Oregon Health & Science University
OHSU Knight Cancer Institute
VA Portland Health Care System
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TITLE: **Phase II study of neoadjuvant pembrolizumab plus intensive androgen axis blockade prior to prostatectomy for high risk localized prostate cancer**

Coordinating Center: Portland VA Research Foundation

Principal Investigator: Mark Garzotto, MD
VA Portland Health Care System, Portland, OR
(503) 220-8262 x 51982
garzotto@ohsu.edu

Clinical Co-Investigators:
Tomasz Beer, MD
Medical Oncology, OHSU, VAPORHCS

Joshi Alumkal, MD
Medical Oncology, OHSU

Julie Graff, MD
Medical Oncology, OHSU, VAPORHCS

Ryan Kopp, MD
Urologic Oncology, OHSU, VAPORHCS

Correlative Science Investigator:
George Thomas, MD
Urologic Pathology, OHSU

Statistician: Motomi Mori, PhD, MBA
Director, Biostatistics Shared Resource, OHSU

Study Coordinator: Wesley Stoller, MA
OHSU, VAPORHCS
(503) 220-8262 x 54868
stoller@ohsu.edu

Supplied Agents:
Pembroluzimab (Keytruda®), Merck
Enzalutamide (Xtandi®), Medivation

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1.0 TRIAL SUMMARY

Abbreviated Title	**Neoadjuvant Pembrolizumab Plus Intensive Androgen Axis Blockade Prior to Prostatectomy**
Trial Phase	Phase II
Clinical Indication	High-risk localized Prostate Cancer
Trial Type	Open Label, single arm
Type of control	Historic controls
Route of administration	Intravenous Pembrolizumab, oral enzalutamide, subcutaneous (SQ) or intramuscular (IM) GNRH agonist
Trial Blinding	none
Treatment Groups	Single arm
Number of trial subjects	32
Estimated enrollment period	*12 months*
Estimated duration of trial*	*1.5 years*
Duration of Participation**	5.5 years
Estimated average length of treatment per patient	4 months

*Duration of trial period includes enrollment and treatment period for all patients.
** Duration of participation as the treatment period plus 5 years of follow-up

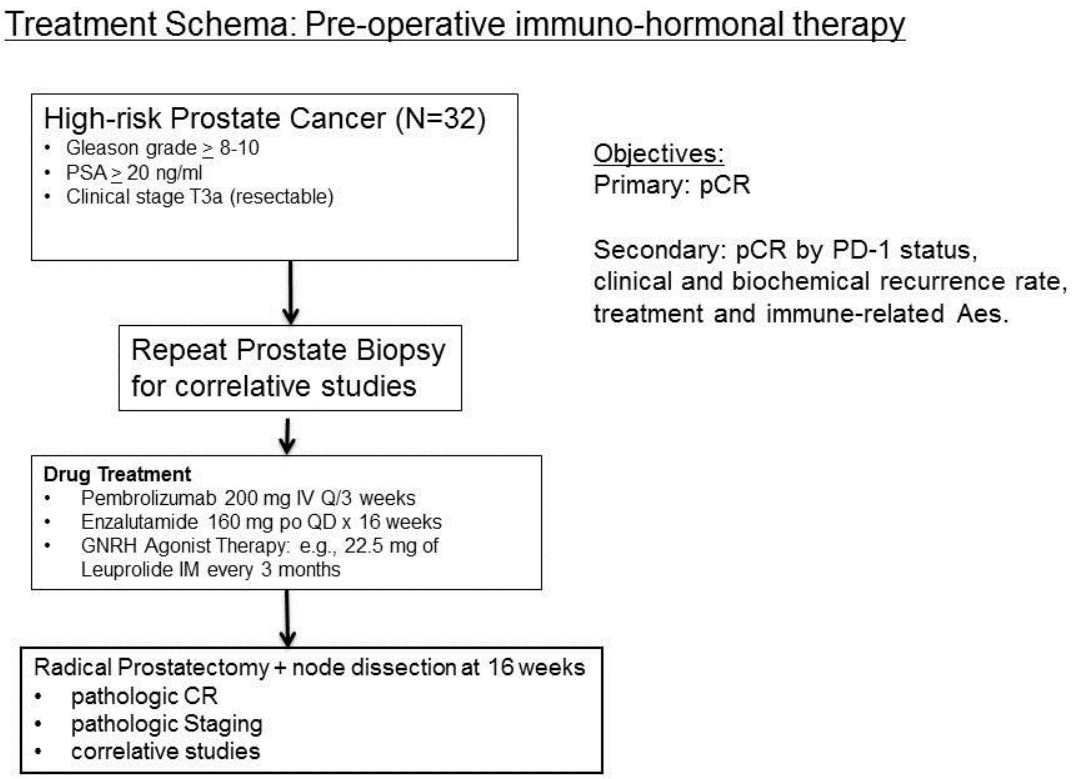
1.1 OBJECTIVES

Trial Concept: Intensive androgen receptor (AR) targeting has effects on prostate cancer in terms of lowering tumor androgen levels and inducing tissue responses, but fails to achieve frequent pathologic complete responses as monotherapy. Agents which incorporate targeting alternative mechanisms of resistance are needed to achieve biologic responses that translate into meaningful and durable clinical benefits. We propose to study the effects of pembrolizumab combined with intensive AR targeting prior to prostatectomy with extended pelvic lymph node dissection in subjects with high-risk localized prostate cancer (HRLPC). A strength of the neoadjuvant model is that biomarkers can be assessed by obtaining rigorously collected tissue specimens from the pre-and post-treatment settings. This will allow for the most valid comparison of drug effectiveness. The successful demonstration of efficacy and safety with this drug combination would promote widespread interest in carrying out trials of this regimen with surgery and radiation for intermediate and high-risk prostate cancers. If successful, these trials could in turn lead to changing the standard of care for the treatment of locally advanced prostate cancer.

2.0 TRIAL DESIGN

2.1 Trial Design- single arm, single-stage open label Phase II of neoadjuvant immune-hormonal therapy in high-risk Localized prostate cancer.

2.2 Trial Diagram



3.0 STUDY HYPOTHESES & OBJECTIVES

3.1 Objectives

To evaluate efficacy and safety of pembrolizumab combined with intensive AR targeting prior to prostatectomy in subjects with high-risk localized prostate cancer (HRLPC).

3.2 Hypotheses

(1) Treatment with the pembrolizumab and intensive AR axis inhibition will result in an increased rate of pathologic complete responses (pCR) for high-risk localized prostate cancer. (2) Neoadjuvant pembrolizumab combined with intensive AR axis inhibition will result in long-term disease control of high-risk localized prostate cancer.

3.3 Primary Endpoint

Pathologic complete response (pCR) defined as absence of detectable malignant cells in the

prostatectomy specimen evaluated by standard histologic techniques.

3.4 Secondary Endpoints

1. Grade 3 or higher treatment related adverse events;
2. Grade 2 or higher Immune-related adverse events (irAEs).
3. pCR in patients stratified by pre-operative prostate infiltrating T-cell PD-1 expression defined as none/low, medium, high (See section 7.3 pg 41)
4. Biochemical complete response rate prior to surgery (i.e. PSA< 0.1 ng/mL).
5. Incidence of surgical complications within 30 days post-surgery using Clavien-Dindo Classification of Surgical Complications.

3.5 Exploratory Endpoints:

1. Five-year incidence of cancer directed treatment.
2. Changes from pre- to post-treatment serum, blood and tissue markers of the immune response.
3. Correlation of changes in markers of inflammatory response with clinical outcomes including pCR, PSA response and disease free survival.
4. Clincal or Biochemical recurrence-free survival at five years (i.e. PSA< 0.1 ng/mL).
5. Changes in health related quality of life (HRQOL) measured by EPIC quality of life) from Week 0 vs. 16.
6. Safety and tolerability of Pembrolizumab, defined as Pembrolizumab related adverse event of any grade and drug dose modification

Type	**Endpoint**	**Definition**
Primary	Pathologic Complete Response (pCR)	No cancer detected on path exam of prostatectomy specimen
Secondary	Grade 3+ Treatment Related Adverse Events (AE)	Grade 3 or higher AE possibly related or related to any of three treatment drugs (pembrolizumab, enzalutamide, GNRH agonist) per CTCAE v4.03, evaluated up until 30 days after prostatectomy
Secondary	Grade 2+ Immune –related AE	Grade 2 or higher immune related AE possibly related or related to any of three treatment drugs (pembrolizumab, enzalutamide, GNRH agonist) per CTCAE v4.03, evaluated up until 30 days after prostatectomy
Secondary	Biochemical Complete Response	PSA < 0.1 ng/mL prior to prostectomy
Secondary	Incidence of Surgical Complications	Clavien-Dindo Classification of Surgical Complications compared to historical department incidence rates.
Exploratory	Safety and tolerability of Pembrolizumab	All AE's that are related or possibily related to Pembrolizumab, drug

		compliance (pharmacy record) and any instance of dose modification.
Exploratory	Five-year incidence of cancer directed therapy	Time from completion of prostatectomy till initiation of new prostate cancer-directed therapy during five years. Those who are alive and free of cancer directed therapy are censored at the date of last contact, and those who die without additional cancer directed therapy are censored at the date of death.
Exploratory	Compare pre- and post-treatment markers of inflammatory response in blood and tissue	Compare baseline to post-treatment markers measured just prior to prostatectomy (see Section 7.3 for a list of biomarkers to be studied).
Exploratory	5-year disease free survival	Period of time from completion of prostatectomy till when patient has a cancer recurrence defined both in terms of clinical or biochemical (PSA) or both.
Exploratory	Change in HRQOL	Intra-patient change in EPIC HRQOL from baseline to 16 weeks prior to surgery.

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4.0 BACKGROUND & RATIONALE

4.1 Background

Use of androgen deprivation is considered standard therapy for advanced prostate cancer and has been intensively studied in the locally advanced prostate cancer setting. In this trial, pembrolizumab will be added to two FDA-approved agents for the treatment of prostate cancer (i.e. enzalutamide and GNRH agonists).

Pembrolizumab is a human, monoclonal antibody directed against the programmed death receptor (PD-1) T-cell surface marker. It is currently indicated for the treatment of advanced melanoma and metastatic non-small cell lung cancer. There are several ongoing studies of this drug in prostate cancer, but no data are available for locally advanced prostate cancer. The goal will be to combine this agent with drugs that target the androgen-receptor axis in a HRLPC cohort that has elected surgery as primary treatment.

Enzalutamide is a potent AR inhibitor that is indicated for the treatment of metastatic

castration-resistant prostate cancer (See Package insert). For this study, this drug will be utilized in an earlier disease state than what it is currently FDA-indicated for.

GNRH agonist therapy (ie leuprolide and goserelin) is a reversible luteinizing hormone receptor agonist which blocks the production of luteinizing hormone and subsequently testosterone production. It is indicated for advanced prostate cancer (See package inserts). The study population in this trial would qualify for this treatment under this general category.

Since both enzalutamide and GNRH agonists are indicated for prostate cancer therapy, there is already a well-established rationale for use of these agents in prostate cancer. The following section will focus on pembrolizumab which has only been studied in a limited fashion in prostate cancer.

4.1.1 Pembrolizumab Pharmaceutical and Therapeutic Background

The importance of intact immune surveillance in controlling outgrowth of neoplastic transformation has been known for decades. Accumulating evidence shows a correlation between tumor-infiltrating lymphocytes (TILs) in cancer tissue and favorable prognosis in various malignancies. In particular, the presence of CD8+ T-cells and the ratio of CD8+ effector T-cells / FoxP3+ regulatory T-cells 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). The structure of murine PD-1 has been resolved. 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. 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. 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. Expression has also been shown during thymic development on CD4-CD8- (double negative) T-cells as well as subsets of macrophages and dendritic cells. The ligands for PD-1 (PD-L1 and PD-L2) are constitutively expressed or can be induced in a variety of cell types, including non-hematopoietic tissues as well as in various tumors . 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. 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). 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 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. Keytruda™ (pembrolizumab) has recently been approved in the United Stated for the treatment of patients with unresectable or metastatic melanoma and disease progression following ipilumumab and, if BRAF V600 mutation positive, a BRAF inhibitor AND platinum-refractory non-small cell lung cancer that expresses PD-L1.

4.1.2 Preclinical and Clinical Trial Data

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

4.2 Rationale

4.2.1 Rationale for the Trial and Selected Subject Population

High-risk Localized Prostate Cancer: an area of unmet need: Technical advances in surgery and radiation therapy have resulted in improved prostate cancer (PC) outcomes; however, disease recurrence and PC-mortality remain a high, particularly for those with HRLPC.[1] Features predictive of adverse outcomes include Gleason grade and disease extent. In a randomized trial of prostatectomy vs. observation, PC-specific mortality was 5-fold higher for high-grade versus low-grade cancers and 14-fold higher for extracapsular disease vs. organ-confined disease.[2] In a study by Stephenson et al., PC-mortality was 34% at 15 years in HRLPC.[3] Novel therapies that improve both systemic and local tumor control are urgently needed in order to reduce the burden of disease in PC.

Androgen deprivation alone is inadequate in HRLPC: AR signaling is a primary means of tumor cell resistance in PC (Chen and Sawyers 2004). Targeting of the AR axis has been established to improve the efficacy of both radiation[4] and chemotherapy[5] in PC. The potential of intensive AR inhibition has been recently demonstrated in a series of pre-prostatectomy studies. For patients with intermediate- and high-risk PC, treatment with the androgen biosynthesis inhibitor abiraterone acetate and LHRH agonist resulted in a 7% pathologic complete response rate.[6] Similarly, in a study using a four drug anti-AR combination, 1 out of 13 patients with primarily intermediate-risk disease achieved a pathologic complete response .[7] The addition of and androgen biosynthesis inhibitor to and anti-androgen has recently been evaluated in the pre-operative setting. In a pre-operative study of abiraterone with enzalutamide there were no pathologic CRs in 44 patients.

Importantly, there have been no adverse safety signals for any of these agents in the pre-operative setting (Efstathiou ASCO 2016). Novel combinations that work in synergy with AR targeting are needed to maximize the potential of these newer agents in use for PC.

PD-1 T-cell upregulation as a mechanism for cancer cell progression: Programmed death receptor-1 (PD-1), is a CD8+ T-cell receptor, which is suppressed as a normal part of T-cell regulation and prevention of autoimmunity.[8] PD-1 interacts with its ligands PD-L1 and PD-L2 or other peptides from the tumor microenvironment, resulting in the blockade of CD8+ T-cell activation and proliferation.[9] Tumor cells stimulate the expression of T-cell PD-1, thereby inhibiting an effective immune response.[10] Pembrolizumab is a fully humanized monoclonal antibody, which binds to the PD-1 receptor, thus blocking the binding of potential ligands from tumor cells or other sources. This agent is FDA-approved for second-line treatment of melanoma and non-small cell lung cancer, but has not been thoroughly investigated in PC. With the support of Merck this agent is presently being studied at OHSU (PI: J Graff) for metastatic castration-resistant prostate cancer (CRPC) patients (NCT02312557).

PD-1 as a target for prostate cancer: Prior studies investigating the potential for immune-based therapies in PC have shown that PD-1 is a promising target. Prostate-infiltrating CD8+ T cells highly express PD-1 in PC specimens but rarely in the non-cancerous prostate gland.[11] Up to 90% of PC infiltrating T-cells highly express the PD-1 receptor, which is consistent with a non-functional "exhausted" phenotype.[10] The investigators concluded that high PD-1 expression renders the "T-cells incapable of mounting an effective anti-tumor immune response" possibly due to production of an inhibitory peptide derived from the H4 histone.[12] Furthermore, in patients with high Gleason scores (≥7), the effect appears to be systemic as peripheral blood T-cells also highly express PD-1 relative to controls. Thus, inhibition of PD-1 could result in the potent anti-neoplastic effects on both local and systemic PC.

Pembrolizumab in combination with enzalutamide -OHSU ongoing study: In an ongoing Phase II single arm study of men with metastatic CRPC, Graff et al. have shown promising signs of anti-PD1efficacy in this population.[13] Three of 10 men treated had PSA reduction to <0.2 ng/mL and 2 of 10 showed partial responses. Adverse events included grade 2 myositis (n=1),grade 3 hypothyroidism (n=1), and one had grade 2 hypothyroidism (n=1). The study is planned for an additional 18 patients to be accrued (total = 28).

4.2.2 Rationale for Pembrolizumab Dose Selection/Regimen/Modification

An open-label Phase I trial (Protocol 001) is being conducted to evaluate the safety and clinical activity of single agent MK-3475. 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 MK-3475 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. 10.0 mg/kg Q2W, the highest dose tested in PN001, will be the dose and schedule utilized in Cohorts A, B, C and

D of this protocol to test for initial tumor activity. Recent data from other clinical studies within the MK-3475 program has shown that a lower dose of MK-3475 and a less frequent schedule may be sufficient for target engagement and clinical activity.

PK data analysis of MK-3475 administered Q2W and Q3W showed slow systemic clearance, limited volume of distribution, and a long half-life (refer to IB). Pharmacodynamic data (IL-2 release assay) suggested that peripheral target engagement is durable (>21 days). This early PK and pharmacodynamic data provides scientific rationale for testing a Q2W and Q3W dosing schedule.

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 MK-3475 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. MK-3475 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 MK-3475 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.

The rationale for further exploration of 2 mg/kg and comparable doses of pembrolizumab in solid tumors is based on: 1) similar efficacy and safety of pembrolizumab when dosed at either 2 mg/kg or 10 mg/kg Q3W in melanoma patients, 2) the flat exposure-response relationships of pembrolizumab for both efficacy and safety in the dose ranges of 2 mg/kg Q3W to 10 mg/kg Q3W, 3) the lack of effect of tumor burden or indication on distribution behavior of pembrolizumab (as assessed by the population PK model) and 4) the assumption that the dynamics of pembrolizumab target engagement will not vary meaningfully with tumor type.

The choice of the 200 mg Q3W as an appropriate dose for the switch to fixed dosing is based on simulations performed using the population PK model of pembrolizumab showing that the fixed dose of 200 mg every 3 weeks will provide exposures that 1) are optimally consistent with those obtained with the 2 mg/kg dose every 3 weeks, 2) will maintain individual patient exposures in the exposure range established in melanoma as associated with maximal efficacy response and 3) will maintain individual patients exposure in the exposure range established in melanoma that are well tolerated and safe.

A fixed dose regimen will simplify the dosing regimen to be more convenient for physicians and to reduce potential for dosing errors. A fixed dosing scheme will also reduce complexity in the logistical chain at treatment facilities and reduce wastage.

Rationale for combination of pembrolizumab with anti-androgen therapy: Targeted anti-androgen therapy can have dramatic effects on the host tumor; however these agents usually result in relatively short term gains of several months in terms of overall survival.[14] At the cellular level these agents may induce cell death and cause the release of highly specific tumor antigens (a process known as "cross-priming").[15] Under the right conditions (such as with anti-PD-1 or PD-L1 therapy) these antigens could be taken up by antigen presenting cells (APCs) which could initiate an effective immune response. Thus, there is a significant potential for synergistic effects between immunologic and anti-androgenic agents in prostate cancer. This strategy holds the potential for long-term tumor remission, a phenomena that has been observed in several tumors including melanoma and non-small cell lung cancer. Currently, numerous trials looking at combinations of novel immunologic therapies in combination with established therapies such as radiation, chemotherapy and small molecule inhibitors are underway.[16] However, to our knowledge, this approach is not being studied in high-risk localized prostate cancer.

4.2.3 Rationale for Endpoints

4.2.3.1 Efficacy Endpoints: The primary efficacy endpoint is the elimination of all tumor cells as observed on standard pathologic sampling in a prostatectomy specimen. Pathologic CR (pCR) is a validated endpoint in a number of solid tumor types. In breast cancer, pCR correlates well with overall survival (OS). Compared to radiographic changes, Funt and Chapman have proposed pCR as a "more biologically rational method of determining treatment effect that may be more likely to predict OS" as compared to radiographic changes.[17] pCR has been considered an endpoint worthy of pursuit in high-risk localized prostate cancer where the rate of pCR is relatively low with current treatment regimens. Thus any significant improvement over the current baseline rate would be of great interest to the research community. This endpoint will give a relatively quick readout for efficacy compared to long-term clinical endpoints which will require several years before it can be attained and may require trial randomization to make valid comparisons. Clinical and biochemical recurrence free survival and time to secondary treatment will also be studied as secondary endpoints. These will be compared to historic controls using established data from clinical nomograms (ie MSKCC).

4.2.3.2 Safety Endpoints: The safety of this combination will be assessed for any unforeseen toxicity in combination or when used in the pre-operative setting. The hormonal agents have previously been given in combination prior to surgery and demonstrate no safety concerns. There are no known overlapping toxicities between so the expectation is that the regimen will be considered safe. Pembrolizumab plus enzalutamide is currently under study in patients with more advanced disease, and there are no unforeseen safety issues to date (J. Graff: personal communication). Further, we will assess the changes in HRQOL before, during and post-treatment using EPIC. We will assess surgical measures of safety and record intraoperative and post-operative metrics using standard case report forms as done previously by our group, the results of which are shown in table 7.[18]

4.2.3.3 Translational Endpoints: Tissues will be examined from biopsy specimens and the levels of PD-1 expression will be assess by independent laboratory assessment by

immunohistochemistry (IHC). PD-1 tissue levels will be done by independent assessment by Merck, who will be blinded to all clinical data. These levels will be correlated with treatment response (pCR: yes vs. no). Additional exploratory markers will be studied for treatment effects in both tissue and serum (see section 7.3). Dr. George Thomas (urologic pathologist, OHSU) will carry out remaining tissue assessments for IHC. Serum samples will be evaluated for pre- and post-treatment changes to evaluate the effect of treatment on serum markers of the immune response (See section 7.3). These analyses will be used to explore possible means of tumor resistance and immune tolerance.

5.0 METHODOLOGY

5.1 Entry Criteria

5.1.1 Diagnosis/Condition for Entry into the Trial

PATIENT POPULATION
Potentially eligible patients will be identified by any of the study investigators in multi-disciplinary clinics at either OHSU or the Portland VAMC. All patients will be required to have at least one high-risk criteria (see inclusion criteria). For each patient the number of high-risk features will be recorded.

5.1.2 Subject Inclusion Criteria

- Be willing and able to provide written informed consent/assent for the trial.
- Capability to understand and comply with the protocol and signed informed consent document.
- Be ≥ 18 years of age on day of signing informed consent.
- Have measurable disease based on RECIST 1.1.
- Be willing to provide tissue from a newly obtained core or excisional biopsy of a tumor lesion. *Newly-obtained is defined as a specimen obtained up to 6 weeks (42 days) prior to initiation of treatment on Day 1. Subjects for whom newly-obtained samples cannot be provided (e.g. inaccessible or subject safety concern) may submit an archived specimen only upon agreement from the Sponsor.*
- Histologically confirmed, non-metastatic adenocarcinoma of the prostate
- Prostatectomy with extended lymph node dissection planned as primary therapy
- 10 year or longer life expectancy based on other co-morbidities
- Have a performance status of 0 or 1 on the ECOG Performance Scale.
- Any one of the following three high risk features:
    - Gleason grade ≥ 8-10
    - PSA ≥ 20 ng/ml
    - Clinical stage T3a (resectable)
- No evidence of lymph nodes ≥ 2 cm in diameter on pelvic CT scan (scan only required in patients with a PSA ≥ 20 ng/ml)
- No evidence of metastases on nuclear bone scan (required on all pts)

- Tumor tissue must be provided for subsequent biomarker analyses. (PD-1 classification not required for study enrollment) A subject must have a PD-1 expression classification (positive, negative, or indeterminate) as determined by the central lab. If insufficient tumor tissue content is provided for analysis, acquisition of additional archived tumor tissue (block and /or slides) for the biomarker analysis is required.
- No other diagnosis of malignancy (with exception of non-melanoma skin cancer or a malignancy diagnosed ≥5 years ago).
- Male subjects of childbearing potential (Section 5.6.2) must agree to use an adequate method of contraception as outlined in Section 5.6.2- Contraception, starting with the first dose of study therapy through the time of surgery. Note: Abstinence is acceptable if this is the usual lifestyle and preferred contraception for the subject.
- Demonstrate adequate organ function as defined in **Error! Not a valid bookmark self-reference.**, all screening labs should be performed within 30 days of treatment initiation.

Table 1 Adequate Organ Function Laboratory Values

System	Laboratory Value
Hematological	
Absolute neutrophil count (ANC)	≥1,500 /mcL
Platelets	≥100,000 / mcL
Hemoglobin	≥9 g/dL or ≥5.6 mmol/L without transfusion or EPO dependency (within 7 days of assessment)
Renal	
Serum creatinine **OR** Measured or calculated[a] creatinine clearance (GFR can also be used in place of creatinine or CrCl)	≤1.5 X upper limit of normal (ULN) **OR** ≥60 mL/min for subject with creatinine levels > 1.5 X institutional ULN
Hepatic	
Serum total bilirubin	≤ 1.5 X ULN **OR**
	Direct bilirubin ≤ ULN for subjects with total bilirubin levels > 1.5 ULN
AST (SGOT) and ALT (SGPT)	≤ 2.5 X ULN **OR** ≤ 5 X ULN for subjects with liver metastases
Albumin	≥2.5 mg/dL
Coagulation	
International Normalized Ratio (INR) or Prothrombin Time (PT) Activated Partial Thromboplastin Time (aPTT)	≤1.5 X ULN unless subject is receiving anticoagulant therapy as long as PT or PTT is within therapeutic range of intended use of anticoagulants ≤1.5 X ULN unless subject is receiving anticoagulant therapy as long as PT or PTT is within therapeutic range

	of intended use of anticoagulants
[a]Creatinine clearance should be calculated per institutional standard.	

5.1.3 Exclusion Criteria

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

- Has significant active medical illness, which in the opinion of the investigator would preclude protocol treatment (e.g. uncontrolled pulmonary, renal, or hepatic dysfunction, uncontrolled infection, cardiac disease).
- 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 treating investigator.
- Has known active Hepatitis B (e.g., HBsAg reactive) or Hepatitis C (e.g., HCV RNA [qualitative] is detected).
- Has a known history of Human Immunodeficiency Virus (HIV) (HIV 1/2 antibodies).
- History of prolonged QT interval or congestive heart failure.
- Prior history of seizures
- Has a history of (non-infectious) pneumonitis that required steroids or current pneumonitis."
- Has known psychiatric or substance abuse disorders that would interfere with cooperation with the requirements of the trial.
- Is currently participating and receiving study therapy or has participated in a study of an investigational agent and received study therapy or used an investigational device within 4 weeks of the first dose of treatment.
- Has had a prior anti-cancer monoclonal antibody (mAb) within 4 weeks prior to study Day 1 or who has not recovered (i.e., ≤ Grade 1 or at baseline) from adverse events due to agents administered more than 4 weeks earlier.
- Has had prior chemotherapy, targeted small molecule therapy, or radiation therapy within 2 weeks prior to study Day 1 or who has not recovered (i.e., ≤ Grade 1 or at baseline) from adverse events due to a previously administered agent.
    - Note: Subjects with ≤ Grade 2 neuropathy are an exception to this criterion and may qualify for the study.
    - Note: If subject received major surgery, they must have recovered adequately from the toxicity and/or complications from the intervention prior to starting therapy.
- Has any other prior therapy for prostate cancer including:
    - Anti-androgen therapy including orchiectomy, LHRH therapy, anti-androgen therapy, abiraterone, ketoconazole, estrogen therapy (Short-term ADT allowed, ≤ 2 months)
    - Radiation (external beam or brachytherapy)
    - Cytotoxic chemotherapy
    - Any experimental therapy for treatment of prostate cancer.

- Received major surgery, they must have recovered adequately from the toxicity and/or complications from the intervention prior to starting therapy.
- Has a known additional malignancy that is progressing or requires active treatment. Exceptions include basal cell carcinoma of the skin or squamous cell carcinoma of the skin that has undergone potentially curative therapy.
- Has known active central nervous system (CNS) metastases and/or carcinomatous meningitis.
- Has a history of or known presence of extensive, disseminated/bilateral or Grade 3 or 4 interstitial fibrosis or interstitial lung disease, including a history of pneumonitis, hypersensitivity pneumonitis, interstitial pneumonia, interstitial lung disease, obliterative bronchiolitis, or pulmonary fibrosis, but not including a history of prior radiation pneumonitis.
- Has a known history of active TB (Bacillus Tuberculosis)
- Has active autoimmune disease that has required systemic treatment in the past 2 years (i.e. with use of disease modifying agents, corticosteroids or immunosuppressive drugs). Replacement therapy (eg., thyroxine, insulin, or physiologic corticosteroid replacement therapy for adrenal or pituitary insufficiency, etc.) is not considered a form of systemic treatment.
- Has a diagnosis of immunodeficiency or is receiving systemic steroid therapy or any other form of immunosuppressive therapy within 7 days prior to the first dose of trial treatment.
- Has an active infection requiring systemic therapy.
- Used systemic corticosteroids within 21 days. Inhaled or topical steroids allowed.
- Has a hypersensitivity to pembrolizumab, Enzalutamide, specific GNRH agonists or any of their excipients.
- Has a gastrointestinal disorder affecting absorption
- Subjects who received prior therapy with interferon, anti-PD-1, anti-PD-L1, anti-PD-L2, anti-CD137, or anti-CTLA-4 antibody
- Is currently using warfarin
- Is using strong CYP2C8 inhibitors (e.g gemfibrozil)
- Is using strong CYP3A4 inhibitors or inducers (e.g., itraconazole, carbamazepine, phenobarbital, phenytoin, rifabutin, rifampin, rifapentine)
- Has a history of allergies (Grade 3 or above) to humanized antibodies.
- Has received a live vaccine within 30 days of planned start of study therapy.
  Note: Seasonal influenza vaccines for injection are generally inactivated flu vaccines and are allowed; however intranasal influenza vaccines (e.g., Flu-Mist®) are live attenuated vaccines, and are not allowed.
- 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.
- Is unable or unwilling to abide by the study protocol or cooperate fully with the investigator or designee
- Is unable to undergo research related pre-treatment biopsy are excluded as this is necessary to complete translational study objectives.

5.2 Trial Treatments

The treatment to be used in this trial is outlined below in Table 2. In this study, pembrolizumab will be added to two FDA-approved drugs routinely used for the treatment of prostate cancer. Surgery will be performed approximately between week 14 and 16 which will allow for a 2-4 week washout for pembrolizumab. Other study medication will continue up until the time of surgery.

Table 2: Study drugs under investigation

Drug	Dose	Dose Frequency	Route of Administration	Regimen/Treatment Period	Use
Pembrolizumab	200 mg	Q3Weeks	IV infusion	Day 1 of each 3 week cycle	Experimental
Enzalutamide	160 mg	Daily	Oral	16 weeks	Experimental
GNRH agonist therapy (ie leuprolide and goserelin)*					
* GNRH Agonist will be given as approved for androgen deprivation at a dose necessary to maintain castrate levels and equivalent to 22.5 mg of Leuprolide IM every 3 months (e.g., leuprolide 45 mg IM every 6 months is equivalent to leuprolide 22.5 mg IM every 3 months).					

5.2.1 Dose Selection/Modifications: Pembroluzimab (Keytruda®)

5.2.1.1 Dose Selection:

The rationale for selection of doses to be used in this trial is provided in Section 4.0 – Background and Rationale. Details on preparation and administration of pembrolizumab (MK-3475) are provided in the Pharmacy Manual.

5.2.1.2 Dose Modification (Escalation/Titration/Other)

Adverse events (both non-serious and serious) associated with pembrolizumab exposure may represent an immunologic etiology. These adverse events may occur shortly after the first dose or several months after the last dose of treatment. Pembrolizumab must be withheld for drug-related toxicities and severe or life-threatening AEs as per 3 below. See Section 5.5.1 for supportive care guidelines, including use of corticosteroids.

Table 3: Dose Modification Guidelines for Drug-Related Adverse Events

Toxicity	Hold Treatment For Grade	Timing for Restarting Treatment	Treatment Discontinuation
Diarrhea/Colitis	2-3	Toxicity resolves to Grade 0-1	Toxicity does not resolve within 12 weeks of last dose or inability to reduce corticosteroid to 10 mg or less of prednisone or equivalent per day within 12 weeks
	4	Permanently discontinue	Permanently discontinue
AST, ALT, or Increased Bilirubin	2	Toxicity resolves to Grade 0-1	Toxicity does not resolve within 12 weeks of last dose
	3-4	Permanently discontinue (see exception below)[a]	Permanently discontinue
Type 1 diabetes mellitus (if new onset) or Hyperglycemia	T1DM or 3-4	Hold pembrolizumab for new onset Type 1 diabetes mellitus or Grade 3-4 hyperglycemia associated with evidence of beta cell failure	Resume pembrolizumab when patients are clinically and metabolically stable
Hypophysitis	2-4	Toxicity resolves to Grade 0-1. Therapy with pembrolizumab can be continued while endocrine replacement therapy is instituted	Toxicity does not resolve within 12 weeks of last dose or inability to reduce corticosteroid to 10 mg or less of prednisone or equivalent per day within 12 weeks
Hyperthyroidism	3	Toxicity resolves to Grade 0-1	Toxicity does not resolve within 12 weeks of last dose or inability to reduce corticosteroid to 10 mg or less of prednisone or equivalent per day within 12 weeks
	4	Permanently discontinue	Permanently discontinue
Hypothyroidism		Therapy with pembrolizumab can be continued while thyroid replacement therapy is instituted	Therapy with pembrolizumab can be continued while thyroid replacement therapy is instituted
Infusion Reaction	2[b]	Toxicity resolves to Grade 0-1	Permanently discontinue if toxicity develops despite adequate premedication
	3-4	Permanently discontinue	Permanently discontinue
Pneumonitis	2	Toxicity resolves to Grade 0-1	Toxicity does not resolve within 12 weeks of last dose or inability to reduce corticosteroid to 10 mg or less of prednisone or equivalent per day within 12 weeks
	3-4	Permanently discontinue	Permanently discontinue
Renal Failure or Nephritis	2	Toxicity resolves to Grade 0-1	Toxicity does not resolve within 12 weeks of last dose or inability to reduce corticosteroid to 10 mg or less of prednisone or equivalent per day within 12 weeks
	3-4	Permanently discontinue	Permanently discontinue
All Other Drug-Related Toxicity[c]	3 or Severe	Toxicity resolves to Grade 0-1	Toxicity does not resolve within 12 weeks of last dose or inability to reduce corticosteroid to 10 mg or less of prednisone or equivalent per day within 12 weeks
	4	Permanently discontinue	Permanently discontinue

Note: Permanently discontinue for any severe or Grade 3 drug-related AE that recurs or any life-threatening event.

[a] For patients with liver metastasis who begin treatment with Grade 2 AST or ALT, if AST or ALT increases by greater than or equal to 50% relative to baseline and lasts for at least 1 week then patients should be discontinued.

[b] If symptoms resolve within one 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 subject should be premedicated for the next scheduled dose; Refer to **Error! Reference source not found.** – Infusion Treatment Guidelines for further management details.

[c] Patients with intolerable or persistent Grade 2 drug-related AE may hold study medication at physician discretion. Permanently discontinue study drug for persistent Grade 2 adverse reactions for which treatment with study drug has been held, that do not recover to Grade 0-1 within 12 weeks of the last dose.

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 3 weeks of the scheduled interruption, unless otherwise discussed with the Sponsor. The reason for interruption should be documented in the patient's study record.

5.2.1.3 Timing of Dose Administration

Trial treatment should be administered on Day 1 of each cycle after all procedures/assessments have been completed as detailed on the Trial Flow Chart (Section 6.0). Trial treatment may be administered up to 3 days before or after the scheduled Day 1 of each cycle due to administrative reasons. All trial treatments will be administered on an outpatient basis.

Pembrolizumab 200 mg will be administered as a 30 minute IV infusion every 3 weeks. Sites should make every effort to target infusion timing to be as close to 30 minutes as possible through an intravenous line containing a sterile, non-pyrogenic, low-protein binding 0.2 micron to 5 micron in-line or add-on filter (See Package Insert). The drug will not co-administer other drugs through the same infusion line. The Pharmacy Manual contains specific instructions for the preparation of the pembrolizumab infusion fluid and administration of infusion solution.

5.2.2 Dose Selection/Modifications: Enzalutamide (Xtandi®)

5.2.2.1 Dose Selection: The dose is derived from the prior trials that lead to the approval of this agent. (See Package insert for details). This will be self-administered as 4 capsules daily for a total of 160 mg per day.

5.2.2.2 Dose Modification (Escalation/Titration/Other). There are no planned dose modifications. If a patient experiences toxicity thought to be attributed to the study drug that is Grade 3 or higher, then the medication will be withheld until resolution or stabilization of symptoms

5.2.2.3 Timing of Dose Administration
Enzalutamide will be self-administered orally on a daily basis from Day 1 of treatment until the day before surgery. The dose will be 160 mg and the drug will be distributed by the Research Pharmacist at each treatment center.

5.3 Trial Blinding/Masking
This is an open-label trial; therefore, the Sponsor, principal investigator and subject will be aware of the treatment administered.

5.4 Concomitant Medications/Vaccinations (allowed & prohibited)

Medications or vaccinations specifically prohibited in the exclusion criteria are not allowed during the ongoing trial. If there is a clinical indication for one of these or other medications

or vaccinations specifically prohibited during the trial, discontinuation from trial therapy or vaccination may be required. The investigator should discuss any questions regarding this with the Merck Clinical team. The final decision on any supportive therapy or vaccination rests with the investigator and/or the subject's primary physician.

5.4.1 Acceptable Concomitant Medications

All treatments that the investigator considers necessary for a subject's welfare may be administered at the discretion of the investigator in keeping with the community standards of medical care. All concomitant medication will be recorded on the case report form (CRF) including all prescription, over-the-counter (OTC), herbal supplements, and IV medications and fluids. If changes occur during the trial period, documentation of drug dosage, frequency, route, and date may also be included on the CRF.

All concomitant medications received within 28 days before the first dose of trial treatment and 30 days after the last dose of trial treatment should be recorded. Concomitant medications administered after 30 days after the last dose of trial treatment should be recorded for SAEs and Event of Clinical Interest ECIs as defined in Section 7.2.

5.4.2 Prohibited Concomitant Medications

Subjects are prohibited from receiving the following therapies during the Screening and Treatment Phase (including retreatment for post-complete response relapse) of this trial:

- Antineoplastic systemic chemotherapy or biological therapy
- Immunotherapy not specified in this protocol
- Chemotherapy not specified in this protocol
- Investigational agents other than pembrolizumab
- Radiation therapy
- Live vaccines within 30 days prior to the first dose of trial treatment and while participating in the trial. Examples of live vaccines include, but are not limited to, the following: measles, mumps, rubella, varicella/zoster, yellow fever, rabies, BCG, and typhoid vaccine.

- Systemic glucocorticoids for any purpose other than to modulate symptoms from an event of clinical interest of suspected immunologic etiology. The use of physiologic doses of corticosteroids may be approved after consultation with the Sponsor.

Subjects who, in the assessment by the 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 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.

5.5 Rescue Medications & Supportive Care for Pembrolizumab

5.5.1 Supportive Care Guidelines

Subjects should receive appropriate supportive care measures as deemed necessary by the treating investigator. Suggested supportive care measures for the management of adverse events with potential immunologic etiology are outlined below. Where appropriate, these guidelines include the use of oral or intravenous treatment with corticosteroids as well as additional anti-inflammatory agents if symptoms do not improve with administration of corticosteroids. Note that several courses of steroid tapering may be necessary as symptoms may worsen when the steroid dose is decreased. For each disorder, attempts should be made to rule out other causes such as metastatic disease or bacterial or viral infection, which might require additional supportive care. The treatment guidelines are intended to be applied when the investigator determines the events to be related to pembrolizumab.
Note: if after the evaluation the event is determined not to be related, the investigator does not need to follow the treatment guidance (as outlined below). Refer to Section 5.2.1 for dose modification.
It may be necessary to perform conditional procedures such as bronchoscopy, endoscopy, or skin photography as part of evaluation of the event.

- **Pneumonitis**:
    - For **Grade 2 events**, treat with systemic corticosteroids. When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks.
    - For **Grade 3-4 events**, immediately treat with intravenous steroids. Administer additional anti-inflammatory measures, as needed.
    - Add prophylactic antibiotics for opportunistic infections in the case of prolonged steroid administration.
- **Diarrhea/Colitis**:

  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).
    - All subjects who experience 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. For Grade 2 or higher diarrhea, consider GI consultation and endoscopy to confirm or rule out colitis.
    - For **Grade 2 diarrhea/colitis,** administer oral corticosteroids.
    - For **Grade 3 or 4 diarrhea/colitis,** treat with intravenous steroids followed by high dose oral steroids.
    - When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks.

- **Type 1 diabetes mellitus (if new onset, including diabetic ketoacidosis [DKA]) or ≥ Grade 3 Hyperglycemia, if associated with ketosis (ketonuria) or metabolic acidosis (DKA)**
    - For **T1DM** or **Grade 3-4** Hyperglycemia
        - Insulin replacement therapy is recommended for Type I diabetes mellitus and for Grade 3-4 hyperglycemia associated with metabolic acidosis or ketonuria.
        - Evaluate patients with serum glucose and a metabolic panel, urine ketones, glycosylated hemoglobin, and C-peptide.
- **Hypophysitis**:
    - For **Grade 2** events, treat with corticosteroids. When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks. Replacement of appropriate hormones may be required as the steroid dose is tapered.
    - For **Grade 3-4** events, treat with an initial dose of IV corticosteroids followed by oral corticosteroids. When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks. Replacement of appropriate hormones may be required as the steroid dose is tapered.
- **Hyperthyroidism or Hypothyroidism**:

  Thyroid disorders can occur at any time during treatment. Monitor patients for changes in thyroid function (at the start of treatment, periodically during treatment, and as indicated based on clinical evaluation) and for clinical signs and symptoms of thyroid disorders.
    - **Grade 2** hyperthyroidism events (and **Grade 2-4** hypothyroidism):
        - In hyperthyroidism, non-selective beta-blockers (e.g. propranolol) are suggested as initial therapy.
        - In hypothyroidism, thyroid hormone replacement therapy, with levothyroxine or liothyroinine, is indicated per standard of care.
    - **Grade 3-4** hyperthyroidism
        - Treat with an initial dose of IV corticosteroid followed by oral corticosteroids. When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks. Replacement of appropriate hormones may be required as the steroid dose is tapered.
- **Hepatic**:
    - For **Grade 2** events, monitor liver function tests more frequently until returned to baseline values (consider weekly).
        - Treat with IV or oral corticosteroids

- For **Grade 3-4** events, treat with intravenous corticosteroids for 24 to 48 hours.
- When symptoms improve to Grade 1 or less, a steroid taper should be started and continued over no less than 4 weeks.

- **Renal Failure or Nephritis**:
  - For **Grade 2** events, treat with corticosteroids.
  - For **Grade 3-4** events, treat with systemic corticosteroids.
  - When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks.
- **Management of Infusion Reactions**: Signs and symptoms usually develop during or shortly after drug infusion and generally resolve completely within 24 hours of completion of infusion.

  Error! Reference source not found. below shows treatment guidelines for subjects who experience an infusion reaction associated with administration of pembrolizumab (MK-3475).

Table 4 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 subject is deemed medically stable in the opinion of the investigator.	None
Grade 2 Requires infusion interruption but responds promptly to symptomatic treatment (e.g., antihistamines, NSAIDS, narcotics, IV fluids); prophylactic medications indicated for < =24 hrs	**Stop Infusion and monitor symptoms.** 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 subject is deemed medically stable in the opinion of the investigator. If symptoms resolve within one hour of stopping drug infusion, the infusion may be restarted at 50% of the original infusion rate	Subject may be premedicated 1.5h (± 30 minutes) prior to infusion of pembrolizumab (MK-3475) with: Diphenhydramine 50 mg po (or equivalent dose of antihistamine). Acetaminophen 500-1000 mg po (or equivalent dose of antipyretic).

NCI CTCAE Grade	Treatment	Premedication at subsequent dosing
	(e.g., from 100 mL/hr to 50 mL/hr). Otherwise dosing will be held until symptoms resolve and the subject should be premedicated for the next scheduled dose. **Subjects who develop Grade 2 toxicity despite adequate premedication should be permanently discontinued from further trial treatment administration.**	
<u>Grades 3 or 4</u> 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: IV fluids Antihistamines NSAIDS Acetaminophen Narcotics Oxygen Pressors Corticosteroids Epinephrine Increase monitoring of vital signs as medically indicated until the subject is deemed medically stable in the opinion of the investigator. Hospitalization may be indicated. **Subject is permanently discontinued from further trial treatment administration.**	No subsequent dosing
Appropriate resuscitation equipment should be available in the room and a physician readily available during the period of drug administration.		

5.5 Androgen Receptor Targeting Study Drugs

5.5.1 Enzalutamide (Xtandi®)

In this study, patients will receive daily oral doses of study drug (enzalutamide 160 mg/day), which will be administered as four capsules/day to be taken at or near the same time each day. Any missed doses will be omitted (Extra doses are not allowed). Enzalutamide can be taken with or without food. Enzalutamide is currently FDA-approved for the treatment of advanced metastatic, castration resistant prostate cancer. It has the chemical name 3-(4-cyano-3-trifluoromethylphenyl)-1-[3-fluoro-4-(methylcarbamoyl) phenyl]-5,5-dimethyl-2-thioxoimidazolin-4-one.

In pre-clinical assays, enzalutamide has been shown to have superior efficacy to bicalutamide in blocking AR activation .[19] Enzalutamide has been shown to inhibit bicalutamide-resistant prostate cancer cells through AR targeting by three distinct molecular processes which include blockade of testosterone binding, inhibition of DNA binding and inhibition of nuclear translocation. In vitro studies have shown that enzalutamide has an 8-fold increased inhibition of the AR than bicalutamide.[19]

Enzalutamide is FDA- approved for the treatment of metastatic CRPC. The efficacy and safety of enzalutamide was initially determined in a randomized Phase III trial of enzalutamide versus placebo in men with metastatic CRPC who were previously treated with docetaxel chemotherapy. 1199 men were randomized in a 2:1 ratio to enzalutamide (n=800) or placebo (n=399). Importantly all patients were maintained on their GnRH agonists throughout the course of the study. At the pre-specified interim analysis when 520 events had occurred median survival was 18.4 mo versus 13.6 mo in the placebo arm (HR 0.63 95%CI 0.53-0.75). Further, in a study of 1,717 chemo-naïve patients with metastatic CRPC (PREVAIL), treatment with enzalutamide versus placebo resulted in a 29% reduction in death and an 83% reduction in radiographic progression ($p<0.0001$ for both endpoints).

Warnings:
Seizure: In the randomized clinical trial, 7 of 800 (0.9%) patients treated with XTANDI 160 mg once daily experienced a seizure. No seizures occurred in patients treated with placebo. Seizures occurred from 31 to 603 days after initiation of XTANDI. Patients experiencing seizure were permanently discontinued from therapy and all seizures resolved. There is no clinical trial experience re-administering XTANDI to patients who experienced seizures.

Posterior Reversible Encephalopathy Syndrome (PRES): in post approval use, there have been reports of PRES in patients receiving XTANDI. PRES is a neurological disorder which can present with rapidly evolving symptoms including seizure, headache, lethargy, confusion, blindness, and other visual and neurological disturbances, with or without associated hypertension. A diagnosis of PRES requires confirmation by brain imaging, preferably MRI. Discontinue XTANDI in patients who develop PRES.

Dose reduction/adjustment: Patients who experience a Grade 3 or greater toxicity that cannot be ameliorated by the use of adequate medical intervention should have their treatment interrupted until the toxicity improves to a Grade 2 or lower severity. Patients may subsequently

be re-started on study drug, including at a reduced dose (80 mg or 120 mg). There are no known adverse interactions with immunologic functions.

Storage and labelling: Drug should be stored in a secure location with limited access at 77°F (25°C), with excursions permitted to 59°F to 86°F (15°C to 30°C). Patients will be instructed to store study drug at room temperature out of the reach of children.

Drug interactions: Concomitant Strong CYP2C8 Inhibitors. The concomitant use of strong CYP2C8 inhibitors should be avoided if possible. Patients who are on gemfibrozil and cannot discontinue this medication will be excluded from trial participation.

5.5.2 GNRH agonist therapy (ie leuprolide and goserelin)

GNRH agonist therapy is standard of care at the VAPORHCS. Dosing will be determined by the attending physician by considering standard therapeutic treatment regimens. GNRH agonists will be sourced by the VAPORHCS pharmacy; drug will not be donated by the manufacturer.

Dose reduction/adjustment: not reported as necessary.

Storage: Goserelin Acetate: The sterile unit will be enclosed in a sealed light and moisture proof package. The package should be stored securely in a dry place at room temperature (not to exceed 25°C or 77°F). Before being opened, each package must be inspected for damage in which case the depot should not be used. Being sterile, the syringe should be removed from its package only immediately before required.

Leuprolide: Store at room temperature 25°C (77°F); excursions permitted to 15-30°C (59-86°F) [see USP Controlled Room Temperature].

Drug interactions: Luteinizing hormone-releasing hormone analogs may diminish the therapeutic effect of antidiabetic agents. No pharmacokinetic-based drug-drug interaction studies have been performed. Because leuprolide is a peptide that is primarily degraded by peptidase and not by Cytochrome P-450 enzymes and the drug is only about 46% protein bound, drug interactions would not be expected to occur

5.6 Diet/Activity/Other Considerations

5.6.1 Diet

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

5.6.2 Contraception

Pembrolizumab may have adverse effects on a fetus in utero. Furthermore, it is not known if pembrolizumab has transient adverse effects on the composition of sperm. For this trial, male subjects will be considered to be of non-reproductive potential if they have azoospermia

(whether due to having had a vasectomy or due to an underlying medical condition).

Subjects should be informed that taking the study medication may involve unknown risks to the fetus (unborn baby) if pregnancy were to occur during the study. In order to participate in the study subjects of childbearing potential must adhere to the contraception requirement from the day of study medication initiation until completion of the prostatectomy. In the event that surgery is not performed, then contraception should be continued up till 120 days after last drug study treatment. If there is any question that a subject of childbearing potential will not reliably comply with the requirements for contraception, that subject should not be entered into the study.

5.6.3 Procedure in Case of Pregnancy:

If a male subject impregnates his female partner the study personnel at the site must be informed immediately and the pregnancy reported to the Sponsor and to Merck and followed as described above and in Section 7.2.2.

The effect of enzalutamide in pregnant and lactating women is not known, and the exposure of a fetus or nursing infant is considered a potential risk. Enzalutamide can cause fetal harm when administered to a pregnant woman based on its mechanism of action. Subjects receiving enzalutamide are advised to use 2 acceptable methods of birth control (one of which must include a condom as a barrier method of contraception) starting at the time of screening for an enzalutamide study and continuing throughout the course of treatment and for at least three months after enzalutamide is discontinued.

If during the conduct of the clinical trial, a male subject impregnates his partner, the subject should report the pregnancy to the Investigator. The Investigator should report the pregnancy to the Sponsor as an SAE within 24 hours of awareness of the event. The expected date of delivery or expected date of the end of the pregnancy, last menstruation, estimated fertility date, pregnancy result and neonatal data etc., should be included in this information.

The Investigator should report the outcome of the pregnancy (independent of outcome, eg. full term delivery, pre-term delivery, spontaneous abortion, induced abortion, stillbirth, death of newborn, congenital anomaly [including anomaly in a miscarried fetus, etc] in accordance with the same reporting procedure as for SAEs. The date of outcome of the pregnancy, gestational age, date of birth and neonatal data etc., should be included in this information.

5.7 Subject Withdrawal/Discontinuation Criteria

Subjects may withdraw consent at any time for any reason or be dropped from the trial at the discretion of the investigator should any untoward effect occur. In addition, a subject may be withdrawn by the investigator or the Sponsor if enrollment into the trial is inappropriate, the trial plan is violated, or for administrative and/or other safety reasons. Specific details regarding discontinuation or withdrawal are provided in Section 7.1.4 – Other Procedures.
A subject must be discontinued from the trial for any of the following reasons:

- The subject or legal representative (such as a parent or legal guardian) withdraws consent.

- Unacceptable adverse experiences as described in Section 5.2.1.2
- Intercurrent illness that prevents further administration of treatment
- Investigator's decision to withdraw the subject
- Noncompliance with trial treatment or procedure requirements
- The subject is lost to follow-up
- Administrative reasons

The End of Treatment and Follow-up visit procedures are listed in Section 6 (Protocol Flow Chart) and Section 7.1.5 (Visit Requirements). After the end of treatment, each subject will be followed for 30 days for adverse event monitoring (serious adverse events will be collected for 90 days after the end of treatment as described in Section 7.2.3.1). Subjects who discontinue for reasons other than progressive disease will have post-treatment follow-up for disease status until disease progression, initiating a non-study cancer treatment, withdrawing consent or becoming lost to follow-up for up to five years.

5.8 Subject Replacement Strategy

The planned sample size is 30 for the trial with a plan to enroll 32 subject to account for subject loss. There is no plan for subject replacement.

5.9 Clinical Criteria for Early Trial Termination

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

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

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

5.10 Duration of Treatment

In the absence of treatment delays due to adverse events, treatment may continue for 16 weeks up until surgical removal of the prostate or until one of the following criteria applies**:**

- Intercurrent illness that prevents further administration of treatment,
- Unacceptable adverse events(s),
- Subject decides to withdraw from the study
- General or specific changes in the subject's condition render the subject unacceptable for further treatment in the judgment of the investigator.
- For any reason, at the Sponsor or Investigator's discretion

In the event of a delay in scheduling of surgery or intercurrent illness that delays completion of surgery, treatment may be continued up till a maximum of 20 weeks.

5.11 Duration of Follow-up

The primary endpoint will be assessed for each patient at the end of the treatment period once the patient has undergone surgical removal of the prostate. Subject will be followed for clinical recurrence for an additional 5 years. Subjects removed from treatment for unacceptable adverse event(s) will be followed until resolution or stabilization of the adverse event.

6.0 STUDY FLOW CHART

Table 5: Schedule of Events.	Pre-recruitment	Screening Visit (up to 45 days before treatment)	Week 0	Every 3 Weeks[1]	Week 16	Week 20[2]	30 day post-surgery Follow-up
Eligibility Checklist	**X**	**X**					
Informed Consent		**X**					
Concomitant Medications		**X**	**X**	**X**		**X**	**X**
Exam / Vital Signs		**X**	**X**	**X**		**X**	
Testosterone		**X**			**X**		
PSA		**X**			**X**		
CBC/differential		**X**		**X**	**X**		**X**
Chemistry Panel		**X**		**X**	**X**		
HRQOL		**X**			**X**		**X**
Thyroid Testing		**X**[3]		**X**	**X**		
AE monitoring		**X**	**X**	**X**	**X**		**X**
Dispensation of Enzalutamide[4]			**X**				
Administer GNRH			**X**				
Administer Pembrolizumab			X	**X**			
TRUS Biopsy[5]		X					
Prostatectomy with LND					**X**		
Imaging (Bone Scan and/or CT/MRI)[6]	X						

[1]For 16 weeks
[2]Surgery will be within 4 weeks of completion of treatment
[3]Will retest at 3 weeks after first cycle of pembrolizumab
[4]Enzalutamide will be taken orally daily for 16 weeks
[5]TRUS Biopsy will be done for research purposes after diagnostic biopsy. Treatment will begin no sooner than 1 week after research biopsy done.

[6]CT or MRI performed at physician discretion, but required if PSA > 20ng/ml

7.0 TRIAL PROCEDURES

7.1 Trial Procedures

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

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

7.1.1 Administrative Procedures

7.1.1.1 Informed Consent

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

7.1.1.1.1 General Informed Consent

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

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

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

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

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

7.1.1.2 Inclusion/Exclusion Criteria

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

7.1.1.3 Medical History

A medical history will be obtained by the investigator or qualified designee. Medical history will include all active conditions, and any condition diagnosed within the prior 5 years that are considered to be clinically significant by the Investigator. Details regarding the disease for which the subject has enrolled in this study will be recorded separately and not listed as medical history.

7.1.1.4 Prior and Concomitant Medications Review

7.1.1.4.1 Prior Medications

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

7.1.1.4.2 Concomitant Medications

The investigator or qualified designee will record medication, if any, taken by the subject during the trial. All medications related to reportable SAEs and ECIs should be recorded as defined in Section 7.2.

7.1.1.5 Disease Details and Treatments

7.1.1.5.1 Disease Details

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

7.1.1.5.2 Prior Treatment Details

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

7.1.1.5.3 Subsequent Anti-Cancer Therapy Status

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

7.1.1.6 Assignment of Screening Number

Each patient that is screened for trial participation will be assigned a study number after informed consent is obtained.

7.1.2 Clinical Procedures/Assessments

7.1.2.1 Adverse Event (AE) Monitoring

The investigator or qualified designee will assess each subject to evaluate for potential new or worsening AEs as specified in the Trial Flow Chart and more frequently if clinically indicated. Adverse experiences will be graded and recorded throughout the study and during the follow-up period according to NCI CTCAE Version 4.0 (see Section 11.2). Toxicities will be characterized in terms regarding seriousness, causality, toxicity grading, and action taken with regard to trial treatment.

Please refer to section 7.2 for detailed information regarding the assessment and recording of AEs.

7.1.2.2 Physical Exam

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

7.1.2.3 Vital Signs

The investigator or qualified designee will take vital signs at screening, prior to the administration of each dose of trial treatment and at treatment discontinuation as specified in the Trial Flow Chart (Section 6.0). Vital signs should include temperature, pulse, respiratory rate, weight and blood pressure. Height will be measured at screening only.

7.1.2.4 Eastern Cooperative Oncology Group (ECOG) Performance Scale

The investigator or qualified designee will assess ECOG status (see Section 10.1) at screening, prior to the administration of each dose of trial treatment and discontinuation of trial treatment as specified in the Trial Flow Chart.

7.1.2.5 Tumor Tissue Collection and Correlative Studies Blood Sampling

Tumor Tissue Collection and Correlative Studies Blood Sampling will be carried out as prescribed in the Flow Sheet of events (Section 6).

7.1.3 Laboratory Procedures/Assessments

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

Table 6: Laboratory tests for hematology, chemistry, urinalysis, and others.

Hematology	Chemistry	Urinalysis	Other
Hematocrit	Albumin	Blood	PT (INR)
Hemoglobin	Alkaline phosphatase	Glucose	aPTT
Platelet count	Alanine aminotransferase (ALT)	Protein	Total thriiodothyronine (T3)
WBC (total and differential)	Aspartate aminotransferase (AST)	Specific gravity	Free tyroxine (T4)
Red Blood Cell Count	Lactate dehydrogenase (LDH)	Microscopic exam *(If dip abnormal)*	Thyroid stimulating hormone (TSH)
Absolute Neutrophil Count	Carbon Dioxide	results are noted	Blood for correlative studies
Absolute Lymphocyte Count	*(CO_2 or biocarbonate)*		
	Uric Acid		
	Calcium		
	Chloride		
	Glucose		
	Phosphorus		
	Potassium		
	Sodium		
	Magnesium		
	Total Bilirubin		
	Direct Bilirubin *(If total bilirubin is elevated above the upper limit of normal)*		
	Total protein		
	Blood Urea Nitrogen		
Table 5			

Laboratory tests for screening or entry into the Second Course Phase should be performed within 30 days prior to the first dose of treatment. After Cycle 1, pre-dose laboratory procedures can be conducted up to 72 hours prior to pembrolizumab dosing. Results must be reviewed by the investigator or qualified designee and found to be acceptable prior to each dose of trial treatment.

7.1.4 Other Procedures

7.1.4.1 Withdrawal/Discontinuation

When a subject discontinues/withdraws prior to trial completion, all applicable activities scheduled for the final trial visit should be performed at the time of discontinuation. Any adverse events which are present at the time of discontinuation/withdrawal should be followed in accordance with the safety requirements outlined in Section 7.2 - Assessing and Recording Adverse Events. After discontinuing treatment following assessment for CR, subjects should return to the site for a Safety Follow-up Visit (described in Section 7.1.5) and then proceed to the Follow-Up Period of the study (described in Section 7.1.6).

7.1.4.2 Screening

Screening for subjects will be carried out by study personnel in clinics at both OHSU and the Portland VA. Screening will be aided by a study checklist that has both inclusion and exclusion criteria.

7.1.4.3 Screening Period

Screening period will last no longer than 45 days from the time of enrollment until administration of drug therapy.

7.1.4.4 Treatment Period

The treatment period begins on day 1 of therapy and ends on the day of surgery. Treatment will be planned for a total of 14 to 16 weeks. This will allow a washout period for pembrolizumab of 2-4 weeks. If in the case there is a delay of surgery due to administrative reasons or intercurrent disease, then treatment will be allowed to continue up until 20 weeks. However, if treatment is delayed due to drug toxicity, treatment with agent felt to be the reason for the AE will be withheld and surgery will be undertaken as soon as the patient is medically fit in the determination of the investigator to undergo surgery.

7.1.4.5 Post-treatment visits

Post-prostatectomy follow-up will be carried out according to institutional policy. Typically the first visit is within 3 weeks of surgery. This is followed by a second visit within 3 months at which time a PSA is obtained. All patients should at a minimum have a PSA done within three months. Thereafter, a minimum of two PSA tests per year should be obtained, although more frequent monitoring per NCCN guidelines (about every 3 months) is recommended. At each visit subjects will be assessed for surgical complications and these will be graded according to the NCI Common Terminology for Adverse Events (CTCAE), version 4.0 and Clavien-Dindo classification (http://www.surgicalcomplication.info/index-2.html.)

7.1.4.6 TRUS-guided research biopsies

Subjects will undergo a mandatory repeat prostate biopsy which will be done for research purposes only. All TRUS procedures will be carried out as previously described by the OHSU/VA research group who has extensive experience with this modality in the research setting.[20] During TRUS, other metrics to be collected include prostate volume and the presence or absence of a hypoechoic lesion in the region of the index lesion (if present). Snap-frozen biopsy specimens will be obtained for future laboratory analysis as previously described by our group.[18] A total of 10 individual samples will be taken under local anesthetic and cryopreserved in OCT at -80C°.

7.1.4.7 Radical Prostatectomy

As a requirement of inclusion, the patient must have already selected prostatectomy with an extended pelvic lymph node as the primary treatment option for prostate cancer. Thus the surgery is considered standard of care and not a study procedure. Because of this, the surgical treatment AEs will be considered separately from the medical therapy AEs. Radical prostatectomy is the most common treatment for localized prostate cancer in the US. An in-depth understanding of anatomical considerations and inter-patient variability have markedly reduced the complication rate for this procedure. The adverse event rates from contemporary series of patients treated with prostatectomy without neoadjuvant therapy are listed below. These historical data will be used as a reference for clinical outcomes on this trial. In the unexpected event that for any reason the patient is deemed to no longer be a surgical candidate after the treatment period, the patient will be counselled about alternative options which would include but not be limited to primary radiotherapy. In our two previous trials done for high risk prostate cancer, no patient failed to undergo surgical treatment for his cancer after the treatment period, so we do not expect this occurrence.[18,21] All metrics related to the surgical procedure including 30-day complication rate will be recorded using a uniform template. All AE's will be graded according to the NCI Common Terminology for Adverse Events (CTCAE), version 4.0 and the Clavien-Dindo Classification of Surgical Complications.

Table 7: Complications from prostatectomy with node dissection	**Rate (%)**
Rectal injury	0.3 – 2.5
Ureteral injury	0.2 – 1.6
Deep venous thrombosis	1.3 – 2.7
Pulmonary embolus	0.7 – 2.0
Cardiovascular	0.7 – 1.0
Lymphocoele	0.1 – 2.0
Urinary leakage	1.2 – 10.0
Wound complication	0.8 – 1.3
Bladder neck contracture	4.0 – 20.5

7.1.5 Safety Follow-Up Visit

The mandatory Safety Follow-Up Visit should be conducted approximately 60 days after the last dose of trial treatment or before the initiation of a new anti-cancer treatment, whichever comes first. All AEs that occur prior to the Safety Follow-Up Visit should be recorded. Subjects with an AE of Grade > 1 will be followed until the resolution of the AE to Grade 0-

1 or until the beginning of a new anti-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.

7.1.6 Follow-up Visits

Subjects who discontinue trial treatment will remain in the study. Subjects will be followed according to institutional policy regarding post-operative follow-up schema. An initial PSA blood test will be done within 3 months of surgery. Then the patients should have a minimum of two PSA blood tests per year. Post-operative imaging will be done at the discretion of the treating physician.

7.2 Assessing and Recording Adverse Events

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

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

Progression of the cancer under study is not considered an adverse event
All adverse events that occur after the consent form is signed but before treatment allocation/randomization must be reported by the investigator if they cause the subject to be excluded from the trial, or are the result of a protocol-specified intervention, including but not limited to washout or discontinuation of usual therapy, diet, placebo treatment or a procedure.

From the time of treatment allocation through 30 days following cessation of treatment, all adverse events must be reported by the investigator. Such events will be recorded at each examination on the Adverse Event case report forms/worksheets. The reporting timeframe for adverse events meeting any serious criteria is described in section 7.2.2 and 7.2.3. The investigator will make every attempt to follow all subjects with non-serious adverse events for outcome.

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.

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

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 within 24 hours to the Sponsor and within 2 working days hours to Merck Global Safety. (Attn: Worldwide Product Safety; FAX 215 993-1220)

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

7.2.2.1 Serious Adverse Events

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

- Results in death;
- Is life threatening;
- Results in persistent or significant disability/incapacity;
- Results in or prolongs an existing inpatient hospitalization;
- Is a congenital anomaly/birth defect;
- Is another important medical event

- **Note:** In addition to the above criteria, adverse events meeting either of the below criteria, although not serious per ICH definition, are reportable to the Merck in the same timeframe as SAEs to meet certain local requirements. Therefore, these events are considered serious by Merck for collection purposes.
    - Is a new cancer (that is not a condition of the study);
    - Is associated with an overdose.

Refer to Table 6 for additional details regarding each of the above criteria.
For the time period beginning when the consent form is signed until treatment

allocation/randomization, any serious adverse event, or follow up to a serious adverse event, including death due to any cause other than progression of the cancer under study (reference Section 7.2.3.3 for additional details) that occurs to any subject must be reported within 24 hours to the Sponsor and within 2 working days to Merck Global Safety if it causes the subject to be excluded from the trial, or is the result of a protocol-specified intervention, including but not limited to washout or discontinuation of usual therapy, diet, placebo treatment or a procedure.

For the time period beginning at treatment allocation through 90 days following cessation of treatment, or 30 days following cessation of treatment if the subject initiates new anticancer therapy, whichever is earlier, any serious adverse event, or follow up to a serious adverse event, including death due to any cause other than progression of the cancer under study (reference Section 7.2.3.3 for additional details), whether or not related to the study drug combination, must be reported within 24 hours to the Sponsor and within 2 working days to Merck Global Safety.

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

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

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

A copy of all 15 Day Reports and Annual Progress Reports is submitted as required by FDA, European Union (EU), Pharmaceutical and Medical Devices agency (PMDA) or other local regulators. Investigators will cross reference this submission according to local regulations to the Merck Investigational Compound Number (IND, CSA, etc.) at the time of submission. Additionally investigators will submit a copy of these reports to Merck & Co., Inc. (Attn: Worldwide Product Safety; FAX 215 993-1220) at the time of submission to FDA.

7.2.2.2 Events of Clinical Interest

Selected non-serious and serious adverse events are also known as Events of Clinical Interest (ECI) and must be reported within 24 hours to the Sponsor and within 2 working days to Merck Global Safety. (Attn: Worldwide Product Safety; FAX 215 993-1220).
For the time period beginning when the consent form is signed until treatment allocation/randomization, any ECI, or follow up to an ECI, that occurs to any subject must be reported within 24 hours to the Sponsor and within 2 working days to Merck Global Safety if it causes the subject to be excluded from the trial, or is the result of a protocol-specified intervention, including but not limited to washout or discontinuation of usual therapy, diet, placebo treatment or a procedure.

For the time period beginning at treatment allocation/randomization through 90 days following cessation of treatment, or 30 days following cessation of treatment if the subject initiates new anticancer therapy, whichever is earlier, any ECI, or follow up to an ECI,

whether or not related to Merck product, must be reported within 24 hours to the Sponsor and within 24 hours to Merck Global Safety.

Events of clinical interest for this trial include:
1. an overdose of Merck product, as defined in Section 7.2.1 - Definition of an Overdose for This Protocol and Reporting of Overdose to the Sponsor, that is not associated with clinical symptoms or abnormal laboratory results.
2. an elevated AST or ALT lab value that is greater than or equal to 3X the upper limit of normal and an elevated total bilirubin lab value that is greater than or equal to 2X the upper limit of normal and, at the same time, an alkaline phosphatase lab value that is less than 2X the upper limit of normal, as determined by way of protocol-specified laboratory testing or unscheduled laboratory testing.*

*Note: These criteria are based upon available regulatory guidance documents. The purpose of the criteria is to specify a threshold of abnormal hepatic tests that may require an additional evaluation for an underlying etiology.

7.2.3 Immediate Reporting of Adverse Events to Astellas/Medivation

Within 24 hours of awareness of a serious adverse event, whether or not related to the study drug, the Investigator will complete and submit a Medwatch 3500A Form to FDA, containing all required information (reference 21 CFR 312.32). The Investigator will submit a copy of this MedWatch 3500A form to Astellas by either e-mail or fax, within the same timeframe. If submission of this SAE to FDA or Astellas or is not possible within 24 hours, the Investigator's local drug safety contact (IRB) should be informed by phone. The SAE documentation, including the Medwatch 3500A Form and available source records should be emailed or faxed to:

Astellas Pharma Global Development – United States
Email: Safety-us@us.astellas.com
Fax number: (847) 317-1241

The following minimum information is required:

· Study number/IIT regulatory identifier
· Subject number, sex and age
· The date of report
· A description of the SAE (event, seriousness of the event)
· Causal relationship to the study drug

Follow-up information for the event should be sent within 7 days as necessary.

7.2.3 Evaluating Adverse Events

An investigator who is a qualified physician or designee will evaluate all adverse events according to the NCI Common Terminology for Adverse Events (CTCAE), version 4.0. Any adverse event which changes CTCAE grade over the course of a given episode will have each change of grade recorded on the adverse event case report forms/worksheets.
All adverse events regardless of CTCAE grade must also be evaluated for seriousness.

7.3 BIOMARKER, CORRELATIVE, AND SPECIAL STUDIES

Biomarker studies

1. PD-1 – Expression of PD-1 CD8+ cells will be performed to assess the intensity of staining. Groups will be ranked in none/low, intermediate and high groups. Clinical responses will be correlated with degree of expression. This will be carried out on the clinical diagnostic biopsy and the post prostatectomy specimens.
2. Compare pre-and post-treatment PD-1 expression comparing individual subjects pre- and post-treatment tumor samples.
3. Tumor PD-L1 expression- pre- and post-treatment tumor samples will be assessed for expression of PD-L1. Treatment responses will be correlated with tumor expression of PD-L1. PD-L1 expression will be carried out by the central lab a Merck. To preserve data integrity they will be blinded to all clinical data.
4. Exploratory biomarkers in biopsy and prostatectomy specimens. IHC will be performed for CD4, CD8 and MDSC. Markers of T-cell activation (CD27, OX-40, 4-1BB) and T-cell exhaustion (CLTA-4, PD-1, TIM-3, LAG-3) will be studied.
5. Serum studies – serum will be collected before and after treatment for use in future studies. Candidate biomarkers would be evaluated as correlates to tissue responses. These include CRP, LDH, ELISPOT (INF-gamma, IL-10). Measured at baseline, pre-op and post-op.
6. Flow cytometry of blood to quantify CD4, CD8 and MDSC, markers of T-cell activation (CD27, OX-40, 4-1BB) and T-cell exhaustion (CLTA-4, PD-1, TIM-3, LAG-3). Measured at baseline, pre-op and post-op.

Table 8: Evaluating Adverse Events

An 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 any use of Merck product that:	
	†**Results in death**; or	
	†**Is life threatening; or** places the subject, in the view of the investigator, at immediate risk of death from the event as it occurred (Note: This does not include an adverse event that, had it occurred in a more severe form, might have caused death.); or	
	†**Results in a persistent or significant disability/incapacity** (substantial disruption of one's ability to conduct normal life functions); or	
	†**Results in or prolongs an existing inpatient hospitalization** (hospitalization is defined as an inpatient admission, regardless of length of stay, even if the hospitalization is a precautionary measure for continued observation. (Note: Hospitalization for an elective procedure to treat a pre-existing condition that has not worsened is not a serious adverse event. A pre-existing condition is a clinical condition that is diagnosed prior to the use of a Merck product and is documented in the patient's medical history.); or	
	†**Is a congenital anomaly/birth defect** (in offspring of subject taking the product regardless of time to diagnosis);or	
	Is a new **cancer** (that is not a condition of the study) **(**although not serious per ICH definition, is reportable to the Sponsor within 24 hours and to Merck within 2 working days to meet certain local requirements); **or**	
	Is an overdose (whether accidental or intentional). Any adverse event associated with an overdose is considered a serious adverse event for collection purposes. An overdose that is not associated with an adverse event is considered a non-serious event of clinical interest and must be reported within 24 hours to the Sponsor and to Merck within 2 working days..	
	Other important medical events that may not result in death, not be life threatening, or not require hospitalization may be considered a serious adverse event when, based upon appropriate medical judgment, the event may jeopardize the 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 Merck product to be discontinued?	
Relationship to Merck Product	Did Merck product cause the adverse event? The determination of the likelihood that Merck product caused the adverse event will be provided by an investigator who is a qualified physician. The investigator's signed/dated initials on the source document or worksheet that supports the causality noted on the AE form, ensures that a medically qualified assessment of causality was done. This initialed document must be retained for the required regulatory time frame. The criteria below are intended as reference guidelines to assist the investigator in assessing the likelihood of a relationship between the test drug and the adverse event based upon the available information. **The following components are to be used to assess the relationship between Merck product and the AE**; the greater the correlation with the components and their respective elements (in number and/or intensity), the more likely Merck product caused the adverse event (AE):	
	Exposure	Is there evidence that the subject was actually exposed to Merck product such as: reliable history, acceptable compliance assessment (pill count, diary, etc.), expected pharmacologic effect, or measurement of drug/metabolite in bodily specimen?
	Time Course	Did the AE follow in a reasonable temporal sequence from administration of Merck product? Is the time of onset of the AE compatible with a drug-induced effect (applies to trials with investigational medicinal product)?
	Likely Cause	Is the AE not reasonably explained by another etiology such as underlying disease, other drug(s)/vaccine(s), or other host or environmental factors

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

7.2.4 Sponsor Responsibility for Reporting Adverse Events

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

8.0 STATISTICAL CONSIDERATIONS

8.1 Study Design

This is a single-arm, single-stage phase II trial to evaluate efficacy and safety of pembrolizumab combined with intensive AR targeting prior to prostatectomy in subjects with high-risk localized prostate cancer (HRLPC). A total of 32 patients will be enrolled during a one-year accrual period at OHSU and PVAMC. All patients will have long-term follow-up of five years.

8.2 Primary and Secondary Endpoints

Primary and secondary endpoints are defined in Section 3. Briefly the primary endpoint is pathologic complete response (pCR) defined as absence of detectable malignant cells in the prostatectomy specimen evaluated by standard histologic techniques. The secondary endpoints include: grade 3 or higher treatment related adverse events; grade 2 or higher immune-related adverse events (irAEs); pCR in patients stratified by pre-operative prostate infiltrating T-cell PD-1 expression; biochemical complete response rate prior to surgery (i.e., PSA< 0.1 ng/mL) ; changes in EPIC health related quality of life from Week 0 vs. 16, and; incidence of surgical complications post-surgery using Clavien-Dindo Classification of Surgical Complications within 30 days of surgery. Exploratory endpoints include: incidence of cancer directed treatment at five years; pre- vs. post-treatment changes in serum, blood and tissue markers of the immune response, and; correlation of changes in markers of inflammatory response with clinical outcomes including pCR, PSA response, biochemical recurrence free survival and Safety and tolerability of Pembrolizumab, defined as Pembrolizumab related adverse event of any grade and drug dose modification.

For the purposes of this study, subjects should be evaluated for pathologic complete response to immuno-hormonal therapy post prostatectomy and biochemical complete response after immuno-hormonal therapy prior to prostatectomy. Time to biochemical recurrence is defined as the time from the date of surgery until the date of a confirmed PSA $\geq$ 0.2 ng/mL on two consecutive values at least a week apart. Those without the documented biochemical recurrence will be censored at the date of last PSA measurement. Quality of Life Assessments will be completed by EPIC quality of life (QOL) questionnaire throughout the course of the treatments.

8.3 Analysis Populations

An intent to treat (IIT) analysis set consists of all patients who consent and are enrolled in the trial

regardless of whether they are exposed to the experimental treatment. A safety analysis set consists of all patients who receive at least one dose of the experimental treatment. An efficacy evaluable analysis set includes patients who receive at last one dose of the experimental treatment and can be assessed for an efficacy endpoint. An IIT analysis set will be used to report the accrual. Safety endpoints will be assessed using the safety analysis set, while the efficacy evaluable analysis set will be used to analyze an efficacy endpoint. Note that the efficacy evaluable analysis set may differ from one endpoint to another depending on availability of efficacy endpoint data.

8.4 Statistical Analysis Plan

8.4.1 Analysis of Primary Endpoint

The primary endpoint is pathologic complete response (pCR) defined as absence of detectable malignant cells in the prostatectomy specimen evaluated by standard histologic techniques. We will perform a one-sided exact binomial test to determine if the pCR rate is significantly better than 6%, which corresponds to the historical rate of the intensive androgen axis targeting alone. An exact two-sided confidence interval will also be estimated and provided.

8.4.2. Analysis of Secondary Endpoints

AEs and irAEs will be tabulated and summarized according to the grade, attribution, and major organ systems according to the NCI CTCAE v4.03. A proportion of patients with the AE event, defined as the number of patients with the AE event divided by the total number of patients, will be provided, as well as the rate defined as the number of AE events divided by the total person-days (the number of patients x the number of days). These measures will be provided for the entire study period, from pre-operative treatment with immune hormonal therapy and separately from surgical therapy post-immunohormonal therapy. Further Clavien-Dindo Classification of Surgical Complications will be tabulated and summarized.

The proportion of patients with pCR will be presented by pre-operative prostate infiltrating T-cell PD-1 expression status based on the median split of the expression level. Exact 95% confidence intervals will also be estimated and provided. The proportion of patients with biochemical (i.e. PSA) complete response rate at 16 weeks will be estimated along with the 95% exact confidence interval. Biochemical recurrence-free survival will be estimated using Kaplan-Meier method. Generalized linear or non-linear mixed effects models will be used to analyze changes over time of health related quality of life measures.

Patients enrolled in this trial will not have radiographically measurable disease as localized prostate cancer is not reliably measurable by available imaging means. Response to treatment will be evaluated by measuring the blood tumor marker PSA and by examining the pathologic specimen at prostatectomy. Descriptive statistics that include maximum PSA reduction on therapy, % of men with a PSA < 0.1 ng/ml after surgery, and others will be used. Standard PSA criteria for response cannot be applied in this short-term therapy setting.

8.4.3. Analysis of Exploratory Endpoints

Time to cancer directed therapy at five years will be estimated using the cumulative incidence

function. Those who are alive and known to be free of any cancer directed therapy and those who died without additional cancer directed therapy are censored at the date of last contact. Generalized linear or non-linear mixed effects models will be used to evaluate changes over time in serum, blood and tissue markers of the immune response. Logistic regression will be used to assess an association with markers of inflammatory response with pCR and PSA response, while Cox regression will be used to assess the association with biochemical recurrent free survival.

8.4.4. Interim Analyses and Stopping Rules

There is no formal statistical interim analysis or stopping rule planned for this trial. However, the Knight Cancer Institute's Data and Safety Monitoring Committee requires regular IIT (investigator initiated trial) report including the information on accrual, protocol status (e.g., amendment, violations) and adverse events.

8.5 Sample Size and Power

A sample size of 30 patients is required to test the null hypothesis of 6% (the historical rate based on the intensive androgen axis targeting alone) vs. an alternative hypothesis of 26% (desired) with one-sided 3.2% significance level and 92% power using the exact binomial test. To accommodate potential 5% dropout and sample insufficiency, a total of 32 patients will be enrolled.

8.6 Randomization Method

Not applicable. No randomization will be performed for this trial.

8.7 Handling of Missing Data

Missing data will not be imputed. Whenever possible, statistical analysis methods that allow incomplete data (e.g., mixed effects models) will be used. In some cases, sensitivity analysis may be conducted under the worst and best case scenarios.

9.0 LABELING, PACKAGING, STORAGE AND RETURN OF CLINICAL SUPPLIES

9.1 Investigational Product

The investigator shall take responsibility for and shall take all steps to maintain appropriate records and ensure appropriate supply, storage, handling, distribution and usage of investigational product in accordance with the protocol and any applicable laws and regulations. Clinical Supplies will be provided by each drug manufacturer as summarized in Table 7.

Table 7 Product Descriptions

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

Product Name & Potency	Dosage Form
Enzalutamide 40 mg	Capsule

9.2 Packaging and Labeling Information

Clinical supplies will be affixed with a clinical label in accordance with regulatory requirements.

9.3 Clinical Supplies Disclosure

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

9.4 Storage and Handling Requirements

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

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

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

9.5 Returns and Reconciliation

The investigator is responsible for keeping accurate records of the clinical supplies received from Merck or designee, the amount dispensed to and returned by the subjects and the amount remaining at the conclusion of the trial.

Upon completion or termination of the study, all unused and/or partially used investigational product will be destroyed at the site per institutional policy. It is the Investigator's responsibility to arrange for disposal of all empty containers, provided that procedures for proper disposal have been established according to applicable federal, state, local and institutional guidelines and procedures, and provided that appropriate records of disposal are kept.

10.0 ADMINISTRATIVE AND REGULATORY DETAILS

10.1 Confidentiality

Patient confidentiality will be protected by keeping all hard copies of subject data in a locked file cabinet in a locked room on a protected floor that only allows access to those with the appropriate badge. Any information stored on a computer will be behind the VA and firewalls. HIPAA procedures will also be followed through the study.

10.2 Compliance with Financial Disclosure Requirements

Funds given to investigators by Merck will be reported for public availability according to the Sunshine Act.

10.3 Compliance with Law, Audit and Debarment

This trial will be conducted in compliance with all applicable institutional, local, and federal regulations. This includes auditing by appropriate regulatory authorities if necessary.

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

10.5 Quality Management System

OHSU Knight Cancer Institute, through the auditing function of the Knight Clinical Trials Office, is responsible for ensuring that all member investigators and affiliate investigators conduct clinical research studies in compliance with local IRB standards, FDA regulations and NIH policies and in accordance with the Data and Safety Monitoring Plan policies and procedures http://ozone.ohsu.edu/cancer/sharedres/kctoresdocs.cfm

Locally initiated studies will be audited by OHSU Knight CI Auditor. Newly approved studies may be audited anytime after enrollment has been initiated. Each OHSU Knight approved treatment protocol will be audited on an annual basis in accordance with the Knight Data and Safety Monitoring Plan. OHSU Knight CI Auditor will review OHSU patients only. VA patients will be audited biennially by Research Compliance Office.

It is the responsibility of each participating site's principal investigator to ensure that the study is conducted in compliance with local IRB standards, FDA regulations, and NIH policies. It is also the responsibility of each site's principal investigator to ensure that quality assurance audits at their site are conducted according to their institution's policies and procedures. The quality assurance audit process provides assurance that reported data accurately reflects the data in the primary subject record.

10.6 Data Management

The investigator will be responsible for developing data collection tools such as Case Report Forms (CRF), Study Management Tools, and a database if appropriate. Data required to meet the objectives of this protocol will be collected via CRF and uploaded to REDcap by the study

coordinator. A Study-Specific Data Monitoring Plan (SsDMP) will be developed as a separate document for both the Lead Site and any participating Sub-sites. Risk based monitoring will be utilized, with remote monitoring of source documentation and protocol compliance. At the discretion of the investigator, monitoring or auditing in addition to that outlined in the SsDMP may be conducted.

11 APPENDICES

11.1 ECOG Performance Status

Grade	Description
0	Normal activity. Fully active, able to carry on all pre-disease performance without restriction.
1	Symptoms, but ambulatory. Restricted in physically strenuous activity, but ambulatory and able to carry out work of a light or sedentary nature (e.g., light housework, office work).
2	In bed <50% of the time. Ambulatory and capable of all self-care, but unable to carry out any work activities. Up and about more than 50% of waking hours.
3	In bed >50% of the time. Capable of only limited self-care, confined to bed or chair more than 50% of waking hours.
4	100% bedridden. Completely disabled. Cannot carry on any self-care. Totally confined to bed or chair.
5	Dead.
* As published in Am. J. Clin. Oncol.: *Oken, M.M., Creech, R.H., Tormey, D.C., Horton, J., Davis, T.E., McFadden, E.T., Carbone, P.P.: Toxicity And Response Criteria Of The Eastern Cooperative Oncology Group. Am J Clin Oncol 5:649-655, 1982.* The Eastern Cooperative Oncology Group, Robert Comis M.D., Group Chair.	

11.2 Common Terminology Criteria for Adverse Events V4.0 (CTCAE)

The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 4.0 will be utilized for adverse event reporting. (http://ctep.cancer.gov/reporting/ctc.html)

11.3 The Clavien-Dindo Classification of Surgical Complications (http://www.surgicalcomplication.info/index-2.html)

Full Scale			Contracted Form	
Grades	Definition		Grades	Definition
Grade I:	Any deviation from the normal postoperative course without the need for pharmacological treatment or surgical, endoscopic and radiological interventions.		**Grade I:**	Same as for Full Scale
	Allowed therapeutic regimens are: drugs as			

	antiemetics, antipyretics, analgetics, diuretics and electrolytes and physiotherapy. This grade also includes wound infections opened at the bedside.			
Grade II:	Requiring pharmacological treatment with drugs other than such allowed for grade I complications. Blood transfusions and total parenteral nutrition are also included.		**Grade II:**	Same as for Full Scale
Grade III:	Requiring surgical, endoscopic or radiological intervention		**Grade III:**	Grades IIIa & IIIb
Grade III-a:	intervention not under general anesthesia			
Grade III-b:	intervention under general anesthesia			
Grade IV:	Life-threatening complication (including CNS complications)‡ requiring IC/ICU-management		**Grade IV:**	Grades IVa & IVb
Grade IV-a:	single organ dysfunction (including dialysis)			
Grade IV-b:	multi organ dysfunction			
Grade V:	Death of a patient		**Grade V:**	Same as for Full Scale
Suffix 'd':	If the patients suffers from a complication at the time of discharge, the			

	suffix "d" (for 'disability') is added to the respective grade of complication. This label indicates the need for a follow-up to fully evaluate the complication.			

‡ brain hemorrhage, ischemic stroke, subarrachnoidal bleeding,but excluding transient ischemic attacks (TIA);IC: Intermediate care; ICU: Intensive care unit.

11.5 Expanded Prostate Cancer Index Composite (http://urologyhealthteam.com/epic.pdf)

11.6 BIOSPECIMEN PROCEDURES

11.6.1 Tissues Samples

Tissue Snap Freezing (Biopsy Samples)

1. Label 1" cardboard squares with pencil (previously cut up from old backs of notebooks) with patient initials, patient id, and sample location
2. Pour Pentane solution over 4-5 pieces of dry ice or about ¾ cup of shaved dry ice until the cup is boiling and has enough liquid to cover the samples
3. Place drop of Cryomatrix on cardboard square (be sure that there aren't any trapped air bubbles)
4. Lay sample in Cryomatrix and cover with another drop of solution
5. Drop into pentane/dry ice solution (can remain in this solution for as long as need be as long as the solution is still boiling)
6. Store at –80 °C in labeled 50 ml tubes

Accessories:

- 10- 1 in^2 cardboard pieces (can be cut out of the back of paper pads)
- Needles and syringes (to transfer biopsy samples from biopsy needle to cardboard)
- Small plastic beaker
- Pentane Solution (2 Methyl butane (Isobutane) – EM Science MXO760-1)
- Cryomatrix (Frozen Specimen Embedding Medium – Shandon 6769006)
- Tweezers (to transfer cardboard out of pentane/dry ice)
- 50 ml plastic tube

Tissue Snap Freezing (Surgical Samples)

Using preoperative biopsy results and gross impression as guides, suspicious areas will be subjected to frozen section evaluation until several areas (6 or more mm in greatest dimension) are confirmed as tumor. Samples are divided in half and one is sent for frozen section and the other cryopreserved. Then bank both the frozen section residual tumor along with the reserved fresh tissue from the same locale. Tissues are then placed in small cryovials and topped off with isopentane. From those areas tissues will be frozen in OCT compound for storage at –80 degrees C. The goal will be to preserve at least three confirmed frozen tumor blocks and at least two frozen blocks of normal prostate from each case. Once everything is frozen (2 tumor and 1 non-tumor, both frozen section residual and reserved fresh tissue for a total of 6 vials) then pack the 6 cryo-tubes in a larger tube and place on dry ice until placed in -80C.

Assuming in the typical case, five frozen tissue samples are collected, they will be labeled accordingly as follows: Study and case number and "FS TUMOR A" (the block from which the frozen section examination was performed); "NON-FS TUMOR B"—when the diagnosis is based on gross impression—and so forth. The confirmed normal prostate tissue will be labeled with Study and case number and "FS NL D" for example, reflecting frozen section confirmation of normalcy with presumptive non-frozen normal tissue also collected and similarly labeled.

Storage: Frozen specimens excluding the Tumor Bank tissue will be stored in the Garzotto laboratory (VA Bldg 103:F218). Two H&E stained slides will be generated as frozen sections; one will remain with the permanent sections slides in the surgical pathology laboratory and the other will be stored in the research laboratory.

As per protocol, a record mapping the physical location (preferably using terms *right/left* and *anterior/posterior* and *mid/apical/bladder neck*) will be made. The frozen samples should not include marginal tissue or otherwise compromise the histologic study of the specimen.

11.6.2 Serum Samples:

Venipuncture at specified time-points will be performed to fill two 10 cc Red Top tubes. Blood will be spun down at approximately 1000 rpm for 10 minutes and serum extracted off the top and aliquoted in a new 1.5 ml Eppendorf tubes until all available serum extracted. Serum will then be frozen down at -80C.

12.0 DATA REPORTING/REGULATORY REQUIREMENTS

12.1 Data Collection and Storage

The following data forms will be used as requested: consent form, eligibility checklist, patient data form, medical history form, physical exam form, screening visit form (including all baseline data as outlined), study drug diary, adverse event report form, and concomitant medication form. Record maintenance will be upheld by the study coordinator. Data accumulated from this study will be forwarded either by completed source documents and/or sent via the networked OCI database to the OHSU Cancer Institute Data Manager on a monthly or as applicable basis unless otherwise informed. The Guardian of records will be:

Wesley Stoller, MA
Clinical Research Coordinator
Portland VA Medical Center
3710 SW US Veterans Hospital Rd.
Mailcode: P3HSR&D
Portland, OR 97239
Telephone (503) 220-8262 x-57758
Fax (503) 721-1479

12.2 Protocol Review

The protocol and informed consent form for this study must be reviewed and approved in writing by the OHSU Knight Cancer Institute (CI) Clinical Research Review Committee (CRRC) and the appropriate Institutional Review Board (IRB) prior to any subject being consented on this study.

12.3 Informed Consent

Written informed consent will be obtained from all subjects, or the legally authorized representative of the subject, participating in this trial, as stated in the Informed Consent section of the case of Federal Regulations, Title 21, Part 50. Documentation of the consent process and a copy of the signed consent shall be maintained in the subject's medical record.

12.4 Changes to Protocol

Any modification of this protocol must be documented in the form of a protocol revision or amendment signed by the principal investigator and approved by the CRRC and IRB, before the revision or amendment may be implemented. The only circumstance in which the amendment may be initiated without regulatory approval is for a change necessary to eliminate an apparent and immediate hazard to the subject. In that event, the investigator must notify the CRRC and IRB in writing within 10 working days after the implementation. Investigators holding the IND must notify FDA of substantive changes to the protocol.

12.5 Maintenance of Records

If the investigator relocates or for any reason withdraws from the study, the study records must be transferred to an agreed upon designee, such as another institution, another investigator, or to OHSU Knight Cancer Institute Clinical Trials Office. Records must be maintained according to sponsor or FDA and VA requirements.

12.6 OHSU IRB Reporting of Unanticipated Problems and Adverse Events

Adverse event lists, guidelines, and instructions for AE reporting can be found in Section 7.0 (Adverse Events: List and Reporting Requirements).

12.8 Inclusion of Women, Minorities and Children

12.8.1 Inclusion of Women and Minorities

No OHSU Cancer Institute or PVAMC study will focus on any particular racial or ethnic subset. No subject will be excluded from the study on the basis of racial or ethnic origin. Ethnic distribution should be similar to Oregon's ethnic distribution, though African Americans may be overrepresented and Asians underrepresented because prostate cancer is more common in African Americans and less common in Asians when compared to the average risk. Minority volunteers will be recruited for this study from the general population and since prostate cancer only affects men, women will not be included in this study.

12.8.2 Inclusion of Children

This protocol does not include children for the following reason: children are not affected by prostate cancer.

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