NCT04809103

03 February 2022

A PHASE I TRIAL OF INTRATUMORAL CISPLATIN FOR EARLY STAGE, RESECTABLE, NON-SMALL CELL LUNG CANCER

A PHASE I TRIAL OF INTRATUMORAL CISPLATIN FOR EARLY STAGE, RESECTABLE, NON-SMALL CELL LUNG CANCER

Principal Investigator:
C. Matthew Kinsey MD, MPH
Director, Interventional Pulmonology, University of Vermont Medical Center
Given D Suite 208, 89 Beaumont Avenue
Burlington, VT 05405
Telephone: 802.656.3521
Fax: 802.656.3526
Email: Matt.Kinsey@med.uvm.edu

Co-Investigators:
Hannah Kooperkamp MD
Assistant Professor of Cardiothoracic Surgery
Department of Surgery, University of Vermont Medical Center

Garth Garrison
Director, Lung Cancer Transdisciplinary Team
Division of Pulmonary and Critical Care, University of Vermont Medical

Bernard Cole Ph.D.
Professor, Department of Mathematics and Statistics, University of Vermont
Director, Cancer Control and Population Health Sciences Section, Vermont Cancer Center

Farrah B. Khan MD
Lung Clinical Trials Lead
Hematology and Oncology Division, University of Vermont Medical Center

Kelly Butnor MD
Lung and Thoracic Pathology
Department of Pathology, University of Vermont Medical Center

Mitch Norotsky MD
Chief of Surgery
Department of Surgery, University of Vermont Medical Center

Pamela C. Gibson MD
Vice Chair, Staff and Faculty Affairs
Department of Pathology, University of Vermont Medical Center

Scott Anderson MD
Director, Cytopathology
Department of Pathology, University of Vermont Medical Center

ABBREVIATIONS:

NSCLC: Non-small cell lung cancer

EBUS: endobronchial ultrasound

ROSE: Rapid on-site cytopathologic examination

CT: computed tomography

DLT: dose limiting toxicity

MTD: maximum tolerated dose

MTV: maximum tolerated volume

CTCAE: Common Terminology Criteria for Adverse Events

STUDY PROTOCOL TABLE OF CONTENTS:

APPENDICES:

1. SYNOPSIS:

TITLE: A PHASE I TRIAL OF INTRATUMORAL CISPLATIN FOR EARLY STAGE, RESECTABLE, NON-SMALL CELL LUNG CANCER

RATIONALE:
Non-small cell lung cancer (NSCLC) is the leading cause of cancer-related death in the United States. Even for early stage disease, the rate of recurrence following surgical resection is as high as 50%. Although neoadjuvant therapy, administered before surgery, for early stage lung cancer is associated with a survival benefit, it is rarely used due to the systemic toxicity of intravenous (IV) cytotoxic chemotherapy. IV immunotherapies are also being evaluated in combination with systemic therapies in the neoadjuvant setting. However, only a minority of patients respond to immunotherapy. One of the most common reasons for failure of immunotherapy is lack of presentation of tumor antigens to the immune system, a problem that may be potentially addressed with cytotoxic agents.

Over the last several years, case series have demonstrated the feasibility and safety of delivering cisplatin directly into lung tumors. For patients that have recurrent lung cancer in a previous radiation field, our prior work demonstrated that a series of four injections results in a 71% complete or partial response rate based on RECIST criteria of the treated volume. The only toxicity seen in this series was nausea and/or vomiting. Further, there are in vitro data that support the idea that cisplatin may modulate the immune system.

Given the current knowledge of safety and tolerability of intratumoral cisplatin, coupled with the potential to achieve immune priming that may help address systemic micrometastases, have led us to postulate that intratumoral cisplatin is a well-tolerated, and potentially effective, neoadjuvant therapy for patients with early stage, resectable, non-small cell lung cancer.

PRIMARY OBJECTIVE:
To identify the maximum tolerated dose (MTD) of intratumoral cisplatin, delivered during a single bronchoscopy with cone-beam CT confirmation, in a dose escalation protocol

DESIGN: 3+3 dose escalation. The amount of delivered cisplatin is based on the volume of the tumor being treated, beginning at a volume of cisplatin of $1/4^{th}$ the volume of the tumor.

2. BACKGROUND:

2.1. **Study Disease**: Non-small cell lung cancer (NSCLC) is the leading cause of cancer-related death in the United States.[1] Even for tumors that are identified at an early stage and resected, the rate of recurrence may be as high as 50%.[2] Direct intratumoral (IT) delivery of cisplatin into early stage lung cancers at the time of diagnosis has several clear potential benefits. First, it reduces time to treatment from time of diagnosis, potentially improving outcomes through this mechanism alone. Second, intratumoral delivery of cisplatin may provide the benefits of intravenous (IV) neoadjuvant chemotherapy while minimizing the systemic, off-target toxicities that have limited the adoption of this approach. Finally, there is increasing evidence that chemotherapy induced tumor cell death may "prime" the immune system by allowing immune effector cells to recognize tumor epitopes and kill undetected micrometastatic tumor cells beyond the resection specimen.[3]

2.2. **Rationale:**

NSCLC Commonly Results in a Prolonged Diagnosis to Treatment Time: Lung cancer remains one of the most challenging diseases to diagnose, stage, and treat. Patients generally undergo CT scans, PET-CT scans, and MRIs to determine the stage and extent of a potential or confirmed NSCLC. This can result in prolonged times to surgical resection. In fact, diagnosis to surgical resection times greater than 8 weeks appear to occur in approximately 30% of patients, and are independently associated with stage progression and a decreased median survival.[4] In a highly homogenous group of patients with Stage 1 squamous cell carcinoma, delayed surgery (>38 days) was associated with a worse overall survival.[5] Initiating therapy earlier has the potential to improve outcomes for patients with early stage NSCLC.

Neoadjuvant Therapy for Resectable Lung Cancer is Effective but Rarely Performed: In early-stage, resectable, lung cancer, neoadjuvant chemotherapy improves survival, with a 13% reduction in the relative risk of death.[6] The majority of neoadjuvant chemotherapy trials have utilized IV cisplatin, or a closely related drug, carboplatin. However, neoadjuvant IV chemotherapy results in significant rates of adverse effects. For instance, neoadjuvant cisplatin plus gemcitabine results in a 32% rate of systemic hematologic toxicity.[7] Intratumoral therapy, delivered at the time of diagnosis, has the potential to immediately initiate therapy and capture the benefits of neoadjuvant chemotherapy while minimizing systemic toxicity.

"Priming" the Immune System May Facilitate Immune Mediated Tumor Destruction: In 2015, the first immunotherapy drug for lung cancer was approved for the treatment of NSCLC.[8] Nivolumab is a humanized antibody that binds the programmed death ligand 1 (PD-L1), one of several immune checkpoint inhibitors (ICIs), that prevent tumor-mediated downregulation of cytotoxic T lymphocytes (CTLs). Nivolumab demonstrated a significant increase in median overall survival compared to docetaxel for patients with advanced squamous NSCLC. This median difference was driven by a significantly prolonged survival in the 20% of patients who responded. Similarly, the ICIs pembrolizumab and durvalumab result in response rates of 45% in treatment-naive advanced NSCLC and 28% in locally advanced non-resectable NSCLC, respectively.[9,10] Within each of these studies, there are groups of patients that experienced durable and

prolonged NSCLC remissions.

The impressive results achieved for a minority of patients with NSCLC have led to considerable interest in additional mechanisms of immune modulation. Some of the most notable ICI responses in fact have occurred in melanoma and lung cancer.[11,12] These are considered carcinogen-driven cancers with high rates of human tumor-specific peptides that arise from somatic mutations in the cancer genome.[13] One reason for failure of ICI therapy is lack of presentation of these antigens to the immune system. This may lead to a relative absence of CTLs in the tumor microenvironment and is associated with failure of ICI therapy.[14] One mechanism for recruiting immune effector cells is tumor cell death which can be induced by cytotoxic agents or radiation.[15]

2.3. **Investigational Agent:**

Cisplatin Delivered IV is Highly Toxic: One of the most commonly used IV cytotoxic medications for the treatment of lung cancer is cisplatin. Cisplatin binds to DNA, cross-linking strands and thereby inhibiting mitosis, which in turn leads to apoptosis of the tumor cell.[16] However, it has more recently been recognized that cisplatin may have immunomodulatory effects that extend beyond those related merely to cell death.[17] In vitro studies implicate cisplatin in upregulation of the major histocompatibility complex (MHC class 1), recruitment and proliferation of effector cells, upregulation of the activity of cytotoxic effector cells, and downregulation of the immunosuppressive environment. Specifically, cisplatin appears to increase recruitment of CTLs and to upregulate Fas receptor, a death signal.[18] Importantly, there is also evidence that cisplatin may work synergistically with PD-1 inhibitors.[19,20] Although many standard regimens for the treatment of NSCLC include an IV platinum agent, this delivery approach is complicated by significant toxicity. Systemic IV cisplatin is associated with greater than 40 untoward effects; the most common is nephrotoxicity, but may commonly include gastrointestinal, hematologic, cardiac, and hepatic events.[16] These off-target effects greatly impair patient quality of life and tolerance of the drug, which in turn limits the dose and frequency at which the medication can be given. In fact, for Stage IV NSCLC there is evidence that platinum-based chemotherapy may hasten death compared to palliative care.[21] **The FDA guidance document for cisplatin highlights a feature of this drug that is atypical for chemotherapeutics, namely that it is more highly absorbed in normal tissues versus tumor tissue**. Direct intratumoral (IT) delivery thus has the potential to maximize the therapeutic effect of cisplatin while minimizing off-target adverse effects.

Safety of IT Cisplatin in Prior Clinical Studies: There is accumulating evidence that cisplatin may be safely delivered directly into a lung tumor with minimal side effects. In general, intratumoral injection for lung cancer dates back to at least 1978.[22] The first Phase III trial of a bronchoscopically delivered agent, the Bacillus Calmette-Guerin vaccine, was completed in 1986.[23] This study established the safety of IT delivery via a needle into early stage lung cancers but did not demonstrate significant clinical effect. The development of modern technologies such as endobronchial ultrasound, navigational bronchoscopy, robotic bronchoscopy, and intraoperative cone-beam CT scan has resulted in the ability to precisely deliver an agent into a tumor, and the opportunity to synergistically augment the effect of modern therapies.

In 2006, Celikuoglu et al used intratumoral injection of a maximum dose of 40mg of cisplatin through a flexible bronchoscope for patients with obstructive, inoperable NSCLC.[24] This same group also performed bronchoscopic injection of cisplatin into centrally located, early-stage lung cancers. They demonstrated endobronchial regression of tumor which enabled subsequent surgical resection.[25] Intratumoral cisplatin has subsequently been used in multiple other studies at doses of 30-40mg.[26] In a Phase III clinical trial, 4 patients with limited-stage small cell lung cancer (SCLC) received intratumoral cisplatin at a dose of 40mg (ClinicalTrials.gov Identifier: NCT01487499). There were insufficient numbers of patients to assess for the primary outcome of progression-free survival, but no reported adverse effects. Our group has utilized intratumoral cisplatin for treatment of late stage disease at a maximum injected dose of 40mg, with only nausea and vomiting identified as adverse effects. At these doses, there appears to be significant efficacy with a 71% complete or partial response rate based on data from two centers (manuscript accepted to the Journal of Thoracic Oncology). We have preliminary data using flow cytometric analyses of lesions sampled 1 week after intratumoral cisplatin administration of cisplatin at these doses that demonstrate up to 90% cell death in the treated volume. Intratumoral cisplatin in the range of 40mg can result in detectable platinum blood levels following administration. Nausea and vomiting were also the most common adverse events documented in a larger series of 36 patients.[27] Serious adverse events have been reported when IT cisplatin is combined with other therapies. Bronchomediastinal fistula has occurred when IT cisplatin was delivered concomitantly with re-irradiation.[27] Similarly, organizing pneumonia occurred in a patient who was simultaneously receiving intravenous cisplatin.[28] Full details of prior studies have been included in the Summary of Prior Human Studies.

Delivery of IT Cisplatin: Despite series demonstrating the overall safety of intratumoral cisplatin, there has been little systematic investigation of the optimal dose or delivery location. For instance, in many studies an identical dose of 40mg of cisplatin has been delivered, regardless of morphology or other tumor characteristics. Further, intraoperative imaging has not been evaluated to guide or assess IT delivery. **It is therefore critical to establish a safe dose for intratumoral cisplatin in a prospective study in patients with early stage resectable NSCLC with appropriate assessment for dose limiting toxicity (DLT).**

Rationale for Dosing: Prior studies have generally treated large tumors (>3cm diameter) and focused on using a fixed dose of cisplatin. However, for smaller tumors the delivery volume of the agent (1mg/ml concentration) may result in reaching the maximum tolerated volume before reaching the MTD. For instance, a 2 cm diameter tumor would have a volume of approximately 4 mL (assuming a spherical morphology). It is clear that delivering a 40mg dose (40mL) is not feasible in this setting. The additional variance of tumor size in this study motivated further analysis to determine feasibility of alternative dosing strategies for smaller tumor volumes. We evaluated 28 cases of patients with recurrent lung cancer in a previously radiated field treated with intratumoral cisplatin at the University of Vermont and the University of Florida. We applied our published computational model of intratumoral cisplatin pharmacokinetics[29] to evaluate a volumetric

dosing strategy. This series include tumors less than 3 cm in diameter, many of which empirically received a lower dose of cisplatin. Each case was evaluated for response using RECIST criteria, v1.1.[30] This model was used to simulate the percentage of tumor cells that reach the threshold intracellular concentration of cisplatin required for cell death (Fig 1). These results demonstrate robust distribution of the agent, and associated cytotoxicity, across a broad range of smaller tumors. For instance, a 4 cm diameter tumor is approximately 30 cm^3 in volume. Delivering one-half of the tumor volume in total drug volume, with a 1/mg/ml concentration, results in a 15mg dose of delivered cisplatin, which would be predicted to lead to cytotoxicity to over 90% of the tumor.

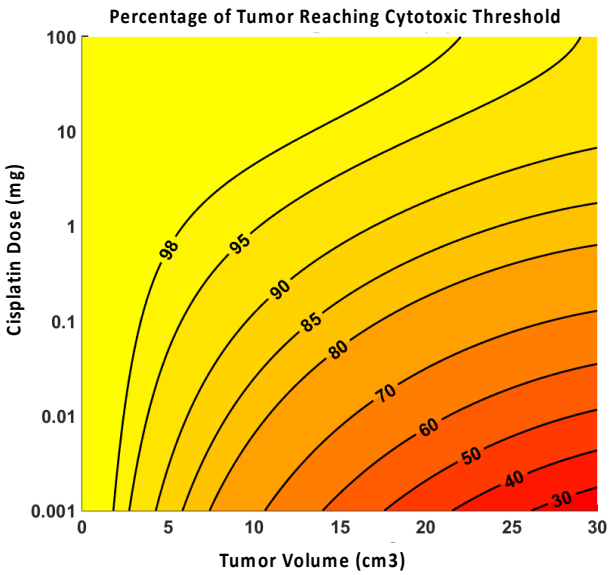

Figure 1: Dosing of Cisplatin Based on Tumor Volume. Isodose lines demonstrate robust coverage of small tumors with low doses of intratumoral cisplatin (1mg/ml). For reference, a 10 cm^3 volume corresponds to a 2.7 cm diameter tumor (assuming a spherical morphology).

The second consideration for dosing in the current study is the maximum administered dose. The majority of prior series have not used doses above 40mg. In our open Phase 1A study of intratumoral cisplatin for metastatic NSCLC, the 20mg dose cohort did not result in any dose limiting toxicity and we are moving to the 40mg dose cohort. Thus, for this protocol we consider 40mg to be the maximum dose. We have outlined three dosing cohorts in the protocol: an Initial cohort at 1/4th tumor volume, a De-escalation cohort at 1/8th tumor volume, and an Escalation cohort at 1/2 tumor volume. If we reached the Escalation cohort (1/2 volume) the maximum dose delivered would be 32.8mg. Tables detailing example doses for each cohort have been included in Section 4.

3. PATIENT SELECTION

3.1. Inclusion criteria

3.1.1. Age ≥18 years.

3.1.2. ECOG performance status 0 or 1

3.1.3. Patients must have adequate organ and marrow function as defined below:

3.1.3.1. Leukocytes ≥3,000/mcL

3.1.3.2. Platelets ≥100,000/mcL

3.1.3.3. Total bilirubin ≤ institutional upper limit of normal (ULN)

3.1.3.4. AST(SGOT)/ALT(SGPT) ≤3 × institutional ULN

3.1.3.5. Creatinine ≤ institutional ULN

3.1.4. Patients with a prior or concurrent malignancy whose natural history or treatment does not have the potential to interfere with the safety or efficacy assessment of the investigational regimen are eligible for this trial

3.1.5. Have known or suspected clinical stage I-IIb NSCLC after CT and/or PET_CT at time of enrollment

3.1.6. Presence of a target lesion with a minimum volume of 1.0 cm^3, (approximately1.2 cm in diameter) and ≤ 5.0 cm in diameter

3.1.7. Agreement from a cardiothoracic surgeon, following review of past medical history, medications, pulmonary function testing, and CT scan that patient is likely to be a surgical candidate and that, after considering known possible adverse events, delivery of intratumoral cisplatin is unlikely to adversely affect surgical feasibility

3.1.8. Rapid on-site cytopathologic examination (ROSE) performed during the procedure returns likely NSCLC (per the determination of a trained, attending, cytopathologist). No research procedures will be performed if ROSE is non-diagnostic

3.1.9. A CT scan of the chest (with or without contrast) within 1 month of the screening visit

3.1.10. Ability to understand and the willingness to sign a written informed consent document

3.2. **Exclusion criteria**

3.2.1. Use of an investigational agent within 30 days of the screening visit

3.2.2. IV chemotherapy within the 30 days of the screening visit

3.2.3. Pregnancy/lactation (pregnancy test to be performed by pre-op as part of standard of care for women of child-bearing age as defined by UVMMC Policy NPREP16)

3.2.4. History of prior radiation to the study lesion

3.2.5. History of allergic reaction to cisplatin or its derivatives

3.2.6. Patients with uncontrolled intercurrent illness

3.2.7. Physician determination that patient would not be appropriate for study

4. TREATMENT PLAN

4.1. Agent Administration

4.1.1. Treatment will be administered intraprocedurally. Reported adverse events and potential risks are described in the Data and Safety Monitoring Plan. No investigational or commercial agents or therapies other than those described below may be concomitantly administered with the intent to treat the patient's malignancy.

4.1.2. The agent will be administered at 3 potential dose levels in a 3+3 design.[31]

4.1.3. IT cisplatin 1mg/ml will be dosed with a delivered volume based on a proportion of the volume of the tumor.

4.1.4. This volumetric dosing approach is being employed to account for the dramatic tumor volume differences seen in this diameter range. Prior studies have most commonly used an empirically derived intratumoral dose of 40mg or less (although have ranged to 100mg intratumorally, Appendix B). To minimize the risk of extravasation of the agent from small tumors we have adopted a volumetric dosing strategy.

4.1.4.1. First dose cohort: The delivered quantity of cisplatin will equal one-fourth ($1/4^{th}$) of the total tumor volume.

4.1.4.2. De-escalation dose cohort : The delivered quantity of cisplatin will equal one-eight ($1/8^{th}$) of the total tumor volume This dose cohort will only be used if the ¼-tumor volume dose cohort is terminated due to DLT.

4.1.4.3. Escalation dose cohort: The delivered quantity of cisplatin will equal one-half (1/2) of the total tumor volume

4.1.4.4. Lower or higher dose cohorts may be pursued if indicated based on the rate of DLT, pending formal review and approval by the appointed Data Safety Monitoring Committee of the UVM Cancer Center. This is a standard approach since the DSMC reviews all AEs and relevant data at any change in dose level (pre-specified or otherwise) before preceding to the next dose level.

4.1.5. Tumor volume will be determined using automated segmentation available in the open-source software platform (https://chestimagingplatform.org) and manually verified.

4.1.6. The minimum volume that is feasible to deliver is 0.5mL.

4.1.7. This leads to the following "INITIAL" *example* dosing regimen for tumors of a specified volume, beginning at 1/4th Volumetric Dosing and assuming that < 2 of the first 3 participants have a dose limiting toxicity (DLT), defined as a Grade 3 by the CTCAE v5.0:

		INITIAL		
Example Tumor Volume(mL)*	**Approximate Tumor Diameter (cm)***	**First Dose Cohort: 1/4 Tumor Volume (mL)**	**First Dose Cohort: IT Cisplatin Delivered Volume (mL)**	**Total Cisplatin (mg)**
1.2	1.3	0.3	Not Eligible**	
1.8	1.5	0.5	0.5	0.5
4.2	2.0	1.1	1.1	1.1
8.2	2.5	2.1	2.1	2.1
14.1	3.0	3.5	3.5	3.5
22.5	3.5	5.6	5.6	5.6
33.5	4.0	8.4	8.4	8.4
47.7	4.5	11.9	11.9	11.9
65.5	5.0	16.4	16.4	16.4
*Calculated based on volume of a sphere				
**Minimum delivery volume is 0.5mL				

4.1.8. The following are *example* doses for the DE-ESCALATION cohort:

		DE-ESCALATION		
Example Tumor Volume(mL)*	**Approximate Tumor Diameter (cm)***	**Second Dose Cohort: 1/8 Tumor Volume(mL)**	**Second Dose Cohort: IT Cisplatin Delivered Volume (mL)**	**Total Cisplatin (mg)**
1.2	1.3	0.2	Not Eligible**	
1.8	1.5	0.2	Not Eligible**	
4.2	2.0	0.5	0.5	0.5
8.2	2.5	1.0	1.0	1.0
14.1	3.0	1.8	1.8	1.8
22.5	3.5	2.8	2.8	2.8
33.5	4.0	4.2	4.2	4.2
47.7	4.5	6.0	6.0	6.0
65.5	5.0	8.2	8.2	8.2
*Calculated based on volume of a sphere				
**Minimum delivery volume is 0.5mL				

4.1.9. The following are *example* doses for the ESCALATION cohort:

		ESCALATION		
Example Tumor Volume(mL)*	Approximate Tumor Diameter (cm)*	Second Dose Cohort: 1/2 Tumor Volume(mL)	Second Dose Cohort	Total Cisplatin (mg)
1.2	1.3	0.6	0.6	0.6
1.8	1.5	0.9	0.9	0.9
4.2	2.0	2.1	2.1	2.1
8.2	2.5	4.1	4.1	4.1
14.1	3.0	7.1	7.05	7.05
22.5	3.5	11.3	11.3	11.3
33.5	4.0	16.8	16.8	16.8
47.7	4.5	23.9	23.9	23.9
65.5	5.0	32.8	32.8	32.8
*Calculated based on volume of a sphere				
**Minimum delivery volume is 0.5mL				

4.1.10. Dose escalation will be performed as follows:

Number of Patients with DLT at a Given Dose Level	Escalation Decision Rule
0 out of 3	Enter 3 patients at the next escalation dose.
≥2	Dose escalation will be stopped. This dose level will be declared the maximally administered dose (highest dose administered). Three (3) additional patients will be entered at the next lowest dose level if only 3 patients were treated previously at that dose. If this is the first dose evaluated, a lower dose level will be established, in concert with DSMC, and 3 patients enrolled to that level.
1 out of 3	Enter at least 3 more patients at this dose level. • If 0 of these 3 patients experience DLT, proceed to the next dose level. • If 1 or more of this group suffer DLT, then dose escalation is stopped, and this dose is declared the maximally administered dose. Three (3) additional patients will be entered at the next lowest dose level if only 3 patients were treated previously at that dose.
≤1 out of 6 at highest dose level below the maximally administered dose	This will be the recommended phase 2 dose. At least 6 patients must be entered at the recommended phase 2 dose.

4.2. **Duration of Study Period:**

4.2.1. There is only a single delivery of the agent, cisplatin.

4.2.2. Primary endpoint adjudication for DLT will occur after the 2 week post-procedure evaluation and laboratory values are completed.

4.2.3. The patient will be monitored for adverse events until surgical resection or for 30 days after cisplatin delivery, whichever occurs first.

4.3. **Treatments and Evaluations by Visit:**

4.3.1. Screening Evaluation:

4.3.1.1. Initial evaluation performed via face-to-face visit or televideo (standard of care)

4.3.1.2. A non-contrast chest CT scan will be performed within 30 days of the screening visit.

4.3.1.3. Baseline blood draws within 14 days of bronchoscopy:

4.3.1.3.1. Samples for the following laboratory tests will be sent if not available:

4.3.1.3.1.1. Hematology Panel: WBC count, RBC count, hemoglobin, hematocrit, platelet count, differential count (neutrophils, eosinophils, basophils, monocytes, lymphocytes, other cells)

4.3.1.3.1.2. Chemistry Panel (serum or plasma): sodium, potassium, chloride, bicarbonate, glucose, creatinine, albumin (pre-op only), calcium, magnesium

4.3.1.3.1.3. Liver panel: ALT, AST, INR, alkaline phosphatase, bilirubin

4.3.2. Bronchoscopic Procedure:

4.3.2.1. Pre-procedure SARS-CoV-2 testing will be performed prior to the procedure (standard of care).

4.3.2.2. Approximately 15mL of blood for research purposes will be drawn.

4.3.2.3. A pre-operative CXR will be performed if no chest imaging (CXR, CT, PET-CT) within 7 days prior to the procedure (research).

4.3.2.4. Patients will be brought to the operating room or procedure suite. Anesthesia staff will be present to administer deep sedation to minimize cough.

4.3.2.5. Standard white light bronchoscopy will be performed. (standard of care)

4.3.2.6. The linear endobronchial ultrasound bronchoscope (EBUS) will be used to perform invasive mediastinal staging. (standard of care)

4.3.2.7. For all participants, all N2 or N3 lymph nodes greater than 5mm in short axis by ultrasound will be aspirated via EBUS-TBNA and evaluated using

ROSE. Two (2) passes with lymphocytes or up to 5 non-diagnostic passes will be considered negative (ROSE charged to research). If ROSE is positive at an N2 or N3 station no agent will be delivered and the patient will be discontinued from the study.

4.3.2.8. The clinically selected bronchoscopic procedure (e.g. EBUS bronchoscope, electromagnetic navigation, robotic platform) will be used to locate the lesion of interest. The lesion will be sampled via needle aspiration. Rapid on-site cytopathologic examination will be used to evaluate needle aspirates unless the target lesion was previously sampled and diagnosed on final pathology as NSCLC. All aspirates necessary for clinical care will be obtained first. This is usually 10-20 aspirates depending on cytopathology requirements. (standard of care)

4.3.2.9. After the confirmation of NSCLC on ROSE, the research portion of the procedure will re-start.

4.3.2.10. An additional IV will be placed by anesthesiology for blood draws (research)

4.3.2.11. Before cisplatin injection, IV ampicillin-sulbactam 3g x 1 will be administered. If the patient has a beta-lactam allergy, clindamycin 900mg will be substituted).(research)

4.3.2.12. Prior to instillation of the cisplatin, 3 needle biopsies with a 21-gauge needle will be obtained for research purposes.. (research)

4.3.2.13. The 21-gauge needle will then be re-inserted into the tumor. The stylet will be removed and a syringe containing the predefined total dose of 1mg/ml cisplatin attached.

4.3.2.14. Intraoperative cone beam CT scan will be used to verify the location. If the needle tip is not located within the lesion, the needle will be repositioned using fluoroscopy. No agent will be delivered if the needle tip is not within the lesion.

4.3.2.15. Injection will then be started. If difficulty is encountered injecting with the 21G needle a 19G needle may be substituted. The same requirements for the location of the needle (4.3.2.13) will be imposed.

4.3.2.16. Following delivery of the agent, a second cone-beam CT scan will be performed to assess for extravasation of the agent.

4.3.2.17. Serial blood draws (maximum 9) for platinum levels will be drawn after delivery of cisplatin and continuing for up to 2 hours later. Each is approximately 2mL and time stamped to allow creation of a titration curve.(research)

4.3.2.18. Research time will be recorded, beginning at completion of acquisition of all samples necessary for clinical care and stopped following delivery of the agent.

4.3.2.19. The patient will be extubated and transferred to the PACU.

4.3.2.20. A follow up CXR will be performed (standard of care)

4.3.3. 24H Post-Bronchoscopy Evaluation

4.3.3.1. A follow up telephone/televideo or in-person evaluation will be performed by a physician, advanced practice practitioner, or nurse and will occur within 36 hours of completion of the procedure. A standard script (Appendix) will be used to assess for AEs. (research)

4.3.3.2. Blood will be drawn for an additional platinum level for patients able to return for an in-person visit.(research)

4.3.4. 1 Week Post-Bronchoscopy Follow Up

4.3.4.1. Participants will undergo an evaluation by a physician or advanced practice provider between 5-9 days post-procedure. Visit will occur in-person. The only exception would be COVID related issues. In that case, telemedicine and local labs will be allowed. No phone evaluations will occur unless there is no other option. A standard script will be used to assess for AEs. (research)

4.3.4.2. A blood draw to assess for AEs will be performed between 5-9 days post-procedure.(research)

4.3.4.2.1. Samples for the following laboratory tests will be sent to UVMMC laboratories for analysis:

4.3.4.2.1.1. Hematology Panel
4.3.4.2.1.2. Chemistry Panel
4.3.4.2.1.3. Liver Panel

4.3.4.3. Blood will be drawn for an additional platinum level.(research)

4.3.5. 2 Week Post-Bronchoscopy Follow Up

4.3.5.1. Participants will undergo an evaluation by a physician or advanced practice provider between 12-16 days post-procedure. Visit will occur in-person. The only exception would be COVID related issues. In that case, telemedicine and local labs will be allowed. No phone evaluations will occur unless there is no other option.A standard script will be used to assess for AEs. (research)

4.3.5.2. A blood draw to assess for AEs will be performed between 12-16 days post-procedure.(research)

4.3.5.2.1. Samples for the following laboratory tests will be sent to UVMMC laboratories for analysis:

4.3.5.2.1.1. Hematology Panel

4.3.5.2.1.2. Chemistry Panel

4.3.5.2.1.3. Liver Panel

4.3.6. A Chest CT will be performed between 12 and 30 days after delivery of the cisplatin.(research)

4.3.7. Surgical Resection, if planned, will be scheduled between 14 and 30 days after delivery of the cisplatin.

4.3.7.1. Approximately 15mL of blood will be drawn for research purposes on the day of (prior to) the resection procedure.

4.3.8. Routine Cancer Care

4.3.8.1. Patients will receive additional therapy at the discretion of the multidisciplinary treatment team. This review generally occurs at the University of Vermont Lung Transdisciplinary Conference (attendance generally includes but is not limited to interventional pulmonary, pulmonary, oncology, radiation oncology, cardiothoracic surgery, pathology, and thoracic radiology).

5. PHARMACEUTICAL INFORMATION:

5.1. A list of the adverse events and potential risks associated with the investigational or commercial agents administered in this study can be found in Section 10.1.

5.2. Formulation of Cisplatin: Cisplatin (cis-diamminedichloroplatinum) will be purchased from Fresenius/APP Pharmaceuticals. Cisplatin is a heavy metal complex containing a central atom of platinum surrounded by two chloride atoms and two ammonia molecules in the cis position. It is a yellow to orange powder with the molecular formula PtCl2H6N2, and a molecular weight of 300.05. It is soluble in water or saline at 1 mg/mL. It has a melting point of 207°C. Cisplatin Injection is a sterile aqueous solution for intravenous use, each mL containing 1 mg cisplatin and 9 mg sodium chloride. Hydro- chloric acid and/or sodium hydroxide added to adjust pH. The pH range of Cisplatin Injection is 3.8-5.9.

5.3. Supply of Study Medication On-Site: Fresenius/APP Cisplatin will be stocked and handled in the Research Pharmacy of the University of Vermont Medical Center. It will be clearly labeled using standard and established protocols. The cisplatin will be delivered in syringes, straight drug (1mg/ml) to the OR/procedure suite.

5.4. Study Drug Dispensing**:** Once the subject has signed the Informed Consent Form to participate in the study, the subject will be associated with the research study using the Research Flag located in the patient banner section of the EPIC. The UVMMC Investigational Drug Service is alerted a study drug order will be placed in the subject's EMR by the principal investigator or designee. A study drug order is placed in EMR

using the unique study identification number, and subject's study ID number.

5.5. Drug Accountability**:** The UVMMC Investigation Drug Service will keep an inventory of all dispensed study drug. Study drug accountability logs are maintained by the IDS. Administration of study drug is recorded in subject's EMR. Study drug not administered is returned to IDS and destroyed.

5.6. Medication Interactions:

5.6.1. No other investigational drugs will be allowed prior to surgical resection.

5.7. Potential Teratogenicity:

5.7.1. Enrolled participants will be counseled that chemotherapeutics including cisplatin could be harmful to the fetus should the patient become pregnant following administration of the drug.

6. STATISTICAL CONSIDERATIONS

6.1. Study design

6.1.1. This is a 3+3 dose escalation protocol.

6.1.2. Enrollment is staggered, and no patient will be enrolled until the prior patient has completed the 1 week assessment.

6.1.3. The Initial dose cohort will be 1/4th tumor volume.

6.1.4. The subsequent dose escalation protocol is dependent on the rate of DLT as noted in the Treatment Plan.

6.2. Criteria for endpoint evaluation

6.2.1. Primary Endpoint: Safety (DLT), as defined by CTCAE v 5.0.

6.2.1.1. Assessment of Adverse Events: The PI will be responsible for evaluating all AEs at the end of the individual patient study period. CTCAE v 5.0 grade 3 or above will be considered a DLT

6.2.1.2. The reporting procedures to be followed are presented in the "CTEP, NCI Guidelines: Adverse Event Reporting Requirements" which can be downloaded from the CTEP web site (http://ctep.cancer.gov/protocolDevelopment/electronic_applications/docs/aeguidelines.pdf).

6.2.1.3. Adverse Laboratory Values

6.2.1.3.1. Not every laboratory abnormality qualifies as an adverse event. A

laboratory test result must be reported as an adverse event if it meets any of the following criteria:

6.2.1.3.2. • Is accompanied by clinical symptoms

6.2.1.3.3. • Results in a change in study treatment (e.g., dosage modification, treatment interruption, or treatment discontinuation)

6.2.1.3.4. • Results in a medical intervention (e.g., potassium supplementation for hypokalemia) or a change in concomitant therapy

6.2.1.3.5. • Is clinically significant in the investigator's judgment

6.2.1.3.6. It is the investigator's responsibility to review all laboratory findings. Medical and scientific judgment will be exercised in deciding whether an isolated laboratory abnormality should be classified as an adverse event.

6.2.1.3.7. Any abnormality of safety laboratories ("laboratory panel") will be considered an AE if is a change from baseline (based on CTCAE v5.0)

6.2.1.4. Adverse Event Characteristics:

6.2.1.5. **CTCAE term (AE description) and grade:** The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 5.0 will be utilized for AE reporting.

6.2.1.6. "**Expectedness;"** AEs can be "Unexpected" or "Expected" for expedited reporting purposes only.

6.2.1.7. **Attribution** of the AE:

Definite – The AE *is clearly related* to the study treatment.
Probable – The AE *is likely related* to the study treatment.
Possible – The AE *may be related* to the study treatment.
Unlikely – The AE *is doubtfully related* to the study treatment.
Unrelated – The AE *is clearly NOT related* to the study treatment.

6.2.2. **Secondary and Exploratory Endpoints:**

6.2.2.1. Safety profile of intralesional cisplatin as classified by the CTCAE, v5.0

6.2.2.2. Proportion of cisplatin-treated subjects proceeding to surgery without extended local injection-related delay, defined as >30 days from preplanned surgery date (21 days from preplanned surgical date with 7-day window to allow for operating room [OR] scheduling)

6.2.2.3. Time to surgery from intralesional therapy.

6.2.3. **Biomarker and Surrogate Endpoints for Research Purposes:**

6.2.3.1. The below assays will be performed from:

6.2.3.1.1. Blood samples, pre-procedure on the date of the bronchoscopy and pre-procedure on the date of the surgical resection

6.2.3.1.2. Three (3) needle biopsies obtained at the time of bronchoscopy before cisplatin delivery

6.2.3.1.3. Seven (7) needle biopsies obtained from the ex-planted resection specimen (3 for platinated DNA, 1 for tumor organoid, 3 for other biomarker assessments described below)

6.2.3.1.4. Approximately 20 sections of fixed tissue from the resected tumor (final amount to be determined by pathology following obtaining all necessary tissue for clinical care, including any additional candidacy for clinical trials)

Objectives	**Endpoints**
Secondary	
To assess pathologic responses (PR) in the resected tumor following a single intra-tumoral administration of cisplatin in subjects with early stage NSCLC	• Proportion of cisplatin-treated subjects with major pathologic response (MPR), i.e., ≥90% reduction in viable tumor cells, at the time of NSCLC resection • Proportion of cisplatin-treated subjects with pathologic complete response (pCR), i.e., no residual viable tumor cells, at the time of NSCLC resection • Change from baseline in percent of residual viable tumor cells (% RVT) in cisplatin-treated subjects at the time of NSCLC resection
Exploratory	
To assess radiologic responses in the tumor following a single intra-tumoral administration of cisplatin in subjects with early stage NSCLC	• Tumor response rate evaluation (Δ tumor size) according to RECIST 1.1 criteria
To explore the change from pre-treatment baseline in measures of immune activation in the blood and tumor (eg, percentages of immune cell subsets, numbers of T-cell clones, concentrations of serum cytokine levels)	Pre- and post-treatment in the bronchoscopic biopsy and surgically resected tumor samples, respectively: • Tissue levels of immune-related gene expression as measured by appropriate techniques such as RNA-sequencing in biopsy samples collected at diagnosis and time of resection • Tissue frequencies of immune cell subsets determined via semi-quantitative immunohistochemistry (IHC) analysis of diagnostic biopsy and surgically-resected tissue specimens • To assess changes in T cell clonality in blood by TCR-sequencing (via DNAseq) at baseline and after therapy, TCR repertoire sequencing will be performed on the tumor to profile in the blood • Tumor mutational burden will be assessed

Objectives	Endpoints
	by next generation sequencing. Pre- and post-treatment blood: • Changes in the concentrations of immune activation and inflammation markers in the serum before and after treatment. Serum cytokine and inflammatory biomarker (e.g., hs-CRP, IL-6, TNF-a) concentrations will be measured by appropriate techniques such as Mesoscale Discovery (MSD). • Serum neutrophil-to-lymphocyte ratio (NLR): Defined as neutrophil count divided by lymphocyte count, as determined by complete blood count (CBC) with differential conducted at baseline and after treatment. • Change in frequencies of immune cell subsets in blood as measured by flow cytometry (eg, CD3, CD8, PD-1) at baseline and after treatment (absolute frequencies and percent change from baseline). • To assess changes in T cell clonality in blood by TCR-sequencing (via DNAseq) at baseline and after therapy. TCR repertoire sequencing will be performed on the biopsy sample of the tumor to ID clones specific to the tumor to profile in the blood.
To evaluate cisplatin pharmacokinetic parameters in the blood and to confirm cisplatin exposure and distribution in the	• Maximum observed serum concentration (Cmax), area under the concentration-time, half-life (T1/2), clearance of drug after administration (CL), volume of distribution (Vd) Cisplatin concentration in the tumor at the site of injection and adjacent tumor regions measured by an ELISA-like method that can detect cisplatinated DNA

6.3. Sample Size

6.3.1. The minimum sample size will be 10. Initial dose ranging will be performed in a 3+3 fashion. Additional patients up to a minimum of 10 patients will be enrolled at the recommended Phase II dose to assess for additional AEs.

6.3.2. The maximum sample size, if six patients at each of two dose levels were enrolled, would be 12 patients.

6.3.3. Accrual would be anticipated to occur over a one year time period

6.4. Power:

6.4.1. Primary endpoint: The sample size in this study is determined by the dose escalation protocol and is thus not driven by considerations of statistical power for between group comparisons.

6.5. Statistical Analysis Plan

6.5.1. Primary Endpoint: Statistical analysis will consist of descriptive summaries of the treated patients along with a summary of the dose-escalation sequence and determination of the MTD. Adverse events, including type and grade, will be tabulated according to dose tier.

7. DATA SAFETY AND MONITORING

7.1. Trial Safety: Identification of risks and plans to minimize risk

Expected Risks	Frequency
• Renal injury/failure (reported with systemic administration, never reported with intratumoral delivery)	☐ Occurs frequently ☐ Occurs infrequently ☐ Occurs rarely ☒ Frequency unknown
• Myelosuppression (reported with systemic administration, never reported with intratumoral delivery)	☐ Occurs frequently ☐ Occurs infrequently ☐ Occurs rarely ☒ Frequency unknown
• Neurologic side effects, including hearing impairment (reported with systemic administration, never reported with intratumoral delivery)	☐ Occurs frequently ☐ Occurs infrequently ☐ Occurs rarely ☒ Frequency unknown
• Hepatic injury (reported with systemic administration, never reported with intratumoral delivery)	☐ Occurs frequently ☐ Occurs infrequently ☐ Occurs rarely ☒ Frequency unknown
• Nausea/vomiting (known but rare adverse event with intratumoral cisplatin)	☐ Occurs frequently ☐ Occurs infrequently ☒ Occurs rarely ☐ Frequency unknown
• Significant bleeding (defined as >100mL is rarely reported with peripheral biopsies	☐ Occurs frequently ☐ Occurs infrequently

<1.0%). Unknown if risk increases with additional research biopsies.	☐ Occurs rarely ☒ Frequency unknown
• Lung or airway injury including pneumothorax. Pneumothorax is reported with peripheral biopsies. Unknown if this risk increases above that associated with performing clinically indicated biopsies since the lesion and needle location will be already established. Other risks to the airway or lung are possible (e.g. pneumonitis) but are not reported or expected.	☐ Occurs frequently ☐ Occurs infrequently ☐ Occurs rarely ☒ Frequency unknown

Other potential risks:

1. Incorrect ROSE diagnosis is very rare, but could lead to cisplatin delivery into a lesion other than a NSCLC. Further, a single, resectable lesion from a metastatic site is also an unusual presentation. Presentation of small cell lung cancer with a single peripheral nodule is also unusual. Although cisplatin is active against both small cell lung cancer, and the most common metastatic lesions (bladder, colorectal, breast, prostate, and head and neck), injection into a tumor other than a NSCLC would be considered an adverse event and reported to the IRB. Similarly, in the extremely rare circumstance that there would be an injection into a lesion with a ROSE diagnosis of NSCLC, but a final diagnosis of a benign disease, this would also be considered an adverse event and reported to the IRB.

2. Delay in surgical resection. Although not clearly documented in the setting of systemic neoadjuvant IV therapy for Stage I or II NSCLC this is possible. However, known adverse events related to a single dose of cisplatin would be anticipated to resolve within 30 days making this unlikely. This would also be considered an adverse event and reported to the IRB.

7.1.1. Planned safety tests/procedures/observations to be performed as measures to protect participants against foreseeable risks.

7.1.1.1. Pre-procedure evaluation by Cardiothoracic Surgery to insure resectability.

7.1.1.2. Follow up phone call, video, or face-to-face visit performed by a physician, advanced practice practitioner (APP), or nurse the day after the bronchoscopy

7.1.1.3. Evaluation (video/telephone or face-to-face) by a physician or APP, 5-9 days post-bronchoscopy

7.1.1.4. Blood draw for laboratory panel at 5-9 days and 12-16 days post-bronchoscopy

7.1.1.5. Only experienced pathology attendings will make the ROSE determination during the procedure (e.g. fellows will not perform the ROSE diagnosis)

7.1.2. Criteria under which an INDIVIDUAL SUBJECT'S study treatment or study participation would be stopped or modified:

7.1.2.1. At subject, PI, or study team member request

7.1.2.2. If the subject had a Serious Adverse Event (SAE) deemed related to study Other: specify

7.1.2.3. During the bronchoscopy, bleeding >100mL or concern for a patient's ability to continue to tolerate the bronchoscopy would result in stopping that procedure (regardless of whether cisplatin had been delivered into the tumor or not). No further intratumoral delivery would be performed.

7.1.3. Criteria under which THE ENTIRE STUDY would need to be stopped.

7.1.3.1. A 30% or greater rate of possibly related, serious, unanticipated events after 6 patients, will trigger halting of the study and initiate IRB and DSMCC review.

7.1.4. Withdrawals/dropout reporting to the IRB prior to study completion will occur at the IRB annual continuing renewal submission

7.1.5. Plan for management of incidental findings:

7.1.5.1. We do not anticipate incidental findings. The pre-bronchoscopy CT scan is obtained as part of clinical care for all patients are undergoing a clinically indicated bronchoscopy. Unanticipated findings on the pre-resection CT will be evaluated as potential AEs.

7.2. Adverse Events:

7.2.1. Definition of adverse events (AE) for this study:

7.2.1.1. An adverse event (AE) will be considered any undesirable sign, symptom or medical or psychological condition even if the event is not considered to be related to the investigational drug/device/intervention. Medical conditions/diseases present before starting the investigational drug/intervention will be considered AEs only if they worsen after starting study treatment/intervention. An AE is also any undesirable and unintended effect of research occurring in human subjects as a result of the collection of identifiable

private information under the research. AEs also include any problems associated with the use of an investigational device that adversely affects the rights, safety or welfare of subjects.

7.2.2. Define of serious adverse events (SAEs):

7.2.2.1. A serious adverse event (SAE) will be considered any undesirable sign, symptom, or medical condition which is fatal, life-threatening, requires or prolongs inpatient hospitalization, results in persistent or significant disability/incapacity, constitutes a congenital anomaly/ birth defect, is medically significant and which the investigator regards as serious based on appropriate medical judgment. An important medical event is any AE that may not result in death, be life-threatening, or require hospitalization but may be considered an SAE when, based upon appropriate medical judgment, it may jeopardize the patient and may require medical or surgical intervention to prevent one of the outcomes listed in the definitions of SAEs.

7.2.2.2. Any serious psychological and emotional distress resulting from study participation (suggesting need for professional counseling or intervention).

7.2.3. ALL AEs above will be collected/ recorded.

7.2.4. A REDCap database will be used to save and manage data.

7.2.4.1. The database is located on the secure UVM server and is firewall protected. Data will be collected from source documents including but not limited to clinic notes, laboratory results, and other EMR available documents. The PI will review all AE data at the end of the study period for each patient. Data will be entered and managed by trained study personnel.

7.2.5. AEs be classified/graded using NCI Common Toxicity Criteria for Adverse Events (CTCAE), Version 5.0

7.2.6. The PI will determine the relationship of AEs to the study using the following scale:

7.2.6.1. Definite: AE is clearly related in time and a direct association can be demonstrated to the study intervention.
7.2.6.2. Probable: AE is reasonably related in time and is more likely explained by the study intervention than by other causes.
7.2.6.3. Possible: AE may be reasonably related in time and the AE can be explained equally well by causes other than the study intervention.
7.2.6.4. Unlikely: A potential relationship could exist with the study intervention, but the AE is most likely explained by other causes.
7.2.6.5. Unrelated:AE is clearly not related to intervention and can be fully explained by another cause. This other cause should be provided.

7.2.7. Recording/reporting of baseline AEs will begin after the subject signs consent. Subsequent AEs will be recorded following initiation of the bronchoscopic procedure.

7.2.8. Recording/reporting of AEs will end following surgical resection or 30 days after cisplatin delivery, whichever occurs first.

7.2.9. The PI will review all AEs following the 1 week follow up visit (occurs at 5-9 days performed by a physician or advanced practice provider), both to assess for unexpected and serious AEs and to ensure that dose escalation occurs consistent with the statistical plan.

8. ADVERSE EVENT REPORTING:

Type of Event	To whom will it be reported	Time Frame for Reporting	How to report?
Death of a research subject unless the death is expected (e.g. due to disease progression).	UVMCC DSMC and UVM IRB	DSMC: Within 24 hours IRB: Within 24 hours of knowledge of the event	DSMC: Email IRB: RNI Click form
Serious Adverse Event (SAE), regardless of relatedness of expectedness	UVMCC DSMC. Per IRB policy, unexpected, related SAEs are reported	DSMC: Within 10 working days from the time the study team received knowledge of the event. IRB: 24 hours	DSMC: Email IRB: RNI Click form
Non-serious AEs	UVMCC DSMC and UVM IRB	At quarterly DSMCC report. Annually at IRB Continuing review.	DSMC: Email IRB: Click form

8.1. Unanticipated Problems and Protocol Deviations

8.1.1. Definition of an unanticipated problem:

8.1.1.1. An unanticipated problem is any event/experience that meets ALL 3 criteria below:

8.1.1.1.1. Unexpected (in terms of nature, severity, or frequency) given: (a) the research procedures that are described in the protocol-related documents, such as the IRB-approved research protocol and informed consent document; and (b) the characteristics of the subject population being studied;

8.1.1.1.2. Related or possibly related to a subject's participation in the research;

8.1.1.1.3. Suggests that the research places subjects or others at a greater risk

of harm (including physical, psychological, economic, or social harm) related to the research than was previously known or recognized.

8.1.1.2. Definition of a protocol deviation:

8.1.1.2.1. A protocol deviation is any variance from the protocol involving a subject or subjects that is not approved by the IRB prior to its initiation or implementation, and occurs when a member of the study team departs from the IRB-approved protocol in any way without the investigator first obtaining IRB approval

8.1.1.3. Recording/reporting of Unanticipated Problems and Protocol Deviations will begin after the subject signs consent

8.1.1.4. Recording/reporting of Unanticipated Problems and Protocol Deviations will end at surgical resection or 30 days after cisplatin delivery, whichever occurs first.

8.1.2. Unanticipated Problem, and Protocol Deviation reporting.

Type of Event	To whom will it be reported	Time Frame for Reporting	How to report?
Unanticipated Problems that are not AEs or protocol deviations.	IRB and DSMC	Within 7 days to both the IRB and DSMC	DSMC: Email IRB: Click form

9. DATA COLLECTIONS METHODS

9.1. Endpoints of the study are described in Section 6 of the protocol

9.2. Endpoints will be collected/recorded using protocol-specific Case Report Forms (CRFs), and source documents and captured in a REDCap database

9.3. The database is located on the secure UVM server and is firewall protected.

9.4. Correlative sub-studies, will link specimens other research data using the following:

9.4.1. All specimens (including CT scans) will be deidentified and scrubbed of HIPPA identifiers. A study number will be assigned. Only the PI and primary study coordinater will have access to the key to link study number with patient information.

9.5. The UVMCC DSM Report will be provided to the UVMCC

9.6. The UVMCC DSMC will be responsible for overseeing the study data

9.7. The PI will perform aggregate review of:

9.7.1. All AEs
9.7.2. Unanticipated Problems
9.7.3. Protocol violations
9.7.4. Audit results
9.7.5. Application of dose finding escalation/de-escalation rules
9.7.6. Application of study designed stopping/decision rules
9.7.7. Early withdrawals
9.7.8. Whether the study accrual pattern warrants continuation/action
9.7.9. Endpoint data

9.8. This aggregate review will occur quarterly

1. TABLE OF STUDY VISITS AND PROCEDURES:

	Screening	Phase 1: Intratumoral Cisplatin Protocol				
Study Visit	**Pre-Bronchoscopy**	**Bronchoscopic Procedure**	**24H Followup**	**1 Week Followup**	**2 Week Followup**	**Surgical Resection****
Informed Consent	X					
History and Evaluation	X		X	X	X	
Research Biopsies		X				
Delivery of Cisplatin		X				
Cisplatin Blood Levels		X	X	X		
CXR	X (pre-bronchoscopy if no imaging within prior 7 days)	X (post-procedure)				
24H Post-Procedure Follow Up			X			
Hematology Panel	X			X	X	
Chemistry Panel	X			X	X	
Liver Panel	X			X	X	
Albumin	X					
Blood Draw for Research		X				X (day of resection)
Review of Adverse Events and Concomitant Medications			X	X	X	
Chest CT Scan (non-contrast if not performed or inadequate will ordered for research purposes)	X				X	
Obtain Research Resected Pathology Specimen						X (fresh and fixed)

* Research biopsies will be evaluated for changes in immunocellular infiltrate vs. resection specimen
**Surgical resection to occur 14-30 days post-procedure

APPENDIX A: DSM REPORTING TABLE FOR ADVERSE EVENTS

Expected Risks	Number	Frequency
• Renal injury/failure (reported with systemic administration, never reported with EBUS-TBNI delivery)		
• Myelosuppression (reported with systemic administration, never reported with EBUS-TBNI delivery)		
• Neurologic side effects, including hearing impairment (reported with systemic administration, never reported with EBUS-TBNI delivery)		
• Hepatic injury (reported with systemic administration, never reported with EBUS-TBNI delivery)		
• Nausea/vomiting (known but rare adverse event with EBUS-TBNI cisplatin)		
• Significant bleeding (defined as >100mL is rarely reported with EBUS for biopsies <0.1%, never for EBUS-TBNI)		

APPENDIX B:
NCI COMMON ADVERSE EVENTS FOR COMMONLY USED ONCOLOGY DRUGS (CAEPR)

Possible Side Effects of Cisplatin (Table Version Date: November 8, 2019)

COMMON, SOME MAY BE SERIOUS In 100 people receiving Cisplatin, more than 20 and up to 100 may have:
• Infection, especially when white blood cell count is low • Bruising, bleeding • Anemia which may cause tiredness, or may require blood transfusions • Kidney damage which may cause swelling, may require dialysis • Hearing loss including ringing in the ears • Nausea, vomiting • Confusion • Numbness and tingling of the arms and legs

OCCASIONAL, SOME MAY BE SERIOUS In 100 people receiving Cisplatin, from 4 to 20 may have:
• Diarrhea • Change in taste • Allergic reaction which may cause rash, low blood pressure, wheezing, shortness of breath, swelling of the face or throat • Hair loss

RARE, AND SERIOUS In 100 people receiving Cisplatin, 3 or fewer may have:
• Brain damage, Posterior Reversible Encephalopathy syndrome, which may cause headache, seizure, blindness • Seizure • A new cancer resulting from treatment of a prior cancer

APPENDIX C:

SCRIPT FOR IDENTIFICATION OF ADVERSE EVENTS BASED ON NCI CAEPR

A PHASE I TRIAL OF INTRATUMORAL CISPLATIN DELIVERED AT DIAGNOSIS FOR EARLY STAGE, RESECTABLE, NON-SMALL CELL LUNG CANCER

SUBJECT NO._____ SUBJECT INITIALS_____

Follow up contact- 1 week post bronchoscpy ____/____/_____
Date

Did the subject report any adverse events? ☐ Yes ☐ No

1. Do you have any health concerns since we saw you on the day of the procedure?

2. Since the procedure, have you had:

Wheezing or shortness of breath?

Chest pain?

Fever or rash?

Easy bruising or bleeding?

Itching?

Hearing loss or ringing in your ears?

Nausea or vomiting?

Confusion?

Numbness or tingling in the arms or legs/

Diarrhea?

Change in taste?

Swelling of the face or throat?

Headache?

Seizure?

Followup Contact Conducted by: _____ ____/____/_____

REFERENCES:

1. Siegel, R. L., Miller, K. D. & Jemal, A. Cancer statistics, 2015. *Ca Cancer J Clin* **65**, 5 29 (2015).

2. Fan, C. *et al.* Risk factors for locoregional recurrence in patients with resected N1 non-small cell lung cancer: a retrospective study to identify patterns of failure and implications for adjuvant radiotherapy. *Radiat Oncol* **8**, 286 (2013).

3. Garg, A. D. *et al.* Trial watch: Immunogenic cell death induction by anticancer chemotherapeutics. *Oncoimmunology* **6**, 00–00 (2017).

4. Samson, P. *et al.* Effects of Delayed Surgical Resection on Short-Term and Long-Term Outcomes in Clinical Stage I Non-Small Cell Lung Cancer. *Ann Thorac Surg* **99**, 1906–1913 (2015).

5. Yang, C.-F. J. *et al.* Impact of Timing of Lobectomy on Survival for Clinical Stage IA Lung Squamous Cell Carcinoma. *Chest* **152**, 1239–1250 (2017).

6. Group, N. M. C. Preoperative chemotherapy for non-small-cell lung cancer: a systematic review and meta-analysis of individual participant data. *Lancet* **383**, 1561–1571 (2014).

7. Scagliotti, G. V. *et al.* Randomized Phase III Study of Surgery Alone or Surgery Plus Preoperative Cisplatin and Gemcitabine in Stages IB to IIIA Non–Small-Cell Lung Cancer. *J Clin Oncol* **30**, 172–178 (2011).

8. Brahmer, J. *et al.* Nivolumab versus Docetaxel in Advanced Squamous-Cell Non-Small-Cell Lung Cancer. *New Engl J Medicine* **373**, (2015).

9. Garon, E. B. *et al.* Pembrolizumab for the treatment of non-small-cell lung cancer. *New Engl J Medicine* **372**, 2018 2028 (2015).

10. Antonia, S. J. *et al.* Durvalumab after Chemoradiotherapy in Stage III Non-Small-Cell Lung Cancer. *New Engl J Medicine* **377**, 1919 1929 (2017).

11. Moreno, B. H., Parisi, G., Robert, L. & Ribas, A. Anti-PD-1 Therapy in Melanoma. *Semin Oncol* **42**, 466 473 (2015).

12. Moreno, B. H. & Ribas, A. Anti-programmed cell death protein-1/ligand-1 therapy in different cancers. *Brit J Cancer* **112**, 1421 1427 (2015).

13. Havel, J. J., Chowell, D. & Chan, T. A. The evolving landscape of biomarkers for checkpoint inhibitor immunotherapy. *Nat Rev Cancer* **19**, 133–150 (2019).

14. Teng, M. W. L., Ngiow, S. F., Ribas, A. & Smyth, M. J. Classifying Cancers Based on T-cell Infiltration and PD-L1. *Cancer Res* **75**, 2139 2145 (2015).

15. Yatim, N., Cullen, S. & Albert, M. L. Dying cells actively regulate adaptive immune responses. *Nat Rev Immunol* **17**, 262 275 (2017).

16. Florea, A.-M. & Büsselberg, D. Cisplatin as an Anti-Tumor Drug: Cellular Mechanisms of Activity, Drug Resistance and Induced Side Effects. *Cancers* **3**, 1351 1371 (2011).

17. Biasi, A. R. de, Villena-Vargas, J. & Adusumilli, P. S. Cisplatin-induced antitumor immunomodulation: a review of preclinical and clinical evidence. *Clin Cancer Res* **20**, 5384 5391 (2014).

18. Merritt, R. E., Mahtabifard, A., Yamada, R. E., Crystal, R. G. & Korst, R. J. Cisplatin augments cytotoxic T-lymphocyte–mediated antitumor immunity in poorly immunogenic murine lung cancer. *J Thorac Cardiovasc Surg* **126**, 1609–1617 (2003).

19. Fournel, L. *et al.* Cisplatin increases PD-L1 expression and optimizes immune check-point blockade in non-small cell lung cancer. *Cancer Lett* (2019) doi:10.1016/j.canlet.2019.08.005.

20. Qin, X., Liu, C., Zhou, Y. & Wang, G. Cisplatin induces programmed death-1-ligand 1(PD-L1) over-expression in hepatoma H22 cells via Erk /MAPK signaling pathway. *Cellular and molecular biology (Noisy-le-Grand, France)* **56 Suppl**, OL1366 72 (2010).

21. Temel, J. S. *et al.* Early palliative care for patients with metastatic non-small-cell lung cancer. *New Engl J Medicine* **363**, 733 742 (2010).

22. Y, H., K, O., I, O. & O, T. Immunotherapy for lung cancer cases using BCG or BCG cell-wall skeleton: intratumoral injections. *Gann Monogr Cancer Res* **21**, 151–160 (1978).

23. Matthay, R. A. *et al.* Intratumoral Bacillus Calmette-Guérin immunotherapy prior to surgery for carcinoma of the lung: results of a prospective randomized trial. *Cancer Res* **46**, 5963–8 (1986).

24. Celikoglu, F., Celikoglu, S. I., York, A. M. & Goldberg, E. P. Intratumoral administration of cisplatin through a bronchoscope followed by irradiation for treatment of inoperable non-small cell obstructive lung cancer. *Lung Cancer* **51**, 225–236 (2006).

25. Celıkoglu, S. I., Celıkoglu, F. & Goldberg, E. P. Endobronchial intratumoral chemotherapy (EITC) followed by surgery in early non-small cell lung cancer with polypoid growth causing erroneous impression of advanced disease. *Lung Cancer* **54**, 339–346 (2006).

26. Zarogoulidis, P. *et al.* Intratumoral chemotherapy for lung cancer: re-challenge current targeted therapies. *Drug Des Dev Ther* **Volume 7**, 571–583 (2013).

27. Mehta, H. J. *et al.* Treatment of isolated mediastinal and hilar recurrence of lung cancer with bronchoscopic endobronchial ultrasound guided intratumoral injection of chemotherapy with cisplatin. *Lung Cancer* **90**, 542 547 (2015).

28. Jabbardarjani, H., Kharabian, S. & Masjedi, M. R. Endobronchial Chemotherapy in Malignant Airway Lesions of the Lung. *J Bronchol* **14**, 242–245 (2007).

29. Mori, V., Roy, G. S., Bates, J. H. T. & Kinsey, C. M. Cisplatin Pharmacodynamics Following Endobronchial Ultrasound-Guided Transbronchial Needle Injection into Lung Tumors. *Sci Rep-uk* **9**, 6819 (2019).

30. Eisenhauer, E. A. *et al.* New response evaluation criteria in solid tumours: Revised RECIST guideline (version 1.1). *Eur J Cancer* **45**, 228–247 (2009).

31. Tourneau, C. L., Lee, J. J. & Siu, L. L. Dose Escalation Methods in Phase I Cancer Clinical Trials. *Jnci J National Cancer Inst* **101**, 708–720 (2009).