**Abbreviated Title: [F-18]-FdCyd Imaging**
**NCI Protocol #: 8865**
**Local Protocol #: 12-C-0014**
**Amendment: G**
**CTEP Protocol #: 8865**
**Title: Phase 0 Trial of [F-18]-5-Fluoro-2'-Deoxycytidine with Tetrahydrouridine**

**Principal Investigator:**

**Karen A. Kurdziel, MD,**
Molecular Imaging Program (MIP)/CCR/NCI
Building 10/ B3B403
10 Center Drive
Bethesda, MD 20892
Tel: 301-443-0622
kurdziek@mail.nih.gov

**Associate Investigators:**

**Alice Chen, M.D.**
DTC/ NCI
31 Center Drive
Bldg 31, Room 3A44
Bethesda, MD 20892
Tel: 301-496-4291
Fax: 301-402-0172
chenali@mail.nih.gov

**Peter L. Choyke, M.D.**
MIP/CCR/NCI
Building 10/1B40
Bethesda, MD 20892
Tel: 301-451-4220
Fax: 301-402-3191
pchoyke@nih.gov

**James H. Doroshow, M.D.**
NCI/DCTD/OD
9000 Rockville Pike, Building 10/12N226
Bethesda, MD 20892
Tel: 301-496-4291
doroshoj@mail.nih.gov

**Maria Liza Lindenberg, M.D.**
MIP, CCR/NCI
Bldg 10/ RMB3B403
Bethesda, MD 20892
301-443-0604
lindenbergml@mail.nih.gov

**Responsible Research Nurse:**

**Yolanda McKinney, RN**
Office of the Clinical Director (OCD), CCR/NCI
Building 10/ Room B3B69E

Bethesda, MD 20892
301-443-6913
ymckinney@mail.nih.gov

**Statistician:** **Joanna H. Shih, Ph.D.**
Biometric Research Branch, DCTD/ NCI
9609 Medical Center Drive Room 4W230
Bethesda, MD 20850-9735
Phone: 240-276-6035
Fax: 240-276-7888
Jshih@mail.nih.gov

## NCI Supplied Investigational Agents:

| | |
|---|---|
| **Drug Name:** | **[F-18]-FdCyd Imaging (NSC 755309), Tetrahydrouridine (NSC 112907)** |
| **IND Number:** | **105843** |
| **Sponsor:** | **CTEP** |

**Identifying words: Imaging, radiopharmaceutical, safety, dosimetry, drug distribution**

## PRÉCIS

***Background:***

- In pre-clinical models, 5-fluoro-2'-deoxycytidine (FdCyd), administered along with tetrahydrouridine (THU; an inhibitor of cytidine/deoxycytidine deaminase), has shown superior anti-tumor activity as compared with 5-fluorouracil.
- FdCyd can be phosphorylated to 5-fluoro-2'-deoxycytidylate (FdCMP) by deoxycytidine kinase and the nucleotide deaminated to FdUMP by deoxycytidylate (dCMP) deaminase. The activity of dCMP deaminase is reported to be higher in human malignancies than in normal tissues, which may result in selective cytotoxicity.
- FdCyd is an inhibitor of DNA methyltransferase and DNA methylation, resulting in re-expression of genes silenced by DNA hypermethylation. It is being evaluated in a phase II multihistology clinical trial at the Developmental Therapeutics Clinic, NCI, Clinical Center, NIH.
- While FdCyd + THU has shown preliminary evidence of activity in early phase trials not all patients show clinical response. The establishment of a radiolabeled form to image the biodistribution *in vivo* at baseline and during therapy may provide insight into the distribution of the therapeutic drug.
- The first step in the development of such an *in vivo* marker is to determine the biodistribution and safety of the radiolabeled form.

***Objectives:***

- Determine the safety of [F-18]-5-fluoro-2'-deoxycytidine (FdCyd) administered intravenously with administration of tetrahydrouridine (THU).
- Estimate the radiation dosimetry of [F-18]-FdCyd in humans.

***Eligibility:***

- Only patients enrolled in NCI Phase II Study evaluating FdCyd with THU (NCI Protocol # 09-C-0214 (CTEP# 8351) or NCI Protocol #12-C-0066 (CTEP# 9127)) at the NIH Clinical Center will be eligible to participate in this study).
- Patients must have a target lesion ≥ 10mm
- May not be pregnant or lactating; must be ≤ 350 lbs; and may not have known allergy to FdCyd or contraindications to PET/CT imaging.

***Design*:**

- There are two arms to this study
    - The first arm will be patients enrolling in the therapeutic Phase II 5-FdCyd/THU study (NCI Protocol # 09-C-0214 (CTEP# 8351) in the NCI Developmental Therapeutics Clinic
    - The second arm will be patients enrolling in the Phase I 5-FdCyd/THU study (NCI Protocol #12-C-0066 (CTEP# 9127)) in the NCI Developmental Therapeutics Clinic.
- Patients will undergo an initial [F-18]-FdCyd + THU PET/CT imaging prior to therapeutic dosing on study NCI Protocol # 09-C-0214 (CTEP# 8351) or NCI Protocol

#12-C-0066 (CTEP# 9127). Repeat imaging will be performed while the patient is receiving FdCyd + THU therapy under the parent therapeutic protocol. This imaging must be completed 2-5 days after cycle start and at least 2 hours after a dose. Upon completion of repeat imaging, patients will be taken off this imaging study 24 hours after the last imaging session.

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# 1 INTRODUCTION

## 1.1 Study Objectives

1.1.1 Primary Objectives:

1.1.1.1 To determine the safety of [F-18]-5-fluoro-2'-deoxycytidine (FdCyd) administered intravenously with administration of tetrahydrouridine (THU)

1.1.1.2 To estimate the radiation dosimetry of [F-18]-FdCyd in humans

1.1.2 Secondary Objectives:

1.1.2.1 To evaluate the pharmacokinetics of [F-18]-FdCyd via PET/CT

1.1.2.2 To evaluate the distribution of FdCyd in tumorous and non-tumorous tissue

# 2 BACKGROUND AND RATIONALE:

5-fluoro-2'-deoxycytidine (FdCyd), administered along with tetrahydrouridine (THU; an inhibitor of cytidine/deoxycytidine deaminase), has shown superior anti-tumor activity as compared with 5-fluorouracil in pre-clinical models. Following administration of FdCyd, it is phosphorylated to 5-fluoro-2'-deoxycytidylate (FdCMP) by deoxycytidine kinase and the nucleotide deaminated to FdUMP by deoxycytidylate (dCMP) deaminase. The activity of dCMP deaminase is reported to be higher in human malignancies than in normal tissues, which may result in selective cytotoxicity. FdCyd is an inhibitor of DNA methyltransferase (DNMT) and DNA methylation, resulting in re-expression of genes silenced by DNA hypermethylation. Co-administration of THU inhibits the metabolism of the parent FdCyd allowing it to circulate in its original form, and intracellularly inhibit DNMT.

The process of intracellular activation creates a charged species that is trapped inside the cell, and since the sensitivity is directly linked to the amount of activated drug that collects inside the cell, the drug itself has very favorable properties as a probe for tumor localization. The biomolecular literature indicates that the pathways for intracellular uptake, retention, and activation of 5FdC to nucleotides are subject to substantial variation in human tumors. Thus, information that could guide the selection of tumors that were more likely to be sensitive or resistant to 5FdCyd would potentially aid in its development.

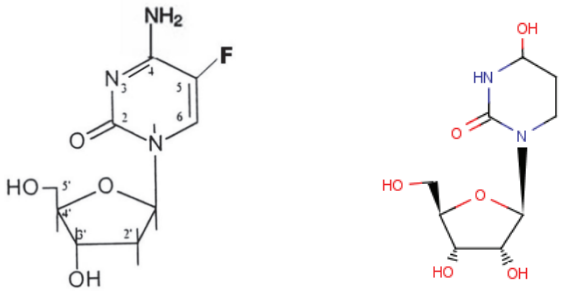



**Figure 1: NSC 48006, 5-F-deoxycytidine (5FdC) and Tetrahydrouridine (THU)**

In general, noninvasive imaging of many pyrimidines has been hindered by the high amounts of background radioactivity produced by catabolism. In the proposed study, co-administration of THU inhibits the catabolism of FdCyd reducing the background.

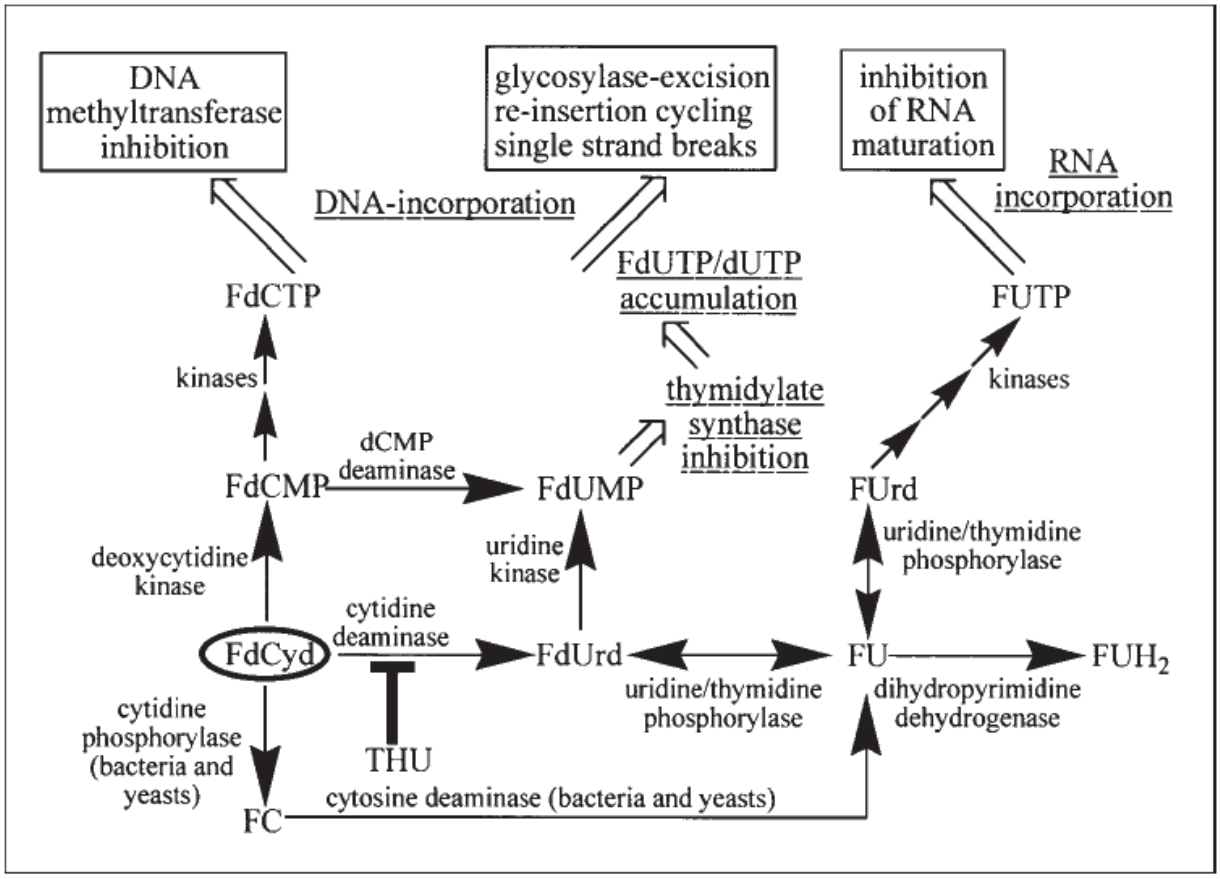



**Figure 2: Metabolism of 5-fluoro-2'-deoxycytidine (FdCyd). Metabolites are 5-fluoro-2'-deoxyuridine (FdUrd), 5-fluorocytosine (FC), 5-fluorouracil (FU), 5-fluorouridine (FUrd), and 5-fluorouridine mono-phosphate (FUMP). Tetrahydrouridine (THU) blocks the conversion of FdCyd to FdUrd by cytidine deaminase. Solid arrows represent metabolic pathways, open arrows represent drug effects (26).**

## 2.1 [F-18]-5-Fluoro-2'-deoxycytidine (FdCyd)

### 2.1.1 Chemistry and Pharmacology:

FdCyd [C9H12FN3O4, mol. wt. 245.21, ε290 (λmax)=11,100 in 0.1 M HCl (23)] is a fluoropyrimidine antimetabolite. The cytotoxicity of FdCyd in culture is associated with inhibition of thymidylate synthase (12) and with incorporation into DNA (14). In addition to being activated by dCyd Kinase (12), FdCyd is a substrate for Cyd/dCyd deaminase (24). The metabolism of the deamination product, 5-FdU, has been well characterized (5). No direct pharmacokinetic studies of FdCyd have been reported. By similarity to other analogs of deoxycytidine, FdCyd is expected to have a very short mean residence time unless an effective inhibitor of Cyd/dCyd deaminase, *e.g.*, THU, is co-administered.

### 2.1.2 Formulation and Stability:

[F-18]-5-Fluoro-2'-deoxycytidine (FdCyd) Injection is formulated in sterile saline and is manufactured by the University of Pennsylvania, Philadelphia, PA.

The radioactive dose is 5 mCi or less at the time of administration.

2.1.3 Toxicity:

The quantity of [F-18]-FdCyd required for imaging is a microdose, and no toxicity is expected to be observed from the radiopharmaceutical. However, patients will be observed particularly for the toxicities noted in the animal and therapeutic studies. Based on the results of animal studies (1), the toxicities of a therapeutic dose of FdCyd are expected to be similar to those produced by 5-FdU and 5-FU, including neutropenia, mucositis, diarrhea, nausea and vomiting. This drug should be considered to have potential carcinogenic and teratogenic effects based on the carcinogenicity/teratogenicity of related drugs and on the teratogenic effects of FdCyd that have been reported in mice, including vertebral malformations, supernumerary limbs, and limb malformations.

During a Phase 1 therapeutic trial, additional toxicities possibly related to the FdCyd/THU were noted at FdCyd doses of 40 mg/m$^2$/day 5 days a week for 2 weeks and above. These toxicities included electrolyte imbalances not immediately corrected with supplementation, elevated liver function tests, and anemia. None of the toxicities to date have had unresolved sequelae.

2.1.4 Pre-clinical Biodistribution for [18F]-FdCyd:

Radiation dosimetry estimates were based on rodent studies performed by IBA (the manufacturer of this radiotracer) under an NCI contract and the results are included in Table 1, and sample images are included in **Figure 3**. Briefly, the estimated ED for the doses proposed in this study (5 mCi FdCyd) and transmission CT for a total of 2 imaging sessions) is 3.52 rem and the organs receiving the highest doses are the urinary bladder, liver, and kidneys.

**Table 1**

| **Radiation Dosimetry and Effective Dose Calculation** | | |
|---|---|---|
| **Material:** | **F-18 FdCyd** | |
| **Dose/study** | **5** | **mCi** |
| **Subject Age:** | **ADULT** | ***DOSE/STUDY*** |
| **Organ** | **rad/mCi** | **rads** |
| **Adrenals** | **0.019** | **0.0955** |
| **Brain** | **0.001** | **0.0065** |
| **Breasts** | **0.004** | **0.01945** |
| **Esophagus** [1] | **0.004** | **0.02085** |
| **Gallbladder Wall** | **0.031** | **0.154** |
| **Lower Large Intestine** | **0.038** | **0.192** |

| **Radiation Dosimetry and Effective Dose Calculation** | | |
|---|---|---|
| **Material:** | **F-18 FdCyd** | |
| **Dose/study** | **5** | **mCi** |
| **Subject Age:** | **ADULT** | ***DOSE/STUDY*** |
| **Organ** | **rad/mCi** | **rads** |
| **Small Intestine** | **0.020** | **0.0975** |
| **Stomach** | **0.009** | **0.0459** |
| **Upper Large Intestine** | **0.018** | **0.091** |
| **Colon** [2] | **0.027** | **0.13443** |
| **Heart Wall** | **0.009** | **0.04625** |
| **Kidney** | **0.125** | **0.625** |
| **Liver** | **0.145** | **0.725** |
| **Lungs** | **0.020** | **0.1015** |
| **Muscle** | **0.016** | **0.079** |
| **Ovaries** | **0.036** | **0.182** |
| **Pancreas** | **0.016** | **0.081** |
| **Red Marrow** [3] | **0.011** | **0.0555** |
| **Bone Surfaces** [4] | **0.007** | **0.03645** |
| **Skin** | **0.006** | **0.02845** |
| **Spleen** | **0.008** | **0.04165** |
| **Testes** | **0.026** | **0.1275** |
| **Thymus** | **0.004** | **0.02085** |
| **Thyroid** | **0.002** | **0.0109** |
| **Urinary Bladder Wall** | **1.250** | **6.25** |

| **Radiation Dosimetry and Effective Dose Calculation** | | |
|---|---|---|
| **Material:** | **F-18 FdCyd** | |
| **Dose/study** | **5** | **mCi** |
| **Subject Age:** | **ADULT** | ***DOSE/STUDY*** |
| **Organ** | **rad/mCi** | **rads** |
| **Uterus** | **0.081** | **0.405** |
| **Salivary Glands**[3] | **0.002** | **0.0109** |
| **REMAINDER AVE** | **0.036** | **0.180544444** |
| **EFFECTIVE DOSE (rem)** | | **4.42E-01** |

## Notes

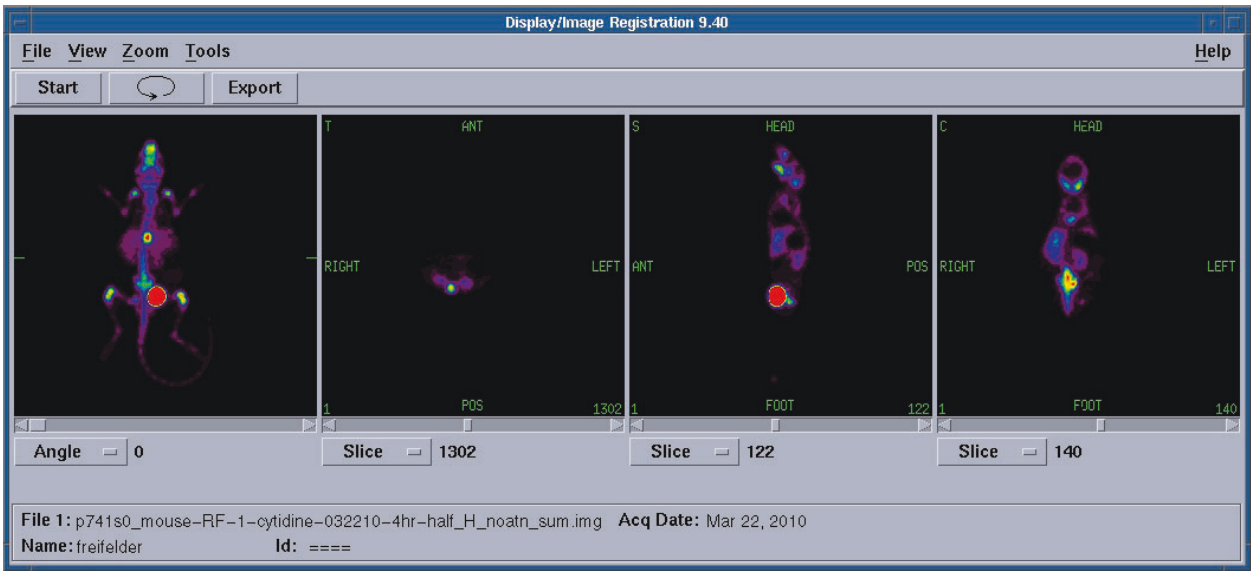
[1] Since no dose is explicitly tabulated for esophagus, thymus dose is used (as per ICRP 80)

[2] Colon Dose estimated by [0.57 ($Dose_{ULI}$) + 0.43 ($Dose_{LLI}$)] (as per ICRP 80).

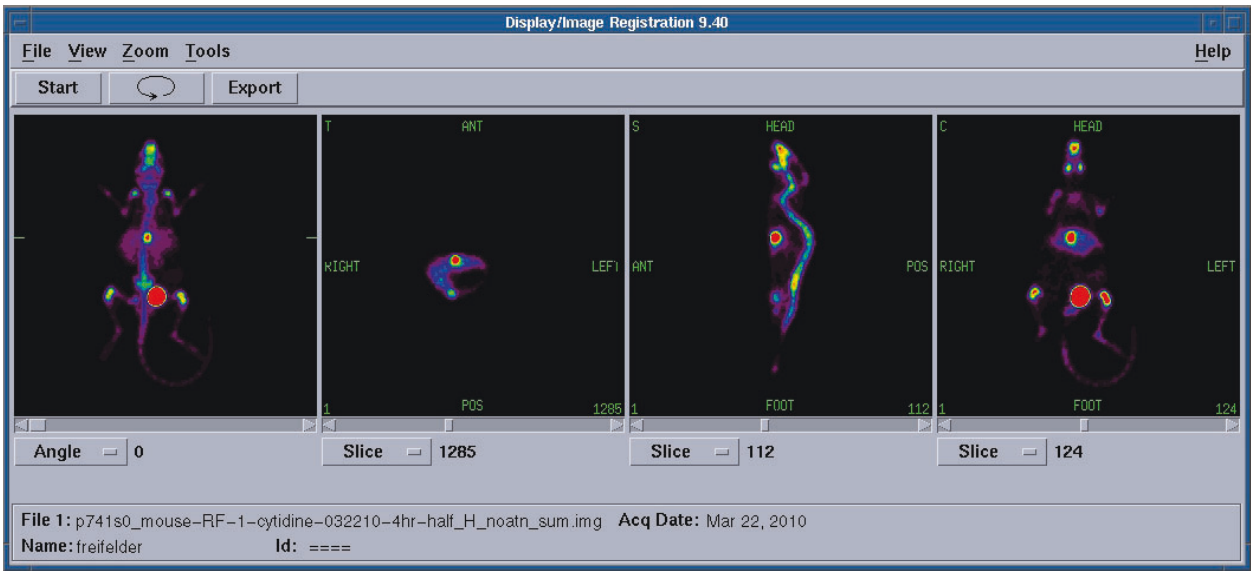
[3]Salivary gland dose is set equal to the thyroid dose

Source: IBA, manufacturer

**Figure 3:**



a) Typical image of the kidneys after 4 hours (MIP, Axial, sagittal, and coronal at the level of the kidneys)




b) Same mouse, typical image of the liver at 4 hours (MIP, Axial, sagittal, and coronal at the level of the liver)

As $^{18}$F FdCyd is chemically identical to the therapeutic drug FdCyd, the preclinical toxicity data from the therapeutic agent is the same as that for this imaging agent.

## 2.2 Tetrahydrouridine (THU)

### 2.2.1 Chemistry and Pharmacology:

THU [C9H16N2O6, mol. wt. 248.2] is an effective inhibitor of Cyd/dCyd deaminase in animals and humans (18, 19). It has a plasma half-life in humans of approximately one hour (19).

### 2.2.2 Toxicity:

At an average total dose of 750 mg/kg (10 to 50 mg/kg/day for up to 23 days), THU alone was not toxic to humans; THU potentiated the toxicity of araC, but no toxicity occurred with the combination that would not have been expected from araC given alone at an equitoxic dose (20). THU has been administered by three-hour infusion in conjunction with araC without producing toxicities other than those expected from the araC (22).

## 2.3 Rationale

FdCyd + THU has shown preliminary evidence of clinical benefit in early phase trial and is being evaluated in a phase II trial for the treatment of refractory head and neck, breast, non-small cell lung and transitional cell carcinoma as part of the CTEP 8351 (NCI Protocol # 09-C-0214) trial. However, the clinical benefit so far has been observed in a minority of patients making it important to design a strategy for patient selection. By establishing a method to evaluate the biodistribution of the drug and its ability to be taken up by the tumor cells, achieving intracellular therapeutic concentrations may provide insight into these variable responses.

Currently an oral form of 5-fluoro-2'-deoxycytidine with oral tetrahydrouridine is being tested in a phase I study (CTEP# 9127, NCI Protocol 12-C-0066) in patients with advanced solid tumors. The goal of the protocol is to achieve oral FdCyd plasma concentrations similar to IV administration concentrations. Protocol 12-C-006 (CTEP 9127) is being performed because one of the main limitations of the IV regimen is the number of days and duration of drug administration in each cycle, which requires multiple patient visits. An oral formulation would improve feasibility and compliance; therefore allowing an additional pool of patients that can participate in this study.

The proposed study with the establishment of a radiolabeled form could image the biodistribution at baseline and during therapy in patients receiving the therapeutic drug. The first step in the development of such an *in vivo* marker is to determine the safety of the radiolabeled form and the radiation dosimetry. The resultant pharmacokinetics of [F-18]-FdCyd will subsequently be evaluated both with and without non- radiolabeled FdCyd concurrently administered using PET/CT. Due to rapid metabolism in the absence of a Cyd/dCyd deaminase inhibitor, a trial of single agent [F-18]-FdCyd alone would be irrelevant to the potential clinical utility of this drug. Therefore, the treatment plan of this protocol incorporates a fixed dose of THU which is known to be safe, and to effectively diminish the activity of Cyd/dCyd deaminase in humans (22).

# 3 PATIENT SELECTION

## 3.1 Eligibility Criteria

3.1.1 Inclusion Criteria

3.1.1.1 Enrolled in the NIH Phase II Clinical protocol evaluating FdCyd with THU (NCI protocol # 09-C-0214 (CTEP# 8351) or NCI protocol #12-C-0066 (CTEP# 9127)) with target lesion measured as ≥ 10mm with spiral CT scan.

3.1.1.2 Written, voluntary, informed consent of the patient must be obtained in compliance with institutional, state and federal guidelines

3.1.1.3 For females: Negative serum pregnancy test OR post-menopausal for at least 2 years OR patient has had a hysterectomy

3.1.2 Exclusion Criteria

3.1.2.1 Participants with severe claustrophobia unresponsive to oral anxiolytics

3.1.2.2 Subjects weighing > 400 lbs (weight limit for scanner table), or unable to fit within the imaging gantry

3.1.2.3 Known allergy to FdCyd

3.1.2.4 The subject is unable to lie still for ~75 minutes

3.1.2.5 Pregnant or lactating women. Pregnant women are excluded from this study because the effects of 18F-FdCyd in pregnancy are not known. Because there is an unknown but potential risk for adverse events in nursing infants secondary to administration of 18F-FdCyd in the mother, breastfeeding should be discontinued if the mother receives 18F-FdCyd

3.1.2.6 Participants with any co-existing medical or psychiatric condition that is likely to interfere with study procedures and/or results

### 3.2 Screening Evaluation

This is a companion study to the therapeutic Phase II 5-FdCyd/THU study (NCI protocol # 09-C-0214: CTEP # 8351) and Phase I 5-FdCyd/THU study in Patients (NCI protocol # 12-C-0066/ CTEP # 9127 open in the NCI Developmental Therapeutics Clinic at the Clinical Center, NIH. Both studies are multicenter studies and only patients enrolled at the Clinical Center will be recruited for this imaging protocol. After determination of enrollment on NCI protocol # 09-C-0214(CTEP# 8351) or NCI protocol # 12-C-0066/CTEP# 9127, patients will be approached for willingness to participate in this imaging study. To ensure eligibility for this imaging study, the following evaluation will be performed:

3.2.1 History and Physical Examination, including vital signs

3.2.2 Evaluation of Performance Status using the ECOG scale

3.2.3 β HCG or urine pregnancy test

3.2.4 Review of previous scans to assure measurable disease

### 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

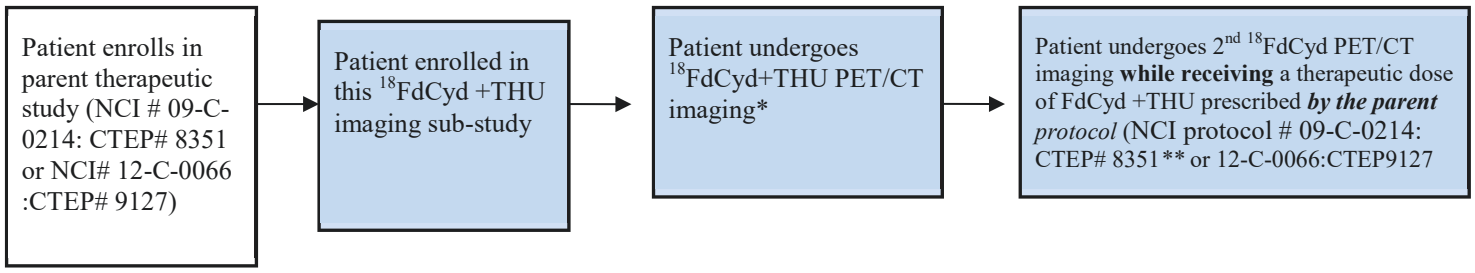
Authorized staff must register an eligible candidate with NCI Central Registration Office (CRO) within 24 hours of signing consent. A registration Eligibility Checklist from the web site (http://home.ccr.cancer.gov/intra/eligibility/welcome.htm) must be completed and faxed to 301-480-0757. After confirmation of eligibility at Central Registration Office, CRO staff will call Molecular Imaging Program (Yolanda McKinney, RN, MIP/CCR/NCI) to advise them of the acceptance of the patient on the protocol prior to the release of any investigational agents. Verification of Registration will be forwarded electronically via e-mail to the research team. A recorder is available during non-working hours.

## 5 STUDY IMPLEMENTATION

### 5.1 Study Design

Only patients who are enrolled in the therapeutic trials NCI Protocol # 09-C-0214 (CTEP# 8351) or NCI Protocol # 12-C-0066 (CTEP # 9127) will be recruited for this trial. The initial [F-18]-FdCyd + THU PET/CT imaging should be completed prior to therapeutic dosing. Repeat imaging will be performed 2-5 days after the initiation of FdCyd + THU therapy and at least 2 hours after dosing.

## 5.2 Study Schema



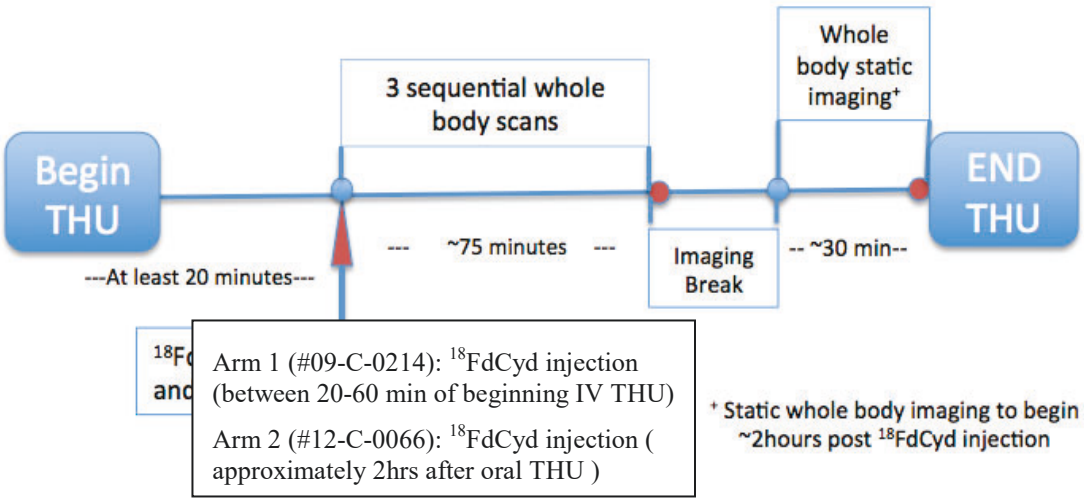




**Figure 4: Imaging session without therapeutic "cold" FdCyd**

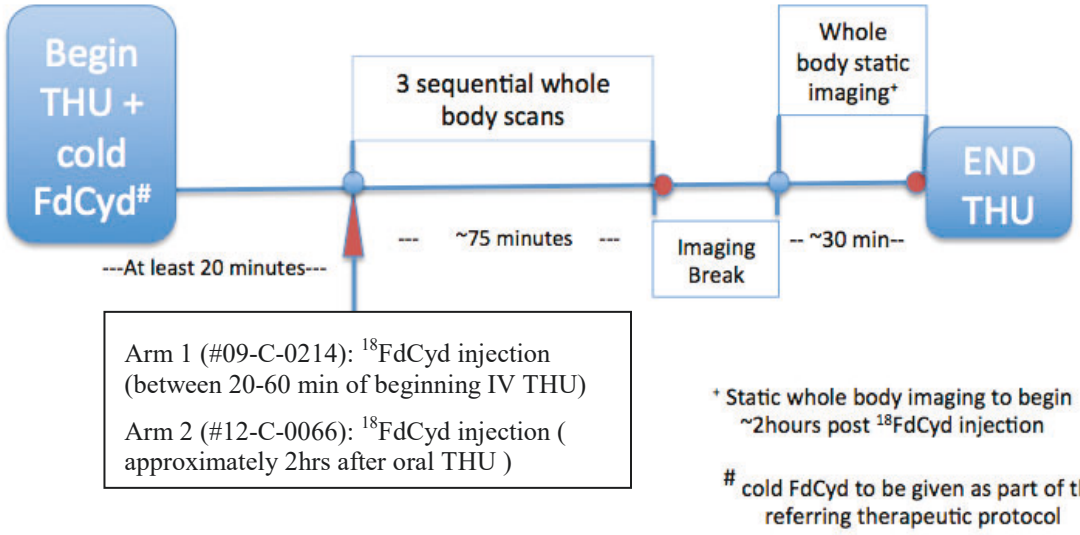



**Figure 5: Imaging session with "cold" FdCyd and administered per NCI protocol # 09-C-0214 (CTEP# 8351) and NCI protocol #12-C-0066 (CTEP #9127)**

5.2.1 Imaging session per NCI protocol #09-C-0214 (CTEP #8351)

For THU administration, an IV bolus of 20% the total dose of tetrahydrouridine (total dose of THU = 350 mg/m$^2$) is administered, followed by the other 80% of the dose, diluted in 250 ml of D5W, which will be infused for 3 hours or until the end of imaging. At least 20 minutes after the tetrahydrouridine IV is begun, the radiopharmaceutical will be administered over ~1 minute. The patient will be imaged continuously from top of head to thigh for 60-75 minutes, followed by whole body static imaging at 2 hours post infusion (approximately 2.5 hours total). A licensed clinical practitioner will be available during the drug administration and imaging session.

**The $^{18}$FdCyd will be administered at least 20 minutes after the THU is begun and concurrent with, or following (within 1 hr of) the start of the FdCyd therapeutic dose.

5.2.2 Imaging session per NCI protocol #12-C-0066 (CTEP #9127)

Patients will receive a dose of THU (total dose of THU is 3000 mg ) for the initial scan approximately 2 hours (+30 minutes) before injection of the F-18 FdCyd radiotracer. The patient will be imaged continuously from top of head to thigh for 60-75 minutes, followed by whole body static imaging at 2 hours post radiotracer injection approximately 2.5 hours total). A licensed clinical practitioner will be available during the drug administration and imaging session.

## 5.3 Stopping Rules

For the purpose of protecting patient safety, the study will be stopped according to the following stopping rule.

**Table 2**

| Number of Patients | Number of Grade 3 or 4 CTCAE toxicity |
|---|---|
| 3-6 | 3 |
| 7-12 | 4 |
| 13-18 | 5 |

The operating characteristic of the stopping rule was assessed by simulations. Based on 10,000 simulations, the probability of stopping early for an excess of grade 3 or 4 CTCAE toxicity over the course of trial is 4.6% when the toxicity rate is 0.10 and 84% when the toxicity rate is 0.35. This is deemed reasonable for protecting patient safety.

## 5.4 CTEP IND Agent Administration

### 5.4.1 5-fluoro-2'-deoxycytidine (FdCyd) and Tetrahydrouridine (THU)

5-fluoro-2'-deoxycytidine (FdCyd) and Tetrahydrouridine (THU) will be administered according to the specifications of the parent protocol NCI protocol # 09-C-0214 (CTEP# 8351) or NCI Protocol #12-C-0066 (CTEP# 9127). The investigational agent administered on this study is the radiolabeled dose of [F-18]-5-Fluoro-2'-deoxycytidine (FdCyd).

### 5.4.2 [F-18]-5-Fluoro-2'-deoxycytidine (FdCyd) (NSC #755309)

Intravenous administration: The imaging dose of [F-18]-FdCyd will be administered to the patient ~20 minutes after initiation of the intravenous bolus dose of THU. It will be administered as an IV injection over ~1 minute.

### 5.4.3 Tetrahydrouridine (THU)

#### 5.4.3.1 Intravenous administration:

An IV bolus of 20% the total dose of tetrahydrouridine (total dose of THU = 350 mg/m$^2$) is administered (over less than 1 minute), followed by the other 80% of the dose, diluted in 250 ml of D5W, which will be infused for 3 hours.

#### 5.4.3.2 Oral Administration:

The dose of THU is fixed at 3000 mg. THU oral solution should be taken on an empty stomach at least 1 hour before or 2 hours after a meal, and 30 minutes before taking FdCyd. THU Oral Solution should be mixed with 2-3 ounces of an acidic juice to aid in masking the taste (i.e., orange juice, apple juice or lemonade) immediately prior to dosing.

### 5.4.4 PET/CT Imaging

A transmission CT of the imaging field of view (top of head to thigh) will be performed immediately prior to radiotracer injection. Three sequential whole body PET emission scans will be performed from top of head to thigh for the first 60-75 minutes. An additional top of the head to thigh scan (~35 minutes) will be acquired at 2 hours post radiotracer injection.

Vital signs and queries for potential adverse events will be performed during the imaging at the following intervals: 0, 15, 30 minutes, 1-hour and 2-hours post injection. A query for potential AEs will also be made 24 +/-8 hrs after (~6 times the t1/2 of the imaging drug) after administration.

### 5.5 General Concomitant Medication and Supportive Care Guidelines

In general, the concomitant medications and supportive care guidelines in the parent protocol NCI protocol # 09-C-0214 (CTEP# 8351) or NCI Protocol #12-C-0066 (CTEP# 9127) will be followed. Based on known toxicity, no reactions are expected; however, supportive care may be necessary in rare instances when a reaction is encountered.

Supportive care will be provided for infusion-related reactions such as allergic or anaphylactic reactions. Allergic reactions requiring therapy will be treated according to standard guidelines by the attending physician after appropriate medical assessment. Subjects with significant infusion related reactions which are atypical and not successfully treated on site in the Molecular Imaging Clinic or which develop later or require closer monitoring may be admitted to the inpatient medical oncology service under Dr. Chen for observation/treatment or the ICU as needed.

### 5.6 Duration of Participation

Participants will be taken off this study 1 day after the repeat imaging session is complete.

### 5.7 Dosing Delays/Dose Modifications

5 mCi of F-18 FdCyd is prescribed. Due to potential unpredictable delays, the total activity administered may be reduced at the discretion of the PI. A corresponding increase in PET bed position acquisition time will be made as needed.

In the unlikely event of complications during the injection (infiltration, adverse reaction), the injection will be discontinued. If the PI determines there are no contraindications, the remaining amount of tracer will be injected following resolution of the cause of discontinuation.

### 5.8 Criteria for Removal from Study

The investigator may withdraw a patient from the study for any of the following reasons:

- A protocol violation occurs,
- A serious or intolerable adverse event occurs
- A clinically significant change in patient status occurs making the patient no longer eligible to receive the radiopharmaceutical or undergo the PET/CT imaging
- The investigator terminates the study

Authorized staff must notify Central Registration Office (CRO) when a subject is taken off-study. An off-study form from the web site (http://home.ccr.cancer.gov/intra/eligibility/welcome.htm) main page must be completed and faxed to 301-480-0757.

## 6 ADVERSE EVENTS: LIST AND REPORTING REQUIREMENTS

Adverse event (AE) monitoring and reporting is a routine part of every clinical trial. The following list of AEs (Section **6.1**) and the characteristics of an observed AE (Section **6.2**) will determine whether the event requires expedited (via CTEP-AERS) **in addition** to routine reporting.

## 6.1 Comprehensive Adverse Events & Potential Risks Lists (CAEPR) for 5-Fluoro-2'-deoxycytidine (FdCyd, NSC 48006) with Tetrahydrouridine (THU, NSC 112907)

The Comprehensive Adverse Event and Potential Risks list (CAEPR) provides a single list of reported and/or potential adverse events (AE) associated with an agent using a uniform presentation of events by body system. In addition to the comprehensive list, a subset, the Specific Protocol Exceptions to Expedited Reporting (SPEER), appears in a separate column and is identified with bold and italicized text. This subset of AEs (SPEER) is a list of events that are protocol specific exceptions to expedited reporting to NCI (except as noted below). Refer to the 'CTEP, NCI Guidelines: Adverse Event Reporting Requirements' http://ctep.cancer.gov/protocolDevelopment/electronic_applications/docs/aeguidelines.pdf for further clarification. *Frequency is provided based on 166 patients.* Below is the CAEPR for 5-Fluoro-2'-deoxycytidine with tetrahydrouridine (FdCyd/THU).

**NOTE**: Report AEs on the SPEER **ONLY IF** they exceed the grade noted in parentheses next to the AE in the SPEER. If this CAEPR is part of a combination protocol using multiple investigational agents and has an AE listed on different SPEERs, use the lower of the grades to determine if expedited reporting is required..

**Version 2.2, January 17, 2014**[1]

| **Adverse Events with Possible Relationship to 5-Fluoro-2'-deoxycytidine with tetrahydrouridine (FdCyd/THU) (CTCAE 4.0 Term) [n= 166]** | | | **Specific Protocol Exceptions to Expedited Reporting (SPEER)** |
|---|---|---|---|
| **Likely (>20%)** | **Less Likely (<=20%)** | **Rare but Serious (<3%)** | |
| BLOOD AND LYMPHATIC SYSTEM DISORDERS | | | |
| Anemia | | | ***Anemia (Gr 2)*** |
| | Febrile neutropenia | | |
| CARDIAC DISORDERS | | | |
| | Sinus tachycardia | | ***Sinus tachycardia (Gr 2)*** |
| GASTROINTESTINAL DISORDERS | | | |
| | Abdominal distension | | |
| | Abdominal pain | | ***Abdominal pain (Gr 2)*** |
| | Constipation | | ***Constipation (Gr 2)*** |
| Diarrhea | | | ***Diarrhea (Gr 2)*** |
| | Dry mouth | | |
| | Dyspepsia | | |
| | Flatulence | | |
| | | Ileus | |
| | Mucositis oral | | ***Mucositis oral (Gr 2)*** |
| Nausea | | | ***Nausea (Gr 2)*** |
| Vomiting | | | ***Vomiting (Gr 2)*** |
| GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS | | | |
| | Chills | | ***Chills (Gr 2)*** |

| | | | |
|---|---|---|---|
| | Edema limbs | | |
| Fatigue | | | ***Fatigue (Gr 2)*** |
| | Fever | | ***Fever (Gr 2)*** |
| | Non-cardiac chest pain | | |
| | Pain | | |
| INVESTIGATIONS | | | |
| | Activated partial thromboplastin time prolonged | | ***Activated partial thromboplastin time prolonged (Gr 2)*** |
| Alanine aminotransferase increased | | | ***Alanine aminotransferase increased (Gr 2)*** |
| Alkaline phosphatase increased | | | ***Alkaline phosphatase increased (Gr 2)*** |
| Aspartate aminotransferase increased | | | ***Aspartate aminotransferase increased (Gr 2)*** |
| | Blood bilirubin increased | | ***Blood bilirubin increased (Gr 2)*** |
| | Cholesterol high | | |
| | Creatinine increased | | ***Creatinine increased (Gr 2)*** |
| Lymphocyte count decreased | | | ***Lymphocyte count decreased (Gr 2)*** |
| Neutrophil count decreased | | | ***Neutrophil count decreased (Gr 2)*** |
| Platelet count decreased | | | ***Platelet count decreased (Gr 2)*** |
| | Weight loss | | ***Weight loss (Gr 2)*** |
| White blood cell decreased | | | ***White blood cell decreased (Gr 2)*** |
| METABOLISM AND NUTRITION DISORDERS | | | |
| Anorexia | | | ***Anorexia (Gr 2)*** |
| | Dehydration | | ***Dehydration (Gr 2)*** |
| | Hypercalcemia | | |
| | Hyperglycemia | | ***Hyperglycemia (Gr 2)*** |
| | Hyperkalemia | | |
| | Hypermagnesemia | | |
| Hypoalbuminemia | | | ***Hypoalbuminemia (Gr 2)*** |
| | Hypocalcemia | | ***Hypocalcemia (Gr 2)*** |
| | Hypokalemia | | ***Hypokalemia (Gr 2)*** |
| | Hypomagnesemia | | ***Hypomagnesemia (Gr 2)*** |
| Hyponatremia | | | ***Hyponatremia (Gr 2)*** |
| | Hypophosphatemia | | ***Hypophosphatemia (Gr 2)*** |
| MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS | | | |
| | Arthralgia | | |
| | Generalized muscle weakness | | |
| | Myalgia | | |
| NERVOUS SYSTEM DISORDERS | | | |
| | Dizziness | | ***Dizziness (Gr 2)*** |
| | Dysgeusia | | |
| | Headache | | ***Headache (Gr 2)*** |
| | Peripheral motor neuropathy | | ***Peripheral motor neuropathy (Gr 2)*** |
| | Peripheral sensory neuropathy | | |
| RENAL AND URINARY DISORDERS | | | |
| | Proteinuria | | |
| RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS | | | |
| | Cough | | |
| | Dyspnea | | ***Dyspnea (Gr 2)*** |
| | Epistaxis | | |

| | | | |
|---|---|---|---|
| SKIN AND SUBCUTANEOUS TISSUE DISORDERS | | | |
| | Alopecia | | |
| | Dry skin | | |
| | Hyperhidrosis | | |
| | Palmar-plantar erythrodysesthesia syndrome | | |
| | Pruritus | | |
| | Rash maculo-papular | | |
| VASCULAR DISORDERS | | | |
| | Hypotension | | ***Hypotension (Gr 2)*** |

[1]This table will be updated as the toxicity profile of the agent is revised. Updates will be distributed to all Principal Investigators at the time of revision. The current version can be obtained by contacting PIO@CTEP.NCI.NIH.GOV. Your name, the name of the investigator, the protocol and the agent should be included in the e-mail.

**Also reported on FdCyd/THU trials but with the relationship to FdCyd/THU still undetermined:**

**CARDIAC DISORDERS** - Aortic valve disease; Atrial fibrillation; Cardiac arrest; Mitral valve disease; Myocardial infarction; Palpitations; Pericardial effusion; Pericarditis; Tricuspid valve disease; Ventricular tachycardia

**EAR AND LABYRINTH DISORDERS** - Ear pain; Middle ear inflammation; Tinnitus

**EYE DISORDERS** - Blurred vision; Conjunctivitis; Dry eye; Eye disorders - Other (right eye blindness [90-95% visual acuity loss]); Eye disorders - Other (subconjuctival hemorrhage); Eye disorders - Other (visual changes); Eye pain; Floaters; Photophobia; Watering eyes

**GASTROINTESTINAL DISORDERS** - Bloating; Colitis; Colonic obstruction; Duodenal ulcer; Dysphagia; Esophagitis; Gastrointestinal disorders - Other (pyloric ulcer); Gastrointestinal pain; Hemorrhoids; Lip pain; Oral pain; Rectal hemorrhage; Typhlitis; Upper gastrointestinal hemorrhage

**GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS** - Edema face; Flu like symptoms; Infusion site extravasation; Injection site reaction; Localized edema; Malaise

**INFECTIONS AND INFESTATIONS** - Enterocolitis infectious; Eye infection; Lung infection; Mucosal infection; Sepsis; Skin infection; Upper respiratory infection

**INJURY, POISONING AND PROCEDURAL COMPLICATIONS** - Bruising; Vascular access complication

**INVESTIGATIONS** - CPK increased; Hemoglobin increased; INR increased; Investigations - Other (bicarbonate)

**METABOLISM AND NUTRITION DISORDERS** - Alkalosis; Hypernatremia; Hypertriglyceridemia; Hyperuricemia; Hypoglycemia

**MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS** - Arthritis; Back pain; Bone pain; Chest wall pain; Joint range of motion decreased; Neck pain; Pain in extremity

**NEOPLASMS BENIGN, MALIGNANT AND UNSPECIFIED (INCL CYSTS AND POLYPS)** - Tumor pain

**NERVOUS SYSTEM DISORDERS** - Cognitive disturbance; Nervous system disorders - Other (neuropathy - cranial); Neuralgia; Syncope; Tremor

**PSYCHIATRIC DISORDERS** - Agitation; Anxiety; Confusion; Depression; Insomnia

**RENAL AND URINARY DISORDERS** - Cystitis noninfective; Hematuria; Hemoglobinuria; Renal and urinary disorders - Other (bladder pain); Renal and urinary disorders - Other (urethra pain); Urinary frequency; Urinary incontinence; Urinary tract pain

**REPRODUCTIVE SYSTEM AND BREAST DISORDERS** - Breast pain; Gynecomastia; Pelvic pain

**RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS** - Allergic rhinitis; Bronchopulmonary hemorrhage; Hiccups; Hypoxia; Pleural effusion; Sore throat

**SKIN AND SUBCUTANEOUS TISSUE DISORDERS** - Bullous dermatitis; Nail discoloration; Periorbital edema; Purpura; Rash acneiform; Skin and subcutaneous tissue disorders - Other (folliculitis); Skin hyperpigmentation; Skin hypopigmentation; Skin ulceration

**VASCULAR DISORDERS** - Flushing; Hot flashes; Hypertension

**Note**: FdCyd/THU in combination with other agents could cause an exacerbation of any adverse event currently known to be caused by the other agent, or the combination may result in events never previously associated with either agent.

### 6.1.1 Adverse Events and Potential Risks of [F-18] FdCyd

This is a first in human study of the radioactive form of FdCyd. [F-18]-FdCyd administration at the chemical dose described for this study is low (≤30mg). Patients on this study will be monitored for adverse events for 10 half-lives (1 day).

No adverse effects are expected as a result of the administration of [F-18]-FdCyd. Potential adverse effects from the tracer injection are listed below:

- Infection at the injection site or systemic infection
- Extravasation of the dose
- Allergic reaction
- Vasovagal reaction

### 6.1.2 Adverse Events and Potential Risks for THU

THU has no expected side effects at the dose used in this study, but THU may increase the risk of experiencing FdCyd side effects.

## 6.2 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.
- **'Expectedness'**: AEs can be 'Unexpected' or 'Expected' (see Section **6.1** above) for expedited reporting purposes only. 'Expected' AEs (the ASAEL) are ***bold and italicized*** in the CAEPR (Section **6.1.1**).
- **Attribution** of the AE:
  - Definite – The AE *is clearly related* to the study treatment.
  - Probable – The AE *is likely related* to the study treatment.
  - Possible – The AE *may be related* to the study treatment.
  - Unlikely – The AE *is doubtfully related* to the study treatment.
  - Unrelated – The AE *is clearly NOT related* to the study treatment.

### 6.2.1 Suspected adverse reaction

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

### 6.2.2 Unexpected adverse reaction

An adverse event or suspected adverse reaction is considered "unexpected" if it is not listed in the investigator brochure or is not listed at the specificity or severity that has been observed; or, if an investigator brochure is not required or available, is not consistent with the risk information described in the general investigational plan or elsewhere in the current application. "Unexpected", also refers to adverse events or suspected adverse reactions that are mentioned in the investigator brochure 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.

### 6.2.3 Serious

An Unanticipated Problem or Protocol Deviation is serious if it meets the definition of a Serious Adverse Event or if it compromises the safety, welfare or rights of subjects or others.

### 6.2.4 Serious Adverse Event

An adverse event or suspected adverse reaction is considered serious if in the view of the investigator or the sponsor, it results in any of the following:

- Death,
- A life-threatening adverse drug experience
- Inpatient hospitalization or prolongation of existing hospitalization
- Persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions
- A congenital anomaly/birth defect
- 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 this definition.

### 6.2.5 Disability

A substantial disruption of a person's ability to conduct normal life functions.

### 6.2.6 Life-threatening adverse drug experience

Any adverse event or suspected adverse reaction that places the patient or subject, in the view of the investigator or sponsor, at immediate risk of death from the reaction as it occurred, i.e., it does not include a reaction that had it occurred in a more severe form, might have caused death.

6.2.7 Protocol Deviation (NIH Definition):

Any change, divergence, or departure from the IRB approved study procedures in a research protocol that does not have a major impact on the subject's rights, safety or well-being, or the completeness, accuracy and reliability of the study data.-approved research protocol.

6.2.8 Non-compliance (NIH Definition)

The failure to comply with applicable NIH Human Research Protections Program (HRPP) policies, IRB requirements, or regulatory requirements for the protection of human research subjects.

6.2.9 Unanticipated Problem

Any incident, experience, or outcome that:

- Is unexpected in terms of nature, severity, or frequency in relation to

  (a) the research risks that are described in the IRB-approved research protocol and informed consent document; Investigator's Brochure or other study documents, and

  (b) the characteristics of the subject population being studied; **AND**
- Is related or possibly related to participation in the research; **AND**
- Suggests that the research places subjects or others at a g*reater risk of harm* (including physical, psychological, economic, or social harm) than was previously known or recognized.

## 6.3 Expedited Adverse Event Reporting

6.3.1 Expedited AE reporting for this study must use CTEP-AERS (CTEP Adverse Event Reporting System), accessed via the CTEP home page (http://ctep.cancer.gov). The reporting procedures to be followed are presented in the "CTEP, NCI Guidelines: Adverse Event Reporting Requirements" which can be downloaded from the CTEP home page (http://ctep.cancer.gov). These requirements are briefly outlined in the table below (Section 6.3.3).

6.3.2 In the rare event when Internet connectivity is disrupted a 24-hour notification is to be made to NCI by telephone at: 301-897-7497, or 301-897-7402 for CIP studies. An electronic report MUST be submitted immediately upon re-establishment of internet connection. Please note that all paper CTEP-AERS forms have been removed from the CTEP website and will NO LONGER be accepted.

6.3.3 Expedited Reporting Requirements for Adverse Events that Occur on Studies under an IND/IDE within 30 Days of the Last Administration of the Investigational Agent/Intervention[1, 2]

**Table 3**

**FDA REPORTING REQUIREMENTS FOR SERIOUS ADVERSE EVENTS (21 CFR Part 312)**

**NOTE:** Investigators **MUST** immediately report to the sponsor (NCI) **ANY** Serious Adverse Events, whether or not they are considered related to the investigational agent(s)/intervention (21 CFR 312.64)

An adverse event is considered serious if it results in **ANY** of the following outcomes:

1) Death
2) A life-threatening adverse event
3) An adverse event results in inpatient hospitalization or prolongation of existing hospitalization for ≥ 24 hours
4) A persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions
5) A congenital anomaly/birth defect.
6) Important Medical Events (IME) that may not result in death, be life threatening, or require hospitalization may be considered serious when, based upon medical judgment, they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition (FDA, 21 CFR 312.32; ICH E2A and ICH E6).

**ALL SERIOUS** adverse events that meet the above criteria **MUST** be immediately reported to the NCI via CTEP-AERS within the timeframes detailed in the table below.

| Grade 1 and 2 Timeframes | Grade 3-5 Timeframes. |
|---|---|
| 10 Calendar Days | 24-Hour 5 Calendar Days |

**Expedited AE reporting timelines are defined as**:

- "24-Hour; 5 Calendar Days" - The AE must initially be reported via CTEP-AERS within 24 hours of learning of the AE, followed by a complete expedited report within 5 calendar days of the initial 24-hour report.
- "10 Calendar Days" - A complete expedited report on the AE must be submitted within 10 calendar days of learning of the AE.

[1]Serious adverse events that occur **more than** 30 days after the last administration of investigational agent/intervention require reporting as follows:

Expedited 24-hour notification followed by complete report within 5 calendar days for **ALL** Grade 4 and 5 AEs and Grade 3 AEs with at least a possible attribution.

[2]For studies using PET or SPECT IND agents, the AE reporting period is limited to 10 radioactive half-lives, rounded UP to the nearest whole day, after the agent/intervention was last administered. Footnote "1" above applies after this reporting period.

Effective Date: May 5, 2011

## 6.4 Protocol-Specific Expedited Adverse Event Reporting Exclusions

For this protocol only, AEs/grades that occur as a result of the therapy or participation in the parent protocol (NCI protocol # 09-C-0214: (CTEP# 8351) or NCI Protocol #12-C-0066: (CTEP# 9127)) are exceptions to the Expedited Reporting Guidelines and do not require expedited reporting (*i.e.*, CTEP-AERS). These AEs will be reported according to the expedited or routine reporting guidelines in NCI protocol # 09-C-0214 (CTEP# 8351) or NCI Protocol #12-C-0066 (CTEP# 9127).

## 6.5 Routine Adverse Event Reporting

All Adverse Events **must** be reported in routine study data submissions. **AEs reported through CTEP-AERS must also be reported in routine study data submissions. All AEs will be reported through CDUS.**

## 6.6 Secondary AML/MDS

AML/MDS events must be reported via CTEP-AERS (in addition to your routine AE reporting mechanisms). In CTCAE v4.0, the event(s) may be reported as either: 1) Leukemia secondary to oncology chemotherapy, 2) Myelodysplastic syndrome, or 3) Treatment-related secondary malignancy.

## 6.7 NCI-IRB Reporting

### 6.7.1 NCI-IRB Expedited Reporting of Unanticipated Problems, and Deaths

The Protocol PI will report to the NCI-IRB:

- All deaths, except deaths due to progressive disease
- All Protocol Deviations
- All Unanticipated Problems
- All serious non-compliance

Reports must be received by the NCI-IRB within 7 working days of PI awareness via iRIS.

### 6.7.2 NCI-IRB Requirements for PI Reporting at Continuing Review

The protocol PI will report to the NCI-IRB:

1. A summary of all protocol deviations in a tabular format to include the date the deviation occurred, a brief description of the deviation and any corrective action.
2. A summary of any instances of non-compliance
3. A tabular summary of the following adverse events:
    - All Grade 2 **unexpected** events that are possibly, probably or definitely related to the research;
    - All Grade 3 and 4 events that are possibly, probably or definitely related to the research;
    - All Grade 5 events regardless of attribution;
    - All Serious Events regardless of attribution.

**NOTE**: Grade 1 events are not required to be reported.

6.7.3 NCI-IRB Reporting of IND Safety Reports

Only IND Safety Reports that require a sponsor recommended change to meet the protocol or the consent form or in the opinion definition of the PI increases risks to study participants an unanticipated problem will need to be reported to the NCI IRB.

# 7 PHARMACEUTICAL INFORMATION

## 7.1 [F-18]-5-Fluoro-2'-deoxycytidine (FdCyd) NSC# 755309

7.1.1 Molecular Formula:

$C_9H_{12}{}^{18}FN_3O_4$

7.1.2 Molecular Weight:

244.2

7.1.3 How Supplied:

5-fluoro-2'-deoxycytidine is supplied as a sterile, single use vial. [F-18]-5-Fluoro-2'-deoxycytidine (FdCyd) Injection is formulated in sterile saline and is manufactured by the University of Pennsylvania, Philadelphia, PA. It is prepared for each patient on the day of administration and shipped to the address below:

Division of Radiation Safety
National Institutes of Health
Building 21 Room 107
21 Wilson Drive
Bethesda, MD 20892-6780

The dose is 5 mCi or less at the time of administration, a mass dose of <30mg

7.1.4 Storage:

Intact vials should be stored at controlled room temperature. An expiration time will be listed on vial label

7.1.5 Route of Administration:

Intravenous

7.1.6 Preparation:

The agent is manufactured according to the University of Pennsylvania Standard Operating Procedures.

7.1.7 Method of Administration:

FdCyd may be mixed tetrahydrouridine (THU NSC 112907) in D5W and is stable for at least 24 hours at room temperature. See Section **5.4.2** for details about [F-18]-FdCyd administration.

7.1.8 Availability

[F-18]-5-Fluoro-2'-deoxycytidine (FdCyd) Injection is an investigational agent supplied to investigators by the University of Pennsylvania.

7.1.9 Agent Ordering

NCI-supplied agents may be requested by the Principal Investigator (or their authorized designee) at each participating institution

$^{18}$F FdCyd will be ordered directly from the University of Pennsylvania.

7.1.10 Agent Accountability

Agent Inventory Records – The investigator, or a responsible party designated by the investigator, must maintain a careful record of the inventory and disposition of all agents received from DCTD using the NCI Drug Accountability Record Form (DARF). (See the CTEP home page at http://ctep.cancer.gov for the Procedures for Drug Accountability and Storage and to obtain a copy of the DARF and Clinical Drug Request form.)

## 7.2 Tetrahydrouridine (THU) (NSC 112907)

7.2.1 Tetrahydrouridine (THU) Injection

**Chemical Name:** 2(1H)-pyrimidinone, tetrahydro-4-hydroxy-1.beta.-D-ribofuranosyl-

**Other Names:** THU, $H_4U$

**CAS Registry Number:** 18771-50-1

**Molecular Formula:** $C_9H_{16}N_2O_6$ **M.W.:** 248

**Approximate Solubility:** The drug is highly soluble in water up to 200 mg/mL at pH 7.0.

**Mode of Action:** Cytidine deaminase inhibitor

**How Supplied:** Tetrahydrouridine Injection is supplied by the DCTD and distributed by the PMB/NCI as a 500 mg vial (10 mg/mL; 50 mL). The clear, colorless solution contains 10 mg tetrahydrouridine per mL with dibasic phosphate, USP, monobasic phosphate, USP used as buffer to maintain pH at 7.4.

**Preparation:** Tetrahydrouridine may be administered without further dilution or may be further diluted in 0.9% Sodium Chloride Injection, USP or 5% dextrose injection, USP to a concentration as low as 2.5 mg/mL.

**Storage:** Intact vials should be stored in the refrigerator ($2^o$ – $8^o$C).

**Stability:** Shelf life stability testing of the intact vials is on-going. Tetrahydrouridine as diluted above is stable for at least 24 hours at room temperature.

CAUTION: The single-use vial contains no antibacterial preservatives. Therefore, it is advised that the product be discarded 8 hours after initial entry.

**Route of Administration:** Intravenous for subjects enrolled from NCI Protocol 09-C-0214 (CTEP# 8351)

7.2.2 Tetrahydrouridine Powder for Oral Solution (NSC 112907)

**Chemical Name:** 2(1H)-pyrimidinone, tetrahydro-4-hydroxy-1.beta.-D-ribofuranosyl-

**Other Names:** THU, $H_4U$

**CAS Registry Number:** 18771-50-1

**Molecular Formula:** $C_9H_{16}N_2O_6$ **M.W.:** 248

**Approximate Solubility:** The drug is highly soluble in water, up to 200 mg/mL.

**Mode of Action:** Cytidine deaminase inhibitor

**How Supplied:** Tetrahydrouridine is supplied by the DCTD and distributed by the PMB/NCI in a single-use amber plastic bottle, with a tamper-evident cellophane seal around the bottle neck, containing 3 grams of Tetrahydrouridine Powder for Oral Solution.

**Preparation for Oral Administration:** Reconstitute each 3 gram bottle with 30 mL water (distilled or tap water may be used). Swirl to dissolve powder.

**Storage:** Store intact bottles and reconstituted solution in the refrigerator (2-8$^{o}$C).

**Stability:** Shelf life stability testing of the intact bottles is on-going. Reconstituted solutions are stable in the amber plastic bottle for up to 3 weeks at refrigerated temperature.

**Route of Administration:** Oral for subjects enrolled from NCI Protocol 12-C-0066 (CTEP# 9127)

**Method of Administration:** THU oral solution should be taken on an empty stomach at least 1 hour before or 2 hours after a meal, and 30 minutes before FdCyd dosing. Immediately prior to dosing, patients should mix the reconstituted THU oral solution with 2-3 ounces of an acidic juice to aid in masking the taste (i.e., orange juice, apple juice or lemonade).

7.2.3 Agent Ordering

NCI-supplied agents may be requested by the Principal Investigator (or their authorized designee) at each participating institution. Pharmaceutical Management Branch (PMB) policy requires that agent be shipped directly to the institution where the patient is to be treated. PMB does not permit the transfer of agents between institutions (unless prior approval from PMB is obtained). The CTEP-assigned protocol number must be used for ordering all CTEP-supplied investigational agents. The responsible investigator at each participating institution must be registered with CTEP, DCTD through an annual submission of FDA Form 1572 (Statement of Investigator), Curriculum Vitae, Supplemental Investigator Data Form (IDF), and Financial Disclosure Form (FDF). If there are several participating investigators at one institution, CTEP-supplied investigational agents for the study should be ordered under the name of one lead investigator at that institution.

Active CTEP-registered investigators and investigator-designated shipping designees and ordering designees can submit agent requests through the PMB Online Agent Order Processing (OAOP) application (https://eapps-ctep.nci.nih.gov/OAOP/pages/login.jspx). Access to OAOP requires the establishment of a CTEP Identity and Access Management (IAM) account (https://eapps-ctep.nci.nih.gov/iam/) and the maintenance of an "active" account status and a "current" password. For questions about drug orders, transfers, returns, or accountability, call

(240) 276-6575 Monday through Friday between 8:30 am and 4:30 pm (ET) or email PMBAfterHours@mail.nih.gov anytime.

### 7.2.4 Agent Accountability

The Principal Investigator or a responsible party designate by the PI will retain a careful record of the inventory and disposition of all agents received from DCTD using the NCI Drug Accountability Record Form per CTEP Policy and Guidelines for Accountability and Storage of Investigational Drugs (http://ctep.cancer.gov/protocolDevelopment/default.htm#agents_drugs).

# 8 STUDY CALENDAR

Patients on the IV FdCyd + THU protocol (NCI Protocol# 09-C-0214: CTEP#8351) will receive THU at least 20 minutes prior to and through the imaging. On their on-therapy scan (repeat imaging session), they will additionally receive cold FdCyd as prescribed by the referring protocol.

Patients on the oral FdCyd + THU protocol (NCI Protocol 12-C-0066: CTEP#9127) will receive THU approximately 2 hrs (±30 minutes) prior to the imaging. On their on-therapy scan (repeat imaging session), the patients will receive their therapeutic FdCyd + THU approximately 2 hrs (±30 minutes) prior to the imaging.

| **PET/CT Imaging Sessions** | | | | | |
|---|---|---|---|---|---|
| | **Baseline Evaluation** | **Day of imaging** | **0h** | **+2.0h** | **+1 day** |
| **$^{18}$F-5-Fluoro-2'-Deoxycytidine[A] ($^{18}$FdCyd)** | | | **X** | | |
| **Informed consent** | **X** | | | | |
| **Demographics** | **X** | | | | |
| **Medical history** | **X** | | | | |
| **Physical exam** | **X** | | | | |
| **Record Concurrent Meds** | **X** | **X** | **X** | | |
| **Performance status** | **X** | | | | |
| **β HCG or urine pregnancy test** | | **X** | | | |
| **Adverse Event evaluation** | **X** | **X** | **X** | | **X** |
| **Vital Signs** | **X** | | | | |
| **3 Dynamic regional Serial whole body PET/CT Imaging[B]** | | | **X** | | |
| **Static Repeat whole body PET/CT Imaging** | | | | **X** | |

**[A.] CTEP IND Agent: ~5mCi $^{18}$F-5-Fluoro-2'-Deoxycytidine single intravenous injection**
**[B.] PET/CT imaging sessions will be performed pre-cycle under the Phase II FdCyd + THU protocol (NCI Protocol # 09-C-0214: CTEP# 8351 or Phase I FdCyd + THU protocol (NCI # 12-C-0066: CTEP 9127)and while receiving FdCyd +THU as prescribed by the protocol (2-5 days after cycle initiation)**

# 9 MEASUREMENT OF EFFECT: IMAGE ANALYSIS

## 9.1 Tumor evaluation

- The location of the target lesion will be identified on the transmission CT

- $^{18}$FdCyd uptake greater than or equal to 20% of the surrounding background (tumor to background ratio >=1.2) will be considered to be qualitatively "positive"
- All regions/volumes of interest will be saved. Region volumes, and SUV values will be output to an Excel spreadsheet using MIM imaging software (www.mimsoftware.com)
- For lesions with uptake greater than background, an estimated smoothed maximum SUV ($SUV_{max}$) will be reported as the mean SUV value of an automated 80% maximum pixel value threshold based volume of interest (i.e. the average SUV of the "hottest" 20% of pixels in the user defined lesion). This method reduces the chances of the $SUV_{max}$ value being dominated by one or more "noisy" pixel values.
- Volumetric ROIs will also be constructed based on manual tumor edge detection on transmission CT and a 50% SUV based threshold and the MIM Vista "PET edge" tool. These volumes will be used to estimate the total tumor tracer uptake and average uptake/gram of tissue.

A small VOI will be defined within the left ventricular cavity (LV), left atrium (LA) or largest vessel in the field of view. This will be used to obtain an image-derived input function. Graphical analyses will be performed using the organ/tumor TAC and the image derived input function and

- Decay corrected time activity curves will be generated.

  Analysis of the uptake will be dependent on whether a "reversible" or "irreversible" tracer uptake pattern is found. In addition to SUV values at discrete time points, either Logan or Patlak graphical analysis will be performed (corrected for metabolites if necessary), using an image based input functions. Kinetic analysis may also be performed if indicated.

## 9.2 Dosimetry Estimates

### 9.2.1 Solid Organs and Tumors

Regions of interest will be drawn within a homogeneous region within the organ of interest (identified on transmission CT if necessary). Anon decay corrected time activity curve (TAC) will be obtained from the serial whole body images

### 9.2.2 Hollow Organs

Regions encompassing the entire organ (i.e. gall bladder, intestine, bladder) will be initially drawn on CT and transposed onto the PET images. The final VOI's may be adjusted for "spillout" based on the PET images. Total activity within the VOIs will be computed for each organ at each time point.

Residence times will be calculated from the time activity curves and dosimetry estimates will be calculated using OLINDA software.

# 10 DATA REPORTING / REGULATORY CONSIDERATIONS

## 10.1 Data Collection

Data will be collected by the Molecular Imaging Program research nurse in a password-protected file in NCI C3D. The data will be entered at least on a weekly basis.

Data collection will include demographics, the site and size of the primary tumor, diagnosis date, biopsy date, and pathological diagnosis including tumor grade. Pharmacokinetic data will be generated and stored in a secure database. Raw image data will be stored on a secure external drive. Reconstructed imaging data will include storage of anonymized images from each time point and other extracted image parameters. This will be stored in a secure, password protected imaging database. Dosimetry (relative organ count rates and per cent injected dose estimates to the major organs) results will be calculated using OLINDA (Vanderbilt University, Nashville, Tennessee).

## 10.2 Data Monitoring

This study will be monitored by the Clinical Data Update System (CDUS) Version 3.0.Data will be entered electronically on C3D and appropriate reports generated for periodic review.

Clinical Data: The investigators will provide quarterly reports of clinical data on the C3D clinical trial software. Cumulative C3D will be submitted quarterly to CTEP by electronic means. Reports are due January 31, April 30, July 31, and October 31.

Imaging Data: DICOM datasets (or digital datasets for non-DICOM studies) - of the PET, PET-CT and pertinent anatomic studies the patients undergo on the trial will be submitted quarterly to CTEP via the NCIA (NCI Imaging Archive). The electronic transfer of image datasets is due January 31, April 30, July 31, and October 31.

## 10.3 Sample Storage, Tracking and Disposition

Blood samples will be collected for screening and on-study clinical laboratory safety assessments. All blood samples will be processed and handled in accordance with standard laboratory procedures. All samples will be analyzed at the NCI Clinical Center laboratory.

# 11 STATISTICAL SECTION

The purpose of the study is to determine the safety of [F-18]-5-fluoro-2'-deoxycytidine (FdCyd) administered intravenously with administration of tetrahydrouridine (THU). As such, the sample size is targeted at 18 evaluable patients, which is adequate to assess the toxicity rate. With 18 evaluable patients, the width of the 80% expected confidence interval of 10% and 20% toxicity rate is 0.18 and 0.24, respectively.

To account for the possibility of a small number of inevaluable patients, the accrual ceiling is set at 20 patients.

Accruing approximately 1 patient/ month, this protocol is expected to be completed within 1.5 years.

# 12 HUMAN SUBJECTS PROTECTIONS

## 12.1 Rationale For Subject Selection

12.1.1 Participants participating in the parent Phase II FdCyd IV therapy study (NCI protocol # 09-C-0214: CTEP# 8351) or the Phase I FdCyd Oral therapy study NCI Protocol #12-C-0066 (CTEP# 9127) will be invited to participate prior to beginning treatment under that protocol.

12.1.2 All ethnicities and races are eligible for participation

12.1.3 Participants older than 18 are included

## 12.2 Participation of Children

The effects of [F-18]-FdCyd have not been studied in children, and children are more sensitive to radiation exposure than adults. Thus, we will not include children in this study.

## 12.3 Evaluation of Benefits and Risks/Discomforts

There is no possibility of direct benefit to participants in this Phase 0 study. The results of this trial could generate generalizable information that may benefit future patients with cancer and therefore, subjects may benefit by knowing they have contributed to scientific knowledge.

The risks for participating are also small. Non-radiolabeled FdCyd has been administered to patients at higher doses under the parent study, and at this lower chemical dose (< 5mg) no FdCyd related adverse events are expected. No adverse effects are expected from tetrahydrouridine infusion.

Specific risks and potential complications will be clearly outlined in a separate consent form at the time of each procedure. Most complications are expected to be minor and require no treatment. Risks and discomforts associated with [F-18]-FdCyd PET/CT imaging are discomfort of an IV placement and the theoretical effects of additional radiation exposure (ED estimated at 3.52 rem). The subject will be required to lay still on their back for ~60 minutes for the longest scan.

A licensed clinical practitioner will be immediately available during the injection and imaging period and they will be backed by the Clinical Center's Code team. If hospitalization is required the participant will be admitted. However, this is unlikely to occur based on what is known of these agents. There is an added inconvenience of having to return for a follow-up scan that the subjects will be made aware of.

Adverse events will be reviewed after every case and a decision will be made about moving forward to the next participant so that each participant will have the benefit of the prior participant's experience.

As this is a new imaging test, the alternative is to choose not to participate.

## 12.4 Consent and Assent Process and Documentation

The participant will be informed of the study by a member of Dr Chen's team and the subject will contact the study research nurse who will explain the study in detail and send the consent form and protocol (if requested) to the participant. If the participant has any questions they will

be answered by telephone. A signed consent will be obtained by the principal investigator or an associate investigator on the day of the study. The original signed consent goes to Medical Records; a copy is placed in the research record.

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# 14 APPENDIX 1: PREPARATION OF TETRAHYDROURIDINE (THU) TUBES

THU-containing tubes can be prepared using Tetrahydrouridine Injection (500 mg/50 ml), NSC 112907. Each single use vial contains THU – 10 mg/ml, Sodium Phosphate dibasic, anhydrous, USP – 1.5 mg/ml, and Sodium Phosphate monobasic, monohydrate, USP – 0.4 mg/ml. The 50-ml vial should be divided into 1-ml aliquots and stored at 2 °C to 8 °C (refrigerated). (The 1-ml aliquots are not suitable for injection.)

Using a 3/10 cc insulin syringe or other similar sized syringe with a fine needle, draw up 20 µl of the THU solution and transfer it to a 2-ml heparinized vacutainer tube (cat # 366664) by piercing the stopper. Do not draw up THU for more than one tube at a time. You will not be able to control the amount of THU solution that leaves the needle, as it is sucked out by the vacuum. Because of the fine needle, you will not lose the vacuum (apart from the volume added) in the collection tube. The THU-containing tubes may be prepared up to a day in advance.

# 15 APPENDIX 2: PERFORMANCE STATUS SCALES/SCORES ECOG OR ZUBROD SCALE KARNOFSKY SCORE

| | | |
|---|---|---|
| 0 | Asymptomatic and fully active | 100% |
| 1 | Symptomatic; fully ambulatory; restricted in physically strenuous activity | 80-90% |
| 2 | Symptomatic; ambulatory; capable of self-care; more than 50% of waking hours are spent out of bed | 60-70% |
| 3 | Symptomatic; limited self-care; spends more than 50% of time in bed, but not bedridden | 40-50% |
| 4 | Completely disabled; no self-care; bedridden | 20-30% |