Official Title: A Pilot Study of 18Fluorine-Fluciclovine Positron Emission Tomography/Computed Tomography for Staging Muscle Invasive Bladder Cancer Preceding Radical Cystectomy.

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A Pilot Study of ^{18}Fluorine-Fluciclovine Positron Emission Tomography/Computed Tomography for Staging Muscle Invasive Bladder Cancer Preceding Radical Cystectomy.

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Responsible Research Nurse: pending

Responsible Data Manager: *pending*

Agent(s): Fluciclovine F18 (Axumin®, Blue Earth Diagnostics Inc; commercial)

Study Exempt from IND Requirements per 21 CFR 312.2(b).

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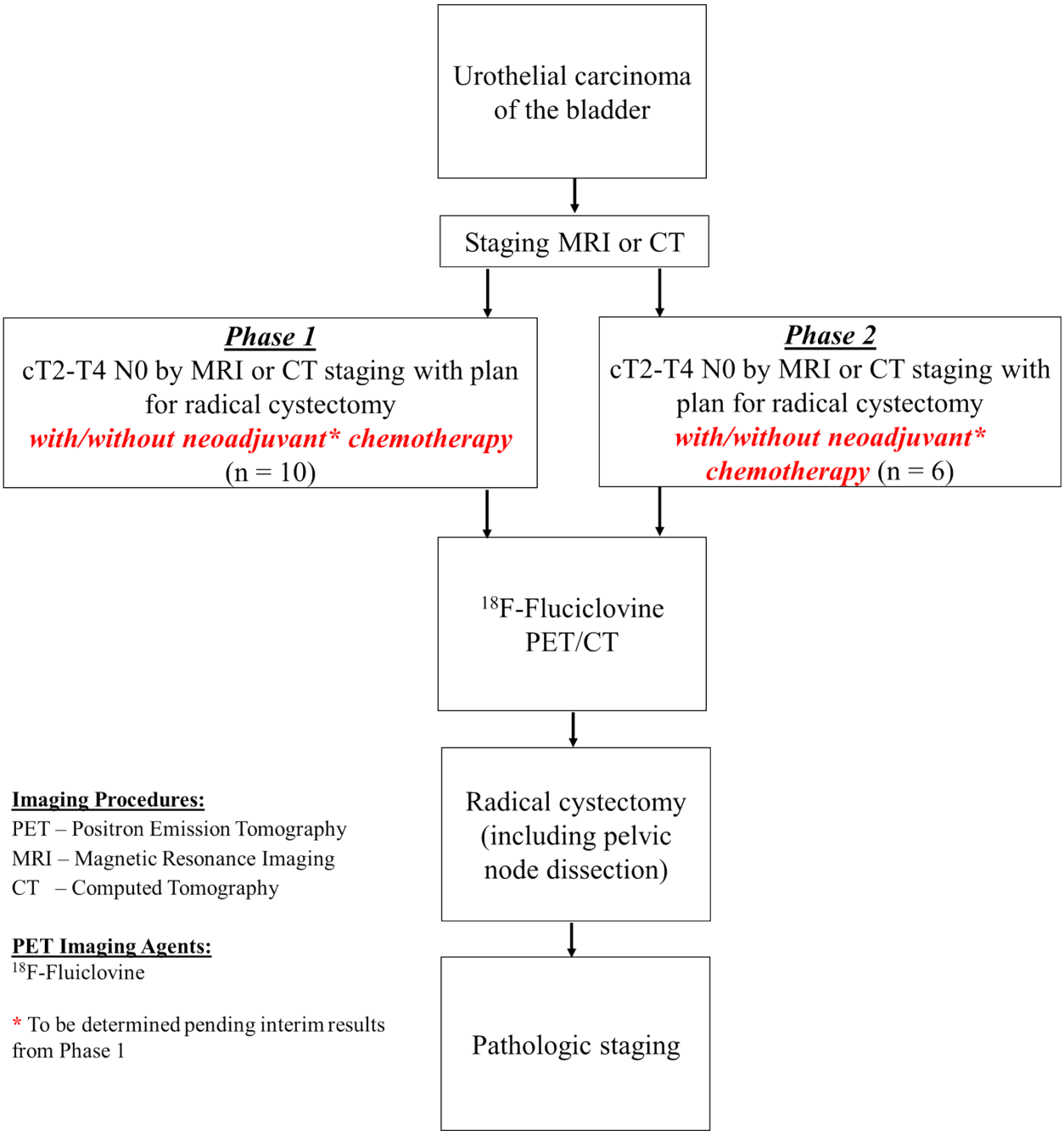
SCHEMA


Urothelial carcinoma of the bladder
Staging MRI or CT
Phase 1
cT2-T4 N0 by MRI or CT staging with plan for radical cystectomy with/without neoadjuvant* chemotherapy (n = 10)
Phase 2
cT2-T4 N0 by MRI or CT staging with plan for radical cystectomy with/without neoadjuvant* chemotherapy (n = 6)
18F-Fluciclovine PET/CT
Radical cystectomy (including pelvic node dissection)
Pathologic staging
Imaging Procedures:
PET – Positron Emission Tomography
MRI – Magnetic Resonance Imaging
CT – Computed Tomography
PET Imaging Agents:
18F-Fluiclovine
* To be determined pending interim results from Phase 1

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1. OBJECTIVES

1.1 Study Design

This is a prospective, pilot study.

1.2 Primary Objectives

To estimate the agreement rate of detecting metastatic disease between ^{18}F-fluciclovine-PET/CT and histopathology from radical cystectomy for detecting locoregional metastatic disease in patients with cT2-T4N0M0 muscle invasive bladder cancer by conventional CT/MRI imaging.

1.3 Secondary Objectives

1.3.1 To estimate the rate of detection of suspected distant metastatic disease by ^{18}F-fluciclovine -PET/CT in patients with cT2-T4N0M0 muscle invasive bladder cancer by conventional CT/MRI imaging.

1.3.2 To investigate the relationship between ^{18}F-fluciclovine uptake on PET/CT and pathologic tumor stage and size of primary bladder tumor after radical cystectomy.

1.3.3 To investigate the relationship between ^{18}F-fluciclovine uptake on PET/CT and the presence/absence of a ASCT2 and LAT1 amino acid transporters in the resected primary bladder tumors.

2. BACKGROUND

2.1 Urothelial carcinoma of the bladder

Urothelial carcinoma of the bladder is the fifth most common malignancy in the United States, with an estimated 75,000 new cases and 15,000 deaths in a year [1]. Approximately 25% of new cases are muscle-invasive bladder cancer (MIBC), while the rest are non-muscle-invasive bladder cancer (NMIBC). Accurate staging of bladder cancer is paramount in deciding the appropriate course of management. While NMIBC is generally managed by cystoscopic resection with intravesical BCG or chemotherapy, the standard treatment for MIBC is radical cystectomy (RC) preceded by neoadjuvant chemotherapy (NAC) in cisplatin-eligible patients [2-4]. It is estimated that ~30% of patients undergoing RC may have nodal metastases [5, 6]. Metastatic disease has exhibited improved outcomes when treated with first-line systemic cisplatin-based chemotherapy and salvage post-platinum therapy using one of multiple programmed death (PD)-1 and PD-ligand (L)-1 inhibitors (pembrolizumab, atezolizumab, durvalumab, nivolumab, avelumab) [7-13]. Additionally, pembrolizumab and atezolizumab are approved for the first-line therapy of cisplatin-ineligible patients based on durable responses [14, 15]. Therefore, pre-operative identification of those with node-positive metastatic disease will enhance prognostic risk stratification and tailoring of neoadjuvant therapy, for example with the consideration of clinical trials of more aggressive and promising neoadjuvant combination

regimens and tailoring the extent of lymph node dissection based on the extent of node-positive disease.

2.2 Current imaging methods to stage bladder cancer

Current imaging methods used to stage bladder cancer predominantly include computed tomography (CT) and magnetic resonance imaging (MRI). While both are useful in locoregional staging, both exhibit limitations. CT and MRI rely on nodal enlargement as an indicator of disease involvement. Non-enlarged lymph nodes harboring disease can be falsely negative by anatomic imaging. Conversely, false positive lymphadenopathy from benign etiologies can preclude eligible patients from RC. CT and MRI sensitivity for detection of nodal metastasis ranges from 31-50% and specificity ranges from 68-100% [6]. Some reports suggest that 40% of those with bladder confined disease by imaging have extravesical and/or pathologically node-positive disease at RC [16-20].

Fluorodeoxyglucose (^{18}FDG)-positron emission tomography (PET) is a functional imaging modality that has excellent sensitivity for detecting metastatic disease when combined with anatomic CT or MRI [21-25]. However, effective use of FDG-PET/CT in bladder cancer is limited by the fact that FDG is excreted in urine. Saturation of the imaging signal from within the bladder and urinary tract can affect accurate assessment of locoregional FDG uptake. Furthermore, elevated FDG uptake can also be seen in benign inflammatory conditions and within the gastrointestinal tract, which can also confound staging assessment. Correspondingly, FDG-PET is not approved by regulatory agencies to stage bladder cancer [4].

Given the limitations of both conventional structural CT/MRI and FDG-PET to stage urinary tract cancers, an alternative functional imaging marker without these limitations is highly desirable and warrants investigation.

2.3 ^{18}Fluorine-Fluciclovine

Anti-1-amino-3-F-18-fluorocyclobutane-1-carboxylic acid (FACBC or ^{18}F-fluciclovine) is a novel radiotracer for imaging amino acid transporters, which can be over-expressed in cancer [26]. ^{18}F-fluciclovine was recently approved by the Food and Drug Administration for the detection of biochemically recurrent prostate cancer [27, 28] and it may have a role in complementing MRI for staging localized prostate cancer [29] and radiation planning [30]. ^{18}F-fluciclovine is predominantly transported in cells via the L-system amino acid transporters, ASCT (alanine-serine-cysteine transporters)-2 and LAT (large neutral amino acid transporters)-1, with peak uptake in tumors 5 to 20 minutes after injection.

In addition to prostate cancer, ^{18}F-fluciclovine has been shown to accumulate in other tumor types, including breast, lung and colon cancer [31]. A unique feature of ^{18}F-fluciclovine biodistribution is low bladder uptake at early post injection timepoints when elevated uptake in tumors is already seen [32, 33]. Additional advantages of ^{18}F-fluciclovine over other naturally occurring amino acid PET imaging agents, such as ^{11}C methionine, include a longer radiolabel half-life (110 minutes for ^{18}F agents vs 20 minutes for ^{11}C) such that an on-site cyclotron is not required [34]. Other ^{11}C agents have also been tested, including ^{11}C choline and ^{11}C acetate

PET/CT have been tested for staging bladder cancer with equivocal results [35].

No adverse effects to patients that is directly attributable to ^{18}F-fluciclovine have been reported to date [36].

2.4 Rationale for staging bladder cancer with ^{18}F-fluciclovine

Amino acid transporters are overexpressed in many cancers [26]. Diets rich in the amino acids L-isoleucine and L-leucine have been shown to promote bladder cancer in rats [37]. The LAT-1 transporter has been implicated as responsible for leucine uptake in T24 bladder cancer cells [38]. Inhibition of LAT-1 can prevent cell growth in bladder cancer cell lines [38], and overexpression of LAT-1 can predict progression of urinary tract cancer [39, 40]. These findings suggest that the amino acid transporters are a viable imaging target in bladder cancer.

^{18}F-fluciclovine has been shown to be a promising imaging agent in multiple cancer types. The biodistribution profile of ^{18}F-fluciclovine shown in prior studies suggests that it may be useful for imaging bladder cancer. Experiments with a rat orthotopic prostate cancer model compared the uptake of ^{18}F-fluciclovine with that of FDG and found that target-to-background ratio was higher for ^{18}F-fluciclovine with only minimal bladder accumulation. Compared to FDG, ^{18}F-fluciclovine is only minimally eliminated by the kidneys during the typical imaging time course suggesting that it may be more optimal than FDG for imaging renal and urinary tract malignancies. ^{18}F- fluciclovine is physiologically found in the pancreas, liver, bone marrow, and muscle, with negligible uptake in kidneys, bowel, and shows delayed urinary excretion even when compared with ^{11}C choline, leading to a more favorable distribution profile in the abdomen and pelvis [32].

Taken together, these studies suggested that ^{18}F-fluciclovine-PET/CT may be a viable imaging strategy to accurately stage muscle invasive bladder cancer.

2.5 Correlative Studies Background

LAT1 and ASCT2 expression are determined by immunohistochemical (IHC) staining with anti-LAT1 and anti-ASCT2 monoclonal antibodies, respectively. Immunostaining with these antibodies are based on standard protocols and have been described in detail previously [41]. Briefly, 5 unstained slides per patient consisting of 5 μM thick sections of formalin fixed paraffin embedded (FFPE) tissue are obtained from deparaffinized and rehydrated sections. These sections are treated with 0.3% hydrogen peroxide (H_2O_2) in methanol for 30 min to block endogenous peroxidase activity. To expose the antigens, sections are autoclaved in ethylenediaminetetraacetic acid (pH 8.0) for 5 min and cooled for 30 min. After rinsing in phosphate-buffered saline, the sections are incubated with anti-LAT1 and anti-ASCT2 antibodies overnight followed by secondary staining with horseradish peroxidase-conjugated antibodies. Visualization of staining is carried out using 0.02% 3,3′-diaminobenzidine tetrahydrochloride and 0.01% H_2O_2 in 0.05 M Tris-HCl (pH 7.4). Negative control tissue sections were stained as described above, except that the primary antibodies are omitted. The LAT1 and ASCT2 expression scores are assessed by the extent of staining as follows: 0, less than 5% of tumor area stained; 1, 5–10% stained; 2, 11–25% stained; 3, 26–50% stained; and 4, 51% or more stained.

3. PARTICIPANT SELECTION

3.1 Eligibility Criteria

Participants must meet the following criteria on screening examination to be eligible to participate in the study:

3.1.1 Participants must have histologically or cytologically confirmed urothelial carcinoma of the bladder.

3.1.2 Participants must have cT2-T4N0 disease at the time of the study, as defined by conventional CT or MRI imaging. Patients must have no definite evidence of locoregional or distant metastatic disease at the time of study eligibility, as defined by conventional imaging.

3.1.3 Radical cystectomy must be planned for the patient after the planned ^{18}F-fluciclovine-PET/CT.

3.1.4 Patients may or may not have had prior neoadjuvant therapy prior to this study.

3.1.5 Age ≥18 years. Since no dosing or adverse event data are currently available on the use of ^{18}F-fluciclovine in participants <18 years of age, and the majority of bladder cancer occur in the adult population [42], children are excluded from this study but will be eligible for future pediatric trials.

3.1.6 ECOG performance status ≤2 (Karnofsky ≥60%, see Appendix A)

3.1.7 Ability and willingness to comply with the study procedures.

3.1.8 The effects of ^{18}F-fluciclovine on the developing human fetus are unknown. For this reason and because radiopharmaceuticals may be teratogenic, women of childbearing potential and men must agree to use adequate contraception (barrier method of birth control; abstinence) prior to study entry and for 24 hours after the PET/CT scan is completed. Should a woman become pregnant or suspect she is pregnant while participating in this study, she should inform her treating physician immediately.

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

3.2 Exclusion Criteria

Participants who exhibit any of the following conditions at screening will not be eligible for admission into the study:

3.2.1 Participants with other known malignancy that has required treatment in the past 3 years.

3.2.2 Pregnant women are excluded from this study because ^{18}F-fluciclovine is a radiopharmaceutical with the potential for teratogenic effects. Because of the radiation exposure to a nursing infant from ^{18}F-fluciclovine, women who are breastfeeding are also excluded from this study.

3.2.3 History of allergic reactions attributed to compounds of similar chemical or biologic composition to ^{18}F-fluciclovine.

3.2.4 Uncontrolled intercurrent illness including, but not limited to, ongoing or active infection, symptomatic congestive heart failure, unstable angina pectoris, cardiac arrhythmia, or psychiatric illness/social situations that would limit compliance with study requirements.

3.2.5 Contraindications for PET/CT including:
- Severe claustrophobia

3.2.6 Any past or current condition that in the opinion of the study investigators would confound the results of the study or pose additional risk to the patient by their participation in the study.

3.3 Inclusion of Women and Minorities

Both men and women of all races and ethnic groups are eligible for this trial.

4. REGISTRATION PROCEDURES

4.1 General Guidelines for DF/HCC Institutions

Institutions will register eligible participants in the Clinical Trials Management System (CTMS) OnCore. Registrations must occur prior to the initiation of any protocol-specific therapy or intervention. Any participant not registered to the protocol before protocol-specific therapy or intervention begins will be considered ineligible and registration will be denied.

An investigator will confirm eligibility criteria and a member of the study team will complete the protocol-specific eligibility checklist.

Following registration, participants may begin protocol-specific therapy and/or intervention. Issues that would cause treatment delays should be discussed with the Overall Principal Investigator (PI). If the subject does not receive protocol therapy following registration, the subject must be taken off-study in the CTMS (OnCore) with an appropriate date and reason entered.

4.2 Registration Process for DF/HCC Institutions

Applicable DF/HCC policy (REGIST-101) must be followed.

4.3 General Guidelines for Other Investigative Sites

N/A

4.4 Registration Process for Other Investigative Sites

N/A

5. IMAGING PLAN

5.1 Summary

All procedures will be performed on an outpatient basis. Expected toxicities and potential risks are described in Section 6 (Expected Toxicities and Dosing Delays/Dose Modification).

This is a pilot study investigating ^{18}F-fluciclovine PET/CT for imaging patients with muscle invasive bladder cancer. The investigational imaging procedure in this study is ^{18}F-fluciclovine-PET/CT.

After the participant meets eligibility criteria for the study and informed written consent is obtained, the participant will undergo the study procedure (^{18}F-fluciclovine-PET/CT imaging). ^{18}F-fluciclovine-PET/CT will be obtained within 6 weeks of the standard-of-care diagnostic staging procedures (MRI and/or diagnostic CT).

Participants will then undergo radical cystectomy per standard-of-care as determined by the medical oncologist. At the time of radical cystectomy, which usually includes pelvic lymph node dissection, standard evaluation of the surgical specimen will determine pathologic staging. We will assess the results of the standard evaluation of the surgical specimen for metastatic disease. These results will be compared with staging as determined by ^{18}F-fluciclovine PET/CT and standard-of-care diagnostic imaging.

The participant will have completed all study requirements once tissue is obtained at radical cystectomy.

5.2 ^{18}F-fluciclovine PET/CT imaging

5.2.1 The ^{18}F-fluciclovine-PET/CT imaging procedure is described in Section 5.4.

Participants will be instructed to refrain from strenuous exercise for the 24 hours preceding the PET/CT exam, since amino acid transporters expression have been

noted to increase in skeletal muscle for that period after exercise [43]. Participants will be NPO for 4 hours prior to administration of ^{18}F-fluciclovine.

5.2.2 Participants will be provided with instructions regarding what precautions to take at home once they receive the study agent. As with all radiopharmaceuticals administered, participants will receive a card stating that they have received a radiopharmaceutical for medical purposes which includes, but is not limited to, radiopharmaceutical, dosage, suggested time of detection, suggested contamination item storage duration, and institutional and departmental contact information.

Given the short half-life and diagnostic dose of ^{18}F-fluciclovine (110 minutes), no special precautions regarding excreta or contact with other humans are necessary.

5.3 Surgery

After completion of the ^{18}F-fluciclovine-PET/CT scan, participants will undergo standard-of-care radical cystectomy surgery as determined by medical oncology and urology. If surgical plans (for example, the presence of unsuspected extensive disease precluding cystectomy) are altered based on clinical findings during surgery, the patient may remain evaluable if a lymph node dissection is performed as determined by the study team. Results from the standard-of-care evaluation of the pathologic specimens will be collected and reviewed. This includes evaluation of pathological samples for metastatic cancer burden.

5.4 Investigational Imaging Agent Administration

10 millicurie (mCi) of ^{18}F-fluciclovine will be administered via slow push over 10 seconds through a peripheral intravenous line. [33]. This is performed immediately prior to imaging. No specific pre-administration lab parameters or procedures for the patient is required prior to agent administration.

5.4.1 Image Acquisition Details:

Immediately after the injection of the radiopharmaceutical, dynamic PET/CT images of the pelvis will be obtained for 15 minutes. Subsequently, PET/CT images will be obtained from the pelvis to the base of skull in the caudal to cranial direction. CT will be obtained for attenuation correction per standard institutional PET/CT policies. PET images will be acquired in 3D mode at 3-5 minutes per bed position.

As noted in Section 8.1, this image acquisition protocol is based on the pharmacokinetics of the agent in male patients with prostate cancer. While we do not anticipate significant differences in the kinetics of the agent in patients with bladder cancer, appropriate adjustments to the imaging acquisition parameters will be made if significant differences are observed on initial imaging studies.

5.4.2 Image Analysis Details:

^{18}F-fluciclovine-PET/CT images will be reviewed for lesions, defined as focal areas of ^{18}F-fluciclovine uptake not correlating with normal anatomy or physiology. Lesions will be scored on a 5-point scale for diagnostic certainty (0, definitely benign; 1, probably benign; 2, equivocal; 3, probably malignant; and 4, definitely malignant).

Three-dimensional regions of interest will be placed in areas of abnormal tracer uptake scored as 3-5 and the level of ^{18}F-fluciclovine uptake recorded. Measures of ^{18}F-fluciclovine will include: maximal standardized uptake value (SUV_{max}, body weight), metabolic tumor volume (MTV; volume of tumor [cm^3] with SUV >42% of SUV_{max}), SUV_{mean} (within the MTV), and total lesion avidity (TLA; calculated as MTV × SUV_{mean}). Tumor to background ratios will also be calculated using blood pool, muscle and bone marrow as reference normal organs.

5.5 Duration of Study Participation

Duration of study participation will be until the time that surgical specimens are obtained at the time of radical cystectomy.

Participants may be removed from study participation prior to the scheduled completion if one of the following events occurs:

- Unacceptable adverse event(s),
- Participant demonstrates an inability or unwillingness to comply with the study procedures,
- Participant decides to withdraw from the study,
- Condition that would result in confounding information for study procedure interpretation as determined by the study investigators,
- General or specific changes in the participant's condition render the participant unacceptable for continuing study procedures in the opinion of the treating investigator.

5.6 Duration of Follow Up

After completing the study procedures, patients will be followed for at least 30 days. Continued standard of care will be provided by the treating physician. Participants removed from the study for unacceptable adverse events determined to be related to the study drug will be followed until resolution or stabilization of the adverse event.

5.7 Criteria for Taking a Participant Off Study

Participants will be removed from study when any of the following criteria apply:

- Lost to follow-up
- Withdrawal of consent for data submission
- Death

The reason for taking a participant off study, and the date the participant was removed, must be documented in the case report form (CRF). In addition, the study team will ensure Off Treatment/Off Study information is updated in OnCore in accordance with DF/HCC policy REGIST-101. If a participant is taken off study prior to undergoing the research ^{18}F-fluciclovine-PET/CT scan, the participant may be replaced.

6. DOSING DELAYS/DOSE MODIFICATIONS

Dose delays and modifications will be made as indicated in the following table(s). The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 4.0 will be utilized for dose delays and dose modifications. A copy of the CTCAE version 4.0 can be downloaded from the CTEP website http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm.

If possible, symptoms should be managed symptomatically. In the case of toxicity, appropriate medical treatment should be used (including anti-emetics, anti-diarrheals, etc.).

The study procedure is ^{18}F-fluciclovine PET/CT imaging. All adverse events experienced by participants will be collected from the time of the administration of the study agent (^{18}F-fluciclovine) until 24 hours after ^{18}F-fluciclovine PET/CT images are obtained, regardless of their attribution/relatedness). Adverse events related to standard-of-care of FDA-approved procedures for comparison with ^{18}F-fluciclovine PET/CT imaging (i.e., Diagnostic CT scans, MR imaging) will not be reported as part of this trial. Beyond 24 hours after ^{18}F-fluciclovine PET/CT imaging, adverse events that are determined to be related to the participant's systemic therapy or underlying condition and are determined not to be related to the agents and procedures in this protocol, will not be reported, as the investigational procedure is a diagnostic imaging procedure.

Adverse events related to radical cystectomy surgery will not be reported as part of this trial. Participants continuing to experience toxicity related to ^{18}F-fluciclovine at the off-study visit may be contacted for additional assessments until the toxicity has resolved or is deemed irreversible.

6.1 Anticipated Toxicities

6.1.1 Expected Adverse Events from ^{18}F-fluciclovine

^{18}F-fluciclovine will be administered in tracer quantities. As such, the tracer dose of ^{18}F-fluciclovine is expected to have no significant effect on normal human physiology and we do not anticipate any adverse events [36].

^{18}F-fluciclovine is an FDA-approved imaging agent for evaluation of metastatic prostate cancer. Adverse reactions were reported in ≤1% of subjects during clinical studies with Axumin (Appendix B). The most common adverse reactions were injection site pain, injection site

erythema and dysgeusia. To date, no adverse events have been reported that have been directly attributable to ^{18}F-fluciclovine [36],

6.1.2 Expected Adverse Events from Intravenous Line Placement

- Minor discomfort
- Bleeding at the injection site
- Infection at the injection site
- Bruising at the injection site
- Claustrophobia
- Discomfort

6.1.3 Expected Adverse Events from Radiation from PET/CT scans

The estimated effective dose from ^{18}F-fluciclovine is approximately effective dose per unit administered activity is 22.1 μSv/MBq [44].

Additional exposure from the CT component of the combined ^{18}F-fluciclovine PET/CT scan is approximately 10 mSv.

Table 1. Dosimetry for ^{18}F-fluciclovine [44]	
Organ	*(μGy/MBq)*
Liver	*33.5±4.2*
Kidneys	*13.7±0.8*
Spleen	*23.8±11.7*
Bladder wall	*25.2±17*
Lungs	*34.5±5.3*
Whole body	*12.6±0.2*
Effective dose	*22.1±2.1*

6.2 Toxicity Management

Allergic reactions and anaphylaxis to ^{18}F-fluciclovine will be managed following standard institutional guidelines for contrast reaction management (DFCI Department of Imaging Policy 5.31).

If a study participant has a severe allergic reaction or anaphylactic reaction to ^{18}F-fluciclovine and PET/CT images are unable to be obtained, the participant will be removed from the study.

All other toxicities and adverse events will be managed as medically necessary. Supportive care will be administered as medically necessary.

6.3 Dose Modifications

The dosage of ^{18}F-fluciclovine is never modified.

7. ADVERSE EVENTS: LIST AND REPORTING REQUIREMENTS

Adverse event (AE) monitoring and reporting is a routine part of every clinical trial. The following list of reported and/or potential AEs (Section 7.1) and the characteristics of an observed AE (Section 7.2) will determine whether the event requires expedited reporting **in addition** to routine reporting.

7.1 Expected Toxicities

Local injection site pain
Local injection site erythema
Dysgeusia

7.2 Adverse Event Characteristics

- **CTCAE term (AE description) and grade:** The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 4.0 will be utilized for AE reporting. All appropriate treatment areas should have access to a copy of the CTCAE version 4.0. A copy of the CTCAE version 4.0 can be downloaded from the CTEP web site http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm.

- **For expedited reporting purposes only:**
  - AEs for the agent(s) that are listed above should be reported only if the adverse event varies in nature, intensity or frequency from the expected toxicity information which is provided.
  - Other AEs for the protocol that do not require expedited reporting are outlined in the next section (Expedited Adverse Event Reporting) under the sub-heading of Protocol-Specific Expedited Adverse Event Reporting Exclusions.

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

7.3 Adverse Event Reporting

7.3.1 In the event of an unanticipated problem or life-threatening complications treating investigators must immediately notify the Overall PI.

7.3.2 Investigators **must** report to the Overall PI any adverse event (AE) that occurs after the initial dose of study treatment, during treatment, or within 30 days of the last dose of treatment on the local institutional SAE form.

7.3.3 DF/HCC Adverse Event Reporting Guidelines

Investigative sites within DF/HCC will report AEs directly to the DFCI Office for Human Research Studies (OHRS) per the DFCI IRB reporting policy.

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7.3.4 Protocol-Specific Adverse Event Reporting Exclusions

For this protocol only, the AEs/grades occurring from the time of ^{18}F-fluciclovine administration until 24 hours after ^{18}F-fluciclovine administration will be reported. However, they still must be reported through the routine reporting mechanism (i.e. case report form).

7.4 Reporting to Hospital Risk Management

Participating investigators will report to their local Risk Management office any participant safety reports, sentinel events or unanticipated problems that require reporting per institutional policy.

7.5 Routine Adverse Event Reporting

All Adverse Events **must** be reported in routine study data submissions to the Overall PI on the toxicity case report forms. **AEs reported through expedited processes (e.g., reported to the IRB, FDA, etc.) must also be reported in routine study data submissions.**

7.6 Additional Adverse Event Reporting

In addition to the reporting of Adverse Events as described in 7.5, any observed Serious Adverse Events that occur following the receipt of ^{18}F-fluciclovine, whether related to the product or not, PI or designee shall notify BED within twenty four (24) hours of knowledge of any SAEs. SAE reports to BED must be recorded and faxed or scanned and emailed to::

Bracco Diagnostics Inc. Drug Safety Unit
E mail: Drugsafetyus@BlueEarthDx.com
Fax: +1-609-514-2522

8. IMAGING AGENT INFORMATION

A list of the adverse events and potential risks associated with ^{18}F-fluciclovine can be found in Section 7.1 and Appendix B

8.1 ^{18}F-fluciclovine

8.1.1 Description

^{18}Fluorine-Fluciclovine is (1r, 3r)-1-amino-3[^{18}F]fluorocyclobutane-1-carboxylic acid. The molecular weight is 132.1.

The following pharmacokinetic information for ^{18}F-fluciclovine is based on experience in male patients with prostate cancer:

Following intravenous administration, ^{18}F-fluciclovine distributes to the liver (14% of administered activity), pancreas (3%), lung (7%), red bone marrow (12%) and myocardium (4%). With increasing time, ^{18}F-fluciclovine distributes to skeletal muscle.

Following intravenous administration, the tumor-to-normal tissue contrast is highest between 4 and 10 minutes after injection, with a 61% reduction in mean tumor uptake at 90 minutes after injection.

Across the first four hours post-injection, 3% of administered radioactivity is excreted in the urine. Across the first 24 hours post-injection, 5% of administered radioactivity is excreted in the urine.

It should be noted that the pharmacokinetics of ^{18}F-fluciclovine is unknown in patients with bladder cancer at this time, and may not follow the distribution pattern described above. While we do not anticipate significant differences for bladder cancer patients, appropriate adjustments to the imaging parameters will be made as necessary if this pattern is dramatically different on initial studies.

8.1.2 Form

^{18}F-fluciclovine is supplied as a clear, colorless injection in a ready to use syringe that contains a single patient-specific dose of ^{18}F-fluciclovine at calibration time and date. ^{18}F-fluciclovine is supplied by PETNET for Blue Earth Diagnostics Ltd. Prior to study opening, Blue Earth Diagnostics will work with Dana Farber Cancer Institute and PETNET to set up a study-specific account for this trial. This account should be used for ordering doses for the study. The BED study number (BED-IIT-355) should referenced when ordering doses.

8.1.3 Storage and Stability

^{18}F-fluciclovine is stored at controlled room temperature (USP) 20°C to 25°C (68°F to 77°F). ^{18}F-fluciclovine does not contain a preservative. ^{18}F-fluciclovine is stored within the original container in radiation shielding.

8.1.4 Compatibility

^{18}F-fluciclovine is being administered in this study in tracer quantities for diagnostic imaging purposes. No drug interactions are expected between ^{18}F-fluciclovine and other drug regimens that each patient may be receiving.

8.1.5 Handling

^{18}F-fluciclovine should be handled and administered only by nuclear medicine personnel trained to handle radioactive material.

8.1.6 Availability

^{18}F-fluciclovine will be provided by Blue Earth Diagnostics Ltd as part of a research agreement for this study.

8.1.7 Administration

10 millicurie (mCi, ± 20%) of ^{18}F-fluciclovine will be administered via slow push over 10 seconds intravenously. A peripheral IV line is preferred. A butterfly line or established IV line may be used as long as the patency is established. The use of a power port needs to be approved by the study team.

8.1.8 Ordering

The study investigators will order the ^{18}F-fluciclovine from PETNET per institutional policies for the ordering of radiopharmaceuticals. Prior to study opening, Blue Earth Diagnostics will work with Dana Farber Cancer Institute and PETNET to set up a study-specific account for this trial. This account should be used for ordering doses for the study. The BED study number (BED-IIT-355) should referenced when ordering doses.

8.1.9 Accountability

A computer-based system for logging receipt and administration of radiopharmaceuticals per DFCI Imaging/Nuclear Medicine will be used for the accountability of the unit doses of the study radiopharmaceutical, ^{18}F-fluciclovine, received from Blue Earth Diagnostics, Ltd.

8.1.10 Destruction and Return

Unused ^{18}F-fluciclovine will be disposed of following institutional policies for the disposal of radioactive waste.

BIOMARKER, CORRELATIVE, AND SPECIAL STUDIES

Correlation of imaging data with pathological specimens obtained at the time of radical cystectomy will be performed as described in Sections 2.5, 5.3 and 13.2.

10. STUDY CALENDAR

Phase	Pre-study	Study day	After Study
		Within 6 weeks of Diagnostic CT/MRI	
Obtain informed consent	x		
Review inclusion/exclusion criteria	x		
Physical exam[1]	x		
Review of prior/concomitant medications[1]	x	x	
Qualitative β-hCG	x[5]		
Height/weight[2]		x	
^{18}F-Fluciclovine PET/CT		x	
Diagnostic CT/ MRI	x[1]		
Collection of surgical specimens			x[6]
Review adverse events[4]	x	x	
[1]Physical exams, review of prior/concomitant medications, serum creatinine, serum or urine β-hCG at baseline, vital signs as part of the physical exam, and diagnostic CT/MRI obtained pre-study are part of the standard-of-care for patients with bladder cancer. We will be collecting and reviewing the data from these standard procedures to compare to ^{18}F-fluciclovine PET/CT imaging			

results. Those with ECOG performance status ≤2 (Karnofsky ≥60%, see Appendix A) will be eligible. A urine pregnancy test will be performed where indicated.
[2]Height and weight will be obtained as part of the PET/CT scan procedures.
[4]Adverse events will be collected from the time of administration of ^{18}F-fluciclovine until 24 hours after ^{18}F-fluciclovine-PET/CT imaging.
[5]Female participants of childbearing potential must have a negative qualitative pregnancy test (β-hCG) at the Study Day. Results of a standard-of-care β-hCG may be used for the injection of ^{18}F-fluciclovine if performed within 24 hours prior. Females who are two years post tubal ligation or postmenopausal for at least one year are not considered to be of childbearing potential and do not need to undergo pregnancy testing.
[6] This is part of the standard-of-care radical cystectomy.

MEASUREMENT OF EFFECT

11.1 Pathological staging of bladder cancer

Pathological staging of bladder cancer is not the primary objective of this study. It will be used for comparison and correlation with imaged-based staging on ^{18}F-fluciclovine PET/CT and standard-of-care MRI and diagnostic CT.

DATA REPORTING / REGULATORY REQUIREMENTS

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

12.1 Data Reporting

12.1.1 Method

The study team in the Department of Imaging will collect, manage, and perform quality checks on the data for this study.

The following databases will be utilized:

· Partners' Research Patient Data Registry (RPDR) – Clinical database that indexes results from the Partners' hospitals, including BWH. This will be used to augment the pathology and laboratory data from the CRIS database.

· Epic – Epic will be used to collect patient medical history as detailed in the "Methods of Data Collection" section below.

· Centricity Patient Archiving and Communication Systems (PACS) – Radiology system for SOC image storage as well as only anatomical imaging analysis on fluciclovine PET/CT studies. This system will be used by the study radiologists to collect and analyze subjects' imaging data and gather tumor measurements.

· Hermes PACS – DFCI Imaging PACS for all of Nuclear Medicine, PET/CT, and non-SOC data of all imaging modalities that can't be stored on Centricity. All fluciclovine project data, including data received from outside hospitals, will be placed into a dedicated archive on Hermes. This data will be read from Hermes storage using Hermes viewing/analysis software.

· DFCI Imaging PET/CT database – A database of patient information and scanning parameters. The database contains data for all PET and PET/CT acquisitions performed at DFCI Imaging. It will be used to retrieve this information when it is needed.

12.1.2 Responsibility for Data Submission

Investigative sites within DF/HCC or DF/PCC are responsible for submitting data and/or data forms to the Office of Data Quality (ODQ) in accordance with DF/HCC policies

12.2 Data Safety Monitoring

The DF/HCC Data and Safety Monitoring Committee (DSMC) will review and monitor toxicity and accrual data from this study. The committee is composed of medical oncologists, research nurses, pharmacists and biostatisticians with direct experience in cancer clinical research. Information that raises any questions about participant safety will be addressed with the Overall PI and study team.

The DSMC will review each protocol up to four times a year with the frequency determined by the outcome of previous reviews. Information to be provided to the committee may include: up-to-date participant accrual; current dose level information; DLT information; all grade 2 or higher unexpected adverse events that have been reported; summary of all deaths occurring within 30 days of intervention for Phase I or II protocols; for gene therapy protocols, summary of all deaths while being treated and during active follow-up; any response information; audit results, and a summary provided by the study team. Other information (e.g. scans, laboratory values) will be provided upon request.

13. STATISTICAL CONSIDERATIONS

13.1 Study Design, Objectives, and Endpoints

This is a prospective pilot study evaluating ^{18}F-fluciclovine-PET/CT for detecting metastatic disease in patients with cT2-T4N0M0 muscle invasive bladder cancer.

The primary objective of this study is to estimate the agreement rate of metastatic disease status between ^{18}F-fluciclovine-PET/CT and histopathology from radical cystectomy for detecting

locoregional metastatic disease in patients with cT2-T4N0M0 muscle invasive bladder cancer by conventional CT/MRI imaging. Secondary objectives include estimating of the rate of detection of suspected distant metastatic disease by ^{18}F-fluciclovine-PET/CT, analysis of the association of ^{18}F-fluciclovine uptake on PE/CT in primary bladder tumor with pathologic tumor grade/size after radical cystectomy, and analysis of ^{18}F-fluciclovine uptake on PET/CT to the presence/absence of a ASCT2 and LAT1 amino acid transporters in the resected primary bladder tumors.

The primary endpoints are metastatic disease status by ^{18}F-fluciclovine-PET/CT and pathologic stage at radical cystectomy. The agreement rate is defined as the number of scans that are considered positive metastatic disease by both ^{18}F-fluciclovine-PET/CT and histopathology and those considered negative metastatic disease by both ^{18}F-fluciclovine-PET/CT and histopathology, out of all scans performed. A positive ^{18}F-fluciclovine-PET/CT is defined as evidence of metastatic disease either regionally in the pelvic lymph nodes or distantly to bone, lungs, viscera, or lymph nodes outside of the pelvis. The ability of ^{18}F-fluciclovine-PET/CT for detecting locoregional metastatic disease will be defined as the presence of disease detected locoregionally on ^{18}F-fluciclovine-PET/CT. Secondary endpoints include visualization (present/absent) and number of distant metastases on ^{18}F-fluciclovine-PET/CT. The amount of ^{18}F-fluciclovine accumulation in the primary bladder tumor on PET will be quantified using the maximum standardized uptake value (SUVmax). Tumor grade/size and the presence of amino acid transporters will be assessed histopathologically with specimens from cystectomy as part of standard-of-care.

13.2 Data Analysis Plan

The primary and first secondary analyses will include all 16 eligible and treated patients with completed ^{18}F-fluciclovine-PET/CT scans and pathology dissection. Among these 16 patients, those who undergo cystectomy will be included for secondary analysis 2 and 3.

We will tabulate the metastatic disease status by ^{18}F-fluciclovine-PET/CT (limited to bladder, locoregional nodes positive, or distant) and pathologic stage at radical cystectomy and compute the agreement rate, along with the corresponding 90% exact binomial confidence interval (CI). The proportion of patients who show positive ^{18}F-fluciclovine-PET/CT will be computed, along with the corresponding 90% exact binomial confidence interval. This similar methodology will be used to evaluate the proportion of agreement compared to histopathology and the rates of locoregional disease and suspected distant metastatic disease detected by ^{18}F-fluciclovine-PET/CT. In addition, treating pathologic and/or biopsy findings as the reference standard, we will estimate the sensitivity and specificity of ^{18}F-fluciclovine-PET/CT for detection of metastatic disease.

The analysis of the relationship of ^{18}F-fluciclovine uptake on PET/CT (SUVmax) in primary bladder tumor to pathologic tumor grade/size after radical cystectomy will be analyzed using the Kruskal-Wallis test. The Wilcoxon-Rank-Sum test will be used to assess the association between SUVmax and the presence/absence of ASCT2 and LAT1 amino acid transporters in the resected primary bladder tumors.

13.3 Sample Size, Accrual Rate and Study Duration

The planned sample size is 16 participants with completed ^{18}F-fluciclovine-PET/CT scans and pathology dissection. Patients who did not complete the scans or without pathology dissection are unevaluable and will be replaced. Considering 4 patients who may be unevaluable and replaced, up to 20 patients can be enrolled. The estimated accrual rate is 1 participant/month, and accrual is expected to complete in another 16 months. Participants who undergo ^{18}F-fluciclovine-PET/CT but have altered surgical plans due to clinical findings during surgery may remain evaluable and not replaced if a lymph node dissection was performed as determined by the study investigators. This will not affect the primary study objective given the primary objective is the agreement rate between PET/CT and pathology for metastases. Participants who do not undergo cystectomy will not be included for secondary objectives (2 and 3).

We will employ a Simon's 2-stage design to test the hypothesis that the agreement rate of metastatic disease status between ^{18}F-fluciclovine-PET/CT and histopathology will be 75%, compared to a null agreement rate of 50%. In the first stage we will enroll 10 patients, and if 6 or more scans among those 10 are in agreement regarding metastatic disease status, we will open the second stage of accrual to enroll an additional 6 patients, making a total sample size of 16 patients. If fewer than 6 scans are in agreement in the first stage then the study may be stopped or the study design may be amended to enroll additional patients. In order to consider the study a success, a total of 11 or more scans need to be in agreement among all 16 scans. This design has a 0.62 probability of stopping early under the null, and has 79.9% power while maintaining an overall one-sided type I error rate of 0.101. The agreement rate between ^{18}F-fluciclovine-PET/CT and histopathology is expected to be 75% (or higher) and the corresponding 90% CI will be (0.52, 0.91).

Accrual Targets					
Ethnic Category	**Sex/Gender**				
	Females		**Males**		**Total**
Hispanic or Latino		+		=	
Not Hispanic or Latino		+		=	
Ethnic Category: Total of all subjects	(A1)	+	(B1)	=	(C1)
Racial Category					
American Indian or Alaskan Native		+		=	
Asian		+		=	
Black or African American		+		=	
Native Hawaiian or other Pacific Islander		+		=	
White		+		=	

Racial Category: Total of all subjects	(A2)	+	(B2)	=	(C2)
	(A1 = A2)		(B1 = B2)		(C1 = C2)

13.4 Stratification Factors

N/A

13.5 Analysis of Primary Endpoints

See section 13.1

13.6 Analysis of Secondary Endpoints

Secondary objectives are to:

1. To determine the rate of detection of suspected distant metastatic disease by ^{18}F-fluciclovine-PET/CT in patients with cT2-T4N0M0 muscle invasive bladder cancer by conventional CT/MRI imaging.

Endpoints:
1) Visualization and number of distant metastases on ^{18}F-fluciclovine-PET/CT

Visualization of distant metastases on ^{18}F-fluciclovine-PET/CT will be binary-categorized as present/absent. In the categorization, a qualitative scale (0-4) may be accounted and an optimal cut-off may be examined. We will compute sensitivity to compare ^{18}F-fluciclovine-PET/CT with standard imaging modalities for distant metastases. The number of distant metastases will be descriptively shown by imaging modalities.

2. To correlate ^{18}F-fluciclovine uptake on PET/CT in primary bladder tumor with pathologic primary tumor stage and size after radical cystectomy.

Endpoints:
- ^{18}F-fluciclovine SUVmax in primary bladder tumor
- Primary tumor stage and size at radical cystectomy

The amount of ^{18}F-fluciclovine accumulation in the primary bladder tumor on PET will be quantified using the maximum standardized uptake value (SUVmax). Tumor grade will be assessed histopathologically with specimens from cystectomy as part of standard-of-care. The correlations between ^{18}F-fluciclovine uptake and tumor grade will be evaluated using Spearman correlation coefficients.

3. To correlate ^{18}F-fluciclovine uptake on PET/CT to the presence/absence of ASCT2 and LAT1 amino acid transporters in the resected primary bladder tumors.

Endpoints:
- ^{18}F-fluciclovine SUVmax in primary bladder tumor
- Presence/absence of ASCT2 and LAT1 amino acid transporter in resected primary bladder tumors

The Wilcoxon-Rank-Sum test will be used to assess the association between SUVmax and the presence/absence of a ASCT2 and LAT1 amino acid transporters in the resected primary bladder tumors.

13.7 Reporting and Exclusions

13.7.1 Evaluation of Toxicity

All participants will be evaluable for toxicity from the time of administration of ^{18}F-fluciclovine.

13.7.2 Evaluation of the Primary Efficacy Endpoint

All participants in who undergo ^{18}F-fluciclovine-PET/CT and radical cystectomy will be evaluable for the primary study endpoint.

14. PUBLICATION PLAN

This is a pilot study and the results will be prepared for publication after all study data answering a specific endpoint has been collected. The plan for publication may first be in abstract form followed by full manuscript publication. The Principal Investigator is primarily responsible for the publication of the study results.

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APPENDIX A PERFORMANCE STATUS CRITERIA

ECOG Performance Status Scale		**Karnofsky Performance Scale**	
Grade	Descriptions	Percent	Description
0	Normal activity. Fully active, able to carry on all pre-disease performance without restriction.	100	Normal, no complaints, no evidence of disease.
		90	Able to carry on normal activity; minor signs or symptoms of disease.
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).	80	Normal activity with effort; some signs or symptoms of disease.
		70	Cares for self, unable to carry on normal activity or to do active 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.	60	Requires occasional assistance, but is able to care for most of his/her needs.
		50	Requires considerable assistance and frequent medical care.
3	In bed >50% of the time. Capable of only limited self-care, confined to bed or chair more than 50% of waking hours.	40	Disabled, requires special care and assistance.
		30	Severely disabled, hospitalization indicated. Death not imminent.
4	100% bedridden. Completely disabled. Cannot carry on any self-care. Totally confined to bed or chair.	20	Very sick, hospitalization indicated. Death not imminent.
		10	Moribund, fatal processes progressing rapidly.
5	Dead.	0	Dead.

APPENDIX B AXUMIN PACKAGE INSERT

HIGHLIGHTS OF PRESCRIBING INFORMATION

These highlights do not include all the information needed to use AXUMIN safely and effectively. See full prescribing information for AXUMIN.

AXUMIN (fluciclovine F 18) injection, for intravenous use
Initial U.S. Approval: 2016

----------INDICATIONS AND USAGE----------

Axumin is a radioactive diagnostic agent indicated for positron emission tomography (PET) imaging in men with suspected prostate cancer recurrence based on elevated blood prostate specific antigen (PSA) levels following prior treatment (1).

----------DOSAGE AND ADMINISTRATION----------

- Use appropriate radiation safety handling measures (2.1).
- Aseptically withdraw Axumin from its container and administer 370 MBq (10 mCi) as a bolus intravenous injection. (2.2).
- Initiate imaging 3-5 minutes after administration. Scanning should start from mid-thigh and proceed to base of skull, with a total scan time of approximately 20-30 minutes (2.4).
- The (radiation absorbed) effective dose associated with 370 MBq (10 mCi) of injected activity of Axumin is approximately 8 mSv (0.8 rem) in an adult (2.6).

----------DOSAGE FORMS AND STRENGTHS----------

Injection: clear, colorless solution in a 30 mL multiple-dose vial containing 335-8200 MBq/mL (9-221 mCi/mL) fluciclovine F 18 at calibration time and date (3).

----------CONTRAINDICATIONS----------

None (4)

----------WARNINGS AND PRECAUTIONS----------

- Image interpretation errors can occur with Axumin imaging (5.1).
- Radiation risk: Axumin contributes to a patient's long-term cumulative radiation exposure. Ensure safe handling to protect patients and health care workers from unintentional radiation exposure (2.1, 5.3).

----------ADVERSE REACTIONS----------

Most commonly reported adverse reactions are injection site pain, erythema, and dysgeusia (6.1).

To report SUSPECTED ADVERSE REACTIONS, contact Blue Earth Diagnostics, Ltd at 1-855-AXUMIN1 (1-855-298-6461) or FDA at 1-800-FDA-1088 or www.fda.gov/medwatch.

See 17 for PATIENT COUNSELING INFORMATION

Revised: 8/2016

FULL PRESCRIBING INFORMATION: CONTENTS*

*Sections or subsections omitted from the full prescribing information are not listed.

FULL PRESCRIBING INFORMATION

1 INDICATIONS AND USAGE

Axumin is indicated for positron emission tomography (PET) in men with suspected prostate cancer recurrence based on elevated blood prostate specific antigen (PSA) levels following prior treatment.

2 DOSAGE AND ADMINISTRATION

2.1 Radiation Safety - Drug Handling

Axumin is a radioactive drug and should be handled with appropriate safety measures to minimize radiation exposure during administration [see *Warnings and Precautions (5.3)*]. Use waterproof gloves and effective shielding, including syringe shields, when handling and administering Axumin.

2.2 Recommended Dose and Administration Instructions

The recommended dose is 370 MBq (10 mCi) administered as an intravenous bolus injection.

- Inspect Axumin visually for particulate matter and discoloration before administration. Do not use the drug if the solution contains particulate matter or is discolored.
- Use aseptic technique and radiation shielding when withdrawing and administering Axumin.
- Calculate the necessary volume to administer based on calibration time and date, using a suitably calibrated instrument. The recommended maximum volume of injection of undiluted Axumin is 5mL.
- Axumin may be diluted with Sodium Chloride Injection, 0.9%.
- After the Axumin injection, administer an intravenous flush of sterile Sodium Chloride Injection, 0.9% to ensure full delivery of the dose.
- Dispose of any unused drug in a safe manner in compliance with applicable regulations.

2.3 Patient Preparation Prior to PET Imaging

- Advise the patient to avoid any significant exercise for at least one day prior to PET imaging.
- Advise patients not to eat or drink for at least 4 hours (other than small amounts of water for taking medications) prior to administration of Axumin.

2.4 Image Acquisition Guidelines

Position the patient supine with arms above the head. Begin PET scanning 3 to 5 minutes after completion of the Axumin injection. It is recommended that image acquisition should start from mid-thigh and proceed to the base of the skull. Typical total scan time is between 20 to 30 minutes.

2.5 Image Display and Interpretation

Localization of prostate cancer recurrence in sites typical for prostate cancer recurrence is based on fluciclovine F 18 uptake in comparison with tissue background. For small lesions (less than 1cm in diameter) focal uptake greater than blood pool should be considered suspicious for prostate cancer recurrence. For larger lesions, uptake equal to or greater than bone marrow is considered suspicious for prostate cancer recurrence.

2.6 Radiation Dosimetry

The radiation absorbed doses estimated for adult patients following intravenous injection of Axumin are shown in Table 1. Values were calculated from human biodistribution data using OLINDA/EXM (Organ Level Internal Dose Assessment/Exponential Modeling) software.

The (radiation absorbed) effective dose resulting from the administration of the recommended activity of 370 MBq of Axumin is 8 mSv. For an administered activity of 370 MBq (10 mCi), the highest-magnitude radiation doses are delivered to the pancreas, cardiac wall, and uterine wall: 38 mGy, 19 mGy, and 17 mGy, respectively. If a CT scan is simultaneously performed as part of the PET procedure, exposure to ionizing radiation will increase in an amount dependent on the settings used in the CT acquisition.

Table 1: Estimated Radiation Absorbed Doses in Various Organs/Tissues in Adults who Received Axumin

Organ/Tissue	Mean Absorbed Dose per Unit Administered Activity (microGy/MBq)
Adrenal glands	16
Brain	9
Breasts	14
Gallbladder wall	17
Lower large intestine wall	12
Small intestine wall	13
Stomach wall	14
Upper large intestine wall	13
Heart wall	52
Kidneys	14
Liver	33
Lungs	34
Muscle	11
Ovaries	13
Pancreas	102
Red bone marrow	25
Osteogenic cells	23
Skin	8
Spleen	24
Testes	17
Thymus gland	12
Thyroid	10
Urinary bladder wall	25
Uterus	45
Total body	13
Effective dose	**22 (microSv/MBq)**

3 DOSAGE FORMS AND STRENGTHS

Injection: supplied as a clear, colorless solution in a 30 mL multiple-dose vial containing 335 to 8200 MBq/mL (9 to 221 mCi/mL) flucicloviue F 18 at calibration time and date.

4 CONTRAINDICATIONS

None

5 WARNINGS AND PRECAUTIONS

5.1 Risk for Image Misinterpretation

Image interpretation errors can occur with Axumin PET imaging. A negative image does not rule out the presence of recurrent prostate cancer and a positive image does not confirm the presence of recurrent prostate cancer. The performance of Axumin seems to be affected by PSA levels *[See Clinical Studies (14)]*. Fluciclovine F 18 uptake is not specific for prostate cancer and may occur with other types of cancer and benign prostatic hypertrophy in primary prostate cancer. Clinical correlation, which may include histopathological evaluation of the suspected recurrence site, is recommended.

5.2 Hypersensitivity Reactions

Hypersensitivity reactions including anaphylaxis may occur in patients who receive Axumin. Emergency resuscitation equipment and personnel should be immediately available.

5.3 Radiation Risks

Axumin use contributes to a patient's overall long-term cumulative radiation exposure. Long-term cumulative radiation exposure is associated with an increased risk for cancer. Ensure safe handling to minimize radiation exposure to the patient and health care providers *[see Dosage and Administration (2.1)]*.

6 ADVERSE REACTIONS

Clinical Trials Experience

Because clinical trials are conducted under widely varying conditions, adverse reaction rates observed in the clinical trials of a drug cannot be directly compared to rates in the clinical trials of another drug and may not reflect the rates observed in practice. The clinical trial database for Axumin includes data from 877 subjects including 797 males diagnosed with prostate cancer. Most patients received a single administration of Axumin, a small number of subjects (n = 50) received up to five administrations of the drug. The mean administered activity was 370 MBq (range, 163 to 485 MBq).

Adverse reactions were reported in <1% of subjects during clinical studies with Axumin. The most common adverse reactions were injection site pain, injection site erythema and dysgeusia.

8 USE IN SPECIFIC POPULATIONS

8.1 Pregnancy

Risk Summary

Axumin is not indicated for use in females and there is no information on the risk of adverse development outcomes in pregnant women or animals with the use of fluciclovine F 18.

8.2 Lactation

Risk Summary

Axumin is not indicated for use in females and there is no information of the presence of fluciclovine F 18 in human milk.

8.3 Pediatric Use

Safety and effectiveness have not been established in pediatric patients.

8.4 Geriatric Use

Of the total number of patients in clinical studies of Axumin, the average age was 66 years with a range of 21 to 90 years. No overall differences in safety or effectiveness were observed between older subjects and younger subjects.

10 OVERDOSAGE

In case of overdose of Axumin, encourage patients to maintain hydration and to void frequently to minimize radiation exposure.

11 DESCRIPTION

11.1 Chemical Characteristics

Axumin contains the fluorine 18 (F 18) labeled synthetic amino acid analog fluciclovine. Fluciclovine F 18 is a radioactive diagnostic agent used with PET imaging. Chemically, fluciclovine F 18 is (1r, 3r)-1-amino-3[^{18}F]fluorocyclobutane-1-carboxylic acid. The molecular weight is 132.1 and the structural formula is:

Axumin is a sterile, non-pyrogenic, clear, colorless, hyperosmolal (approximately 500 - 540 mOsm/kg) injection for intravenous use. Each milliliter contains up to 2 micrograms of fluciclovine, 335 to 8200 MBq (9 to 221 mCi) fluciclovine F 18 at calibration time and date, and 20 mg trisodium citrate in water for injection. The solution also contains hydrochloric acid, sodium hydroxide and has a pH between 4 and 6.

11.2 Physical Characteristics

Fluorine 18 (F 18) is a cyclotron produced radionuclide that decays by positron emission (ß+ decay, 96.7%) and orbital electron capture (3.3%) to stable oxygen 18 with a physical half-life of 109.7 minutes. The positron can undergo annihilation with an electron to produce two gamma rays; the energy of each gamma ray is 511 keV (Table 2).

Table 2: Principal Radiation Produced from Decay of Fluorine 18 Radiation

	Energy (keV)	Abundance (%)
Positron	249.8	96.7
Gamma	511.0	193.5

11.3 External Radiation

The point source air-kerma coefficient for F 18 is 3.75×10^{-17} Gy m^2/(Bq s). The first half-value thickness of lead (Pb) for F 18 gamma rays is approximately 6 mm. The relative reduction of radiation emitted by F 18 that results from various thicknesses of lead shielding is shown in Table 3. The use of 8 cm of Pb will decrease the radiation transmission (i.e., exposure) by a factor of about 10,000.

Table 3: Radiation Attenuation of 511 keV Gamma Rays by Lead Shielding

Shield Thickness cm of Lead (Pb)	Coefficient of Attenuation
0.6	0.5
2	0.1
4	0.01
6	0.001
8	0.0001

12 CLINICAL PHARMACOLOGY

12.1 Mechanism of action

Flucicloviне F 18 is a synthetic amino acid transported across mammalian cell membranes by amino acid transporters, such as LAT-1 and ASCT2, which are upregulated in prostate cancer cells. Flucicloviне F 18 is taken up to a greater extent in prostate cancer cells compared with surrounding normal tissues.

12.2 Pharmacodynamics

Following intravenous administration, the tumor-to-normal tissue contrast is highest between 4 and 10 minutes after injection, with a 61% reduction in mean tumor uptake at 90 minutes after injection.

12.3 Pharmacokinetics

Distribution

Following intravenous administration, flucicloviне F 18 distributes to the liver (14% of administered activity), pancreas (3%), lung (7%), red bone marrow (12%) and myocardium (4%). With increasing time, flucicloviне F 18 distributes to skeletal muscle.

Excretion

Across the first four hours post-injection, 3% of administered radioactivity was excreted in the urine. Across the first 24 hours post-injection, 5% of administered radioactivity was excreted in the urine.

13 NONCLINICAL TOXICOLOGY

13.1 Carcinogenesis, Mutagenesis, Impairment of Fertility

Carcinogenesis

No long term studies in animals have been performed to evaluate the carcinogenic potential of flucicloviне.

Mutagenesis

Flucicloviне was not mutagenic *in vitro* in reverse mutation assay in bacterial cells and in chromosome aberration test in cultured mammalian cells, and was negative in an *in vivo* clastogenicity assay in rats after intravenous injection of doses up to 43 mcg/kg. However, flucicloviне F 18 has the potential to be mutagenic because of the F 18 radioisotope.

Impairment of Fertility

No studies in animals have been performed to evaluate potential impairment of fertility in males or females.

14 CLINICAL STUDIES

The safety and efficacy of Axumin were evaluated in two studies (Study 1 and Study 2) in men with suspected recurrence of prostate cancer based on rising PSA levels following radical prostatectomy and/or radiotherapy.

Study 1 evaluated 105 Axumin scans in comparison to histopathology obtained by biopsy of the prostate bed and biopsies of lesions suspicious by imaging. PET/CT imaging generally included the abdomen and pelvic regions. The Axumin images were originally read by on-site readers. The images were subsequently read by three blinded independent readers. Table 4 shows the performance of Axumin in the detection of recurrence in each patient scan and, specifically, within the prostatic bed and extra-prostatic regions, respectively. The results of the independent read were generally consistent with one another and confirmed the results of the on-site reads.

Table 4: Performance of Axumin in Patients with Biochemically Suspected Recurrent Prostate Cancer, at the Patient Level and at the Prostate Bed and Extraprostatic Region Levels

	Reader 1	Reader 2	Reader 3
Patient	N = 104	N = 105	N = 99
True Positive	75	72	63
False Positive	24	23	13
True Negative	5	7	15
False Negative	0	3	8
Prostate Bed	N = 98	N = 97	N = 96
True Positive	58	56	47
False Positive	29	26	15
True Negative	10	12	24
False Negative	1	3	10
Extraprostatic	N = 28	N = 28	N = 25
True Positive	25	26	22
False Positive	2	2	2
True Negative	0	0	0
False Negative	1	0	1

N = number of patient scans evaluated

The detection rate of Axumin seems to be affected by PSA levels *[see Warnings and Precautions (5.1)]*. In general, patients with negative scans had lower PSA values than those with positive scans. The detection rate (number with positive scans/total scanned) for patients with a PSA value of less than or equal to 1.78 ng/mL (1st PSA quartile) was 15/25, of which 11 were histologically confirmed as positive. In the remaining three PSA quartiles, the detection rate was 71/74, of which 58 were histologically confirmed. Among the 25 patients in the first PSA quartile, there were 4 false positive scans and 1 false negative scan. For the 74 patients with PSA levels greater than 1.78 ng/mL, there were 13 false positive scans and no false negative scans.

Study 2 evaluated the concordance between 96 Axumin and C11 choline scans in patients with median PSA value of 1.44 ng/mL (interquartile range 0.78 to 2.8 ng/mL). The C 11 choline scans were read by on-site readers. The Axumin scans were read by the same three blinded independent readers used for Study 1. The agreement values between the Axumin and C11 choline reads were 61%, 67% and 77%, respectively.

16 HOW SUPPLIED/STORAGE AND HANDLING

16.1 How Supplied

Axumin is supplied as a clear, colorless injection in a 30 mL multiple-dose glass vial containing approximately 26 mL solution of 335-8200 MBq/mL (9-221 mCi/mL) fluciclovine F 18 at calibration time and date.

30 mL sterile multiple-dose vial: NDC 69932-001-30

16.2 Storage and Handling

Store Axumin at controlled room temperature (USP) 20°C to 25°C (68°F to 77°F). Axumin does not contain a preservative. Store Axumin within the original container in radiation shielding.

This preparation is approved for use by persons under license by the Nuclear Regulatory Commission or the relevant regulatory authority of an Agreement State.

17 PATIENT COUNSELING INFORMATION

- Instruct patients to avoid significant exercise for at least a day before the PET scan.
- Instruct patients not to eat or drink for at least 4 hours before the PET scan (other than small amounts of water for taking medications).

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