Evaluation of Endometriosis with 18F-fluoroestradiol PET / MRI

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Evaluation of Endometriosis with 18F-fluoroestradiol PET / MRI

Drug Name(s): 18F-fluoroestradiol (FES)

Sponsor: The University of North Carolina at Chapel Hill

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Amendment 3 Date:

Amendment 4 Date:

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PROTOCOL TITLE:

Evaluation of Endometriosis with 18F-fluoroestradiol PET / MRI

Short Title: FES PET/MRI of Endometriosis

Lead Investigator:

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University of North Carolina at Chapel Hill

Protocol Version: 1.2

Version Date: August 27, 2020

The signature below constitutes the approval of this protocol and the attachments, and provides the necessary assurances that this trial will be conducted according to all stipulations of the protocol, including all statements regarding confidentiality, and according to local legal and regulatory requirements and applicable U.S. federal regulations and ICH guidelines.

Principal Investigator Name: _____

Principal Investigator Signature: _____

Date: _____

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ABBREVIATIONS AND DEFINITIONS OF TERMS

Abbreviation	**Definition**
FES	Fluoro-Estradiol
PET	Positron Emission Tomography
MRI	Magnetic Resonance Imaging
PACS	Picture Archiving and Communication System

PROTOCOL SYNOPSIS

Study Title	Evaluation of Endometriosis with 18F-fluoroestradiol PET / MRI
Funder	The University of North Carolina
Clinical Phase	Pilot Study
Study Rationale	Current techniques to diagnose endometriosis (apart from laparoscopy) lack the necessary sensitivity to detect the disease. As endometriosis expresses estrogen receptors, a radiotracer that binds to estrogen receptors may be able to detect it in a noninvasive fashion.
Study Objective(s)	The primary objective is to evaluate the sensitivity and specificity of 18F-fluoroestradiol (FES) PET/MR for evaluating endometriosis. Secondary objectives include comparing PET to conventional MRI, using histopathology from laparotomy as the gold standard and evaluating association of uptake values (SUV-max) with EHP-30 and pain rating scales, controlling for covariates.
Investigational Drug	This study will be an evaluation of the radiotracer, FES, which binds to estrogen receptors and has previously been used to study estrogen receptor expression in tumors, to detect endometriosis.
Study Design	This is a prospective, one arm, single center study of 12 subjects with clinically suspected endometriosis to demonstrate FES PET-MR's clinical utility for diagnosis of endometriosis.
Subject Population key criteria for Inclusion and Exclusion:	**Inclusion Criteria** • Age 18 or older • Clinically suspected endometriosis. • Scheduled for planned operative laparoscopy with no hormone treatment for at least two cycles (Note: if women are amenorrhoeic, 56 days will be used as a substitute for 2 cycles.) • Able to provide informed consent

	Exclusion Criteria
	• Male patients
	• Patients under 18
	• Institutionalized subject (prisoner or nursing home patient)
	• Patients with known endometrial, breast, or ovarian cancer.
	• Pregnant or breast-feeding women
	  • Contraindication to MRI (pacemaker, metallic implants, metallic fragments or foreign bodies, body piercings that are unable to be removed, and certain cardiac devices) Subjects will be screened for any contraindications prior to the MRI.
	  • Claustrophobia that would prevent subjects from being inside the MRI scanner for the duration of the scan.

Number Of Subjects	A total of 12 participants will be recruited from individuals with clinically suspected endometriosis.
Study Duration	This study is anticipated to last approximately 1 year.
Statistical And Analytic Plan	The sensitivity of FES PET /MR is defined as the ability of readers (radiologists) to detect endometriosis in patients. Diagnostic accuracy will be defined by the histologic presence of endometriosis and by symptomatic improvement as defined clinically by the referring physician within 3 months of surgery.
DATA AND SAFETY MONITORING PLAN	The Principal Investigator will provide continuous monitoring of patient safety in this trial with periodic reporting to an independent Medical Monitor.

1. BACKGROUND AND RATIONALE

1.1 Introduction

Endometriosis is a major cause of infertility and pain for many women of childbearing age. Estimates of the frequency of endometriosis in infertile women range from 20-50%, with an estimated overall prevalence of 0.8-6% in the female population. [1]

Imaging is often used for evaluation of endometriosis. Endometriosis is divided by location into ovarian, peritoneal, and deep; ovarian endometriosis is located in the ovary (often known as endometrioma), peritoneal endometriosis is in the peritoneum penetrating less than 5 mm, and a deep infiltrating endometriotic (DIE) lesion penetrates into the retroperitoneal space or pelvic organ walls to at least 5 mm, affecting 4-37% of women with endometriosis. Transvaginal ultrasound is usually the first-line imaging modality used, with MRI as a second line to look for deep implants.[2] In general, these are both quite effective, with transvaginal ultrasound being effective for genital DIE and MRI for the diagnosis of pelvic genital and extragenital DIE; however, even MRI can have difficulty distinguishing between surgical scar and active lesions if the patient has been previously operated, and questions still remain about its efficacy in evaluating nerves and bowel.[3]

While DIE is often amenable to detection by conventional methods, more superficial lesions, even those with extensive coverage of the pelvic endometrium, are frequently invisible to currently-used imaging techniques.[4] The lack of non-surgical diagnostic or even screening tests is one factor leading to the average five to nine-year delay between symptom onset and diagnosis.[5,6] The ability to detect earlier-stage endometriosis could play a key role in preventing unnecessary surgery on the one hand and prevention of progression on the other.[7] The overlap in symptoms between endometriosis and other causes of chronic pelvic pain also makes recurrence difficult to assess, leading to potentially preventable repeat surgery.[7] Finally, without non-surgical diagnostic techniques, innovations in medical therapy are very difficult to develop. These factors have led to an international consensus that non-invasive diagnostic testing is a key research priority for the field.[7]

One potential option for imaging endometriosis is a radiotracer, 16α-[18F]fluoro-17β-estradiol or more commonly 18F-fluoroestradiol (FES). This involves an estradiol molecule which has been labeled with a radioactive fluorine atom, which binds to estrogen receptors and can be detected by positron emission tomography (PET). The tracer correlates with receptor expression in biopsy material[8,9], and can be administered with minimal (<50 micrograms) estrogenic material[10] and doses similar to those in other nuclear medicine studies.[11–13] This tracer has already been demonstrated for use with estrogen-receptor positive breast, endometrial, and ovarian cancers, correlating with expression in tumors, response to endocrine therapy, assessment of tumor burden and heterogeneity of disease, and pharmacodynamics.[14] Other applications have included differentiating endometrial hyperplasia from endometrial cancer and differentiating

benign uterine leiomyomas from malignant uterine sarcomas.[14] Given that endometriosis contains estrogen receptors, we hope to use FES to identify endometriosis using non-invasive imaging approaches.

1.2 Existing work with FES

Current use of FES: At the present time, FES has only been used to find endometriosis in one study of 4 patients with extragenital endometriosis, specifically of the deep infiltrating variety. This study showed multiple lesions detectable on FES-PET that were not detectable on MRI; of the 9 lesions found by histology, 4 were found by MRI, 8 by FES-PET, and the histology agreed with PET in all cases.[3] There was also a limited relationship between SUVmax and chronic pelvic pain.[3] Furthermore, this study was performed using PET/CT, which increases the radiation exposure to patients relative to PET/MR.

Our innovation: Previous work with FES has involved breast cancer or other malignancies which express estrogen receptors, such as endometrial cancer. We are hoping to build on prior work by Cosma et al.[3] which shows that endometriosis, which also displays endometrial receptors, can be detected by FES-PET.

In addition, we will utilize PET/MR rather than PET/CT, to allow us to avoid the radiation dose associated with the CT. This is particularly important in women of childbearing age. In addition, the superior soft tissue contrast of MRI vis-à-vis CT can help to localize lesions in the pelvis. Furthermore, we will utilize novel MR imaging approaches to potentially enhance the sensitivity of MR-only studies to endometriosis. Utilizing PET/MR will also improve the co-registration for the development of MR based approaches. If endometriosis could be reliably detected through noninvasive imaging, infertility patients who did not display FES uptake, particularly those with prior surgical history who might be difficult to stage using MRI, could avoid the expense and pain of exploratory laparotomy.

2. STUDY OBJECTIVES

The aim of this study is to demonstrate FES PET/MR's clinical utility for diagnosis of endometriosis.

2.1 Primary Objectives

Aim 1: The primary objective is to evaluate the sensitivity and specificity of FES PET/MR for evaluating endometriosis.

2.2 Secondary Objectives

Aim 2: To compare PET to conventional MRI, using histopathology from laparotomy as the gold standard.

Aim 3: To evaluate the association of uptake values (SUV-max) with EHP-30 and pain rating scales, controlling for BMI, race, age, and physician.

3. INVESTIGATIONAL PLAN

3.1 Study Design

This is a prospective, one arm, single center study of 12 subjects with clinically suspected endometriosis to demonstrate PET-MR's clinical utility for diagnosis of endometriosis.

3.2 Study Duration, Enrollment and Number of Subjects

We will include a total of 12 participants in this trial. These subjects will be followed for up to 3 months after initial imaging with PET-MR.

3.3 Study Population

3.3.1 Inclusion Criteria

- Age 18 or older
- Female of childbearing age
- Clinically suspected (symptomatic) endometriosis.
- Scheduled for planned operative laparoscopy with no hormone treatment for at least two cycles (Note: if women are amenorrhoeic, 56 days will be used as a substitute for 2 cycles.)
- Able to provide informed consent

3.3.2 Exclusion Criteria

- Male
- Institutionalized subject (prisoner or nursing home patient)
- Known history of breast, ovarian or endometrial cancer.
- Pregnant or breast-feeding women
- Contraindication to MRI (pacemaker, metallic implants, metallic fragments or foreign bodies, body piercings that are unable to be removed, and certain cardiac devices) Subjects will be screened for any contraindications prior to the MRI.
- Claustrophobia that would prevent subjects from being inside the MRI scanner for the duration of the scan.

4. STUDY PROCEDURES

Patients with suspected (but not confirmed) endometriosis and suspicion of extragenital DIE who are scheduled for planned operative laparoscopy with no hormone treatment for at least two cycles will be recruited from the Minimally Invasive Gynecological Surgery Clinic (Director, E. Carey, Co-I) at UNC. If women are amenorrhoeic, 56 days will be used as a substitute for 2 cycles. The Endometriosis Health Profile-30 (EHP-30), a quality of life questionnaire used in women with endometriosis, a pain numeric rating scale, and information about the last menstrual period will be obtained. Patients will undergo FES PET/MR within 4 weeks prior to surgery. Prior to surgery, suspected areas of endometriosis will be identified on imaging. These imaging findings will then be

compared to surgical findings and histopathology from exploratory laparotomy. Surgical documentation of endometriosis location will rely on published, consensus staging methods.[15,16] Surgeons will not see the results from the FES-PET/MRI prior to surgery. The results of FES-PET MRI will not be used to guide surgery. The surgeons will be performing exploratory laparoscopy as per standard of care.

4.1 Screening/Baseline Visit procedures

A total of 12 participants will be enrolled to this study. The study subjects will be consecutively recruited from individuals that have clinically suspected endometriosis and have planned exploratory laparoscopy. Eligible patients will be identified by research staff review in coordination with the UNC MIGS group.

Once a patient has been referred, the patient will be approached by a coordinator from Radiology to assess interest in participation.

All eligible participants who agree to participate in the study will be asked to come to their scheduled appointment thirty minutes early to complete the informed consent process.

Review of the consent will take place in the privacy of an exam room, or when possible, a sample consent form will be sent to the patient via email prior to the patient's visit to allow for ample review.

Patients will complete the EHP-30 and pain scale questionnaire. In addition, patients will be asked the date of their last menstrual period. These questionnaires may be completed in person, via the phone, or via REDCap, whichever is preferred by the participant.

4.2 Research Imaging Procedure

Participants who consent for the study will be escorted by the research coordinator to the dedicated study room for the imaging exam. For our female patients who are of child bearing age, a pregnancy test will be performed prior to the administration of the radiotracer.

The subject will have the PET-MR scan performed in a similar manner to a conventional clinical PET-MR scans. The imaging agent will be injected approximately one hour prior to imaging after dosimetry and radiotracer has been verified per protocol. The research technologist will assist in positioning the patient in the PET-MR unit. Once positioned, the total scan time is approximately 1 hour. The length of time for the positioning and examination of a subject's pelvis may vary but it is expected that the entire visit will take about 150 minutes. We will not utilize gadolinium contrast agent for this study.

We have extensive experience in the synthesis of radiotracers at the UNC Biomedical Research Imaging Center (BRIC), with dedicated radiochemists and radiopharmacists. We thus expect to be able to synthesize the radiotracer without much difficulty, as there is an established IND available for cross-filing on the NCI website [17], which contains a

full set of manufacturing and QC documents. A manufacturing IND has already been obtained.

4.3 Follow-Up Phone Call
Patients will be called 3 to 4 weeks following the FES PET scan to determine the last menstrual period following the scan.

4.4 Medical Record Abstraction
Participants' medical record will be reviewed for up to 3 months following their initial research imaging to meet the primary aim. The operative surgeon will be asked to fill out a specific post-operative form defining the areas in which they identified visible endometriosis within the pelvis.

4.5 Interpretation
Upon completion of all study image data collection, a reader study will be performed with a fellowship-trained abdominal radiologist and nuclear radiologist (separate individuals). The abdominal and nuclear reads will be performed separately, without discussion between radiologists. MR information will be used for anatomic localization of the PET scan, as is commonly performed. Differences between the two will then be discussed in the eventual publication. All images will be reconstructed using standard algorithms, and nuclear medicine studies presented on MIM workstations for nuclear medicine viewing, with PACS viewing software with conventional zoom/pan/window and level tools. Locations of endometriosis will be recorded with the same classification system as the surgical system. This research classification will be made by radiologists who are blinded to the surgical findings of endometriosis.

4.6 Variables of Interest
Primary Variables of Interest:

- Presence of endometriosis based on imaging (true/false)
- Localization of endometriosis (American Society for Reproductive Medicine Revised Classification of Endometriosis 1997 and World Endometriosis Foundation System)[15]
- Standardized Uptake values (positive real number)
- EHP-30 value (integer)
- Pain rating (integer)

5. STATISTICAL CONSIDERATION
5.1 Primary Endpoint
The sensitivity of FES PET /MR is defined as the ability of readers (radiologists) to detect endometriosis in patients. Diagnostic accuracy will be defined by the histologic presence of endometriosis and by symptomatic improvement as defined surgically by the referring physician within 3 months of surgery.

5.2 Statistical Methods

Subjects will be recruited until 12 subjects complete the research imaging. For subjects that are enrolled and do not complete the research PET/MR, they will be considered a screen fail and replaced. Two radiologists, a nuclear radiologist and an abdominal radiologist, will be recruited to conduct the reader study. Both the radiologists and surgeons will utilize the Revised Endometriosis Classification System.

Sensitivity and specificity of 18F-fluoroestradiol (FES) PET/MR for evaluating endometriosis will be computed by comparing imaging against surgical findings and histopathology in order to answer Aim 1. We will compute point estimates, along with 95% confidence intervals and precision estimates that may be used to plan future studies.

We will address Aim 2 in the following fashion. After imaging, we will compare the PET results against conventional MRI, using histopathology obtained at exploratory laparotomy as the gold standard. We will do this in 2 phases, first, at the patient level, and then, as an ancillary analysis, at the endometriotic implant site level. For each phase, we will report sensitivity, specificity, and accuracy of the PET results, along with associated 95% confidence intervals. We will also report sensitivity, specificity, and accuracy, along with associated 95% confidence intervals, using conventional MRI. We will use McNemar's test, constructing the 2x2 table among all patients with a surgical diagnosis of positive to compare sensitivity for PET versus conventional MRI. Similarly, we will use McNemar's test among all patients with a surgical diagnosis of negative to compare specificity for PET versus conventional MRI. P-values <0.05 for the comparison of sensitivity and specificity will be construed as evidence that sensitivity/specificity differ between PET and conventional MRI. P-values >0.05 will be considered inconclusive.

To evaluate Aim 3, whether uptake values are associated with EHP-30 and pain rating scales, we will implement a random effects linear regression model, modeling SUV-max as a function of EHP-30 and the pain rating, while controlling for patient-level covariates (BMI, race, age). We will include physician as a random effect to account for physician-level correlation. P-values <0.05 for the EHP-30 and pain rating parameter estimates will be taken as evidence of association with uptake values. P-values >0.05 will be considered inconclusive.

As a sensitivity analysis for Aim 3, we will compute Cook's distance (D) for all observations, remove observations where D >1, and recalculate the above specified model, comparing model conclusions to the original analysis.

Budgetary constraints limit sample size for this study. For Aim 2 and Aim 3, precision of estimators will be low, power levels of tests will be low, and confidence intervals will be wide. However, study results are expected to be useful in a grant proposal for a future study.

5.3 Sample Size Rationale

With 12 patients, we will have 80% power at alpha=.05 to detect an odds ratio of 21.91 for comparing sensitivities between PET/MRI versus conventional MRI. The detectable OR will be >10000000 for comparing specificities between PET/MRI versus conventional MRI.

Given our analysis strategy, McNemar's test, the following information was needed for the power calculation for detecting differences in sensitivity between 2 modalities: the total number of patients and the anticipated proportion of discordant pairs. We used 12 for the number of patients due to budget constraints for the pilot study. The proportion of discordant pairs is the percentage of test results that do not agree between PET/MRI and conventional MRI. We estimated this to be 55% based on published studies about MRI and the Cosmal et al. study. From this, we estimated an odds ratio of 21.91. Power calculations are made using GPower, version 3.1.9.2.

5.4 Interim Analysis

Interim analyses will not be conducted.

6. ADVERSE EVENTS (DRUGS- CONTRAST AGENTS)

6.1 Definitions

6.1.1 Adverse Event (AE)

An adverse event (AE) is any untoward medical occurrence (e.g., an abnormal laboratory finding, symptom, or disease temporally associated with the use of a drug) in a patient or clinical investigation subject administered a pharmaceutical product and which does not necessarily have a causal relationship with this treatment. An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal product, whether or not related to the medicinal product.

Hospitalization for elective surgery or routine clinical procedures that are not the result of an AE (e.g., surgical insertion of central line) need not be considered AEs and should not be recorded as an AE. Disease progression should not be recorded as an AE, unless it is attributable by the investigator to the study therapy.

6.1.2 Suspected Adverse Reaction (SAR)

A suspected adverse reaction (SAR) is any AE for which there is a reasonable possibility that the drug is the cause. Reasonable possibility means that there is evidence to suggest a causal relationship between the drug and the AE. A suspected adverse reaction implies a lesser degree of certainty about causality than adverse reaction, which means any adverse event caused by a drug.

Causality assessment to a study drug is a medical judgment made in consideration of the following factors: temporal relationship of the AE to study drug exposure, known mechanism of action or side effect profile of study treatment, other recent or

concomitant drug exposures, normal clinical course of the disease under investigation, and any other underlying or concurrent medical conditions. Other factors to consider in considering drug as the cause of the AE:

Single occurrence of an uncommon event known to be strongly associated with drug exposure (e.g., angioedema, hepatic injury, Stevens-Johnson Syndrome)

One or more occurrences of an event not commonly associated with drug exposure, but otherwise uncommon in the population (e.g., tendon rupture); often more than once occurrence from one or multiple studies would be needed before the sponsor-investigator could determine that there is reasonable possibility that the drug caused the event.

An aggregate analysis of specific events observed in a clinical trial that indicates the events occur more frequently in the drug treatment group than in a concurrent or historical control group

6.1.3 Unexpected AE or SAR

An AE or SAR is considered unexpected if the specificity or severity of it is not consistent with the applicable product information (e.g., Investigator's Brochure (IB) for an unapproved investigational product or package insert/summary of product characteristics for an approved product). Unexpected also refers to AEs or SARs that are mentioned in the IB as occurring with a class of drugs or as anticipated from the pharmacological properties of the drug, but are not specifically mentioned as occurring with the particular drug under investigation.

6.1.4 Serious AE or SAR

An AE or SAR is considered serious if, in the view of the sponsor- investigator, it results in any of the following outcomes:

- Death;
- Is life-threatening (places the subject at immediate risk of death from the event as it occurred);
- Requires inpatient hospitalization (>24 hours) or prolongation of existing hospitalization;
- Results in congenital anomaly/birth defect;
- Results in a persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions;
- Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered a serious adverse drug experience when, based upon appropriate medical judgment, they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in the definition. For reporting purposes, also consider the occurrences of pregnancy as an event which must be reported as an important medical event**.**

Pregnancy that occurs during the study must also be reported as an SAE.

6.2 Documentation of non-serious AEs or SARs
For non-serious AEs or SARs, documentation must begin from day 1 of study treatment and continue through the 30-day follow-up period after treatment is discontinued.

Collected information should be recorded in the Case Report Forms (CRF) for that patient. Please include a description of the event, its severity or toxicity grade, onset and resolved dates (if applicable), and the relationship to the study drug. Documentation should occur at least monthly.

6.3 SAEs or Serious SARs
6.3.1 Timing
After informed consent but prior to initiation of study medications, only SAEs caused by a protocol-mandated intervention will be collected (e.g. SAEs related to invasive procedures such as biopsies, medication washout.

For any other experience or condition that meets the definition of an SAE or a serious SAR, recording of the event must begin from day 1 of study treatment and continue through the 30 day follow-up period after treatment is discontinued.

6.3.2 Documentation and Notification
These events (SAEs or Serious SARs) must be recorded within 24 hours of learning of its occurrence.

6.3.3 Reporting
IRB Reporting Requirements:

The UNC-IRB will be notified of all SAEs that qualify as an Unanticipated Problem as per the UNC IRB Policies using the IRB's web-based reporting system within 7 days of the Investigator becoming aware of the problem.

<u>Pregnancy</u>

Pregnancies and suspected pregnancies (including a positive pregnancy test regardless of age or disease state) of a female subject occurring while the subject is on study should be recorded as SAEs. The patient is to be discontinued immediately from the study. The female subject should be referred to an obstetrician-gynecologist, preferably one experienced in reproductive toxicity for further evaluation and counseling.

The Investigator will follow the female subject until completion of the pregnancy, and must document the outcome of the pregnancy (either normal or abnormal outcome). If the outcome of the pregnancy was abnormal (e.g., spontaneous or therapeutic abortion), the Investigator should report the abnormal outcome as an AE. If the abnormal outcome meets any of the serious criteria, it must be reported as an SAE.

Funding Source (e.g. Manufacturer) Reporting Requirements:

If an investigator deems that an event is both a serious SAR AND unexpected, it must also (in addition to REDCap) be recorded on the MedWatch Form 3500A as per 21 CFR 312.32. The MedWatch 3500a form can be accessed at: http://www.fda.gov/Safety/MedWatch/HowToReport/DownloadForms/default.htm. (Please be sure and access form 3500a, and not form 3500). The Sponsor-investigator of the study will make the final determination regarding FDA submission**.**

Once the UNC Principal Investigator determines an event is a serious SAR AND unexpected, the MedWatch 3500A form will be submitted to the FDA.

7. Data and Safety Monitoring Plan

The Principal Investigator will provide continuous monitoring of patient safety in this trial with periodic reporting to an independent Medical Monitor. The medical monitor will review any reported Unanticipated Adverse Drug Effects after patients 5 and 10.

Meetings/teleconferences will be held at a frequency dependent on study accrual, and in consultation with the study Biostatistician. These meetings will include the investigators and any other relevant personnel the principal investigators may deem appropriate. At these meetings, the research team will discuss all issues relevant to study progress, including enrollment, safety, regulatory, data collection, etc.

The team will produce summaries or minutes of these meetings. These summaries will be available for inspection when requested by any of the regulatory bodies charged with the safety of human subjects and the integrity of data including, but not limited to, the oversight (Office of Human Research Ethics (OHRE) Biomedical IRB, the Scientific Review Committee (SRC), the Office of Clinical Trials (OCT), or the North Carolina TraCS Institute Data and Safety Monitoring Board (DSMB).

The PI will be responsible for submitting the following information for review by the independent medical monitor: 1) safety and accrual data including the number of study participants imaged; 2) significant developments reported in the literature that may affect the safety of participants or the ethics of the study; 3) preliminary response data; and 4) summaries of team meetings that have occurred since the last report. Findings of the medical monitor review will be disseminated by memo.

7.1 Adverse Events Safety Assessments

A licensed provider will observe subject vitals following the administration of the radiotracer on through the completion of the imagining scan. Study personnel will contact subjects 1 day (+3 days window) after to inquire about delayed onset complications. The subjects will also be given a phone number that they can call to reach a nurse if they believe they have developed a complication of the procedure.

8. STUDY MANAGEMENT

8.1 Institutional Review Board (IRB) Approval and Consent

It is expected that the IRB will have the proper representation and function in accordance with federally mandated regulations. The IRB should approve the consent form, assent form, and protocol.

In obtaining and documenting informed consent, the investigator should comply with the applicable regulatory requirement(s), and should adhere to Good Clinical Practice (GCP) and to ethical principles that have their origin in the Declaration of Helsinki.

Before recruitment and enrollment onto this study, the patient will be given a full explanation of the study and will be given the opportunity to review the consent form. Each consent form must include all the relevant elements currently required by the FDA Regulations and local or state regulations. Once this essential information has been provided to the patient and the investigator is assured that the patient understands the implications of participating in the study, the patient will be asked to give consent to participate in the study by signing an IRB-approved consent form.

Prior to a patient's participation in the trial, the written informed consent form should be signed and personally dated by the patient and by the person who conducted the informed consent discussion.

For patients under the age of 18, appropriate assent must be obtained in addition to parental permission prior to participation in the study.

8.2 Registration Procedures
Study participants will be registered into CRMS, a web based clinical research platform by one of the Study Coordinators.

8.3 Data Management and Monitoring/Auditing
The FES PET-MR scan that is obtained of all eligible enrolled subjects will be de-identified for inclusion in the appropriate readers study. Copies of the clinical report forms as well as the de-identified images described in the preceding will be submitted for each case to the Study Coordinators for maintaining the study record and entering data in preparation for the reader study.

The online REDCap software system provided by UNC's TraCS Institute will be used to collect and store research data. Information regarding why a data value is missing will be documented in the study database.

Coded copies of subject study images will be received at the time of the scan and will be stored in the participant file in a locked file cabinet.

8.4 Adherence to the Protocol
Except for an emergency situation in which proper care for the protection, safety, and well-being of the study patient requires alternative treatment, the study shall be conducted exactly as described in the approved protocol.

8.5 Emergency Modifications

UNC investigators may implement a deviation from, or a change of, the protocol to eliminate an immediate hazard(s) to trial subjects without prior UNC's IRB/IEC approval/favorable opinion.

For any such emergency modification implemented, an IRB modification form must be completed by UNC Research Personnel within five (5) business days of making the change.

8.6 Protocol Deviations/Violations

According to UNC's IRB, a protocol deviation is any unplanned variance from an IRB approved protocol that:

- Is generally noted or recognized after it occurs
- Has no substantive effect on the risks to research participants
- Has no substantive effect on the scientific integrity of the research plan or the value of the data collected
- Did not result from willful or knowing misconduct on the part of the investigator(s).

An unplanned protocol variance is considered a violation if the variance meets any of the following criteria:

- Has harmed or increased the risk of harm to one or more research participants.
- Has damaged the scientific integrity of the data collected for the study.
- Results from willful or knowing misconduct on the part of the investigator(s).
- Demonstrates serious or continuing noncompliance with federal regulations, State laws, or University policies.

If a deviation or violation occurs please follow the guidelines below:

Protocol Deviations: UNC personnel will record the deviation and report to any data and safety monitoring committee in accordance with their policies. Deviations should be summarized and reported to the IRB according to the UNC IRB reporting requirements.

Protocol Violations: Violations should be reported by UNC personnel within one (1) week of the investigator becoming aware of the event using the same IRB online mechanism used to report Unanticipated Problems.

Unanticipated Problems:

Any events that meet the criteria for "Unanticipated Problems" as defined by UNC's IRB must be reported by the study team using the IRB's web-based reporting system.

8.7 Amendments to the Protocol

Should amendments to the protocol be required, the amendments will be originated and documented by the Principal Investigator at UNC. It should also be noted that when an amendment to the protocol substantially alters the study design or the potential risk to the patient, a revised consent form might be required.

The written amendment, and if required the amended consent form, must be sent to UNC's IRB for approval prior to implementation.

8.8 Record Retention
Study documentation includes all eCRFs, data correction forms or queries, source documents, Sponsor-Investigator correspondence, monitoring logs/letters, and regulatory documents (e.g., protocol and amendments, IRB correspondence and approval, signed patient consent forms).

Source documents include all recordings of observations or notations of clinical activities and all reports and records necessary for the evaluation and reconstruction of the clinical research study.

Government agency regulations and directives require that all study documentation pertaining to the conduct of a clinical trial must be retained by the study investigator. In the case of a study with a drug seeking regulatory approval and marketing, these documents shall be retained for at least two years after the last approval of marketing application in an International Conference on Harmonization (ICH) region. In all other cases, study documents should be kept on file until three years after the completion and final study report of this investigational study.

8.9 Obligations of Investigators
The Principal Investigator is responsible for the conduct of the clinical trial at the site in accordance with Title 21 of the Code of Federal Regulations and/or the Declaration of Helsinki. The Principal Investigator is responsible for personally overseeing the treatment of all study participants. The Principal Investigator must assure that all study site personnel, including sub-investigators and other study staff members, adhere to the study protocol and all FDA/GCP/NCI regulations and guidelines regarding clinical trials both during and after study completion.

8.10 Conflict of Interest
Any investigator who has a conflict of interest (COI) with this study as defined by the policies of the University of North Carolina will have the conflict reviewed by a properly constituted Conflict of Interest Review Committee with a committee-sanctioned conflict management plan that has been reviewed and approved by the IRB prior to participation in this study. All University of North Carolina investigators will follow the University conflict of interest policy.

9. PLANS FOR PUBLICATION
Study results will be submitted to a peer-reviewed journal for publication. This study will also be listed on Clinicaltrials.gov and study results will be posted in accordance with appropriate regulations and ICJME requirements.

Neither the complete nor any part of the results of the study carried out under this protocol will be published or passed on to any third party without the consent of the study sponsor-investigator. Any investigator involved with this study will be obligated to

provide the sponsor-investigator with complete results and all data derived from the study.

10. REFERENCES

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11. APPENDIX

Appendix A: FES

Most of this information is taken directly from the NCI Investigator's Brochure.

11.1 Chemical name

16α-[18F]-fluoro-17β-estradiol (FES)

11.2 Pharmacology and toxicology

16α-[18F]-fluoro-17β-estradiol (FES) is a radiopharmaceutical designed for imaging estradiol binding to estrogen receptors (ERs) *in vivo*. Its molecular weight is 290.4 Daltons. FES has chemical properties very similar to estradiol. The relative binding affinity (RBA, FES/estradiol) for the estrogen receptor is 0.8[1]. The metabolism of FES and estradiol are similar[2, 3] with elimination primarily by conjugation in the liver, followed by renal clearance of the glucuronide. Measurements of the relative binding affinity for the blood transport protein, sex hormone binding globulin (SHBG)[4], was 10% relative to estradiol[5]. An average of 45% of circulating FES is bound to SHBG, similar to estradiol[6]. In the sections that follow, the pharmacology and toxicity of estradiol in addition to FES is reviewed.

FES is produced with a specific activity greater than 170 Ci/mmol and the injected mass dose is less than or equal to 5 μg (17 nmole). The requirement for high specific activity, low mass dose, assures that only a small fraction of the estrogen receptors (ER) are occupied during a PET imaging study. If the receptor approaches saturation, then FES uptake would no longer reflects receptor concentration. A 5-μg dose is far below any known toxicity for fluoroestradiol or other ER ligands.

The pharmacology of FES is best understood by analogy to estradiol. Estradiol is a naturally occurring steroid that comes from two sources: (1) synthesis in the ovary in pre-menopausal women and (2) conversion from adrenal steroids, largely through aromatization (and aromatase enzymes)[7, 8] in a variety of tissues, most notably fat, breast tissue, and breast cancers. Pre-menopausal levels of estradiol vary widely depending upon the phase of the menstrual cycle, reaching levels as high as 500 pg/ml (1.7 nM) mid-cycle. In post-menopausal women, and in men, levels are generally less than 30 pg/ml (0.1 nM).

Estradiol is very lipophilic and is generally present in slightly higher concentration in tissues with higher fat content. Circulating estradiol is largely protein bound with high affinity but low capacity to SHBG and with low affinity but high capacity to albumin[6, 9]. Much of circulating estradiol is bound to SHBG and the remainder is bound to albumin[6]. FES is an estrogen analog used as a diagnostic agent to image regional estradiol binding to ER and is closely related to estradiol[1]. Estradiol exerts its physiologic effect by binding to ER, a nuclear

receptor. ER is selectively expressed in a variety of tissues, most notably the breast, uterus, ovaries, bone, and pituitary.

Ovarian synthesis of estradiol is a key component of female endocrine function in a complex feedback loop with the pituitary. Estradiol also promotes new bone formation and is important in maintaining bone mineral density, especially in women. Estrogens affect the cardiovascular system, largely through their beneficial effect on serum lipids. In the breast, estradiol promotes ductal epithelial cell proliferation and is a key component stimulating lactation. Estrogens are established growth factors for endometrial and many breast cancers. Approximately 60% of breast cancers express ER, and estradiol and other estrogens provide a key stimulus for tumor growth and an opportunity for endocrine-based therapy. This last effect is the impetus for developing a diagnostic agent for imaging ER expression in breast cancer patients that led to our investigation of FES for use in PET diagnostic imaging[10].

FES metabolism has also been studied in humans. Metabolite analysis of blood and urine was performed in patients undergoing [^{18}F]FES PET studies[2]. Results were similar to rat data, showing that FES is rapidly metabolized to polar species, with less than 20% of blood radioactivity in the form of [^{18}F]FES by 60 minutes after injection. There is also net clearance of both FES and labeled metabolites from the blood via hepatic uptake, biliary excretion, and urinary excretion of polar conjugates. By 120 minutes after injection, circulating FES is less than 5% of peak values, and the total of FES and labeled metabolites is less than 40% of the peak. Clearance rates of intravenous FES and intravenous estradiol are similar; for both compounds circulating levels have decreased to less than 5% of peak levels by 60 minutes after injection. Analysis of metabolites excreted in the urine sampled 90 – 120 minutes after injection has been done using glucuronidases to dissociate the glucuronide conjugates and acid hydrolysis to break the sulfate conjugation bond. These experiments recovered mostly [^{18}F]FES, with a small percentage of a more polar substance not identified in the studies[2]. These results suggest that, on the time scale of PET imaging, FES is metabolized primarily to non-oxidized conjugated FES.

In summary, FES biochemistry, ER binding affinity, and metabolism are very similar to estradiol, suggesting that data on estradiol biochemistry and pharmacology are applicable to FES. Any differences between results for FES and estradiol appear to arise from the fact that only short-term (1 – 2 hours), transient kinetics and metabolism of the radiolabeled [^{18}F]FES are relevant to its use in PET. Studies of estradiol physiology suggest that exposures of several hours to days are needed to elucidate physiologic effects and thus longer-term, equilibrium kinetics and metabolism are most relevant. Because of the ^{18}F half-life limitation, oxidation plays only a minor role in [^{18}F]FES metabolism and liver conjugation is responsible for enterohepatic circulation and prompt excretion in urine over the life of ^{18}F.

Biodistribution studies in humans further support the concept that FES metabolism is similar to that of estradiol. The liver rapidly takes up FES with subsequent excretion into bile[2, 11] . From sequential images of the biodistribution of FES using PET, it was shown that FES passed into the bile and moved through the small intestine[11]. Very little, if any, radioactivity was seen in the large intestine, suggesting highly efficient enterohepatic circulation, similar to that of estradiol. Similar results were found using 16-α-radioiodo-17-β-estradiol in a swine model [12].

11.3 Toxicity of FES in humans

Estradiol is a naturally occurring substance with biochemical and pharmacologic properties nearly identical to FES. It is important to interpret toxicity data for FES relative to reported toxicity for estradiol in the context of the intended use of FES as a single-dose-administration agent for diagnostic imaging. In this setting, FES reaches physiologic levels (i.e., greater than post-menopausal levels) only on a transient basis. This must be viewed in the context of many years of exposure to physiologic levels of estradiol in most women. [^{18}F]FES could potentially exert toxic effects through one of three mechanisms: (1) radiation exposure to tissues from the radioactive label[11], (2) physiologic actions mediated through the ER, and (3) directly toxic or mutagenic effects of FES metabolites. Radiation exposure from [^{18}F]FES at activity doses used in PET (6 mCi, typical) is low, and is comparable to other nuclear medicine procedures[11]. Radiation exposure is discussed in detail in Section 10.4. With respect to the other two mechanisms of toxicity, FES injected as a bolus for PET imaging transiently reaches physiologic concentrations, but returns to sub-physiologic levels within an hour after injection. As such, toxic effects due to actions mediated through the ER and directly toxic effects of metabolites will be far less than those of natural ER ligands.

Given the biochemical and pharmacologic similarity between FES and estradiol, the low mass administered and short-term exposure to FES resulting from PET studies, the estradiol toxicity literature serves as an appropriate gauge for any potential toxicity of FES.

11.4 Dosimetry

The uptake of [^{18}F]FES in normal human tissues has been measured and used to estimate the radiation absorbed dose associated with the imaging procedure. Dosimetry studies were performed at the University of Washington and have been peer-reviewed and published in the *Journal of Nuclear Medicine*[11]. For more details, the reader is referred to the investigator's brochure at NCI.

Table 1. Human dosimetry estimates

(source: investigator's brochure)

Organ	Mean mrad/mCi	Mean mGy/MBq	SD* mGy/MBq	25% mGy/MBq	75% mGy/MBq
Adrenals	85	0.023	0.003	0.021	0.025
Brain	36	0.010	0.001	0.009	0.010
Breasts	32	0.009	0.002	0.008	0.010
Gall Bladder Wall	379	0.102	0.041	0.075	0.134
Lower Large Intestine	45	0.012	0.001	0.011	0.013
Small Intestine	99	0.027	0.015	0.017	0.038
Stomach	50	0.014	0.001	0.013	0.014
Upper Large Intestine	110	0.030	0.016	0.019	0.042
Heart Wall	96	0.026	0.004	0.024	0.029
Kidney	128	0.035	0.004	0.032	0.038
Liver	466	0.126	0.030	0.105	0.149
Lungs	61	0.017	0.002	0.015	0.018
Muscle	79	0.021	0.001	0.021	0.022
Ovaries	66	0.018	0.002	0.016	0.019
Pancreas	84	0.023	0.002	0.021	0.024
Red Marrow	48	0.013	0.002	0.012	0.014
Bone Surface	53	0.014	0.001	0.014	0.015
Skin	18	0.005	0.000	0.005	0.005
Spleen	54	0.015	0.003	0.012	0.017
Testes	44	0.012	0.001	0.011	0.012
Thymus	50	0.014	0.001	0.013	0.014
Thyroid	45	0.012	0.001	0.012	0.013
Urinary Bladder Wall	186	0.050	0.020	0.036	0.066
Uterus	145	0.039	0.013	0.031	0.049
Lens	33	0.009	0.000	0.009	0.009

***SD = Standard Deviation**

Effective Dose Equivalent = 0.022 mSv/MBq (0.004 SD). For a 6 mCi (222 MBq) dose, this would be about 4.8 mSv.

11.5 Previous FES human imaging studies

Early studies established a correlation between FES uptake and *in vitro* assay of ER expression[13] and documented the biodistribution and radiation dosimetry of FES[11]. Several studies documented the metabolism, clearance, and serum protein binding of FES in humans[5, 11]. Other studies demonstrated heterogeneous uptake of FES in advanced breast cancer as a reflection of heterogeneous ER expression[14, 15]. One paper measured FES uptake in meningiomas[16] Finally, other studies have measured FES uptake in patients treated with hormonal therapy[17, 18, 19]. The general conclusion from the studies summarized above is that [^{18}F]FES PET images identified estrogen receptor

positive tissue that was heterogeneously distributed within human tumors. These data may be helpful in identifying patients who will benefit from endocrine therapy for their cancer and predict the likelihood of response to specific treatment hormonal regimens.

In a paper published in 2010, Tsujikawa et al[20] reported on the correlation between the uptake of 16α-[^{18}F]fluoro-17β-estradiol (FES) and expression of estrogen receptors, as well as other related immunohistochemistry markers. Nineteen patients with endometrioid adenocarcinoma underwent preoperative PET studies with FES and FDG. Standardized uptake values (SUVs) for each tracer and the regional FDG to FES SUV ratio were calculated using images after coregistration. FES uptake showed a significantly positive correlation with expression of estrogen receptor α (ERα). The FDG to FES ratio showed a significantly negative correlation with expression of ERα and progesterone receptor B (PR-B). The FES uptake and FDG to FES ratio did not correlate with expression of ERβ, Ki-67 or glucose transporter 1 (GLUT1). FDG uptake was not correlated with any of the immunohistochemical scores. The PR-B score was strongly correlated with the ERα score. Well-differentiated carcinoma (grade 1) showed a significantly higher FES uptake and significantly lower FDG to FES ratio than moderately or poorly differentiated carcinoma (grade 2 – 3). None of the PET parameters were significantly different between advanced-stage carcinoma (≥ stage IB) and early-stage carcinoma (IA) based on the Féderation International de Gynécologie et d'Obstétrique (FIGO) staging classification. Differentiation grade was the most closely correlated parameter to FES uptake and FDG to FES ratio by multivariate analyses. The authors concluded FES PET combined with FDG would be useful for non-invasive evaluation of ERα distribution, as well as ERα function, which reflects differentiation grade in endometrial carcinoma[20].

In a study of 16 female healthy volunteers published in 2007, Tsuchida et al[21] administered a single dose of FES to investigate the relationship between endometrial and myometrial FES uptake and menstrual phase or endogenous estrogen level.

Endometrial SUV was significantly higher in the proliferative phase than in the secretory phase (6.03 ± 1.05 vs. 3.9 ± 1.29, *P* = .022). In contrast, there was no significant difference in myometrial SUV when the proliferative and secretory phases were compared (*P* = .23). Further, there was no correlation between SUV and endogenous estrogen level in the proliferative phase. The authors concluded that the change of ER concentration relative to menstrual cycle as characterized by FES PET was consistent with those from previous reports that used an immunohistochemical technique. These data suggest that FES PET is a feasible, noninvasive method for characterizing changes in ER concentration[21].

In a study published in 2008 by Tsujikawa et al[22] FES and FDG PET studies were performed in 38 patients with benign and malignant uterine tumors to compare differences in tracer accumulation. Regional values of tracer uptake were

evaluated by using standardized uptake value (SUV). Patients with endometrial carcinoma showed significantly greater mean SUV for FDG (9.6 ± 3.3) than for FES (3.8 ± 1.8) (*P* < .005). Patients with endometrial hyperplasia showed significantly higher mean SUV for FES (7.0 ± 2.9) than for FDG (1.7 ± 0.3) (*P* < .05). Patients with leiomyoma showed significantly higher mean SUV for FES (4.2 ± 2.4) than for FDG (2.2 ± 1.1) (*P* < .005), and patients with sarcoma showed opposite tendencies for tracer accumulation. Tracer uptake in patients with endometrial carcinoma was significantly higher for FDG (*P* < .001) and significantly lower for FES (*P* < .05) when compared with values in patients with endometrial hyperplasia. On the other hand, patients with sarcoma showed a significantly higher uptake for FDG (*P* < .005) and a significantly lower uptake for FES (*P* < .05) compared with patients with leiomyoma. The authors concluded that ER expression and glucose metabolism of uterine tumors measured by using PET showed opposite tendencies, and that PET studies utilizing both FES and FDG could provide pathophysiologic information for the differential diagnosis of uterine tumors. These results demonstrate the potential predictive capability of FES PET[22].

In another study by Peterson et al published in 2008[23], [^{18}F]fluoroestradiol uptake was compared with ER expression assayed *in vitro* by immunohistochemistry (IHC) with both qualitative and semiquantitative measures. Seventeen patients with primary or metastatic breast cancer were studied with dynamic [^{18}F]FES PET; cancer tissue samples, collected close to the time of imaging, were assayed for ER expression by IHC. For each tumor, partial-volume-corrected measures of [^{18}F]FES uptake were compared with ER expression measured by three different ER scoring methods: qualitative scoring (0 – 31), the Allred score (0 – 10), and a computerized IHC index. The authors noted that there was excellent agreement (r^2 = 0.99) between observers using IHC as well as the different methods of measuring ER content (*P* < 0.001), and they concluded that there is good agreement between [^{18}F]FES PET and ER expression measured by IHC, and that [^{18}F]FES imaging may be a useful tool for aiding in the assessment of ER status, especially in patients with multiple tumors or for tumors that are difficult to biopsy[23].

In a study by Dehdashti et al published in 2009[24], 51 post-menopausal women with advanced estrogen receptor positive breast cancer were studied. Patients underwent FES PET and FDG PET at baseline and repeat FDG PET after 30 mg estradiol. Tracer uptake was measured as the standardized uptake value (SUV). Patients were subsequently treated with either an aromatase inhibitor or fulvestrant. PET results were correlated with responsiveness to endocrine therapy. Per study criteria, 17 patients responded and 34 patients did not respond to endocrine therapy. Four responders and one non-responder had a clinical flare reaction, while only the responders demonstrated metabolic flare. After estradiol challenge, a significantly higher mean (± SD) percent change in SUV for FDG was noted in responders (20.9 ± 24.2) compared with non-

responders (-4.3 ± 11.0, $P < 0.0001$). On FES PET, a higher tumor SUV was noted in responders (3.5 ± 2.5) compared with non-responders (2.1 ± 1.8, $P = 0.0049$). There was significantly longer overall survival in patients with metabolic flare than in those without flare regardless of type of endocrine therapy ($P = 0.0062$). The authors concluded that baseline tumor FES uptake and metabolic flare after an estradiol challenge are both predictive of responsiveness to endocrine therapy in ER+ breast cancer[24].

In a study by Kumar et al published in 2007[25] an improved automated radiosynthesis methodology for [^{18}F]FES was developed. Stability studies of the resulting injectable form were performed up to 24 hours after dose formulation under normal storage conditions. A comparison of FES versus FDG PET imaging was then conducted in ER+ breast cancer patients. The results of the improved synthesis methodology were favorable and the subsequent PET imaging suggested specificity of FES for ER+ tumors versus FDG[25].

In a 2008 paper Tsujikawa et al[26] reported two postmenopausal patients under suspicion of endometrial carcinoma on the basis of cytology and/or magnetic resonance imaging (MRI), who were on tamoxifen treatment since undergoing surgery for breast cancer. Pelvic MRI suggested endometrial carcinomas, whereas FDG and FES-PET showed no abnormal tracer accumulation. A postoperative histopathologic examination revealed that the lesions were endometrial hyperplasias with no malignant findings. They concluded that FES PET enabled them to evaluate endometrial ER expression noninvasively. The evaluation of ER expression using FES PET requires careful attention regarding the influence of hormonal therapy because tamoxifen greatly affects FES accumulation of even endometrial hyperplasia, which should be an FES-avid lesion[26].

Another study published by Tsujikawa et al in 2009[27] investigated whether [^{18}F]FES and [^{18}F]FDG PET reflect clinic-pathologic features in patients with endometrial tumors[27]. A total of 22 patients with endometrial adenocarcinoma and nine with endometrial hyperplasia underwent [^{18}F]FES PET for estrogen receptor imaging and [^{18}F]FDG PET. The diagnostic accuracy of MRI findings for clinical staging was also compared. They found that although the SUV for [^{18}F]FDG was significantly lower in endometrial hyperplasia than in carcinoma, a statistically significant difference between high-risk and low-risk carcinoma was observed only in SUV for [^{18}F]FES. High-risk carcinoma showed a significantly greater [^{18}F]FDG to [^{18}F]FES ratio (3.6 ± 2.1) than did low-risk carcinoma (1.3 ± 0.5, $P < 0.01$) and hyperplasia (0.360.1, $P < 0.005$). Low-risk carcinoma showed a significantly higher [^{18}F]FDG to [^{18}F]FES ratio than hyperplasia ($P < 0.0001$). In receiver operating-characteristic (ROC) analysis, the most accurate diagnostic PET parameter for predicting high-risk and low-risk carcinoma was the [^{18}F]FDG to [^{18}F]FES ratio. The optimal [^{18}F]FDG/[^{18}F]FES cutoff value of 2.0, determined by ROC analysis, revealed 73% sensitivity, 100% specificity, and 86% accuracy, which was better than the 77% accuracy for MRI. The [^{18}F]FDG to [^{18}F]FES ratio

of 0.5 yielded a correct diagnosis for carcinoma from hyperplasia with 100% accuracy. They concluded that endometrial carcinoma reduces estrogen dependency with accelerated glucose metabolism as it progresses to a higher stage or grade, that the [^{18}F]FDG to [^{18}F]FES ratio reflects tumor aggressiveness, and that this index will be useful for making noninvasive diagnoses and deciding the appropriate therapeutic strategy for patients with endometrial carcinoma[27].

11.6 Reported adverse events and potential risks

Approximately 1500 subjects are represented in the published studies. Other than infrequent transient intravenous site discomfort and an “alcohol taste”, there have been no adverse events related to [^{18}F]FES administration. Although lab values have not been routinely measured pre- and post-FES PET scans as part of the PET procedure, many patients at the University of Washington have undergone serial measurements of renal and liver function, differential blood counts and assay of electrolytes as part of their clinical management. To estimate toxicity risk, 109 consecutive patients who underwent FES PET scans between 2002 and 2005 were examined. Of these 109, 30 patients had hematology and serum chemistry values measured both before FES PET scanning (median 16 days prior) and within 21 days after the infusion of [^{18}F]FES (median 10.5 days post scan).

Measurements of renal and liver function (serum creatinine, SGOT, SGPT, and alkaline phosphatase) showed no clinically significant changes pre- versus post-FES infusion in this group of patients. Three patients had elevated alkaline phosphatase prior to FES infusion, due to extensive bony metastatic disease, and these patients continued to have elevated levels post FES infusion with no clinically significant change. Differential blood counts (platelet counts, WBC, neutrophils, hemoglobin, and hematocrit) were examined. These showed a number of patients with abnormal blood values prior to FES PET scanning; however, this was expected in a heavily pre-treated population undergoing salvation therapy for metastatic breast cancer. There were no clinically significant changes in blood counts seen post-[^{18}F]FES infusion compared with the preimaging values.

11.7 FES Administered Dose

The [^{18}F]fluoroestradiol is a sterile, IV injectable solution with a volume of ≤ 20 ml containing 0.15 M phosphate buffered saline: < 15% ethanol (v:v). The injected dose of [^{18}F]FES is generally 6 mCi (185 MBq) with an allowable range of 3 to 6 mCi of [^{18}F]fluoroestradiol. The drug product solution is stored at room temperature in a gray butyl septum sealed, sterile, pyrogen-free glass vial with an expiration time of 8 hours. The mass of injected drug is ≤ 5 µg (≤ 17 nmol) of FES.

11.8 Agent Availability

[18F]FLT will be provided by the Biomedical Research Imaging Radiopharmaceutical Core under an IND held by the Cancer Imaging Program (CIP)/NCI.

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