – 99,999	Decrease by 1 dose level. This dose reduction *is not* permanent	Treat on schedule. No modification.
<500 or	<50,000	Omit gemcitabine and reduce by 1 dose level on day 15. This dose reduction *is* permanent	• If this is Dose 1 or 2, OR no prior permanent gemcitabine dose level reductions: Treat as scheduled. No modification. • If at least one prior cycle gemcitabine dose level reduction or pembrolizumab-interval increase: Delay pembrolizumab and give on Day 1 of next cycle. Treat to 5 total doses.
<500 AND	<50,000	Omit gemcitabine and reduce by 1 dose level on day 15. This dose reduction *is* permanent	Delay pembrolizumab and give on Day 1 of next cycle. Treat to 5 total doses. Treat to 5 total doses.

Cohort II (and Cohort I-p); Arm B; <u>Toxicity Guidelines for Day 15 or Day 22 Dose/Interval Changes</u>

ANC Day 15 or 22	Platelets Day 15 or 22	Gemcitabine Dose Modification Day 15 only	Is Pembrolizumab Treatment Due? Days 15 or 22
≥ 750 AND	≥75,000	If day 8 ANC was ≥1,000 and platelet count was ≥75,000 then the day #15 gemcitabine dose is the same as day 8. If day 8 ANC was 750-999 and the platelet count was ≥75,000 then the day #15 gemcitabine dose is the same as day 8. If day 8 ANC was 500-749 or Platelet 50,000-74,999 then the day #15 gemcitabine dose is reduced one dose level from the day 1 dose. This reduction *is not* permanent. If day 8 ANC < 500 or platelet < 50,000 then the day #15 gemcitabine dose is permanently reduced one dose level from the day 1 dose.	Treat on schedule. No modification.
500-749 AND	50,000-74,999	If day 8 ANC was ≥1,000 and platelet count was75,000-99,999 then the day #15 gemcitabine dose is reduced one dose level from the day 8 dose. This dose reduction is not permanent. If day 8 ANC was 750-999 and the platelet count was ≥75,000 then the day #15 gemcitabine dose is the same as the day 8 dose. If day 8 ANC was 500-749 or Platelet 50,000-74,999 then the day #15 gemcitabine dose is reduced one dose level from the day 1 dose. This reduction is not permanent. If day 8 ANC was < 500 or platelet < 50,000 then the day #15 gemcitabine dose is permanently reduced one dose level from the day 1 dose. .	Treat on schedule. No modification.
<500 or	< 50,000	Omit gemcitabine. If the dose of gemcitabine was also omitted on Day 8, then reduce the dose of gemcitabine on Day 1 of the following cycle by one dose level. This dose reduction is permanent.	Dose 1 or 2, OR no permanent gemcitabine dose level change: Treat as scheduled. Prior cycle permanent gemcitabine or pembrolizumab modification: Delay pembrolizumab and give on Day 1 of next cycle. Treat to 5 total doses.

<500 AND	<50,000	Omit gemcitabine and reduce by 1 dose level on day 15. This dose reduction is permanent	Delay pembrolizumab and give on Day 1 of next cycle. Treat to 5 total doses.

6.3.3 Cohort II (and Cohort I-p); Arm B; Pembrolizumab Dose Modifications for Hematologic Toxicity

In general, dose modifications for hematologic toxicity will follow chemotherapy-driven management guidelines. While there may be hematologic parameter related delays for pembrolizumab, the future dose level and interval does not change.
It is possible, however, to have a pembrolizumab attributable hematologic toxicity, including immune related events where a work up is warranted. If suspected, the treating physician is encouraged to work up a suspected immune related hematologic toxicity and discuss a treatment plan with the sponsor-investigator. A total of five pembrolizumab doses should be given, and may require additional dosing beyond chemotherapy in the case of delays. Permanent discontinuation should be considered for any severe or life-threatening event.

Delays at start of cycle

If initiation of a new gemcitabine cycle was:

- Delayed for up to one week: the pembrolizumab dose (as well as gemcitabine) should be delayed until ANC ≥ 1,500 AND platelets ≥ 100,000 and the subject should restart pembrolizumab as originally described within that specific gemcitabine cycle (see Figure 3 or Table 4). Subsequent intervals should revert to the prior dosing interval of every 3 weeks.
    - For example, if was scheduled to be dosed on Day 15 of a cycle where Day 1 was delayed for 6 days due to neutropenia; pembrolizumab should be dosed as originally sequenced on Day 15 once the new cycle begins (even though this means that the interval was 3 weeks and 6 days from the last dose, the subsequent interval should be 3 weeks).

Delayed more than 1 week and less than 4 weeks: the pembrolizumab dose should be delayed until ANC ≥ 1,500 AND platelets ≥ 100,000. Pembrolizumab should resume to the "sequence" within the current gemcitabine cycle (see Figure 3), subsequent intervals should remain the same as prior to the delay.

- Delayed >4 weeks due to hematologic toxicity, discontinue all therapy and continue workup to optimize the subject for surgical consideration.

6.3.4 Febrile Neutropenia

For febrile neutropenia (defined here as temperature ≥ 38.5° C [101° F] sustained for more than one hour concomitant with ANC ≤ 500/mm^3) reduce gemcitabine and cisplatin by one dose level for this and subsequent cycles. Consider filgrastim or peg-filgrastim in subsequent cycles.

6.3.5 Hepatic Dysfunction (See Table 6 below)

Hepatic dysfunction should prompt consideration of workup for etiology such as medication and supplement review, infection, imaging, pancreatico-biliary evaluation, and amylase, lipase.

Consideration should be given to evaluation of concomitant pancreatitis in the appropriate setting. This evaluation and workup is at the treating physician discretion.

For bilirubin > 1.5 x ULN (or > 3 x ULN for subjects with Gilbert's syndrome), delay pembrolizumab treatment until bilirubin ≤ 1.5 x ULN (< 3 x ULN for subjects with Gilbert's syndrome), then resume with one dose level reduction of gemcitabine and at the previous dose of cisplatin.

If bilirubin is > 1.5 x ULN despite two gemcitabine dose reductions, gemcitabine should be discontinued but cisplatin and pembrolizumab may be redosed when total bilirubin ≤ 1.5. For subjects with Gilbert's syndrome, if bilirubin is > 3 x ULN despite two gemcitabine dose reductions, gemcitabine and cisplatin should be permanently discontinued; treatment with pembrolizumab may continue once ≤ 3.

Table 6: Guidelines for Dose Modifications due to LFT abnormalities.

LFT	Gemcitabine[2]	Cisplatin[2]	Pembrolizumab
AST/ALT >ULN to 2 x ULN AND Total bilirubin >1.5 x ULN OR 3 x ULN for subjects with Gilbert's syndrome[1]	Delay treatment until AST/ALT ≤ 2 x ULN and total bilirubin ≤ 1.5 x ULN (≤ 3 x ULN for subjects with Gilbert's syndrome[1]), then resume with one dose level reduction of gemcitabine. After 2 dose level reductions, gemcitabine should be discontinued.	Delay treatment until AST/ALT ≤ 2 x ULN and total bilirubin ≤ 1.5 x ULN (≤ 3 x ULN for subjects with Gilbert's syndrome[1]), then resume at previous dose of cisplatin. For subjects with Gilbert's, discontinue both gemcitabine and cisplatin after 2 occurrences of total bilirubin >1.5.	Delay treatment until AST/ALT ≤ 3 x ULN and total bilirubin ≤ 1.5 x ULN (≤ 3 x ULN for subjects with Gilbert's syndrome[1]), then resume at full dose for total of 5 doses.
AST/ALT > 2 x ULN to ≤ 8 x ULN AND Total bilirubin ≤ 1.5 x ULN OR ≤ 3 for subjects with Gilbert's syndrome[1]	Delay treatment until AST/ALT ≤ 2 x ULN and total bilirubin ≤ 1.5 x ULN (≤ 3 x ULN for subjects with Gilbert's syndrome[1]), then resume with one dose level reduction of gemcitabine. Consider hepatologist consult.	Delay treatment until AST/ALT ≤ 2 x ULN and total bilirubin ≤ 1.5 x ULN (≤ 3 x ULN for subjects with Gilbert's syndrome[1]), then resume at previous dose of cisplatin. Consider hepatologist consult.	Hold or discontinue pembrolizumab per below criteria. Monitor LFTs every 48 hours until decreasing and then follow weekly. If not improving consider biopsy and additional immunosuppressive agents. For grade 3, permanently discontinue and administer corticosteroids (initial dose of 1-2mg/kg/day prednisone or equivalent) followed by taper over at least 2-4 weeks. For grade 2, administer corticosteroids (initial dose of 0.5-1mg/kg/day prednisone or equivalent) followed by taper over 2-4 weeks.

			Criteria to restart after discussion with sponsor-investigator include resolution to Grade 0 or 1 and prednisone ≤10mg/day.
AST/ALT > 2 x ULN to ≤ 8 x ULN AND Total bilirubin > 1.5 x ULN OR 3 for subjects with Gilbert's syndrome[1]	Delay treatment until AST/ALT ≤ 2 x ULN and total bilirubin ≤ 1.5 x ULN (≤ 3 x ULN for subjects with Gilbert's syndrome[1]), then resume with one dose level reduction of gemcitabine. After 2 dose level reductions, gemcitabine should be discontinued. Consider hepatologist consult.	Delay treatment until AST/ALT ≤2 x ULN and total bilirubin ≤ 1.5 x ULN (≤3 x ULN for subjects with Gilbert's syndrome[1]), then resume at previous dose of cisplatin. For subjects with Gilbert's, discontinue both gemcitabine and cisplatin after 2 occurrences of total bilirubin >1.5. Consider hepatologist consult.	Hold or discontinue pembrolizumab per below criteria. Monitor LFTs every 48 hours until decreasing and then follow weekly. If not improving consider biopsy and additional immunosuppressive agents. For grade 3, permanently discontinue and administer corticosteroids (initial dose of 1-2mg/kg/day prednisone or equivalent) followed by taper over at least 2-4 weeks. For grade 2, administer corticosteroids (initial dose of 0.5-1mg/kg/day prednisone or equivalent) followed by taper over 2-4 weeks. Criteria to restart after discussion with sponsor-investigator include resolution to Grade 0 or 1 and prednisone ≤10mg/day.

AST/ALT > 8 x ULN AND Total Bilirubin any level	Delay treatment until AST/ALT ≤ 2 x ULN and total bilirubin ≤ 1.5 x ULN (≤ 3 x ULN for subjects with Gilbert's syndrome[1]), then resume with one dose level reduction of gemcitabine. Consider hepatologist consult.	Delay treatment until AST/ALT ≤ 2 x ULN and total bilirubin ≤ 1.5 x ULN (≤3 x ULN for subjects with Gilbert's syndrome[1]), then resume at previous dose of cisplatin. For subjects with Gilbert's, discontinue both gemcitabine and cisplatin after 2 occurrences of total bilirubin > 1.5. Consider hepatologist consult.	Discontinue pembrolizumab. Consider inpatient admission. If not improving consider biopsy and additional immunosuppressive agents. For grade 3 or 4, permanently discontinue and administer corticosteroids (initial dose of 1-2mg/kg/day prednisone or equivalent) followed by taper over at least 2-4 weeks. *Any missed dose(s) will not be made up*.

[1]Subject must have a previous diagnosis of Gilbert's syndrome documented in their medical record prior to study initiation.

For grade 3 sensory or motor neuropathy, skip cisplatin until the toxicity resolves to ≤grade 2 and then resume therapy with one dose level reduction of cisplatin on Day 1 of the next scheduled cycle. If cisplatin is skipped for two consecutive cycles, permanently discontinue cisplatin. Treatment with gemcitabine and pembrolizumab may continue.

For grade 4 sensory or motor neuropathy, skip gemcitabine and cisplatin therapy until resolution to ≤ grade 2. Finish the current cycle planned doses of pembrolizumab. If sensory neuropathy improves to grade 3, but not grade 2 or better, within 4 weeks, resume gemcitabine at the previous dose and pembrolizumab without cisplatin. If the sensory neuropathy remains at grade 4 within 4 weeks, dose remaining pembrolizumab treatments and discontinue gemcitabine and cisplatin, then prepare the subject for surgery within 2-7 weeks after the final dose of pembrolizumab.

6.3.6 Gastrointestinal Toxicity

For grade 3 or 4 nausea or vomiting despite maximal antiemetic therapy (including 5HT-3 antagonist, corticosteroids up to 5 days, and aprepitant), discontinue cisplatin. Continue gemcitabine and pembrolizumab at the previous dose when symptoms resolve to ≤ grade 1.

6.4 Cohort I; Cisplatin-Eligible; Subjects becoming Cisplatin-Ineligible Kidney Dysfunction

A Ccr from properly collected timed (24 hours; 18-24 hours acceptable) urine supersedes a calculated value for determination of cisplatin eligibility and dosing. Accuracy of timed urine collection, however, is dependent on patient compliance; proper collection adherence should be confirmed with the patient. Improper collection methods disqualify use of the timed collection results and a calculated serum value should be used.

For creatinine clearance < 50 mL/min (measured or calculated) on Day 1 of a cycle, delay all treatment until creatinine clearance improves to ≥ 50 mL/min as follows:

- If creatinine clearance improves to ≥ 60 mL/min within 1 week, resume with one dose level reduction for cisplatin and gemcitabine for all subsequent doses, and continue pembrolizumab at the same dose and schedule.
- If creatinine clearance improves to ≥ 50 and < 60 mL/min within 1 week, resume with one dose level reduction of cisplatin for this and all subsequent cycles. Administer cisplatin as a split dose on Days 1 and 2 (preferred) or Days 1 and 8 of this cycle. Resume gemcitabine at the same dose level and continue pembrolizumab at the same dose and schedule.
- If creatinine clearance does not improve to ≥ 50 mL/min within 1 week, skip cisplatin for this cycle only. Treat with cisplatin at one lower dose level for this and all subsequent cycles with split dosing over Days 1&2 (or Days 1 and 8, per treating physician discretion as previously described) and pembrolizumab at the same dose and schedule. For a subsequent cycle, if creatinine clearance is again < 50 mL/min, discontinue cisplatin [**Cohort "I-no platinum" (or, cohort I-p)** will be used to refer to those Cohort I subjects that have permanently discontinued cisplatin] and treat as follows:
    - If the subject becomes cisplatin ineligible (for renal, neurologic, auditory, or other reason per site investigator) *after 1 or 2 cycles on Arm A*, he or she may continue on study in this manner: switched to cohort I-p and follow Arm B treatment and guidelines for the remaining treatments: maximum 3 cycles with gemcitabine weekly for three weeks and repeated every 4 weeks. For instance, if a subject completes 1 cycle on Arm A and then continues their treatment according to Arm B guidelines, they are referred to as "Cohort I-p" and should complete an additional 2 cycles per the Arm B regimen. Five doses of pembrolizumab should still be given. A discussion with the HCRN project manager to determine the proper transition is encouraged.
    - If the subject becomes cisplatin ineligible (for renal, neurologic, auditory, or other reason per site investigator) *after completion of 3 cycles of therapy on Arm A*, he or she may continue on study in this manner: switched to cohort I-p and follow Arm A *and withhold cisplatin for the final fourth cycle*. Treatment should continue with gemcitabine on Day 1 and 8 at the prior dose, and pembrolizumab should continue for all 5 doses.

For Cohort I cisplatin eligible subjects: Grade 3 or 4 acute kidney injury will be attributed to cisplatin. Subject may continue treatment per guidelines as a Cohort I-p subject as described above. The pembrolizumab dose/schedule will not be modified unless an immune related nephritis is determined with serology, urine studies, and/or supporting biopsy and discussed with the sponsor-investigator. Patients with renal dysfunction will be attributed to cisplatin and not pembrolizumab for the purposes of drug modifications (and in this case, re-assignment from cohort I to I-p). Table 5 pembrolizumab dose modifications and discontinuation for renal toxicity should only be followed if the etiology is determined to be immune-mediated with serology, urine studies, and/or supporting biopsy.

On Day 2 or 8 renal labs are not required though may be obtained per treating physician discretion during split-dose cisplatin. If creatinine clearance < 50 mL/min (measured or calculated), skip cisplatin for this day (applies only if split-dose cisplatin is indicated for this

cycle). Reduce cisplatin by one lower dose level for all subsequent doses. Refer **to** section 5.8.2 above for dose modifications for Day 1 dosing for the next cycle.

6.4.1 Gemcitabine Non-Hematologic Toxicity

Grade	Dose Modification
0-2	Full dose
3-4	Hold until resolution to ≤ Grade 2, then decrease by 1 dose level from current dose. This dose reduction is not permanent. If toxicity does not resolve within 4 weeks, discontinue gemcitabine treatment.

Grade 3/4 nausea or vomiting only requires dose modifications if it persists > 24 hours despite adequate antiemetic medication. Grade 3/4 adverse events not related to treatment such as a thrombosis, pulmonary embolus or non-neutropenic infection do not require dose reductions when treatment is resumed.

For suspected > Grade 2 pneumonitis, suspect pembrolizumab per Table 5 (despite gemcitabine a known confounder) and initiate treatment per Table 8 below.

6.5 Supportive Care for Other Toxicities

Subjects should receive appropriate supportive care measures as deemed necessary by the site investigator including but not limited to the items outlined below:

6.5.1 Diarrhea

Subjects should be carefully monitored for signs and symptoms of enterocolitis (such as diarrhea, abdominal pain, blood or mucus in stool, with or without fever) and of bowel perforation (such as peritoneal signs and ileus). In symptomatic subjects, infectious etiologies should be ruled out, and if symptoms are persistent and/or severe, endoscopic evaluation should be considered.

- In subjects with severe enterocolitis (Grade 3), pembrolizumab will be permanently discontinued and treatment with systemic corticosteroids should be initiated at a dose of 1 to 2 mg/kg/day of prednisone or equivalent. When symptoms improve to Grade 1 or less, corticosteroid taper should be started and continued over at least 1 month.
- In subjects with moderate enterocolitis (Grade 2), pembrolizumab should be withheld and anti-diarrheal treatment should be started. If symptoms are persistent for more than one week, systemic corticosteroids should be initiated (e.g., 0.5 mg/kg/day of prednisone or equivalent). When symptoms improve to Grade 1 or less, corticosteroid taper should be started and continued over at least 1 month. Regarding guidelines for continuing treatment with pembrolizumab.
- All subjects who experience diarrhea 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.

6.5.2 Nausea and Vomiting

Nausea and vomiting should be treated aggressively, and consideration should be given in subsequent cycles to the administration of prophylactic antiemetic therapy according to standard institutional practice. Subjects should be strongly encouraged to maintain liberal oral fluid intake.

6.5.3 Anti-Infectives

Subjects with a documented infectious complication should receive oral or IV antibiotics or other anti-infective agents as considered appropriate by the site investigator for a given infectious condition, according to standard institutional practice.

6.5.4 Management of Infusion Reactions

Acute infusion reactions (which can include cytokine release syndrome, angioedema, or anaphylaxis) are different from allergic/hypersensitive reactions, although some of the manifestations are common to both AEs. Signs and symptoms of acute infusion reactions usually develop during or shortly after drug infusion and generally resolve completely within 24 hours of completion of infusion. Signs/symptoms may include: Allergic reaction/hypersensitivity (including drug fever); Arthralgia (joint pain); Bronchospasm; Cough; Dizziness; Dyspnea (shortness of breath); Fatigue (asthenia, lethargy, malaise); Headache; Hypertension; Hypotension; Myalgia (muscle pain); Nausea; Pruritus/itching; Rash/desquamation; Rigors/chills; Sweating (diaphoresis); Tachycardia; Tumor pain (onset or exacerbation of tumor pain due to treatment); Urticaria (hives, welts, wheals); Vomiting. See Table 7 below:

Table 7: Infusion Reaction Treatment Guidelines

NCI CTCAE Grade	Treatment	Premedication at Subsequent Dosing
Grade 1 Mild reaction; infusion interruption not indicated; intervention not indicated	Increase monitoring of vital signs as medically indicated until the participant is deemed medically stable in the opinion of the investigator.	None
Grade 2 Requires therapy or infusion interruption but responds promptly to symptomatic treatment (e.g., antihistamines, NSAIDs, narcotics, IV fluids); prophylactic medications indicated for ≤24 hrs	**Stop Infusion.** Additional appropriate medical therapy may include but is not limited to: IV fluids Antihistamines NSAIDs Acetaminophen Narcotics Increase monitoring of vital signs as medically indicated until the participant is deemed medically stable in the opinion of the investigator. If symptoms resolve within 1 hour of stopping drug infusion, the infusion may be restarted at 50% of the original infusion rate (e.g. from 100 mL/hr to 50 mL/hr). Otherwise dosing will be held until symptoms resolve and the participant should be premedicated for the next scheduled dose. **Participants who develop Grade 2 toxicity despite adequate premedication should be permanently discontinued from further study drug treatment**	Participant may be premedicated 1.5h (± 30 minutes) prior to infusion of _____ 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 v4.0 (CTCAE) at http://ctep.cancer.gov		

6.5.5 Pneumonitis

Subjects with symptomatic pneumonitis should immediately stop receiving pembrolizumab and have an evaluation. The evaluation may include bronchoscopy and pulmonary function tests to rule out other causes such as infection. If the subject is determined to have pembrolizumab associated pneumonitis, the suggested treatment plan is detailed in Table 8.

Table 8 Recommended Approach to Pneumonitis

Pembrolizumab associated pneumonitis	Withhold/ Discontinue pembrolizumab?	Supportive Care
Grade 1 (asymptomatic)	No action	Intervention not indicated
Grade 2	Withhold pembrolizumab, may return to treatment if improves to Grade 1 or resolves within 12 weeks	Systemic corticosteroids are indicated with an initial dose of 1-2mg/kg/day prednisone or equivalent followed by taper.
Grade 3 and Grade 4	Discontinue pembrolizumab	Systemic corticosteroids are indicated with an initial dose of 1-2mg/kg/day prednisone or equivalent followed by taper. Consider additional non-corticosteroid immunosuppression if warranted. Add prophylactic antibiotics for opportunistic infections. Refer to the Event of Clinical Interest and Immune-related Adverse Event Guidance Document for additional recommendations.

For Grade 2 pneumonitis that improves to ≤ Grade 1 within 12 weeks, the following rules should apply:

- First episode of pneumonitis
  - May increase dosing interval by one week in subsequent cycles
- Second episode of pneumonitis
  - Permanently discontinue pembrolizumab if, upon rechallenge, the subject develops pneumonitis ≥ Grade 2

7. TREATMENT DISCONTINUATION

Subjects may withdraw consent at any time for any reason or be dropped from the trial at the discretion of the site investigator should any untoward effect occur. In addition, a subject may be withdrawn by the site investigator or the sponsor-investigator/HCRN if enrollment into the trial is inappropriate, the trial plan is violated, or for administrative and/or other safety reasons.

If subjects discontinue gemcitabine/cisplatin or gemcitabine chemotherapy treatment due to toxicity commonly associated with cytotoxic chemotherapy (such as neuropathy, hearing loss),

the patient may elect to receive gemcitabine with pembrolizumab (as in Cohort I-p) or alone per the protocol schedule every 3 weeks for a total of 5 doses, or may go off study and choose second-line treatment, after consultation with the treating physician and HCRN/sponsor-investigator. This does not apply due to gemcitabine related grade 3 or 4 myelopsuppression or those with persistent/refractory myelosuppression.

7.1. Reasons for Discontinuing Study Treatment

A subject will be discontinued from the treatment under the following circumstances:

- Biopsy confirmed radiographic disease progression
- If the treating physician thinks a change of therapy would be in the best interest of the subject.
- Intercurrent illness that prevents further administration of treatment
- If the drug(s) exhibit(s) an unacceptable adverse event.
- If a subject becomes pregnant or is unwilling to use appropriate birth control techniques as outlined in the inclusion criteria
- If there is a treatment interruption for greater than 4 weeks due to treatment related adverse event.
- Subjects can stop participating at any time. However, if they decide to stop participating in the study, subjects will continue to be followed for disease progression and survival.

7.2. Reasons for Withdrawal from Study

- Subject withdraws consent for participation
- Termination of the study
- Lost to follow-up
- Death

8. STUDY SCHEDULE OF EVENTS:

8.1. Phase Ib/Cohort I and Phase II/Cohort I/Arm A: Cisplatin-eligible (cycle = 21 days)

Trial Period:	**Screen**	**Treatment Cycles**											**End of Treatment[8]**	**Surgery**	**Post-Treatment**	
															Safety Follow-up[8]	Survival Follow-Up[9]
Treatment Cycle/Title:	Screen	Cycle 1				Cycles 2-4				Cycle 5				2-7wks post pembro	30 days post EOT	Every 12 weeks
		1	2	8	15	1	2	8	15	1	8	15				
Scheduling Window (Days):	-28 to -1	±3	±1	±1	±1	±3	±1	±1	±1	±1	±1	±1			-7/+14	±14
REQUIRED ASSESSMENTS																
Informed Consent	X															
Inclusion/Exclusion Criteria	X															
Demographics and Medical History	X															
Physical Examination	X	X				X					X		X		X	X
Vital Signs and Weight	X	X				X					X		X		X	X
ECOG Performance Status	X	X				X					X		X		X	X
Prior and Concomitant Medication Review	X	X				X					X		X		X	
Review Adverse Events	X	X				X					X		X		X	X
Post-study outcome: RFS, OS																X
LABORATORY ASSESSMENTS																
Pregnancy Test – Urine or Serum β-HCG[1]	X															
PT/INR and aPTT	X															
CBC with Differential	X	X		X		X		X			X		X	X	X	X
Comprehensive Metabolic Panel and LDH	X	X				X					X		X	X	X	X
TSH[2]	X														X	
Urinalysis	X													X		
DISEASE ASSESSMENT																
Tumor Imaging	X													< 21 days pre surgery		X

Trial Period:	**Screen**	**Treatment Cycles**											**End of Treatment[8]**	**Surgery**	**Post-Treatment**	
															Safety Follow-up[8]	Survival Follow-Up[9]
Treatment Cycle/Title:	Screen	Cycle 1				Cycles 2-4				Cycle 5				2-7wks post pembro	30 days post EOT	Every 12 weeks
		1	2	8	15	1	2	8	15	1	8	15				
Scheduling Window (Days):	-28 to -1	±3	±1	±1	±1	±3	±1	±1	±1	±1	±1	±1			-7/+14	±14
TREATMENT EXPOSURE																
Gemcitabine		X		X		X		X								
Cisplatin (Ccr of ≥ 60 mL/min)		X				X										
Cisplatin (Ccr of 50-59 mL/min)		X	X	opt		X	X	opt								
Pembrolizumab[7]				X				X			X[7]					
CORRELATIVE STUDIES																
Unstained Slides-Mandatory[3]		X														
Whole blood collection for PBMC analysis OPTIONAL[4]		X		pre&~ D11[3]												
Serum collection for autoimmune analysis MANDATORY[5]		X														
Tissue collection from radical cystectomy- OPTIONAL[6]														X		

Calendar footnotes:

opt: See section 5.5.4- Split-dose cisplatin is on a Day 1 and 2 regimen to avoid overlap of dexamethasone with pembrolizumab treatment. With documentation of extenuating circumstances, an alternative Day 1 and 8 regimen (and not Day 1 and 2) is acceptable.

1: For females of child bearing potential

2: TSH will be collected at screening and safety follow up visit. Monitoring of additional thyroid tests (ex. T3 and FT4) as well as monitoring of thyroid function at additional time points are at the site investigator's discretion.

3: MANDATORY: Identify for submission unstained slides from archival tissue collection for PD-L1 expression, basal and luminal subtype analysis. Subjects should be encouraged to undergo a biopsy prior to starting therapy if archived tumor tissue is not available and sufficient neoplastic tissue is thought to exist. See SPM for collection, labeling and shipping instructions.

4: OPTIONAL whole blood collection for future PBMC extraction and analysis to be collected at: at pre-treatment Cycle 1 Day 1, pretreatment Cycle 1 Day 8 and Cycle 1 Day 11 to Day 15 (although closer to Day 11 is preferred). See SPM for collection, processing and shipping instructions.

5: Serum collection for subsequent autoimmune analysis. These samples will be banked, and analyzed if a subject develops autoimmune phenomenon on study. If the subject does not develop an autoimmune phenomenon the sample will be stored for future cancer related research

6: OPTIONAL tissue from radical cystectomy or NU to be collected at the time of surgery. Subjects participating at UH Seidman Cancer Center/Case Comprehensive Cancer Center will submit fresh frozen cores for analysis. Other participating centers may submit FFPE block or unstained slides if unable to submit fresh frozen cores. See SPM for additional details.

7: The last dose of Pembrolizumab falls on what would be day 8 of a 5th cycle, however gemcitabine-cisplatin is NOT GIVEN.

8: Post Treatment Safety Follow-Up Visit: This visit should be conducted approximately 30 days (-7/+14) after the last dose of trial treatment or before the initiation of a new anti-cancer treatment, whichever comes first. All subjects should have a safety follow up visit after discontinuation of study treatment whether for progression, toxicity or other reasons. All AEs that occur prior to the Safety Follow-Up Visit should be recorded. Subjects with a treatment related AE of Grade > 1 will be followed until the resolution of the AE to Grade 0-1 or until the beginning of a new anti-neoplastic therapy, whichever occurs first. SAEs that occur within 90 days of the end of treatment or before initiation of a new anti-cancer treatment should also be followed and recorded. The pre-cystectomy surgery visit and post treatment safety follow up may be combined on the same day but the treating medical oncologist must do the toxicity assessment.
9: Post-Treatment Survival Follow-up: Subjects will be followed for 18 months post-surgery. If the subject experiences POD, biopsy should be considered to confirm. Tumor imaging to be performed within 21 days prior to surgery and every 12 weeks thereafter for 18 months from end of treatment while on study. Follow up may be done ± 14 days. After 18 months, subjects will be followed for survival every 6 months for 5 years from the end of treatment or until the study is terminated, whichever occurs first.

8.2 **Phase II/Cohort II/Arm B:** Cisplatin-Ineligible: (cycle = 28 days)

Trial Period:	**Screen**	**Treatment Cycles**												**End of Treatment[8]**	**Surgery**	**Post-Treatment**	
		Cycle 1				Cycle 2-3				Cycle 4						Safety Follow-up[8]	Survival Follow-Up[9]
Treatment Cycle/Title:	Screen	1	8	15	22	1	8	15	22	1	8	15	22		2-7wks post pembro	30 days post EOT	Every 12 weeks
Scheduling Window (Days):	-28 to -1	±3	±1	±1	±1	±3	±1	±1	±1	±1	±1	±1	±1			-7/+14	±14
REQUIRED ASSESSMENTS																	
Informed Consent	X																
Inclusion/Exclusion Criteria	X																
Demographics and Medical History	X																
Physical Examination	X	X				X					X			X		X	X
Vital Signs and Weight	X	X				X					X			X		X	X
ECOG Performance Status	X	X				X					X			X		X	X
Prior and Concomitant Medication Review	X	X				X										X	
Review Adverse Events	X	X				X					X			X		X	X
Post-study outcome: RFS, OS																	X
LABORATORY ASSESSMENTS																	
Pregnancy Test: Urine or Serum β-HCG[1]	X																
PT/INR and aPTT	X																
CBC with Differential	X	X	X	X		X	X	X			X			X	X	X	
Comprehensive Metabolic Panel and LDH	X	X				X		3D15	2D22		X			X	X	X	X
TSH[2]	X															X	
Urinalysis	X														X		
DISEASE ASSESSMENT																	
Tumor Imaging	X														<21d pre cystectomy		X
TREATMENT EXPOSURE																	
Gemcitabine		X	X	X		X	X	X									
Pembrolizumab			X			2D1		3D15	2D22		X[7]						

Trial Period:	Screen	Treatment Cycles												End of Treatment[8]	Surgery	Post-Treatment	
																Safety Follow-up[8]	Survival Follow-Up[9]
Treatment Cycle/Title:	Screen	Cycle 1				Cycle 2-3				Cycle 4					2-7wks post pembrolizumab	30 days post D/C	Every 12 weeks
		1	8	15	22	1	8	15	22	1	8	15	22				
Scheduling Window (Days):	-28 to -1		±1	±1	±1	±3	±1	±1	±1	±1	±1	±1	±1			-7/+14	±14
CORRELATIVE STUDIES																	
Unstained Slides-Mandatory[3]		X															
Whole blood Collection for PBMC analysis- OPTIONAL[4]		X	pre&~ D11[3]														
Serum collection for autoimmune analysis MANDATORY[5]		X															
Tissue collection from radical cystectomy- OPTIONAL[6]															X		

Calendar footnotes:
1: For females of child bearing potential
2: TSH will be collected at screening and safety follow up visit. Monitoring of additional thyroid tests (ex. T3 and FT4) as well as monitoring of thyroid function at additional time points are at the site investigator's discretion.
3: MANDATORY: Identify for submission unstained slides from archival or newly obtained tissue collection for PD-L1 expression, basal and luminal subtype analysis. Subjects should be encouraged to undergo a biopsy prior to starting therapy if archived tumor tissue is not available and sufficient neoplastic tissue is thought to exist. See SPM for collection, labeling and shipping instructions.
4: OPTIONAL whole blood collection for future PBMC extraction and analysis to be collected at: at pre-treatment Cycle 1 Day 1, pretreatment Cycle 1 Day 8 and Cycle 1 Day 11 to Day 15 (closer to Day 11 preferred). See SPM for collection, processing and shipping instructions.
5: Serum collection for subsequent autoimmune analysis. These samples will be banked, and analyzed if a subject develops autoimmune phenomenon on study. If the subject does not develop an autoimmune phenomenon the sample will be stored for future cancer related research.
6: OPTIONAL tissue from radical cystectomy or NU to be collected at the time of surgery. Subjects participating at UH Seidman Cancer Center/Case Comprehensive Cancer Center will submit fresh frozen cores for analysis. Other participating centers may submit FFPE block or unstained slides if unable to submit fresh frozen cores. See SPM for additional details.
7: The last dose of pembrolizumab falls on what would be day 8 of a 4th cycle, however gemcitabine is NOT GIVEN.
8: Post Treatment Safety Follow-Up Visit: This visit should be conducted approximately 30 days (-7/+14) after the last dose of trial treatment or before the initiation of a new anti-cancer treatment, whichever comes first. All subjects should have a safety follow up visit after discontinuation of study treatment whether for progression, toxicity or other reasons. All AEs that occur prior to the Safety Follow-Up Visit should be recorded. Subjects with a treatment related AE of Grade > 1 will be followed until the resolution of the AE to Grade 0-1 or until the beginning of a new anti-neoplastic therapy, whichever occurs first. SAEs that occur within 90 days of the end of treatment or before initiation of a new anti-cancer treatment should also be followed and recorded. The pre-cystectomy surgery visit and post treatment safety follow up may be combined on the same day but the treating medical oncologist must do the toxicity assessment.
9: Post-Treatment Survival Follow-up: Subjects will be followed for 18 months post-surgery. If the subject experiences POD, biopsy should be considered to confirm. Tumor imaging to be performed within 21 days prior to surgery and every 12 weeks thereafter for 18 months from end of treatment while on study. Follow up may be done ± 14 days. After 18 months, subjects will be followed for survival every 6 months for 5 years from the end of treatment or until the study is terminated, whichever occurs first.

8.3 Phase Ib/Cohort I and Phase II/Cohort I/Arm A: Cisplatin eligible

8.3.1 Screening (within 28 days prior to registration for study unless otherwise specified)

- Informed consent
- Physical exam
- Concomitant medication review
- Review adverse events and events of clinical interest
- Vital signs, weight and height (height during Screening ONLY)
- ECOG performance status
- Laboratory Assessments
    - Pregnancy test- urine or serum BHCG for FOCBP
    - PT/INR and aPTT
    - CBC with differential and platelet
    - Comprehensive metabolic profile to include: albumin, alk phos, ALT, AST, BUN and creatinine, CO2 or bicarbonate (if SOC at site), calcium, chloride, glucose, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bili is > ULN), total protein,
    - Lactate Dehydrogenase (LDH),
    - Urinalysis (to include blood, glucose, protein, specific gravity, microscopic (if abnormal results are noted
    - T3, FT4 and TSH
- Imaging studies: CT abdomen/pelvis at minimum. A pelvic MRI may be done in lieu of CT abdomen/pelvis per treating physician discretion and local conventional practice. CT of the chest should be done only if clinically indicated at the discretion of the site investigator.

8.3.2 Cycle 1 Day 1

- Physical Exam
- Concomitant medication review
- Review adverse events and events of clinical interest
- Vital Signs including weight
- ECOG performance status
- Laboratory Assessments
    - CBC with differential and platelet count
    - Comprehensive metabolic profile to include: albumin, alk phos, ALT, AST, BUN and creatinine, CO2 or bicarbonate (if SOC at site), calcium, chloride, glucose, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bili is > ULN), total protein,
    - Lactate Dehydrogenase (LDH),
    - MANDATORY: Serum collection for subsequent autoimmune analysis. These samples will be banked for future cancer related research if not analyzed for an autoimmune phenomenon. Permission will be obtained per informed consent.
    - OPTIONAL: PRE dose whole blood collection for future PBMC extraction and analysis
- Gemcitabine administration
- Cisplatin administration: For Ccr ≥ 50 and < 60 mL/min Cisplatin given in split dose

- MANDATORY: Identify for submission unstained slides from archival tissue collection when available and strongly encourage re-biopsy if the subject agrees and has sufficient retained neoplastic tissue available

8.3.3 Cycle 1 Day 2 (± 3 day)

- Cisplatin administration: For Ccr ≥ 50 and < 60 mL/min Cisplatin given in split dose

8.3.4 Cycle 1 Day 8 (±1 day)

- Laboratory Assessment
  - CBC with differential and platelet count
  - OPTIONAL: PRE dose whole blood collection for future PBMC extraction and analysis
- Gemcitabine administration
- OPTIONAL: If Cisplatin NOT GIVEN on C1D2; may be given C1D8 with appropriate documentation why C1D2 is not feasible
- Pembrolizumab administration

8.3.5 Cycle 1 Day 11-15

- Laboratory Assessment
  - OPTIONAL: PRE dose whole blood collection for future PBMC extraction and analysis. Time point closer to Day 11 is preferred when able.

8.3.6 Cycle 1 Day 15 (± 1 day)

- No scheduled procedures or treatments

8.3.7 Cycle 2-4 Day 1 (± 3 days)

- Physical Exam
- Concomitant medication review
- Review adverse events and events of clinical interest
- Vital Signs including weight
- ECOG performance status
- Laboratory Assessments
  - CBC with differential and platelet count
  - Comprehensive metabolic profile to include: albumin, alk phos, ALT, AST, , BUN and creatinine, CO2 or bicarbonate (if SOC at site), calcium, chloride, glucose, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bili is > ULN), total protein,
  - Lactate Dehydrogenase (LDH)
- Gemcitabine administration
- Cisplatin administration: For Ccr ≥ 50 and < 60 mL/min Cisplatin given in split dose

8.3.8 Cycles 2-4 Day 2 (± 1 day)

- Cisplatin administration: For Ccr ≥ 50 and < 60 mL/min Cisplatin given in split dose

8.3.9 Cycles 2-4 Day 8 (± 1 day)

- Laboratory Assessment
  - CBC with differential and platelet count
- OPTIONAL: If Cisplatin NOT GIVEN on C1D2; may be given C1D8 with appropriate documentation why C1D2 is not feasible
- Pembrolizumab administration

8.3.10 Cycles 2-4 Day 15 (± 1 day)

- No scheduled procedures or treatments

8.3.11 Cycle 5 Day 1 (± 1 day)

- No scheduled procedures or treatments

8.3.12 Cycle 5 Day 8 (± 1 day)

- Physical Exam
- Concomitant medication review
- Review adverse events and events of clinical interest
- Vital Signs including weight
- ECOG performance status
- Laboratory Assessments
  - CBC with differential and platelet count
  - Comprehensive metabolic profile to include: albumin, alk phos, ALT, AST, , BUN and creatinine, CO2 or bicarbonate (if SOC at site), calcium, chloride, glucose, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bili is > ULN), total protein
  - Lactate Dehydrogenase (LDH)
- Pembrolizumab administration

8.3.13 End of Treatment (EOT)

- Physical Exam
- Review adverse events and events of clinical interest
- Vital Signs including weight
- ECOG performance status
- Laboratory Assessments
  - CBC with differential and platelet count
  - Comprehensive metabolic profile to include: albumin, alk phos, ALT, AST, , BUN and creatinine, CO2 or bicarbonate (if SOC at site), calcium, chloride, glucose, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bili is > ULN), total protein
  - Lactate Dehydrogenase (LDH)

8.3.14 Surgery (2-7 weeks post last dose Pembrolizumab)

- Laboratory Assessments
  - CBC with differential and platelet count
  - Comprehensive metabolic profile to include: albumin, alk phos, ALT, AST, , BUN and creatinine, CO2 or bicarbonate (if SOC at site), calcium, chloride, glucose, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bili is > ULN), total protein,

  - Lactate Dehydrogenase (LDH),
  - Urinalysis (to include blood, glucose, protein, specific gravity, microscopic (if abnormal results are noted
- Optional tissue from radical cystectomy or NU to be collected at the time of surgery. Subjects participating at UH Seidman Cancer Center/Case Comprehensive Cancer Center will submit fresh frozen cores for analysis, and is an option for other institutions where resources exist. Other participating centers will submit FFPE block, or unstained slides
- Tumor Imaging-CT Abdomen/ Pelvis is obtained post chemotherapy and within 21 days prior to consolidative surgery. A pelvic MRI may be obtained instead per treating physician standard practice

8.3.15 Safety follow up visit; 30 days (-7/+14) after last dose of pembrolizumab

- Review adverse events and events of clinical interest: all AEs that occur prior to the Safety Follow-Up Visit should be recorded. Subjects with a treatment related AE of Grade > 1 will be followed until the resolution of the AE to Grade 0-1 or until the beginning of a new anti-neoplastic therapy, whichever occurs first. SAEs that occur within 90 days of the end of treatment or before initiation of a new anti-cancer treatment should also be followed and recorded.
- Physical Exam
- Vital Signs including weight
- ECOG performance status
- Laboratory Assessments
  - CBC with differential and platelet count
  - Comprehensive metabolic profile to include: albumin, alk phos, ALT, AST, , BUN and creatinine, CO2 or bicarbonate (if SOC at site), calcium, chloride, glucose, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bili is > ULN), total protein,
  - Lactate Dehydrogenase (LDH),
  - TSH, FT_4, T_3

8.3.16 Relapse and Survival follow up (± 14 days) every 12 weeks up to 18 months post-surgery

- Physical Exam
- Review adverse events and events of clinical interest
- Vital Signs including weight and height
- ECOG performance status
- Laboratory Assessments
  - Comprehensive metabolic profile to include: albumin, alk phos, ALT, AST, , BUN and creatinine, CO2 or bicarbonate (if SOC at site), calcium, chloride, glucose, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bili is > ULN), total protein,
  - Lactate Dehydrogenase (LDH),
- Restaging with a CT chest/abdomen/pelvis to be performed. If the subject experiences POD, a biopsy should be considered to confirm.
- After 18 months, subjects will be followed for survival every 6 months for 5 years from the end of treatment or until the study is terminated, whichever occurs first.

8.4 Phase II/Cohort II/Arm B: Cisplatin Ineligible:

8.4.1 Screening (within 28 days prior to registration for study unless otherwise specified)

- Informed consent
- Physical exam
- Concomitant Medication Review
- Review adverse events and events of clinical interest
- Vital signs, weight and height (height during Screening ONLY)
- ECOG performance status
- Laboratory Assessments
  - Pregnancy test- urine or serum BHCG for FOCBP
  - PT/INR and aPTT
  - CBC with differential and platelet
  - Comprehensive metabolic profile to include: albumin, alk phos, ALT, AST, BUN and creatinine, CO2 or bicarbonate (if SOC at site), calcium, chloride, glucose, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bili is > ULN), total protein,
  - Lactate Dehydrogenase (LDH),
  - Urinalysis (to include blood, glucose, protein, specific gravity, microscopic (if abnormal results are noted
  - T3, FT4 and TSH
- Imaging studies: CT abdomen/pelvis at minimum. A pelvic MRI may be done in lieu of CT abdomen/pelvis per treating physician discretion and local conventional practice. CT of the chest should be done only if clinically indicated at the discretion of the site investigator.

8.4.2 Cycle 1 Day 1

- Physical Exam
- Concomitant medication review
- Review adverse events and events of clinical interest
- Vital Signs including weight
- ECOG performance status
- Laboratory Assessments
  - CBC with differential and platelet count
  - Comprehensive metabolic profile to include: albumin, alk phos, ALT, AST, , BUN and creatinine, CO2 or bicarbonate (if SOC at site), calcium, chloride, glucose, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bili is > ULN), total protein
  - Lactate Dehydrogenase (LDH)
  - MANDATORY: Serum collection for subsequent autoimmune analysis. These samples will be banked for future cancer related research if not analyzed for autoimmune phenomenon. Permission will be obtained per informed consent.
  - OPTIONAL: PRE dose whole blood collection for future PBMC extraction and analysis
- Submission of unstained slides from archival tissue collection; MANDATORY
- Gemcitabine administration

8.4.3 Cycle 1 Day 8 (±1 day)

- Laboratory Assessments
  - CBC with differential and platelet count
  - OPTIONAL: PRE dose whole blood collection for future PBMC extraction and analysis
- Gemcitabine administration
- Pembrolizumab administration

8.4.4 Cycle 1 Day 11-15

- Laboratory Assessment
  - OPTIONAL: PRE dose whole blood collection for future PBMC extraction and analysis. Time point closer to Day 11 is preferred when able.

8.4.5 Cycle 1 Day 15 (± 1 day)

- Laboratory Assessment
  - CBC with differential and platelet count
- Gemcitabine administration

8.4.6 Cycle 1 Day 22 (± 1 day)

- No scheduled procedures or treatments

8.4.7 Cycle 2-3 Day 1 (± 3 days)

- Physical Exam
- Concomitant medication review
- Review adverse events and events of clinical interest
- Vital Signs including weight
- ECOG performance status
- Laboratory Assessments
  - CBC with differential and platelet count
  - Comprehensive metabolic profile to include: albumin, alk phos, ALT, AST, , BUN and creatinine, CO2 or bicarbonate (if SOC at site), calcium, chloride, glucose, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bili is > ULN), total protein
  - Lactate Dehydrogenase (LDH)
- Gemcitabine administration
- CYCLE 2 DAY 1 ONLY: Pembrolizumab administration

8.4.8 Cycles 2-3 Day 8 (± 1 day)

- Laboratory Assessment
  - CBC with differential and platelet count
- Gemcitabine administration

8.4.9 Cycles 2-3 Day 15 (± 1 day)

- Laboratory Assessments
  - CBC with differential and platelet count
  - CYCLE 3 DAY 15 ONLY: Comprehensive metabolic profile to include: albumin, alk phos, ALT, AST, BUN and creatinine, CO2 or bicarbonate (if SOC at site), calcium, chloride, glucose, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bili is > ULN), total protein
  - CYCLE 3 DAY 15 ONLY: Lactate Dehydrogenase (LDH),
- Gemcitabine administration
- CYCLE 3 DAY 15 ONLY: Pembrolizumab administration

8.4.10 Cycle 2-3 Day 22 (± 1 day)

- Laboratory Assessments
  - CYCLE 2 DAY 22 ONLY: Comprehensive metabolic profile to include: albumin, alk phos, ALT, AST, BUN and creatinine, CO2 or bicarbonate (if SOC at site), calcium, chloride, glucose, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bili is > ULN), total protein
  - CYCLE 2 DAY 22 ONLY: Lactate Dehydrogenase (LDH)
- CYCLE 2 DAY 22 ONLY: Pembrolizumab administration

8.4.11 Cycle 4 Day 1 (± 1 day)

- No scheduled procedures or treatments

8.4.12 Cycle 4 Day 8 (± 1 day)

- Physical Exam
- Concomitant medication review
- Review adverse events and events of clinical interest
- Vital Signs including weight
- ECOG performance status
- Laboratory Assessments
  - CBC with differential and platelet count
  - Comprehensive metabolic profile to include: albumin, alk phos, ALT, AST, , BUN and creatinine, CO2 or bicarbonate (if SOC at site), calcium, chloride, glucose, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bili is > ULN), total protein
  - Lactate Dehydrogenase (LDH)
- Pembrolizumab administration

8.4.13 End of Treatment (EOT)

- Physical Exam
- Review adverse events and events of clinical interest
- Vital Signs including weight
- ECOG performance status

- Laboratory Assessments
  - CBC with differential and platelet count
  - Comprehensive metabolic profile to include: albumin, alk phos, ALT, AST, , BUN and creatinine, CO2 or bicarbonate (if SOC at site), calcium, chloride, glucose, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bili is > ULN), total protein
  - Lactate Dehydrogenase (LDH)

8.4.14 Surgery (2-7 weeks post last dose pembrolizumab)

- Laboratory Assessments
  - CBC with differential and platelet count
  - Comprehensive metabolic profile to include: albumin, alk phos, ALT, AST, , BUN and creatinine, CO2 or bicarbonate (if SOC at site), calcium, chloride, glucose, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bili is > ULN), total protein,
  - Lactate Dehydrogenase (LDH),
  - Urinalysis (to include blood, glucose, protein, specific gravity, microscopic (if abnormal results are noted
- Tumor Imaging-CT Abdomen/ Pelvis is obtained post chemotherapy and within 21 days prior to consolidative surgery. A pelvic MRI may be obtained instead per treating physician standard practice
- OPTIONAL tissue from radical cystectomy or NU to be collected at the time of surgery. Subjects participating at UH Seidman Cancer Center/Case Comprehensive Cancer Center will submit fresh frozen cores for analysis, and is an option for other institutions where resources exist. Other participating centers will submit FFPE block or unstained slides

8.4.15 Safety follow up visit; 30 days (-7/+14) after last dose of pembrolizumab

- Review adverse events and events of clinical interest: all AEs that occur prior to the Safety Follow-Up Visit should be recorded. Subjects with a treatment related AE of Grade > 1 will be followed until the resolution of the AE to Grade 0-1 or until the beginning of a new anti-neoplastic therapy, whichever occurs first. SAEs that occur within 90 days of the end of treatment or before initiation of a new anti-cancer treatment should also be followed and recorded.
- Vital Signs including weight
- ECOG performance status
- Laboratory Assessments
  - CBC with differential and platelet count
  - Comprehensive metabolic profile (albumin, alk phos, ALT, AST, LDH, C02 or bicarbonate (if standard of care at site), uric acid, calcium, chloride, glucose, phosphorus, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bilirubin is >ULN), total protein, BUN, creatinine
  - TSH, FT_4, T_3

8.4.16 Relapse and Survival follow up (± 14 days) every 12 weeks up to 18 months post-surgery

- Physical Exam
- Review adverse events and events of clinical interest
- Vital Signs including weight and height

- ECOG performance status
- Laboratory Assessments
  - Comprehensive metabolic profile to include: albumin, alk phos, ALT, AST, , BUN and creatinine, CO2 or bicarbonate (if SOC at site), calcium, chloride, glucose, potassium, sodium, magnesium, total bilirubin, direct bilirubin (if total bili is > ULN), total protein
  - Lactate Dehydrogenase (LDH)
- Restaging with a CT chest/abdomen/pelvis to be performed. If the subject experiences POD, a biopsy should be considered to confirm.
- After 18 months, subjects will be followed for survival every 6 months for 5 years from the end of treatment or until the study is terminated, whichever occurs first.

8.5 Pre-operative and Post-Operative Restaging Evaluation

CT Abdomen, Pelvis is obtained post neoadjuvant chemotherapy within 21 days prior to consolidative surgery. A pre-operative pelvic MRI may be done instead per site investigator's discretion. CT Chest, Abdomen, Pelvis 12 weeks post operatively and every 3 months thereafter for restaging follow-up. Subjects with restaging scans that are concerning for relapse should be considered for pathologic confirmation. RECISTv1.1 and irRC evaluation.

8.6 End of Treatment (EOT)

Vital signs, history and physical exam, ECOG performance status, CBC and differential, comprehensive metabolic profile with LFTs, and review of adverse events should all be performed at end of treatment or discontinuation of treatment. This may occur after dose 5 pembrolizumab and surgery, or if treatment is discontinued for toxicity.

8.7 Follow up

Subject should have a history and physician exam with treating oncologist every 12 weeks (±14 days) including evaluation of restaging scans, delayed toxicities and performance status.

9 CRITERIA FOR DISEASE EVALUATION

Primary efficacy endpoint response is determined at radical cystectomy and lymph node dissection (or nephroureterectomy) where the pTNM is compared to the initial clinical TNM stage (cTNM) assessed at study entry. Investigators should work with their team of pathologists and urologists to determine the most accurate cTNM and pTNM stage. It is recognized that cTNM staging requires integration of imaging, pathologic, and physical exam findings, and reproducibility can be difficult. To limit variation between investigators and institutions, the table below outlines cTNM staging. Investigators may also refer to AJCC staging manual, 7th edition (2009) or the NCCN guidelines page ST-2 as an additional reference when considering how to distinguish cT2 (muscle invasion) from cT3 (bladder wall thickening) from cT4 (fixed mass on exam under anesthesia or extra-vesicle extension to perivesicle fat).[68,69]

Table 9a: Summary of clinical TNM staging of bladder urothelial cancer to guide primary tumor stage assessment. EUA, exam under anesthesia

	Pathology	Cross Sectional Imaging	EUA
cT1	Tumor does not invade muscularis	May or may not be visible, bladder wall not thickened	Bladder and or mass may or may not be appreciated and not fixed
cT2	Tumor invades muscularis propria, depth of penetration is sometimes described to involve inner or outer half (cT2a vs cT2b)	Bladder wall not thickened; a mass may or may not be visible	Mass is Mobile
cT3	Tumor invades muscularis, depth of penetration is sometimes described to include perivesical tissue	Bladder wall is thickened; a mass may or may not be visible	Mass is mobile
cT4a	Tumor invades muscularis, depth of penetration is sometimes described	Bladder wall is thickened; a mass is often visible with attention for extravesicular extension and may be seen to invade prostate, uterus, vagina	Mass is fixed
cT4b	Tumor invades muscularis, depth of penetration is sometimes described	Mass visibly extends to the pelvic wall or abdominal wall	Mass is fixed

Response assessments will be made both using the Immune Related Response Criteria (irRC) for the pre-surgery restaging scan, and using RECIST v1.1 for all additional post-surgery follow up scans. The same measurable and non-measurable lesions will be followed by both RECIST v1.1 & irRC. Post Neoadjuvant chemotherapy/pre-surgery clinical TNM staging should be based on RECIST v1.1.

9.1 Immune related response criteria

This study will utilize the Immune Related Response Criteria (irRC) during the time the patient is treated with pembrolizumab: *ie*, *the post-chemotherapy/pre-surgical restaging evaluation. Those subjects that are not surgical candidates and remain on study should also be followed by both RECIST v1.1 and irRC.* These response criteria were developed to overcome the variable and unusual patterns of response to immunotherapeutic agents, in particular, ipilimumab[70]. The development of the guidelines were prompted by observations, mostly in subjects with metastatic melanoma, of initial disease progression followed by later response, late responses, and mixed responses with an overall decrease in tumor burden.

9.2 Antitumor response based on total measurable tumor burden

For the irRC, only index and measurable new lesions are taken into account (in contrast to conventional WHO criteria, which do not require the measurement of new lesions, nor do they include new lesion measurements in the characterization of evolving tumor burden). At the baseline tumor assessment, the sum of the products of the two largest perpendicular diameters (SPD) of all index lesions (five lesions per organ, up to 10 visceral lesions) is calculated. At each

subsequent tumor assessment, the SPD of the index lesions and of new, measurable lesions (up to 5 new lesions per organ; 10 visceral lesions) are added together to provide the total tumor burden:

$$\text{Tumor Burden} = SPD_{\text{index lesions}} + SPD_{\text{new, measurable lesions}}$$

Table 9b: Comparison of WHO and irRC criteria

	WHO	irRC
New, measurable lesions	Always represent PD	Incorporated into tumor burden
New, nonmeasurable lesions	Always represent PD	Do not define progression (but preclude irCR)
Non-index lesions	Changes contribute to defining BOR of CR, PR, SD, and PD	Contribute to defining irCR (complete disappearance required)
CR	Disappearance of all lesions in two consecutive observations not less than 4 wk apart	Disappearance of all lesions in two consecutive observations not less than 4 wk apart
PR	≥50% decrease in SPD of all index lesions compared with baseline in two observations at least 4 wk apart, in absence of new lesions or unequivocal progression of non-index lesions	≥50% decrease in tumor burden compared with baseline in two observations at least 4 wk apart
SD	50% decrease in SPD compared with baseline cannot be established nor 25% increase compared with nadir, in absence of new lesions or unequivocal progression of non-index lesions	50% decrease in tumor burden compared with baseline cannot be established nor 25% increase compared with nadir
PD	At least 25% increase in SPD compared with nadir and/or unequivocal progression of non-index lesions and/or appearance of new lesions (at any single time point)	At least 25% increase in tumor burden compared with nadir (at any single time point) in two consecutive observations at least 4 wk apart

9.3 Time-point response assessment using irRC

Percentage changes in tumor burden per assessment time point describe the size and growth kinetics of both conventional and new, measurable lesions as they appear. At each tumor assessment, the response in index and new, measurable lesions is defined based on the change in tumor burden (after ruling out irPD). Decreases in tumor burden must be assessed relative to baseline measurements (i.e., the SPD of all index lesions at screening). The irRC were derived from WHO criteria and, therefore, the thresholds of response remain the same. However, the irRC response categories have been modified from those of WHO criteria as detailed in Tables 10 and 11.

9.4 Overall response using the irRC

The sum of the products of diameters at tumor assessment using the immune-related response criteria (irRC) for progressive disease incorporates the contribution of new measurable lesions. Each net Percentage Change in Tumor Burden per assessment using irRC criteria accounts for the size and growth kinetics of both old and new lesions as they appear.

Definition of Index Lesions Response Using irRC

- **irComplete Response (irCR):** Complete disappearance of all *index* lesions. This category encompasses exactly the same subjects as "CR" by the mWHO criteria.
- **irPartial Response (irPR):** Decrease, relative to baseline, of 50% or greater in the sum of the products of the two largest perpendicular diameters of all *index* and all new measurable lesions (ie., Percentage Change in Tumor Burden). Note: the appearance of new measurable lesions is factored into the overall tumor burden, but does not automatically qualify as progressive disease until the SPD increases by ≥25% when compared to SPD at nadir.
- **irStable Disease (irSD):** Does not meet criteria for irCR or irPR, in the absence of progressive disease.
- **irProgressive Disease (irPD):** At least 25% increase Percentage Change in Tumor Burden (i.e., taking sum of the products of all *index* lesions and any new lesions) when compared to SPD at nadir.

Definition of Non-Index Lesions Response Using irRC

- **irComplete Response (irCR):** Complete disappearance of all *non-index* lesions. This category encompasses exactly the same subjects as "CR" by the mWHO criteria.
- **irPartial Response (irPR) or irStable Disease (irSD):** *non-index* lesion(s) are not considered in the definition of PR, these terms do not apply.
- **irProgressive Disease (irPD):** Increases in number or size of *non-index* lesion(s) does not constitute progressive disease unless/until the Percentage Change in Tumor Burden increases by 25% (i.e., the SPD at nadir of the index lesions increases by the required amount).

Impact of New Lesions on irRC

New lesions in and by themselves do not qualify as progressive disease. However, their contribution to total tumor burden is included in the SPD which in turn feeds into the irRC criteria for tumor response. Therefore, new non-measurable lesions will not discontinue any subject from the study.

Definition of Overall Response Using irRC

Overall response using irRC will be based on these criteria **(see Table 10**):

- **Immune-Related Complete Response (irCR):** Complete disappearance of *all* tumor lesions (index and nonindex together with no new measurable/unmeasurable lesions) for at least 4 weeks from the date of documentation of complete response.
- **Immune-Related Partial Response (irPR):** The sum of the products of the two largest perpendicular diameters of all index lesions is measured and captured as the SPD baseline. At each subsequent tumor assessment, the sum of the products of the two largest perpendicular diameters of all index lesions and of new measurable lesions are added together to provide the Immune Response Sum of Product Diameters (irSPD). A decrease, relative to baseline of the irSPD compared to the previous SPD baseline, of 50% or greater is considered an immune Partial Response (irPR).
- **Immune-Related Stable Disease (irSD):** irSD is defined as the failure to meet criteria for immune complete response or immune partial response, in the absence of progressive disease.

- **Immune-Related Progressive Disease (irPD):** It is recommended in difficult cases to confirm PD by serial imaging. Any of the following will constitute progressive disease:
  - At least 25% increase in the sum of the products of all index lesions over nadir SPD calculated for the index lesions.
  - At least a 25% increase in the sum of the products of all index lesions and new measurable lesions (irSPD) over the baseline SPD calculated for the index lesions.

Immune-Related Best Overall Response Using irRC (irBOR)

irBOR is the best confirmed irRC overall response over the study as a whole, recorded between the date of first dose until the last tumor assessment before subsequent therapy (except for local palliative radiotherapy for painful bone lesions) for the individual subject in the study. For the assessment of irBOR, all available assessments per subject are considered.

irCR or irPR determinations included in the irBOR assessment must be confirmed by a second (confirmatory) evaluation meeting the criteria for response and performed no less than 4 weeks after the criteria for response are first met.

Table 10: Derivation of irRC overall responses

Measurable response	Nonmeasurable response		Overall response
Index and new, measurable lesions (tumor burden),*%	Non-index lesions	New, nonmeasurable lesions	Using irRC
↓100	Absent	Absent	irCR†
↓100	Stable	Any	irPR†
↓100	Unequivocal progression	Any	irPR†
↓≥50	Absent/Stable	Any	irPR†
↓≥50	Unequivocal progression	Any	irPR†
↓<50 to <25↑	Absent/Stable	Any	irSD
↓<50 to <25↑	Unequivocal progression	Any	irSD
≥25	Any	Any	irPD†

*Decreases assessed relative to baseline (scan prior to start of any protocol therapy), including measurable lesions only

†Assuming response (irCR) and progression (irPD) are confirmed by a second, consecutive assessment at least 4 wk apart.

9.5 Definitions Associated with Response Evaluation Criteria in Solid Tumors (RECIST) version 1.1

9.5.1 Measurable disease

The presence of at least one measurable lesion. If the measurable disease is restricted to a solitary lesion, its neoplastic nature should be confirmed by cytology/histology.

9.5.2 Measurable lesions

Measurable lesions are defined as those that can be accurately measured in at least one dimension (longest diameter to be recorded) as ≥20 mm by chest x-ray, as ≥10 mm with CT

scan, or ≥10 mm with calipers by clinical exam. All tumor measurements must be recorded in millimeters (or decimal fractions of centimeters).

9.5.3 Non-measurable lesions

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

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

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

9.5.4 Malignant lymph nodes

To be considered pathologically enlarged and measurable, a lymph node must be ≥15 mm in short axis when assessed by CT scan (CT scan slice thickness recommended to be no greater than 5 mm). At baseline and in follow-up, only the short axis will be measured and followed.

9.5.5 Target lesions

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

9.5.6 Non-target lesions

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

9.6 Response Criteria

9.6.1. Evaluation of target lesions

Complete Response (CR)	Disappearance of all target lesions. Any pathological lymph nodes (whether target or non-target) must have reduction in short axis to <10 mm.
Partial Response (PR)	At least a 30% decrease in the sum of the diameters of target lesions, taking as reference the baseline sum diameters
Progressive Disease (PD)	At least a 20% increase in the sum of the diameters of target lesions, taking as reference the smallest sum on study (this includes the baseline sum if that is the smallest on study). In addition to the relative increase of 20%, the sum must also demonstrate an absolute increase of at least 5 mm. (Note: the appearance of one or more new lesions is also considered progressions).
Stable Disease (SD)	Neither sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD, taking as reference the smallest sum diameters while on study

9.6.2. Evaluation of non-target lesions

Complete Response (CR)	Disappearance of all non-target lesions and normalization of tumor marker level. All lymph nodes must be non-pathological in size (<10 mm short axis) Note: If tumor markers are initially above the upper normal limit, they must normalize for a subject to be considered in complete clinical response.
Non-CR/ Non-PD	Persistence of one or more non-target lesion(s) and/or maintenance of tumor marker level above the normal limits
Progressive Disease (PD)	Appearance of one or more new lesions and/or unequivocal progression of existing non-target lesions. Unequivocal progression should not normally trump target lesion status. It must be representative of overall disease status change, not a single lesion increase.

Although a clear progression of "non-target" lesions only is exceptional, the opinion of the treating physician should prevail in such circumstances, and the progression status should be confirmed at a later time by the sponsor-investigator.

9.6.3 Evaluation of best overall response

Target Lesions	Non-Target Lesions	New Lesions	Overall Response
CR	CR	No	CR
CR	Non-CR/ Non-PD	No	PR
	Not evaluated	No	PR
PR	Non-CR/ Non-PD/ not evaluated	No	PR
SD	Non-CR/ Non-PD/ not evaluated	No	SD
PD	Any	Yes or No	PD
Any	PD*	Yes or No	PD
Any	Any	Yes	PD

*In exceptional circumstances, unequivocal progression in non-target lesions may be accepted as disease progression.

Subjects with a global deterioration of health status requiring discontinuation of treatment without objective evidence of disease progression at that time should be classified as having "symptomatic deterioration". Every effort should be made to document the objective progression even after discontinuation of treatment.

In some circumstances it may be difficult to distinguish residual disease from normal tissue. When the evaluation of complete response depends on this determination, it is recommended that the residual lesion be investigated (fine needle aspirate/biopsy) to confirm the complete response status.

9.7 Definitions for Response Evaluation – RECIST version 1.1

9.7.1 First Documentation of Response

The time between initiation of therapy and first documentation of PR or CR.

9.7.2 Confirmation of Response

To be assigned a status of complete or partial response, changes in tumor measurements must be confirmed by repeat assessments performed no less than four weeks after the criteria for response are first met.

9.7.3 Duration of Response

Duration of overall response—the period measured from the time that measurement criteria are met for complete or partial response (whichever status is recorded first) until the date that recurrent or progressive disease is objectively documented (taking as reference for progressive disease the smallest measurements recorded since treatment started).

9.7.4 Duration of Overall Complete Response

The period measured from the time that measurement criteria are met for complete response until the first date that recurrent disease is objectively documented.

9.7.5 Objective response rate
The objective response rate is the proportion of all subjects with confirmed PR or CR according to RECIST v1.1, from the start of treatment until disease progression/recurrence (taking as reference for progressive disease the smallest measurements recorded since the start of treatment).

9.7.6 Time to Progression:
A measurement from the date of randomization until the criteria for disease progression is met as defined by RECIST 1.1. Subjects who have not progressed or have died due to any cause will be right-censored at the date of the last disease evaluation or date of death.

9.7.7 Relapse Free Survival
A measurement from the date of randomization until the criteria for disease progression is met as defined by RECIST 1.1 or death occurs. Radiographic suggestion of relapse should be followed with biopsy to document disease recurrence versus another process. Subjects who have not progressed or relapsed will be right-censored at the date of the last disease evaluation.

9.7.8 Overall Survival
Overall survival is defined by the date of randomization to date of death from any cause.

10. BIOLOGICAL SPECIMEN PARAMETERS FOR CORRELATIVES
Please see Study Procedures Manual for additional information regarding collection, labeling and shipping instructions.

10.1 Prior to Treatment Archival Tissue or Biopsy

10.1.1 MANDATORY; PD-L1 Testing
Submission of 5 unstained slides from an archival tissue collection for PD-L1 expression analysis. If archival tissue is not available for slides, subjects are strongly encouraged to undergo a biopsy prior to starting. M1 subjects are not eligible on this trial. Additionally, tumor tissue from bone metastases is not evaluable for PD-L1 expression and is therefore not acceptable.

Tumor tissue should be of good quality based on total and viable tumor content. Fine-needle aspiration, brushing, cell pellet from pleural effusion, bone metastases, and lavage samples are not acceptable. For core needle biopsy specimens, at least three cores or via TURBT should be submitted for evaluation.

10.1.2 MANDATORY; Tumor Subtype Testing
Submission of 6 unstained slides from archival or, when feasible, newly obtained tissue collection for IHC testing of basal and luminal urothelial subtypes. IHC markers that will be analyzed are CD44, CD20 and CD5/6. Subjects are strongly encouraged to undergo a biopsy prior to starting therapy if archived tumor tissue is not available.

10.2 On Study Surgical Biopsy

10.2.1 OPTIONAL; Fresh Frozen Cores from Surgical Specimen

Seidman Cancer Center/Case Comprehensive Cancer Center sites *as well as* other interested sites will provide frozen tissue cores (3) collected at the time of radical cystectomy or NU. Tissue will include muscle and obtained from anywhere along the luminal side of the specimen. Analysis will include histology, gene and protein analysis for stromal lymphocyte characterization.

10.2.2 OPTIONAL; Formalin Fixed Paraffin Embedded (FFPE)

Encouraged for all sites. Luminal side tissue that includes muscle measuring at least 0.5cm x 0.5cm x 0.5cm will be collected from a random site of the radical cystectomy or NU as a FFPE block, or 10 unstained slides post sectioning. Analysis will include histology, and a tissue microarray for protein analysis, and stromal lymphocyte characterization. Data from these samples may be limited by ability to characterize lymphocytes in FFPE tissue and assays are being optimized.

10.3 Whole Blood Collection

10.3.1 OPTIONAL; Whole Blood Collection for Future PBMC Analysis

- Pre-treatment Cycle 1 Day 1
- Pre-treatment Cycle 1 Day 8
- One collection during Cycle 1 Day 11-15, with date and time recorded. Time point closer to Day 11 is preferred when able.

10.3.2 MANDATORY; Whole Blood Collection for Banked Serum

Whole blood for serum collection will be drawn at Pre-dose Cycle 1 Day 1 and will be banked for the duration of the study. The serum sample will be used to determine if a subject who develops autoimmune phenomenon had pre-existing antibodies and/or other serum findings that are consistent with the subject's specific autoimmune diagnosis. If the subject does not develop an autoimmune phenomenon the sample will be stored for future cancer related research.

11. DRUG INFORMATION

11.1 Pembrolizumab

Please refer to the current version of the Investigator's Brochure for additional information on this medication.

11.1.1 Other Names

Pembrolizumab; MK-3475 [Anti-PD-1 Antibody MK-3475]

11.1.2 Chemical Name and Properties

Humanized X PD-1_mAb (H409A11) IgG4

11.1.3 Availability and Distribution

Merck will supply pembrolizumab directly to sites at no charge to subjects participating in this clinical trial.

Clinical Supplies will be provided by Merck as summarized below.

Product Descriptions

Product Name & Potency	Dosage Form
Pembrolizumab 50 mg	Lyophilized Powder for Injection
Pembrolizumab 100 mg/ 4mL	Solution for Injection

11.1.4 Preparation and Administration

Please refer to the Pharmacy Manual for a comprehensive description of pembrolizumab preparation.

Pembrolizumab is provided as a white to off-white lyophilized powder (50 mg/vial) or as a liquid solution (100 mg/vial) in Type I glass vials intended for single use only. Pembrolizumab Powder for Solution for Infusion, 50 mg/vial, is reconstituted with sterile water for injection prior to use. Pembrolizumab is formulated with L-histidine as buffering agent, polysorbate 80 as surfactant, sucrose as stabilizer/tonicity modifier, and hydrochloric acid (HCl) and/or sodium hydroxide (NaOH) for pH adjustment (if necessary). The drug product is stored as a stable lyophilized powder or liquid solution under refrigerated conditions (2°C - 8°C).

The product after reconstitution with sterile water for injection and the liquid drug product is a clear to opalescent solution, which may contain extraneous and proteinaceous particulates. The reconstituted product and liquid product is intended for IV administration. The reconstituted drug product solution and liquid drug product can be further diluted with normal saline in IV containers made of polyvinyl chloride (PVC) or non-PVC material. Reconstituted vials should be immediately used to prepare the infusion solution in the IV bag and the infusion solution should be immediately administered. If not used immediately, vials and/or IV bags may be stored at 2-8 °C for up to a cumulative time of 20 hours. If refrigerated, the vials and/or IV bags should be allowed to equilibrate to room temperature prior to subsequent use. Pembrolizumab solutions may be stored at room temperature for a cumulative time of up to 4 hours. This includes room temperature storage of reconstituted drug product solution and liquid drug product in vials, room temperature storage of infusion solution in the IV bag and the duration of infusion.

11.1.5 Adverse Events

Please see current Investigator's Brochure for a complete list of adverse events.

Pembrolizumab is generally well tolerated and demonstrates a favorable safety profile in comparison to chemotherapy. Pembrolizumab is an immunomodulatory agent, and based on this mechanism of action, immune mediated adverse events are of primary concern. Important identified risks for pembrolizumab are of an immune mediate nature, including: pneumonitis, colitis, hypophysitis (including hypothryoidism/hyperthyroidism), hepatitis, Type I diabetes mellitis, uveitis, and nephritis, myositis, Guillain-Barre syndrome, pancreatitis, and severe skin

reaction toxic epidermal necrolysis (TEN), some with fatal outcome). A new important risk of myocarditis has been identified; cases with fatal outcome have been reported.

The majority of immune-mediated adverse events were mild to moderate in severity, were manageable with appropriate care, and rarely required discontinuation of therapy.

In addition to the previously noted identified risks, infusion-related reactions are a risk but are not considered immune mediated; these are also further described in the current IB.

11.2 Gemcitabine

Please see package insert for detailed information regarding this medication

11.2.1 Other Names

2'-Deoxy-2',2'-difluorocytidine monohydrochloride, Gemzar

11.2.2 Classification

Antimetabolite (nucleoside pyrimidine analogue)

11.2.3 Mode of Action

Gemcitabine exhibits cell phase specificity, primarily killing cells undergoing DNA synthesis (S phase) and also blocking the progression of cells through the G1/S phase boundary. Gemcitabine is metabolized intracellularly by nucleoside kinases to the active diphosphate (dFdCDP) and triphosphate (dFdCTP) nucleosides. The cytotoxic effect of gemcitabine is attributed to a combination of two actions of the diphosphate and the triphosphate nucleosides, which leads to inhibition of DNA synthesis. First, gemcitabine diphosphate inhibits ribonucleotide reductase, which is responsible for catalyzing the reactions that generate the deoxynucleoside triphosphates for DNA synthesis. Inhibition of this enzyme by the diphosphate nucleoside causes a reduction in the concentrations of deoxynucleotides, including dCTP. Second, gemcitabine triphosphate competes with dCTP for incorporation into DNA. The reduction in the intracellular concentration of dCTP (by the action of the diphosphate) enhances the incorporation of gemcitabine triphosphate into DNA (self-potentiation). After the gemcitabine nucleotide is incorporated into DNA, only one additional nucleotide is added to the growing DNA strands. After this addition, there is inhibition of further DNA synthesis. DNA polymerase epsilon is unable to remove the gemcitabine nucleotide and repair the growing DNA strands (masked chain termination).

11.2.4 Storage and Stability

Unreconstituted drug vials are stored at controlled room temperature (15ºC to 30ºC, 59ºF to 86ºF). Reconstituted solution should be stored at controlled room temperature and used within 24 hours. Solutions of gemcitabine should not be refrigerated; as crystallization may occur. The unused portion should be discarded.

11.2.5 Preparation

Gemcitabine may be further diluted with normal saline as per institutional standards.

11.2.6 Route of Administration
IV infusion.

11.2.7 Availability
Gemcitabine is commercially available in 200 mg and 1 gm vials

11.2.8 Side Effects
Please see package insert for detailed information regarding side effects related to Gemcitabine.

Neutropenia, anemia, thrombocytopenia, and leukopenia are reported. Rash with pruritis and injection-site reactions are can occur. Nausea and vomiting, diarrhea, constipation and mucositis have been reported. Abnormalities of hepatic transaminase enzymes occur in two-thirds of subjects, but they are usually mild, non-progressive, and rarely necessitate stopping treatment. Bronchospasm and/or dyspnea within a few hours of infusion of the drug, cough, rhinitis, pneumonitis may occur. Somnolence, insomnia, paresthesia, pain. Peripheral edema is reported in about 30% of subjects. Flu-like symptoms are reported for about 20% of subjects. This includes fever, headache, back pain, chills, myalgia, asthenia, and anorexia.

11.3 Cisplatin
Please see package insert for detailed information regarding side effects related to this drug.

11.3.1 Other Names
Cisdiaminedichloroplatinum, Cis-diaminedichloroplatinum (II), diaminedichloroplatinum, cis-platinum, platinum, Platinol, Platinol-AQ, DDP, CDDP, DACP, NSC 119875.

11.3.2 Classification
Alkylating agent

11.3.3 Mode of Action
Inhibits DNA synthesis by forming inter- and intra-strand crosslinks. Other possible mechanisms include chelation of DNA and binding to cell membranes thereby stimulating immune mechanisms.

11.3.4 Storage and Stability
Inhibits DNA synthesis by forming inter- and intra-strand crosslinks. Other possible mechanisms include chelation of DNA and binding to cell membranes thereby stimulating immune mechanisms.

11.3.5 Preparation
The desired dose of cisplatin is diluted with 250-1000 mL of saline and/or dextrose solution. Varying concentrations of 0.225-5% sodium chloride and 5% dextrose may be used. To maintain stability of cisplatin, a final sodium chloride concentration of at least 0.2% is recommended.

11.3.6 Administration
Cisplatin solution must be administered over a 60 to 120 minute period.

11.3.7 Incompatibilities
Amsacrine, cefepime, gallium nitrate, mesna, piperacillin, sodium bicarbonate, and thiotepa. Cisplatin may react with aluminum which is found in some syringe needles or IV sets, forming a black precipitate.

11.3.8 Availability
Commercially available as a mg/mL solution in 50 and 100 mg vials.

11.3.9 Side Effects
Please see package insert for detailed information regarding side effects related to this drug.

A dose-related ototoxicity, manifested by high-frequency hearing loss and tinnitus, occurs in about 30% of subjects. Mild leukopenia and thrombocytopenia occur in 25-30% of subjects, but are rarely dose-limiting. Nausea and vomiting occur in almost 100% of subjects unless adequate antiemetic prophylaxis is given. Allergic reactions are reported in up to 20% of subjects. Symptoms include: rash, facial edema, wheezing, hypotension, and tachycardia.

12 ADVERSE EVENTS

12.1 Definitions of Adverse Events
The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) v4 will be utilized for AE assessment. A copy of the CTCAE v4 can be downloaded from the CTEP website at http://ctep.cancer.gov

12.1.1 Adverse Event
Adverse event (AE) means any untoward medical occurrence associated with the use of a drug in humans, whether or not considered drug related. An adverse event (also referred to as an adverse experience) can be any unfavorable and unintended sign (e.g., an abnormal laboratory finding), symptom, or disease temporally associated with the use of a drug, and does not imply any judgment about causality. An adverse event can arise with any use of the drug (e.g., off-label use, use in combination with another drug) and with any route of administration, formulation, or dose, including

A site investigator who is a qualified physician will evaluate all adverse events according to the NCI Common Terminology for Adverse Events (CTCAE), v4. Any adverse event which changes CTCAE grade over the course of a given episode will have each change of grade recorded on the adverse event case report forms/worksheets.

12.1.2 Serious Adverse Event
A serious adverse event is any untoward medical occurrence resulting in one or more of the following:

- Results in death **NOTE:** Death due to progressive disease is not considered a SAE unless the event was related to the study drug.
- Is life-threatening (defined as an event in which the subject was at risk of death at the time of the event; it does not refer to an event which hypothetically might have caused death if it were more severe)

- Requires inpatient hospitalization or prolongation of existing hospitalization
- Results in persistent or significant disability/incapacity
- Is a congenital anomaly or birth defect
- Is an important medical event (defined as a medical event(s) that may not be immediately life-threatening or result in death or hospitalization but, based upon appropriate medical and scientific judgment, may jeopardize the subject or may require intervention (e.g., medical, surgical) to prevent one of the other serious outcomes listed in the definition above). Examples of such events include, but are not limited to, intensive treatment in an emergency room or at home for allergic bronchospasm; blood dyscrasias or convulsions not resulting in hospitalization; or the development of drug dependency or drug abuse.

12.1.3 Suspected Adverse Reaction

Suspected adverse reaction means any adverse event for which there is a reasonable possibility that the drug caused the adverse event. For the purposes of IND safety reporting, 'reasonable possibility' means there is evidence to suggest a causal relationship between the drug and the adverse event. A suspected adverse reaction implies a lesser degree of certainty about causality than adverse reaction, which means any adverse event caused by a drug.

12.1.4 Adverse Reaction

An adverse reaction means any adverse event caused by a drug. Adverse reactions are a subset of all suspected adverse reactions where there is reason to conclude that the drug caused the event.

12.1.5 Unexpected Adverse Event

For this study, an AE is considered unexpected when it varies in nature, intensity or frequency from information provided in the current AE list, the IB, the package insert or when it is not included in the informed consent document as a potential risk.

12.1.6 Attribution

Attribution is the relationship between an AE or SAE and the study treatment. Attribution will be assigned as follows:

Unrelated	The Adverse Event is ***clearly not related*** to the study drug
Unlikely	The Adverse Event is ***doubtfully related*** to the study drug
Possible	The Adverse Event ***may be related*** to the study drug
Probable	The Adverse Event is ***likely related*** to the study drug
Definite	The Adverse Event is ***clearly related*** to the study drug

12.2 Adverse Event Reporting

12.2.1 Site Requirements for Recording Adverse Events

Adverse events (AEs) will be recorded from the time of consent and for at least 30 days after treatment discontinuation, regardless of whether or not the event(s) are considered related to trial medications. All AEs considered related to trial medication will be followed until resolution, return to baseline, or deemed clinically insignificant, even if this occurs post-trial.

Adverse events will not be collected for subjects during the pre-screening period (for determination of archival tissue status) as long as that subject has not undergone any protocol-specified procedure or intervention. If the subject requires a blood draw, fresh tumor biopsy etc., the subject is first required to provide consent to the main study and AEs will be captured according to guidelines for standard AE reporting.

12.2.2 SAE Reporting from Sites to HCRN

Sites will report to Hoosier Cancer Research Network (HCRN) any SAE, or follow up to a SAE, including death due to any cause that occurs to any subject from the time the consent is signed through 90 days following the last dose of study drug, or the initiation of new anti-cancer therapy, whichever is earlier, whether or not related to Merck product.

All SAEs will be reported on the SAE/ECI Submission Form and submitted to HCRN per guidelines outlined in this section. In addition, All AEs and SAEs will be recorded on the appropriate study specific eCRF within the EDC system. The original copy of the SAE/ECI Submission Form and the e-mail correspondence or the fax confirmation sheet must be kept within the Trial Master File at the study site.

Sites must report any SAEs occurring during the course of the study to HCRN **within one business day** of discovery of the event. The completed SAE/ECI Submission Form must be sent electronically to safety@hoosiercancer.org. The site investigator is responsible for informing the IRB and/or the Regulatory Authority of the SAE as per local requirements.

Follow-up information will be sent electronically to HCRN to safety@hoosiercancer.org using a new SAE/ECI Submission Form. Each re-occurrence, complication, or progression of the original event should be reported as a follow-up to that event regardless of when it occurs.

12.2.3 HCRN Requirements for Reporting SAEs to Merck

HCRN will report any SAE from the time the consent is signed through 90 days following the last dose of study drug, or the initiation of new anti-cancer therapy, whichever is earlier, whether or not related to Merck product. The report will be sent to Merck **within one business day** of receipt of the SAE/ECI Submission Form. Hoosier Cancer Research Network will fax follow up information as reasonably requested to Merck.

SAE reports and any other relevant safety information are to be forwarded to the Merck Global Safety facsimile number: +1-215-993-1220.

12.3 Sponsor-Investigator Responsibilities

HCRN will send a SAE summary to the sponsor-investigator **within 1 business day** of receipt of SAE/ECI Submission Form from a site. The sponsor-investigator will promptly review the SAE summary and assess for expectedness and relatedness.

12.4 Reporting to the Food and Drug Administration (FDA)

The FDA concluded this protocol is exempt from the requirements of an IND on 1.15.15. HCRN will continue to facilitate compliance of applicable requirements for the sponsor-investigator in relation to this study. This includes but is not limited to 21 CFR 50.20 informed consent, 21 CFR Part 56 IRB, and pertinent sections of the Public Health Service Act and FDAAA.

12.5 Definition of an Overdose for This Protocol and Reporting of Overdose

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

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

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

All reports of overdose with and without an adverse event must be reported from sites to HCRN **within one business day**. HCRN will report such events to Merck Global Safety **within one business day** of notification of the event. (Attn: Worldwide Product Safety; FAX 215 993-1220)

12.6 Reporting of Pregnancy and Lactation

Although pregnancy and lactation are not considered adverse events, it is the responsibility of the site investigators or their designees to report any pregnancy or lactation in a subject (spontaneously reported to them), including the pregnancy of a male subject's female partner that occurs during the trial or within 120 days of completing the trial. All subjects and female partners of male subjects who become pregnant must be followed to the completion/termination of the pregnancy. Pregnancy outcomes of spontaneous abortion, missed abortion, benign hydatidiform mole, blighted ovum, fetal death, intrauterine death, miscarriage and stillbirth must be reported as serious events (Important Medical Events). If the pregnancy continues to term, the outcome (health of infant) must also be reported.
Such events must be reported **within one business day** to HCRN. HCRN will report such events to Merck Global Safety **within one business day** of receiving notification of the event. (Attn: Worldwide Product Safety; FAX 215-993-1220).

12.7 Events of Clinical Interest

Selected non-serious and serious adverse events are also known as Events of Clinical Interest (ECI) and must be recorded as such on the SAE/ECI Submission Form. ECIs are reported **within one business day** by sites to HCRN who will then report to Merck **within one business day** to Merck Global Safety. (Attn: Worldwide Product Safety; FAX 215 993-1220). Events of clinical interest for this trial include:

- An overdose of Merck product, as defined above that is not associated with clinical symptoms or abnormal laboratory results.
- An elevated AST or ALT lab value that is greater than or equal to 3X the upper limit of normal and an elevated total bilirubin lab value that is greater than or equal to 2X the upper limit of normal and, at the same time, an alkaline phosphatase lab value that is less than 2X the upper limit of normal, as determined by way of protocol-specified laboratory testing or unscheduled laboratory testing.*
- Progression or pseudo-progression at post neoadjuvant chemotherapy that impacts decision making for definitive surgery is considered an event of clinical interest and should be discussed with the HCRN project manager/sponsor-investigator.
-

*Note: These criteria are based upon available regulatory guidance documents. The purpose of the criteria is to specify a threshold of abnormal hepatic tests that may require an additional evaluation for an underlying etiology. The trial site guidance for assessment and follow up of these criteria can be found in the SPM

Table 11 Guidance for evaluating adverse events

A site investigator who is a qualified physician, will evaluate all adverse events as to:

V4.0 CTCAE Grading	**Grade 1**	**Mild; asymptomatic or mid symptoms; clinical or diagnostic observations only; intervention not indicated.**
	Grade 2	**Moderate; minimal, local or noninvasive intervention indicated; limiting age-appropriate instrumental ADL.**
	Grade 3	**Severe or medically significant but not immediately life-threatening; hospitalization or prolongation or hospitalization indicated; disabling; limiting self-care ADL.**
	Grade 4	**Life threatening consequences; urgent intervention indicated.**
	Grade 5	**Death related to AE**
Seriousness	A serious adverse event is any adverse event occurring at any dose or during treatment with study drug	
	†**Results in death**; or	
	†**Is life threatening; or** places the subject, in the view of the site investigator, at immediate risk of death from the event as it occurred (Note: This does not include an adverse event that, had it occurred in a more severe form, might have caused death.); or	
	†**Results in a persistent or significant disability/incapacity** (substantial disruption of one's ability to conduct normal life functions); or	
	†**Results in or prolongs an existing inpatient hospitalization** (hospitalization is defined as an inpatient admission, regardless of length of stay, even if the hospitalization is a precautionary measure for continued observation. (Note: Hospitalization [including hospitalization for an elective procedure] for a preexisting condition which has not worsened does not constitute a serious adverse event.); or	
	†**Is a congenital anomaly/birth defect** (in offspring of subject taking the product regardless of time to diagnosis);or	
	Is a new cancer; (that is not a condition of the study) **or**	
	Is an overdose (whether accidental or intentional). Any adverse event associated with an overdose is considered a serious adverse event. An overdose that is not associated with an adverse event is considered a non-serious event of clinical interest and must be reported within 24 hours.	
	Other important medical events that may not result in death, not be life threatening, or not require hospitalization may be considered a serious adverse event when, based upon appropriate medical judgment, the event may jeopardize the subject and may require medical or surgical intervention to prevent one of the outcomes listed previously (designated above by a †).	
Duration	Record the start and stop dates of the adverse event. If less than 1 day, indicate the appropriate length of time and units	
Action taken	Did the adverse event cause the study drug to be discontinued?	
Relationship to test drug	Did the study drug cause the adverse event? The determination of the likelihood that the Merck product caused the adverse event will be provided by a site investigator who is a qualified physician. The site investigator's signed/dated initials on the source document or worksheet that supports the causality noted on the AE form, ensures that a medically qualified assessment of causality was done. This initialed document must be retained for the required regulatory time frame. The criteria below are intended as reference guidelines to assist the site investigator in assessing the likelihood of a relationship between the test drug and the adverse event based upon the available information. **The following components are to be used to assess the relationship between the study drug and the AE**; the greater the correlation with the components and their respective elements (in number and/or intensity), the more likely the Merck product caused the adverse event (AE):	
	Exposure	Is there evidence that the subject was actually exposed to the study drug such as: reliable history, acceptable compliance assessment (pill count, diary, etc.), expected pharmacologic effect, or measurement of drug/metabolite in bodily specimen?
	Time Course	Did the AE follow in a reasonable temporal sequence from administration of the study drug? Is the time of onset of the AE compatible with a drug-induced effect (applies to trials with investigational medicinal product)?
	Likely Cause	Is the AE not reasonably explained by another etiology such as underlying disease, other drug(s)/vaccine(s), or other host or environmental factors

Relationship to study drug (continued)	**The following components are to be used to assess the relationship between the test drug and the AE: (continued)**	
	Dechallenge	Was the study drug discontinued or dose/exposure/frequency reduced? If yes, did the AE resolve or improve? If yes, this is a positive dechallenge. If no, this is a negative dechallenge. (Note: This criterion is not applicable if: (1) the AE resulted in death or permanent disability; (2) the AE resolved/improved despite continuation of the study drug; or (3) the trial is a single-dose drug trial); or (4) Merck product(s) is/are only used one time.)
	Rechallenge	Was the subject re-exposed to the study drug in this study? If yes, did the AE recur or worsen? If yes, this is a positive rechallenge. If no, this is a negative rechallenge. (Note: This criterion is not applicable if: (1) the initial AE resulted in death or permanent disability, or (2) the trial is a single-dose drug trial); or (3) Study drug(s) is/are used only one time). NOTE: IF A RECHALLENGE IS PLANNED FOR AN ADVERSE EVENT WHICH WAS SERIOUS AND WHICH MAY HAVE BEEN CAUSED BY THE STUDY DRUG, OR IF REEXPOSURE TO THE MERCK PRODUCT POSES ADDITIONAL POTENTIAL SIGNIFICANT RISK TO THE SUBJECT, THEN THE RECHALLENGE MUST BE APPROVED IN ADVANCE BY THE U.S. CLINICAL MONITOR AS PER DOSE MODIFICATION GUIDELINES IN THE PROTOCOL.
	Consistency with Trial Treatment Profile	Is the clinical/pathological presentation of the AE consistent with previous knowledge regarding the Merck product or drug class pharmacology or toxicology?
The assessment of relationship will be reported on the case report forms /worksheets by a site investigator who is a qualified physician according to his/her best clinical judgment, including consideration of the above elements.		
Record one of the following	**Use the following scale of criteria as guidance (not all criteria must be present to be indicative of a study drug relationship).**	
Yes, there is a reasonable possibility of study drug relationship.	There is evidence of exposure to the Merck product. The temporal sequence of the AE onset relative to the administration of the Merck product is reasonable. The AE is more likely explained by the Merck product than by another cause.	
No, there is not a reasonable possibility study drug relationship	Subject did not receive the study drug OR temporal sequence of the AE onset relative to administration of the study drug is not reasonable OR there is another obvious cause of the AE. (Also entered for a subject with overdose without an associated AE.)	

13 STATISTICAL METHODS

13.1 Study Design and Stratification

Subjects who are appropriate for surgical intent must meet eligibility criteria, and are then stratified into cohort I: cisplatin eligible and treated on Phase Ib or Phase II, Arm A; or cohort II: cisplatin-ineligible and treated on Phase II, Arm B. See Section 3.3.1 for details of how cisplatin -eligible or –ineligible subjects are defined. Note that though subjects may fit cisplatin-ineligibility, they still must be fit enough to qualify for surgery per surgeon's documentation stating surgical intent (and therefore NYHA class III and IV are not permitted).

13.2 Study Phases

Phase Ib: This is a 3+3 study design in the cisplatin-eligible, cohort I, group. Two pembrolizumab dose levels of 200mg, and 120mg will be investigated with gemcitabine/cisplatin. The MTD will be the highest pembrolizumab dose level in combination with gemcitabine/cisplatin at which no more than 1 of 6 subjects experiences a non-gemcitabine/cisplatin related DLT. Once the MTD is established, this portion of the study will close and the Phase II will open to cohort I and II. The recommended Phase II dose (RP2D) will be based on the MTD and discussed with and approved by the sponsor-investigator. Including a ~4% patient drop out, the maximal sample size is 14.

Phase II: Cohort I and II subjects will be treated at the RP2D in an independent Simon two-stage design. For cohort I, the null hypothesis that the true pathologic muscle invasive response rate (PaIR) is 23% will be tested against a one-sided alternative. In the first stage, 15 subjects will be accrued. If there are 4 or fewer responses in these 15 subjects, cohort I will be stopped. Otherwise, 17 additional subjects will be accrued for a total of 32. The null hypothesis will be rejected if 12 or more responses are observed in 32 subjects. This yields a type I error rate of 4% and power of 86% when the true response rate is 48%. For cohort II, the null hypothesis that the true PaIR rate is 18% will be tested against a one-sided alternative. In the first stage, 19 subjects will be accrued. If there are 4 or less responses in these 19 subjects, the cohort will be stopped. Otherwise, 16 additional subjects will be accrued for a total of 35. The null hypothesis will be rejected if 11 or more responses are observed in 35 subjects. This yields a type I error rate of 4% and power of 86% when the true response rate is 40%.

13.3 Definition of Primary Endpoint

Pathologic muscle invasion response (PaIR), or ≤ypT1N0M0, rate is assessed from the consolidative surgery (radical cystectomy (RC) / nephroureterectomy (NU) - lymph node dissection (LND)) specimen.

13.4 Definitions of Secondary Endpoints

Relapse-free survival (RFS) and OS will be measured from the date of randomization to date of relapse or death due to any cause, whichever occurs first. Relapse after curative intent surgery usually requires biopsy for confirmation. Pathologic confirmation is per site investigator, however the date of relapse should be recorded as the date of the imaging that prompted the biopsy. Response will be defined per RECIST v1.1, see section 9.7.
Safety and tolerability of pembrolizumab in combination with gemcitabine – cisplatin will occur during Phase Ib and II, and in combination with gemcitabine alone will occur during Phase II.

Acute and longer term safety and tolerability data will be collected per adverse event documentation performed during, as well as after, neoadjuvant therapy in the follow up assessments.

Consolidation surgery after combination neoadjuvant therapy includes completed radical cystectomy with pelvic lymph node dissection or nephroureterectomy. The rate of pT0N0 will be determined. The rate of consolidation surgery amongst the evaluable subjects will be determined.

13.5 Analysis Plan for Primary and Secondary Objectives

All evaluable subjects will be included in data safety analysis if they complete at least 1 cycle of the neoadjuvant portion of treatment as described in the treatment plan, or discontinue treatment early for defined toxicity criteria (criteria in section 6 above). Toxicity data will be tabulated. Subjects will be evaluable for the primary efficacy endpoint if they complete at least 2 cycles of neoadjuvant therapy and 2 doses of pembrolizumab, then have a radical cystectomy with pelvic lymph node dissection, or nephroureterectomy with lymph node dissection, where a pathologic T and N stage can be assigned (and subjects with a "X" notation for a T or N will not be included in the efficacy analysis). The true response rate (PaIR) of the evaluable subjects will be estimated using binomial distribution theory. The rate will be estimated for each cohort. The confidence intervals for them will be estimated using Wilson's method[71]. The predictive value of biomarkers, such as PD-L1 on response rate will be estimated using logistic regression after controlling the effects of confounders, such as demographics, baseline histology.

The probabilities of RFS and OS and their median survival times will be estimated by Kaplan-Meier method for each cohort separately. Kaplan-Meier estimates for RFS and OS for those downstaged or not to PaIR and pT0 will also be compared. Factors, such as age, gender, baseline histology and lab correlates including PD-L1, that predict survivals (RFS, OS) will be identified by Cox model or extended Cox model. χ^2 tests will be used to evaluate the relationship between prior BCG use and PaIR.

13.6 Sample Size/Accrual/Study Duration/Replacement Rules

Phase Ib: This is a 3+3 dose design study in the cisplatin-eligible, cohort I, group. Two pembrolizumab dose levels of 200mg, and 120mg will be investigated with gemcitabine/cisplatin. The MTD will be the highest pembrolizumab dose level in combination with gemcitabine/cisplatin at which no more than 1 of 6 subjects experience a dose limiting toxicity (DLT) related to pembrolizumab (and not of gemcitabine or cisplatin). Once the MTD is established, this portion of the study will close and the Phase II will open to cohort I and II at the recommended phase II dose (RP2D). Including a ~4% subject drop out, the maximal sample size is 14.

Phase II: Cohort I and II subjects will be treated at RP2D from the phase 1b portion in independent Simon two-stage designs. For cohort I, the null hypothesis that the true pathologic muscle invasive response rate (PaIR) is 23% will be tested against a one-sided alternative. In the first stage, 15 subjects will be accrued. If there are 4 or fewer responses in these 15 subjects, cohort I will be stopped. Otherwise, 17 additional subjects will be accrued for a total of 32. The null hypothesis will be rejected if 12 or more responses are observed in 32 subjects. This yields

a type I error rate of 4% and power of 86% when the true response rate is 48%. For cohort II, the null hypothesis that the true PaIR rate is 18% will be tested against a one-sided alternative. In the first stage, 19 subjects will be accrued. If there are 4 or less responses in these 19 subjects, the cohort will be stopped. Otherwise, 16 additional subjects will be accrued for a total of 35. The null hypothesis will be rejected if 11 or more responses are observed in 35 subjects. This yields a type I error rate of 4% and power of 86% when the true response rate is 40%. Subjects who are not evaluable for the primary efficacy endpoint (see section 13.5) will be replaced.

Historical Controls

Current level one evidence for neoadjuvant chemotherapy is based on the SWOG8710 neoadjuvant MVAC trial[24] and the EORTC/MRC neoadjuvant CMV trial[72]. Though CMV/MVAC was considered, it is not predicted to be a favorable combination with an immune checkpoint blockade in this trial design (see section 1.2.3). Based on data from the metastatic setting that show similar survival with MVAC, GC[28] has become a standard alternative for use in the neoadjuvant setting as well. Pathologic response correlates with outcome in subjects treated with neoadjuvant therapy.

Subject demographics in this trial reflects those used in prior neoadjuvant trials and results will be similarly summarized with ability to comment on primary outcome similarities or differences. Similar subject characteristics include histology (urothelial carcinoma, transitional cell carcinoma, mixed histologies with urothelial carcinoma background) and TNM stage. Gemcitabine-cisplatin in the neoadjuvant setting has been evaluated in a prospective phase II trial[29] as well as in retrospective cohorts[30,73] which have reported a PaIR (similarly defined and reported as pCR, or p≤T1) rates in the table below

Author	Neoadjuvant Regimen (NAC)	Analysis	PaIR	Median Time to Cystectomy	comment
Herchenhorn et al[29]	GC x4 cycles	prospective Phase II	23%	6.7w from end of NAC	standard practice
Galsky et al[73]	GC x4 cycles	retrospective	27%	N/A	
Weight et al[30]	GC x4 cycles	retrospective	7%	NAC: 29.7 w from diagnosis RC only: 6.9w from diagnosis	
Plimack et al[74]	dose dense GC	prospective Phase II	45%	N/A	closed early (tox)

Timing of RC after neoadjuvant therapy is an important consideration for this multimodality intervention and subjects on this trial will receive RC similar to historical controls. Delivery of cystectomy within 10 weeks from the last dose of a combination neoadjuvant chemotherapy regimen of 9-12 weeks in duration was found to not compromise survival in a retrospective cohort of 153 subjects[67]. This HCRN GU14-188 trial delivers pembrolizumab 3 weeks after the last dose of gemcitabine (G) or gemcitabine-cisplatin (GC), and therefore requires delivery of surgery (RC or NU) within 2 to 7 weeks after this dose. In the context of Herchenhorn *et al* as an historical control, this neoadjuvant pembrolizumab trial delivers surgery 5 to 10 weeks after standard practice gemcitabine/cisplatin chemotherapy which is within the recommended 10 weeks and can be compared to this group. In the context of Weight *et al*, surgery was delivered a median of ~30weeks from diagnosis. It's reasonable to assume a 4-8 week 'setup time' which

may include path resulting time, arrangement of oncology referral, lab and radiographic evaluation, and port placement from the time of diagnosis; and an additional 12 weeks of gemcitabine/cisplatin chemotherapy. Therefore, delivery of cystectomy in this cohort is estimated to have been ~10-14 weeks after the last chemotherapy and well within the timeframe for this neoadjuvant pembrolizumab trial for eventual comparison of results.

Cohort II subjects treated with gemcitabine alone have historical references for response in the advanced and metastatic setting where cystectomy was not performed. It is estimated that the primary endpoint for a cohort II patient receiving gemcitabine monotherapy in the neoadjuvant setting will have a PaIR ~5% less than their counterparts receiving GC. We do not yet have adequate historical data from randomized trials for RC alone. RC alone has been reviewed in a retrospective cohort of ~1000 subjects and found a pathologic complete response (pCR) rate of 6.3%. A prospective evaluation of single modality RC pCR rate was determined to be 15% in the comparator arm of the Phase III SWOG 8710 trial[24,44]. These subjects had prior diagnostic (and occasionally maximal) transurethral resection of bladder tumor (TURBT) to yield a pCR rate of ~15% at RC. It's important, however, to understand this rate should only be viewed within the context of being a comparator group against which a difference with neoadjuvant therapy was assessed within the pre-specified controls of the clinical trial. A pathologic CR rate for RC-only would require a clinical trial that controls for extent of TURBT prior to RC, and controls for reproducible pathologic specimen sectioning (detailed sectioning of whole-mounts may reveal Tis or T1 lesions).

14 TRIAL MANAGEMENT

14.1 Data and Safety Monitoring Plan

The study will be conducted in accord with Case Comprehensive Cancer Center's Data and Safety Monitoring Plan.

HCRN oversight activities include:

- Review all adverse events requiring expedited reporting as defined in the protocol
- Provide trial accrual progress, safety information and data summary reports to the sponsor-investigator
- Submit data summary reports to the lead institution Data Safety Monitoring Committee according to Case Comprehensive Cancer Center's DSMP.

14.2 Case Comprehensive Cancer Center's Data Safety Monitoring Committee

HCRN will provide the following for the Case Comprehensive Cancer Center's DSMC to review:

- Adverse event summary report
- Audit results if applicable
- Data related to stopping/decision rules described in study design
- Study accrual patterns
- Protocol deviations

14.3 Data Quality Oversight Activities

14.3.1 Onsite Monitoring

Monitoring visits to the trial sites may be made periodically during the trial to ensure key aspects of the protocol are followed. During onsite monitoring visits, source documents will be reviewed for verification of agreement with data entered into the EDC system. Additional for cause visits may occur as necessary. It is important for the site investigator and their relevant personnel to be available for a sufficient amount of time during the monitoring visits or audit, if applicable. The site investigator and institution guarantee access to source documents by HCRN or its designee.

The trial site may also be subject to quality assurance audit by Merck or its designee as well as inspection by appropriate regulatory agencies.

14.3.2 Remote Monitoring

Remote validation of the EDC system data will be completed on a continual basis throughout the life cycle of the study. The data entry completion status of each participating site will be reviewed on a weekly basis to ensure timeliness of data entry. Electronic Case Report Form (eCRF) constraints will be utilized to prevent data entry errors and ensure complete data entry. Automated bi-weekly edit check programs will check for errors in accuracy, validity, and consistency in the data. Listings from the edit check programs will be used to generate queries in the EDC system. Corrections will be made in a timely manner by the study site personnel.

14.4 Compliance with Trial Registration and Results Posting Requirements

Under the terms of the Food and Drug Administration Modernization Act (FDAMA) and the Food and Drug Administration Amendments Act (FDAAA), the sponsor-investigator of the trial is solely responsible for determining whether the trial and its results are subject to the requirements for submission to the Clinical Trials Data Bank, http://www.clinicaltrials.gov. All results of primary and secondary objectives must be posted to CT.gov within a year of completion. The sponsor-investigator has delegated responsibility to HCRN for registering the trial and posting the results on clinicaltrials.gov. Information posted will allow subjects to identify potentially appropriate trials for their disease conditions and pursue participation by calling a central contact number for further information on appropriate trial locations and study site contact information.

15 DATA HANDLING AND RECORD KEEPING

15.1 Data Management

HCRN will serve as the Clinical Research Organization for this trial. Data will be collected through a web based clinical research platform, EDC system, a system compliant with Good Clinical Practices and Federal Rules and Regulations. HCRN personnel will coordinate and manage data for quality control assurance and integrity. All data will be collected and entered into EDC system by study site personnel from participating institutions.

15.2 Case Report Forms and Submission

Generally, clinical data will be electronically captured in EDC system and correlative results will be captured in the EDC system or other secure database(s). If procedures on the study calendar are performed for standard of care, at minimum, that data will be captured in the source document. Select standard of care data will also be captured in EDC system according to study-specific objectives.

The completed dataset is the sole property of the sponsor-investigator's institution and should not be exported to third parties, except for authorized representatives of appropriate Health/Regulatory Authorities, without permission from the sponsor-investigator and HCRN.

15.3 Record Retention

To enable evaluations and/or audits from Health Authorities/HCRN, the site investigator agrees to keep records, including the identity of all subjects (sufficient information to link records; e.g., hospital records), all original signed informed consent forms, copies of all source documents, and detailed records of drug disposition. All source documents are to remain in the subject's file and retained by the site investigator in compliance with the site contract with HCRN. No records will be destroyed until HCRN confirms destruction is permitted.

15.4 Confidentiality

There is a slight risk of loss of confidentiality of subject information. All records identifying the subject will be kept confidential and, to the extent permitted by the applicable laws and/or regulations, will not be made publicly available.

Information collected will be maintained on secure, password protected electronic systems. Paper files that contain personal information will be kept in locked and secure locations only accessible to the study team. Samples that are collected will be identified by a subject study number assigned at the time of registration to the trial. Any material issued to collaborating researchers will be anonymized and only identified by the subject study number.

Subjects will be informed in writing that some organizations including the sponsor-investigator and his/her research associates, HCRN, Merck & Co., IRB, or government agencies, like the FDA, may inspect their medical records to verify the information collected, and that all personal information made available for inspection will be handled in strictest confidence and in accordance with local data protection laws.
If the results of the study are published, the subject's identity will remain confidential.

15.5 Changes to the Protocol

Study procedures will not be changed without the mutual agreement of the sponsor-investigator, HCRN, and Merck & Co.

If it is necessary for the study protocol to be amended, the amendment or a new version of the study protocol (amended protocol) will be generated by the HCRN and must be approved by each IRB, Merck & Co., and if applicable, also the local regulatory authority. Local requirements must be followed.

If a protocol amendment requires a change to the Informed Consent Form, site investigators must notify their local IRB. Approval of the revised Informed Consent Form by the IRB is required before the revised form is used.

The site investigator is responsible for the distribution of these documents to his or her IRB, and to the staff at his or her center.

Merck & Co's willingness to supply study drug is predicated upon the review of the protocol. HCRN agrees to provide written notice to Merck & Co. of any modifications to the protocol or informed consent.

16 ETHICS

16.1 Ethics Review

The final study protocol, including the final version of the Informed Consent Form, must be approved in writing by an IRB. The site investigator must submit written approval to the HCRN office before he or she can enroll any subject into the study.

The site investigator is responsible for informing the IRB of any amendment to the protocol in accordance with local requirements. In addition, the IRB must approve all advertising used to recruit subjects for the study. The protocol must be re-approved by the IRB annually, as local regulations require.

16.2 Ethical Conduct of the Study

The study will be performed in accordance with ethical principles originating from the Declaration of Helsinki, which are consistent with ICH/Good Clinical Practice, and applicable regulatory requirements.

16.3 Informed Consent Process

The site investigator will ensure the subject is given full and adequate oral and written information about the nature, purpose, possible risks and benefits of the study. Subjects must also be notified they are free to discontinue from the study at any time. The subject should be given the opportunity to ask questions and allowed time to consider the information provided.

The subject's signed and dated informed consent must be obtained before conducting any procedure specifically for the study. The site investigator must store the original, signed Informed Consent Form. The site investigator must provide the subject with a copy of the signed Informed Consent Form.

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