February 6, 2018

Martha Kruhm, MS RAC
Head, Protocol and Information Office
Quality Assurance Section
CTEP, DCT, NCI
6130 Executive Blvd, EPN Room 7000
Bethesda, MD 20892

Dear Ms. Kruhm:

Enclosed is Addendum #3 to EAI141, *Early Assessment of Treatment Response in AML using FLT PET/CT Imaging.*

The below comments were received on November 22, 2017 from CTEP's review of Addendum #3. PI responses to each comment appear in bold below.

I. Comments Requiring a Response– Major Issues:

#	Section	Comments
1.	Global	Even though the investigators only expect to have ten patients for the new exploratory objectives involving the reinduction FDG-PET/CT scans, they should include the specific summary statistics they plan to compute. For instance, do they plan to compute the median SUVs (or median changes in SUV) between patients experiencing complete remission versus those that do not? **PI Response: The study team is no longer moving forward with the new exploratory objectives involving FLT PET after reinduction.**
2.	Global	Multiple Sections: The abbreviations AP, NPIVR, IVR, RA, etc, all new in the A05 document, need to be explained. **PI Response: These abbreviations are included in the template language provided by CTSU. The expansions for each acronym listed above are included in section 4 "CTEP Investigator Registration Procedures" prior to first acronym use.**
3.	Global	Regarding the reinduction scan and timing of imaging. 1. Please explain the reasoning for the adding optional post reinduction FLT PET CT at this time, half way through this Trial's accrual plan? **PI Response: The study team is no longer moving forward with the new exploratory objectives involving FLT PET after reinduction.** 2. How many patients are expected to undergo both induction and reinduction moving forward?

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#	Section	Comments
		PI Response: The study team is no longer moving forward with the new exploratory objectives involving FLT PET after reinduction. Based on the statistics of the patients accrued so far, we would expect less than 10 patients that would receive re-induction and were candidates for a potential post re-induction scan. 3. What are the biologic and clinical differences between those patients who do and do not receive reinduction? **PI Response: Patients with resistant disease that requires re-induction chemotherapy often have high risk features such as poor risk cytogenetics, poor risk molecular findings (i.e. FLT3), or age > 60 years.** 4. What is the anticipated accrual into each of the 2 optional FLT arms and will either reach any statistical ignificance? **PI Response: The study team is no longer moving forward with the new exploratory objectives involving FLT PET after reinduction.** **For the requested statistics, please reference the supplemental data provided separate from this resubmission. With regard to the optional baseline FLT scan: we initially projected that half of the cohort would be scanned at T=0 (or baseline) and powered up the study for the corresponding aim. However, the monitoring of our data showed that the actual rate was lower than expected, thus, this won't reach statistical significance (the power calculation was mainly aimed to provide statistical significance for the primary endpoint). However, we anticipate that there will be a sufficient number of patients for providing a strong rationale for future follow-up studies specifically focusing on these exploratory imaging endpoints.** 5. Will the Bone Marrow Biopsy at Day 28-35 always be post reinduction FLT (for those patients in which this late FLT scan is performed)? Section 4.8 (and in Section 5; multiple places) suggests that this is not necessarily the case. Is it not possible that FLT scan could be positive simply due to effects of recent bone marrow biopsy? **PI Response: The study team is no longer moving forward with the new exploratory objectives involving FLT PET after reinduction.** 6. What is the expected time period from induction to reinduction? **PI Response: Reinduction chemotherapy is started soon after the day 14 marrow if there is persistent disease. The day 14 (nadir) marrow is performed 14 days after the initial chemotherapy is started.**

3.	Global	7. What is the maximal allowable time period between these for Trial purposes of obtaining a late FLT scan? **PI Response: The protocol does not have a maximum allowable time period and we think this is best to keep in place to stay in line with institutional practice. However, the study team is no longer moving forward with the post reinduction FLT PET CT.** 8. What are the statistical accrual needs for the T = 0 scan? **PI Response: Time T=0 (pre-treatment) has been revised to be exploratory as it won't reach statistical significance (the power calculation was aimed to provide statistical significance for the primary endpoint). However, we anticipate that there will be a sufficient number of patients for providing a strong rationale for future follow-up studies, specifically focusing on these exploratory imaging endpoints.** 9. Is the new plan powered to account for the several patients that are likely to accrue into the T = 0 scan and therefore be inelegible for the late FLT scan? **PI Response: The study team is no longer moving forward with the new exploratory objectives involving FLT PET after reinduction.** 10. Will the new plan lead to insufficient accrual into the T = 0 day or baseline scan due to the accrual needs for the late scan? **PI Response: The study team is no longer moving forward with the new exploratory objectives involving FLT PET after reinduction.** 11. Is the new plan powered to account for the several deaths that are likely to occur during induction? **PI Response: The study team is no longer moving forward with the new exploratory objectives involving FLT PET after reinduction.** 12. Can the PI or ECOG ACRIN provide current accrual information for the following: - Number of patients who received the baseline or T = 0 day FLT scan - Number of patients who received the T = 14 day (day 10-17) FLT scan - A complete listing and correlation of the patients in the T = 0 and T = 14 groups - Number of patients that have had the bone marrow biopsy and the dates (T=?) for these biopsies? Reasons why some patients did not receive a bon e marrow biopsy. A separate explanation for each patient that did not receive both the T = 0 and the T = 14 day FLT scans. **PI Response: For the requested statistics, please reference the supplemental data provided separate from this resubmission.**

#	Section	Comments
4.	Schema	Lettering is too small to read. Day 10-17 FLT scan should be listed as mandatory. Schema notations for figure 1 and figure 2 could be added. **PI Response: In the original amendment submission, the Post-Induction Imaging (occurs day 10-17) is noted as required in the schema. However, the study team is no longer moving forward with the new exploratory objectives involving FLT PET after reinduction.** **Footnote 3 has been amended to clarify that Day 10-17 FLT scan is mandatory.**
5.	Objectives	Exploaratory Objectives: 1. What is purpose for this amendment for obtaining the post reinduction scan? 2. Although technically correct, the terms induction and reinduction are very easy to confuse. Perhaps for the sake of maximal clarity it would be best to use completely different terminology for the post reinduction scan mentioned in this A5 document (Late scan, day ? 28 scan, or other similar name, etc). Adding a timeline with expected dates, eg T=0, T=14, etc, to the items noted on the schema would also add needed clarity. **PI Response: The study team is no longer moving forward with the new exploratory objectives involving FLT PET after reinduction.**
6.	2	Detail appears lacking especially in Section 2.3.2. Many wording changes are noted in Section 2 in keeping with PIs desire to perform an optinal late FLT scan. **PI Response: The study team is no longer moving forward with the new exploratory objectives involving FLT PET after reinduction.**
7.	4.8	Section 4.8 changes require discussion, especially as FLT scan should not occur with a few days following a bone marrow biopsy. In general molecular imaging should occur before biopsies or surgery, not immediately after. **PI Response: study team is no longer moving forward with the new exploratory objectives involving FLT PET after reinduction.**
8.	5	Section 5. A major change of the study design appears to be the introduction of chemotherapy choice number 2. Was this previously agreed to by CTEP Oncology? If so, please provide the details of this previous approval. Such requested therapy changes are subject to review via CTEP and possible review at PRC. Can the PI explain how this therapy choice might change the findings:

#	Section	Comments
		- on 18F FLT scan - of bone marrow biopsy. Is the Trial plan powered to allow for this change in therapy / choice? **PI Response: This change is no longer included in the proposed addendum, but will be addressed in a future addendum.**
9.	5.7	Section 5.7. Last sentence is not clear. A bone marrow biopsy will take place after induction. Please make clear the potential timeline differences between marrow biopsy date following induction (only) vs timeline in those patients who receive both induction and reinduction. For PIO: This is not noted on the Changes List. **PI Response: Please see item #24 on the change list of the original submission for this proposed change. However, the study team is no longer moving forward with the new exploratory objectives involving FLT PET after reinduction, so no changes will be made at this time.**
10.	7.1.6	Section 7.1.6 There are many problems with the timelines and wording used throughout the A05 document. This is why the schema should have a very clear timeline from registration, to induction, to reinduction, to bone marrow biopsy, etc. Using terms like Day 14, 7-10 days after…. can lead to needless confusion. **PI Response: The study team is no longer moving forward with the new exploratory objectives involving FLT PET after reinduction.**
11.	7.1.6	Section 7.1.8 Table. There is no Footnote 8 in the footnotes on p 64 of 103, it is on p 65 of 103 pgs in the submitted pdf. **PI Response: Footnote 8 was included in the original amendment submission. Please reference the clean version of the protocol. Additionally, no PDF versions of the protocol were submitted.** **However, the study team is no longer moving forward with the new exploratory objectives involving FLT PET after reinduction, so footnote 8 will no longer be included.**

II. Comments Requiring a Response– Administrative & Editorial Issues:

#	Section	Comments
12.	ICD	**Optional Studies:** The optional studies section of the informed consent document is not formatted per NCI Consent Template requirements and missing several sections. Please see copy of NCI Informed Consent Template under: http://ctep.cancer.gov/protocolDevelopment/default.htm#informed_consent Specifically, the following sections were omitted and needs to be added: • It is unclear as to who will pay for the extra FLT PET/CT imaging scan following the second round of chemotherapy. This was omitted in both the main consent and the optional consent section. The cost information currently listed in the optional section only pertains to the research samples collected. The below section should also be addressed for the FLT PET/CT imaging that might be obtained for the optional section. • What is involved? • What are the possible risks? • How will information about me be kept private? • What are the possible benefits? • Are there any costs or payments? (*please clearly state who will be responsible for the cost of all the optional studies requested*) • What if I change my mind? • What if I have more questions? • **PI Response: The study team is no longer moving forward with the new exploratory objectives involving FLT PET after reinduction, so no changes will be made at this time.**

III. Recommendations:

#	Section	Comments
13.	4	**In Section 4 *IRB Approval,* please delete the text in RED and replace with the text in BLUE:** Each investigator or group of investigators at a clinical site must obtain IRB approval for this protocol and submit IRB approval and supporting documentation to the CTSU Regulatory Office before they can be approved to enroll patients. Assignment of site registration status in the CTSU Regulatory Support System (RSS) uses extensive data to make a determination of whether a site has fulfilled all regulatory criteria including but not limited to the following: • An active Federal Wide Assurance (FWA) number • An active roster affiliation with the Lead Network or a participating organization • A valid IRB approval • Compliance with all protocol specific requirements. In addition, the site-protocol Principal Investigator (PI) must meet the following criteria: • Active registration status • The IRB number of the site IRB of record listed on their Form FDA 1572 • An active status on a participating roster at the registering site.

#	Section	Comments
		~~This study is supported by the NCI Cancer Trials Support Unit (CTSU). Each investigator or group of investigators at a clinical site must obtain IRB approval for this protocol and submit IRB approval and supporting documentation to the CTSU Regulatory Office before they can be approved to enroll patients. Assignment of site registration status in the CTSU Regulatory Support System (RSS) uses extensive data to make a determination of whether a site has fulfilled all regulatory criteria including but not limited to: an active Federal Wide Assurance (FWA) number, an active roster affiliation with the Lead Network or a participating organization, a valid IRB approval, and compliance with all protocol specific requirements.~~ **PI Response: This change has been made.**
14.	4.1	**Please delete Section 4.1, in accordance with the new Registration and Credentialing Repository language:** ~~**CTEP Associate Registration Procedures / CTEP-IAM Account**~~ ~~The Cancer Therapy Evaluation Program (CTEP) Identity and Access Management (IAM) application is a web-based application intended for use by both Investigators (i.e., all physicians involved in the conduct of NCI-sponsored clinical trials) and Associates (i.e., all staff involved in the conduct of NCI-sponsored clinical trials).~~ ~~Associates will use the CTEP-IAM application to register (both initial registration and annual re-registration) with CTEP and to obtain a user account.~~ ~~Investigators will use the CTEP-IAM application to obtain a user account only. (See CTEP Investigator Registration Procedures above for information on registering with CTEP as an Investigator, which must be completed before a CTEP-IAM account can be requested.)~~ ~~An active CTEP-IAM user account will be needed to access all CTEP and CTSU (Cancer Trials Support Unit) websites and applications, including the CTSU members' website.~~ ~~Additional information can be found on the CTEP website at <http://ctep.cancer.gov/branches/pmb/associate_registration.htm>. For questions, please contact the ***CTEP Associate Registration Help Desk*** by email at <ctepreghelp@ctep.nci.nih.gov>.~~ **PI Response: This change has been made. Subsequent sections have been renumbered accordingly.**

The following revisions to EAI141 protocol have been made in this addendum:

	Section	Change
1.	Cover Page	Updated Version Date. Updated Update Date.
2.	Global Changes	General formatting, grammatical, and spelling corrections made throughout.
3.	Contact Page	Revised contact information for Ryan Mattison, MD, Fenghai Duan, PhD, and Dona Alberti, BSN, RN, MSM.
4.	Study Objectives	Revised third and fourth secondary objectives to be exploratory objectives. Increased accrual goal to 83 patients accrued over a minimum of 21 months.
5.	Schema	Revised footnote 3 to clarify that Day 10-17 FLT scan is mandatory.
6.	2.2, 2.3	Revised third and fourth secondary objectives to be exploratory objectives. Inserted section 2.3 “Exploratory Objectives” in its entirety.

	Section	Change
7.	4	Revised CTEP Registration Procedures per updated CTSU language. Subsequent sections renumbered accordingly
8.	4.1	Revised IRB Approval information per updated CTSU language.
9.	4.5	Revised CTSU Regulatory Office Address.
10.	4.6	Revised Required Protocol Specific Regulatory Documents information per updated CTSU language.
11.	4.7	Revised Patient Enrollment information per updated CTSU language.
12.	4.9.3.4	Revised data collection information per updated CTSU language.
13.	4.10.1	Inserted final sentence to note RAs may submit SOC imaging via TRIAD.
14.	5	Increased accrual goal to 83 patients.
15.	5.4.2	Removed ninth bullet for the “Serious Adverse Event (SAE)” definition, as it was included in duplicate.
16.	5.4.3	Revised ECOG-ACRIN AE team phone number. Removed “Secondary Malignancy” definition, as it was included in duplicate to section 5.4.4.
17.	7.1.2	Revised first bullet to note collection of total bilirubin and creatinine instead of serum bilirubin and serum creatinine. Revised fourth bullet to note AE evaluation with the “PET Adverse Event Assessment” form.
18.	7.1.3	Revised section title to “Visit 2: Mandatory FLT PET Imaging Study (FLT-post induction).” Revised first bullet to note collection of total bilirubin and creatinine instead of serum bilirubin and serum creatinine. Revised third bullet to note AE evaluation with the “PET Adverse Event Assessment” form.
19.	7.1.5	Revised first paragraph to clarify timing of remission bone marrow biopsy in cases where reinduction therapy is administered.
20.	7.1.6	Revised Study Procedures Table footnote 1 to note collection of total bilirubin and creatinine instead of serum bilirubin and serum creatinine.
21.	8.2.7	Revised FLT administration information to eliminate ≤ 10 mL requirement.
22.	8.2.9.3	Inserted hyperlink to access the CTEP Drug Accountability Record form.
23.	10	Revised sixth paragraph and inserted table to provide rationale for accrual goal increase to 83 patients.
24.	10.2.3, 10.2.4, 10.3	Relocated third and fourth secondary objectives to 10.3.1 and 10.3.2 in new section 10.3 “Exploratory Objectives.” Subsequent sections renumbered accordingly.
25.	10.4	Revised gender and ethnicity distribution table to reflect increased accrual goal of 83 patients.
26.	Appendix II	Removed scenario #4.

The following revisions to EAI141 informed consent document have been made in this addendum:

	Section	Change
1.	Cover Page	Updated Version Date. Updated Update Date.
2.	Why is this study being done?	Revised first paragraph to reflect increased accrual goal of 83 patients.

3.	During the research study	Revised first bullet to clarify optional FLT PET/CT imaging study.

If you have any questions regarding this addendum, please contact Colin Burnett at cburnett@ecog-acrin.org or 857-504-2900.

We request review and approval of this addendum to EAI141 so ECOG-ACRIN may activate it promptly.

Thank you.

Sincerely,

Pamela Cogliano

Protocol Development Manager

Enclosure:
CC:

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ECOG-ACRIN
cancer research group
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EAI141

Early Assessment of Treatment Response in AML using FLT PET/CT Imaging

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STUDY CO-CHAIR: Ryan J. Mattison, MD
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PATHOLOGY CO-CHAIR: Daniel Arber, MD
LABORATORY CO-CHAIR: Elisabeth Paietta, PhD

Version Date: February 6, 2018
Update Date: July 26, 2017

STUDY PARTICIPANTS

Rev. 10/16

ALLIANCE / Alliance for Clinical Trials in Oncology
NRG / NRG Oncology
SWOG / SWOG

Participating sites will be required to obtain imaging qualification and have access to the FLT agent

NOTE: This study is supported by the NCI Cancer Trials Support Unit (CTSU). Institutions not aligned with ECOG-ACRIN will participate through the CTSU mechanism.

ACTIVATION DATE

December 9, 2015

PRE-ACTIVATION DATE

October 7, 2015
Update #1 – Incorporated Prior to Activation
Addendum #1 – 10/16
Addendum #2 – 7/17

Update #2 – 7/17
Addendum #3

Agents	IND#	NSC#	Supply
^{18}F-FLT		NSC#743144	Academic site supply and approved commercial vendors authorized in the IND

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Rev. 10/16
Rev. 7/17

CANCER TRIALS SUPPORT UNIT (CTSU) ADDRESS AND CONTACT INFORMATION

For regulatory requirements:	**For patient enrollments:**	**For study data submission:**
Regulatory documentation must be submitted to the CTSU via the Regulatory Submission Portal. Regulatory Submission Portal: (Sign in at www.ctsu.org, and select the Regulatory Submission sub-tab under the Regulatory tab.) Institutions with patients waiting that are unable to use the Portal should alert the CTSU Regulatory Office immediately at 1-866-651-2878 to receive further instruction and support. Contact the CTSU Regulatory Help Desk at 1-866-651-2878 for regulatory assistance.	Please refer to the patient enrollment section of the protocol for instructions on using the Oncology Patient Enrollment Network (OPEN) which can be accessed at https://www.ctsu.org/OPEN_SYSTEM/ or https://OPEN.ctsu.org. Contact the CTSU Help Desk with any OPEN-related questions at ctsucontact@westat.com.	Data collection for this study will be done exclusively through Medidata Rave. Please see the data submission section of the protocol for further instructions.
The most current version of the **study protocol and all supporting documents** must be downloaded from the protocol-specific Web page of the CTSU Member Web site located at https://www.ctsu.org. Access to the CTSU members' website is managed through the Cancer Therapy and Evaluation Program - Identity and Access Management (CTEP-IAM) registration system and requires user log on with CTEP-IAM username and password.		
For clinical questions (i.e., patient eligibility or treatment-related) Contact the Study PI of the Coordinating Group.		
For non-clinical questions (i.e., unrelated to patient eligibility, treatment, or clinical data submission) contact the CTSU Help Desk by phone or e-mail: CTSU General Information Line – 1-888-823-5923, or ctsucontact@westat.com. All calls and correspondence will be triaged to the appropriate CTSU representative.		
The CTSU Web site is located at https://www.ctsu.org		

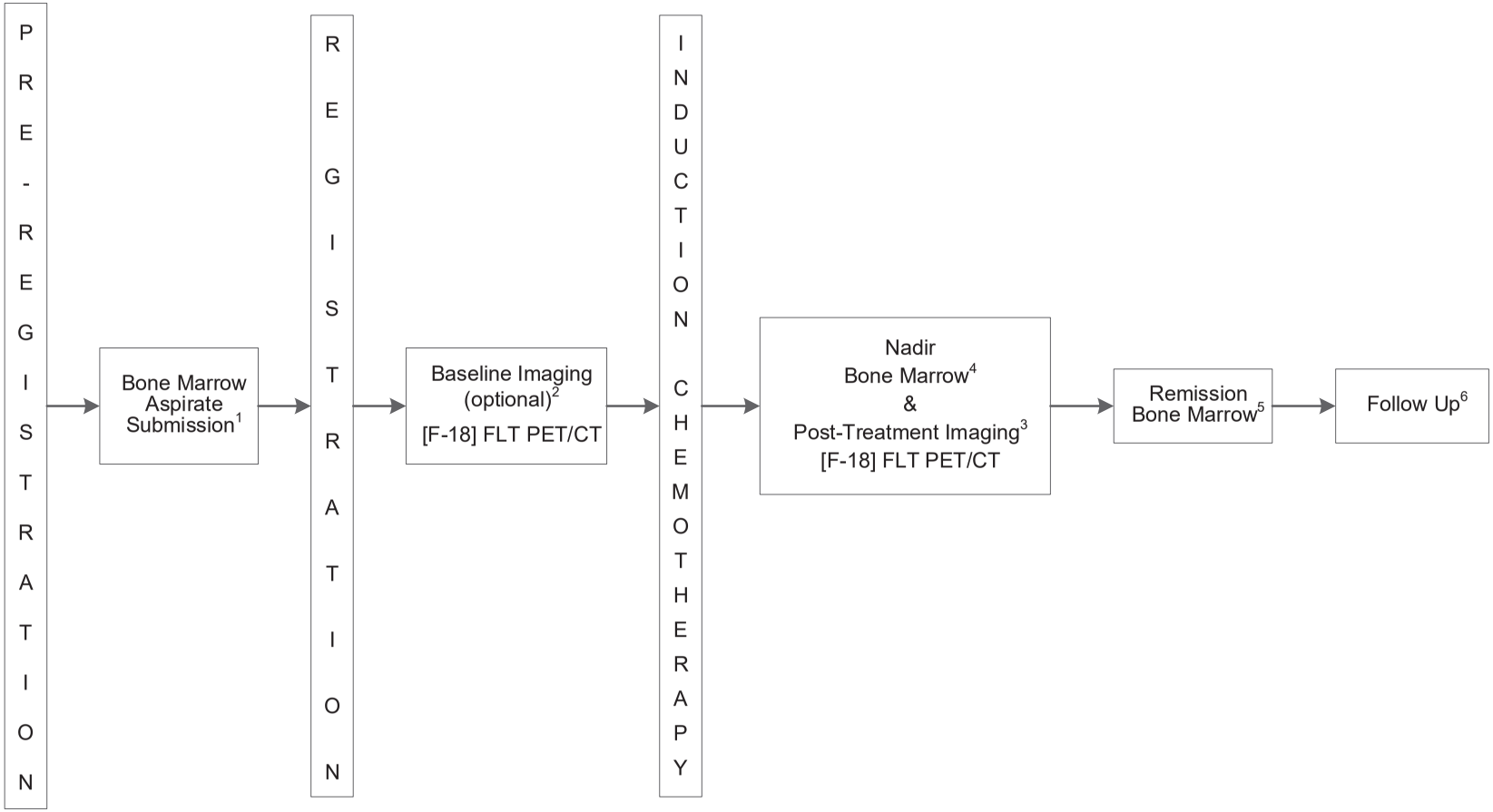
Rev. 10/16

ABBREVIATIONS

AE: Adverse event
APL: acute promyelocytic leukemia
AML: acute myeloid leukemia
BMBX: bone marrow biopsy
CDUS: CTEP Data Update System
CR: complete remission
CRi: complete remission with incomplete platelet recovery
CTEP: Cancer Therapy Evaluation Program
CTSU: Cancer Trials Support Unit
ECOG-ACRIN: Eastern Cooperative Oncology Group-American College of Radiology Imaging Network
FLT: ^{18}F-fluoro-3'deoxy-3'-D-fluorothymidine
FLT-base: optional baseline FLT PET/CT
FLT-post: post-treatment FLT PET/CT
IMAC: Image Analysis Core
LFS: leukemia-free state
LTB: Leukemia Tissue Bank
LTRL: Leukemia Translational Research Laboratory
LVEF: left ventricular ejection fraction
MRD: minimal residual disease
NCCN: National Comprehensive Cancer Network
NPV: negative predictive value
OPEN: Oncology Patient Enrollment Network
PET/CT: positron emission tomography/computed tomography
PPV: positive predictive value
PR: Partial Remission
RD: residual disease
RSS: Regulatory Support System
STS: Sample Tracking System
SUV: Standardized Uptake Value
SUV_{max}: maximum FLT uptake across the total bone marrow compartment
SUV_{mean}: mean FLT uptake across the total bone marrow compartment
SUV_{hetero}: coefficient of variation of FLT uptake across the total bone marrow compartment

Rev. 10/16

Schema



1. Pre-treatment bone marrow aspirate, or peripheral blood should be sent for central review.
2. Optional Imaging must be done within 1 week prior to initiation of therapy.
3. Post-Treatment Imaging is mandatory and must be completed 10-17 days after initiation of first induction cycle and prior to reinduction.
4. Nadir Bone Marrow should be completed 7-10 days after completion of induction therapy. Remission Bone Marrow should be completed 28-35 days after initiation of first induction therapy
5. Remission bone marrow aspirate should be sent for central analysis.
6. Participants will be followed for up to 1 year beyond the end of the accrual period.

Rev. 10/16

STUDY OBJECTIVES

Primary Objective

To evaluate the negative predictive value (NPV) of post-treatment FLT PET/CT imaging for complete remission (CR) in patients receiving induction chemotherapy for acute myeloid leukemia (AML).

Secondary Objectives

- To evaluate the positive predictive value (PPV) of post-treatment FLT PET/CT imaging for complete remission;
- To estimate the sensitivity and specificity of post-treatment FLT PET/CT imaging for detecting complete remission;
- To correlate FLT PET/CT imaging with biologic correlates (MRD Assessment)
- To correlate FLT PET/CT imaging with relapse-free survival and overall survival.

Rev. Add3

Exploratory Objectives

- To evaluate pre-treatment FLT PET/CT imaging as a predictor of complete remission;
- To evaluate the change between pre-treatment and post-treatment FLT PET/CT imaging as a predictor of complete remission;

ELIGIBILITY *(see Section 3 for details)*

Patients must be 18 years old and have previously untreated AML and be candidates for intensive induction chemotherapy. Patients are allowed to have had prior hydroxyurea. Patients must not have acute promyelocytic leukemia (APL) and must not have evidence of t(15;17)(q22;q21). Patients must have an ECOG performance status of 0-3 (restricted to ECOG PS 0-2 if age > 70 years).

SAMPLE SIZE

Rev. Add3

A total of 83 eligible AML patients who are candidates for induction chemotherapy will be enrolled into this trial from approved NCTN institutions. Anticipated accrual is four participants per month, to be accrued over a minimum of 21 months. All participants will receive the post-treatment FLT PET/CT within three days before or after the nadir bone marrow biopsy (between Days 10-17 after initiation of first induction cycle and prior to reinduction). Eligible participants will be actively involved in the trial until they complete all trial procedures, specifically the remission bone marrow biopsy (Day 28-35 biopsy). Patients will be followed for survival outcomes for at least 1 year (1 year beyond the end of the study accrual period).

1. Introduction

1.1 Abstract

In patients with acute myeloid leukemia (AML), developing a more accurate early predictor of chemotherapy effectiveness is needed to avoid unnecessary therapy, to identify refractory AML patients earlier, and to improve overall clinical outcomes. One potential tool is to incorporate fluorothymidine positron emission tomography/computed tomography (FLT PET/CT) imaging as a surrogate of response during treatment. FLT PET/CT is a molecular imaging technique that has the ability to measure proliferative activity in the bone marrow before and after chemotherapy. As a whole body imaging modality, PET/CT provides the ability to assess to entire bone marrow compartment rather than the relatively small sample obtained during a bone marrow aspirate or biopsy. Given the need for a better method to assess chemotherapy effectiveness in AML, and the pilot results that indicate FLT PET/CT as a potential method, we plan to conduct a multicenter trial assessing the predictive value of FLT PET/CT in AML through ECOG-ACRIN.

1.2 FLT PET/CT in AML: A Potential Early Predictor of Response

Acute myeloid leukemia (AML) is a devastating, clinically heterogeneous disease for which improved treatment strategies are needed. Long-term cure depends upon the ability to induce a complete remission, followed by consolidation treatment with either chemotherapy or stem cell transplantation. Standard induction chemotherapy is administered over 7 days, after which treatment response is usually assessed at two time points after initiation of chemotherapy: the nadir bone marrow at approximately Day 14, and at the time of peripheral blood count recovery, which is at approximately Day 28.

If the Day 14 marrow has a significant number of residual leukemia blasts, a second induction therapy course can be administered. If the Day 14 marrow is hypoplastic, the patient will receive no further treatment at that time and the counts will be allowed to recover. However, the Day 14 marrow has limitations as a marker of the success of induction therapy. A substantial number of patients with no blasts in the nadir marrow will not achieve remission (a false negative result). On the other hand, some patients who have residual disease in the marrow at Day 14 will eventually achieve a complete remission (CR) even without another induction chemotherapy course (a false positive result). A review of the literature showed the nadir marrow as a test has a false negative rate ranging from 21% to 33% and a false positive rate of 19% to 36%. Possible explanations for these error rates include a fairly limited sample of an extensive marrow space, heterogeneity of AML involvement in the bone marrow, and lack of clarity about whether residual blasts are in the process of dying at the time of assessment or whether they are blasts resistant to chemotherapy.

A better early predictor of residual disease that assesses chemotherapy effectiveness would give clinicians and patients an opportunity to modify therapeutic strategies. Preliminary, single-center data support proliferation imaging using an investigational agent known as 3'-deoxy-3'-^{18}F-fluorothymidine (FLT) in positron emission tomography/computed tomography (PET/CT) as a method of assessing the entire burden of disease and avoid the confounding

effects of marrow blasts that are still viable but no longer proliferative and to marrow heterogeneity, which may overcome the limitations of the Day 14 bone marrow. The goal of the proposed trial is to validate these promising early results in a prospective, multi-center trial of FLT PET/CT as a predictor of CR for induction therapy in AML.

The investigational agent FLT is a structural analog of thymidine, a DNA constituent. FLT is an ^{18}F radiolabeled imaging agent that has been used in conjunction with PET/CT. FLT is trapped in the cell due to phosphorylation by thymidine kinase as part of the proliferation pathway. As such, FLT is a marker of proliferating tumor tissues in proportion to DNA synthesis rates. Prior studies have shown that FLT kinetics are particularly favorable in detecting proliferating normal marrow and leukemia cells, making FLT a potentially very sensitive tool for residual proliferating leukemia after induction therapy.

FLT PET/CT thus has the potential for providing a whole-body assessment of leukemia and associated bone marrow proliferation in contrast to the point sample provided during bone marrow aspirate and biopsy. Agool et al. have investigated differences in spatial uptake patterns of FLT to distinguish different hematological disorders. Buck et al. have explored the use of FLT PET to stage patients with lymphoma by determining bone marrow involvement and have analyzed bone marrow uptake and extramedullary uptake in patients with AML, suggesting quantification of FLT uptake could lead to treatment response biomarkers. The most relevant to this proposal is a pilot study using FLT PET/CT in patients with AML conducted at the University of Wisconsin. The results of that study have been extremely promising (see Preliminary Data), but only included a limited cohort of patients. We now plan to open a larger multi-center trial to validate the single-center results and make this tool available to guide future AML therapy trials.

A positive trial would contribute multi-center data on the validity of this predictive biomarker as a clinical assessment tool towards induction therapy and for further study as a potentially practice-changing metric for guiding treatment of AML. FLT PET/CT imaging would provide indication about which patients would benefit from post-remission allogeneic stem cell transplantation versus consolidation chemotherapy. It also would allow early identification of those in whom a standard anthracycline plus cytarabine therapeutic approach will be insufficient or completely ineffective. As such, FLT PET/CT could potentially provide an integral biomarker for future AML trials if validated as a predictor of induction CR.

This trial will also have important consequences for a better understanding of AML. Demonstrating the phenotypic behavior of a disease through FLT PET/CT imaging will complement molecular, genotypic abnormalities in helping clinicians choose appropriate post-remission therapy (currently chemotherapy vs. transplant).

1.3 FLT PET/CT in AML: Preliminary Data

In a pilot study by the University of Wisconsin group (Jeraj, Juckett PIs), 8 adult subjects with AML receiving induction chemotherapy underwent FLT PET/CT scans prior to therapy and at different time points during induction chemotherapy. A dramatic difference in the bone marrow FLT uptake in those patients who achieved a complete remission (CR) versus those who had residual *disease* (RD) present at count recovery was found (Figure 1).

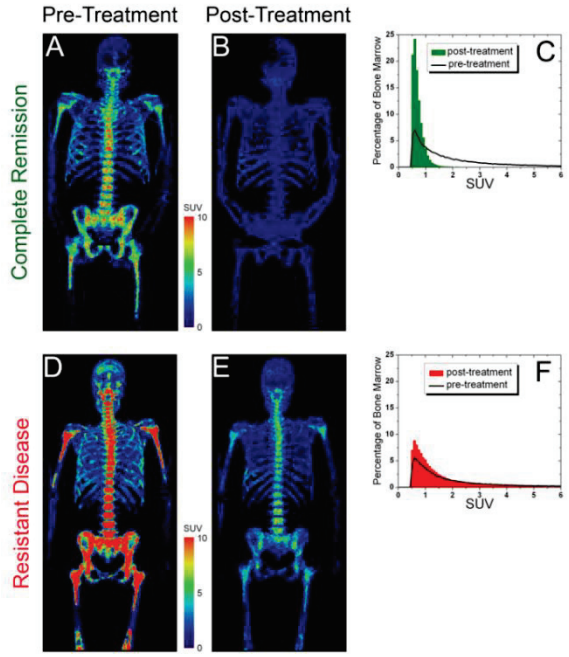


Figure 1: Remarkably different bone marrow proliferation response between complete response (CR) and residual disease (RD) in patients, pre- and post-treatment. Pre-treatment, FLT PET maximum intensity projection image of bone marrow of RD patient (D) exhibited markedly greater uptake than CR patient (A). Post-treatment, FLT PET image of CR patient (B) revealed very low uptake, suggesting successful bone marrow ablation. However, post-treatment FLT PET image of RD patient (E) displayed significant uptake, indicating substantial remaining proliferative bone marrow and poor treatment response. Post-treatment, bone marrow Standardized Uptake Value (SUV) distribution of CR patient (C) was completely within the SUV < 2 range, while that of RD patient contained a significant portion in the SUV > 2 range (F).

Interestingly, the significant differences were observed in pre-treatment FLT PET/CT scans between patients with AML and normal subjects; however *only* for RD patients; the CR patients had "normal" looking proliferation characteristics (Figure 2).

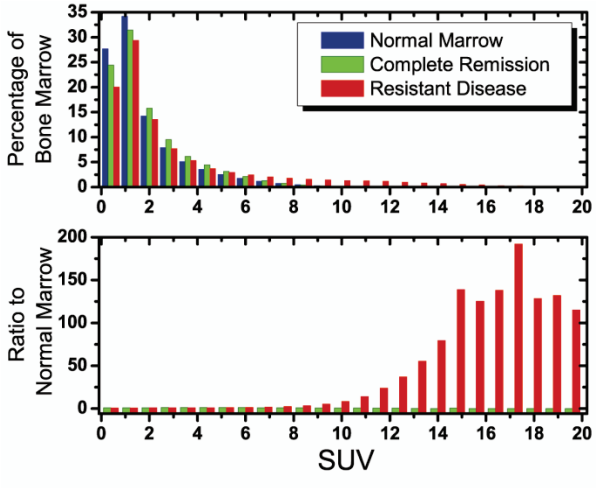


Figure 2: Significantly different pre-treatment bone marrow proliferation characteristics in normal subjects with that of patients with AML pre-treatment. FLT Standardized Uptake Value (SUV) distribution reveals similarity of normal marrow (blue) and marrow of CR patient pre-treatment (green), with almost all uptake in SUV < 8 range. Distribution is quite different for RD patient (red) who displayed significant uptake in the SUV > 8 range (top panel). At all SUV, the ratio of CR patients' pre-treatment marrow to normal marrow uptake was approximately unified. At SUV > 8, RD patients' pre-treatment marrow often displayed uptake *over 100 times greater* than that of normal marrow (bottom panel).

These data provide strong rationale for FLT PET as a test of residual AML that moves beyond the current dichotomy of presence/absence of blasts in marrow samples. These data support FLT PET as a marker for early AML response assessment and therapeutic guidance that deserves further evaluation.

1.4 ^{18}F-fluoro-3'deoxy-3'-D-fluorothymidine (FLT)

3'-deoxy-3'-^{18}F fluorothymidine ([F-18]FLT) (half life = 110 mins) is a structural analog of the DNA constituent, thymidine (Figure 3). It is a radiolabeled PET imaging agent that has been proposed for investigating cellular proliferation as an analogue of thymidine. Although FLT is not incorporated into DNA, it is

trapped in the cell due to phosphorylation by thymidine kinase 1. It is also relatively resistant to *in vivo* degradation and accumulates predominantly in proliferating tissues in proportion to the DNA synthesis rate. Hence it is considered as a potential marker of tumor cellular proliferation. Therefore, FLT is proposed as a radiolabeled imaging probe for *in vivo* assessment of cellular proliferation in malignant tumors using PET.

Figure 3 (from Investigator's Brochure, Edition Number 7; Edition Date: 2009)

Briefly, FLT is taken up by cells and follows the salvage pathway of DNA synthesis and, like thymidine, undergoes phosphorylation by thymidine kinase 1 (TK1), which leads to intracellular trapping within the cell. FLT is a selective substrate for TK1. In quiescent cells, TK1 activity is virtually absent but in proliferating cells, it is increased about 10-fold, as cells enter the DNA synthetic phase of the cell cycle.[1-3] The uptake of FLT has been found to increase with increasing TK1 activity in human tumor cell lines. As FLT is not incorporated into DNA, FLT uptake has been validated with an independent measure of DNA synthesis in tissue. The protein Ki-67 identified by MIB-1 antibody staining is a histopathologic measure of proliferation, which correlates with the level of FLT uptake measured by PET.

1.5 Previous Human FLT Imaging Studies

Many clinical studies have used FLT PET imaging (UCLA, University of Washington in Seattle, Wayne State University, University of Wisconsin). The imaging protocols were pre-approved by their respective regulatory committees and conducted under the RDRC process or under NCI IND. FLT PET/CT has been used previously in the ACRIN 6688 multi-center clinical trial (Phase II Study of 3'-Deoxy-3'-18F Fluorothymidine [FLT] in Invasive Breast Cancer) under the NCI IND to be used for this study. ACRIN 6688 was successfully completed without any safety issues.

1.6 Rationale

In patients with AML undergoing intensive induction treatment with anthracycline and infusional cytarabine, the nadir bone marrow evaluation can be useful as an early measure of chemotherapy effectiveness. However, the results of this evaluation are often inaccurate. In a review of six clinical trials, the nadir marrow's ability to predict CR was highly variable [2]. The test characteristics of sensitivity and specificity ranged from 67-94% and 43-81%, respectively. Similarly, the negative predictive value (NPV) that predicts CR after a hypocellular nadir marrow ranged from 67-84%. The predictive value that

predicts lack of remission when persistent blasts were seen at the nadir ranged from 64-81%. One hypothesis that could partially explain the inaccuracy of the nadir marrow is that it is difficult to tell whether an intermediate blast count at the nadir represents refractory disease, or the dying leukemia cells. In addition, the marrow aspirate and biopsy is a relatively small tissue sample that may miss areas of active disease. Thus, there is a risk of undertreating patients who will not achieve a remission despite a negative nadir bone marrow, while on the other hand, overtreating others who would eventually achieve CR despite having persistent blasts during the early marrow evaluation.

In a review of six Eastern Cooperative Oncology Group (ECOG) leukemia studies, Rowe and colleagues found that patients who entered a CR with induction chemotherapy had 5-year and 10-year survival rates that were similar, regardless of whether one of two induction cycles were required to achieve remission [1]. According to the treatment protocols, all patients with persistent disease during the nadir marrow were treated with a second chemotherapy course. Therefore, it is not known whether some patients with an intermediate blast count at the nadir would have achieved CR without a second course of treatment. Designing a study to randomize patients with an intermediate nadir blast burden to either observation until count recovery or to a second course of chemotherapy is technically possible, but would be difficult to conduct given clinicians' reluctance to break from the standard treatment paradigm. In addition, spatial analysis of the FLT PET/CT response conducted at the University of Wisconsin indicated high spatial heterogeneity of response to chemotherapy. This response heterogeneity may help explain the weak predictive power of the biopsy, which typically samples only one small region of the pelvic bone marrow. Given the need for a better method to assess chemotherapy effectiveness in AML, and the pilot results that indicate FLT PET/CT may be one potential method, we plan to conduct this trial through ECOG-ACRIN.

2. Hypothesis

The negative predictive value (NPV) of the post-treatment FLT PET/CT imaging for complete remission (CR) in patients receiving induction chemotherapy for AML will be higher than the NPV of the Day 14 bone marrow biopsy for CR.

Objectives

2.1 Primary Objectives

To evaluate the negative predictive value (NPV) of post-treatment FLT PET/CT imaging for complete remission (CR) in patients receiving induction chemotherapy for acute myeloid leukemia (AML).

Three imaging parameters (SUV_{mean}, SUV_{max}, SUV_{hetero}) will be measured from an FLT PET/CT scan and SUV_{max} will be the primary endpoint.

Note that the CR designation requires that the patient achieve the morphologic leukemia-free state and have an absolute neutrophil count of more than 1,000/uL and platelets of greater than 100,000/uL. Hemoglobin concentration or hematocrit has no bearing on remission status, although the patient must be independent of transfusions. Occasionally, a rare peripheral blood blast may be identified during regeneration; however, if the patient is in CR, the bone marrow would have less than 5% blasts and no Auer rods [4].

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2.2 Secondary Objectives

2.2.1 To evaluate the positive predictive value (PPV) of post-treatment FLT PET/CT imaging for complete remission;

2.2.2 To estimate sensitivity and specificity of post-treatment FLT PET/CT imaging for detecting complete remission;

2.2.3 To correlate FLT PET/CT imaging with biologic correlates (MRD assessment);

2.2.4 To correlate FLT PET/CT imaging with relapse-free survival and overall survival.

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2.3 Exploratory Objectives

2.3.1 To evaluate pre-treatment FLT PET/CT imaging as a predictor of complete remission;

2.3.2 To evaluate the change between pre-treatment and post-treatment FLT PET/CT imaging as a predictor of complete remission;

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3. Selection of Patients

Each of the criteria in the checklist that follows must be met in order for a patient to be considered eligible for this study. Use the checklist to confirm a patient's eligibility. For each patient, this checklist must be photocopied, completed and maintained in the patient's chart.

In calculating days of tests and measurements, the day a test or measurement is done is considered Day 0. Therefore, if a test is done on a Monday, the Monday four weeks later would be considered Day 28.

ECOG-ACRIN Patient No. _____

Patient's Initials (L, F, M) _____

Physician Signature and Date _____

NOTE: This study involves preregistration (Step 0, see Section 4). Bone marrow or peripheral blood specimens must be submitted to the ECOG-ACRIN Leukemia Translational Research Laboratory (LTRL). If bone marrow aspirate was performed prior to preregistration/patient consent, peripheral blood can be submitted after registration to Step 1, prior to protocol defined imaging.

3.1 Eligibility Criteria

_____ 3.1.1 Age ≥ 18 years

_____ 3.1.2 Patients must have previously untreated AML and be candidates for intensive induction chemotherapy. Patients are allowed to have had prior hydroxyurea

_____ 3.1.3 Patients must not have acute promyelocytic leukemia (APL) and must not have evidence of t(15;17)(q22;q21)

_____ 3.1.4 Patients must have an ECOG performance status of 0-3 (restricted to ECOG PS 0-2 if age >70 years)

_____ 3.1.5 Patients must have left ventricular ejection fraction (LVEF) > 45% or within institutional normal limits

_____ 3.1.6 Patients must be able to lie still for a 1.5 hour PET scan

_____ 3.1.7 Patient must NOT have a history of allergic reaction attributable to compounds of similar chemical or biologic composition to ^{18}F-fluorothymidine

_____ 3.1.8 Patient must NOT weigh more than the maximum weight limit for the PET/CT table for the scanner(s) to be used at each center

_____ 3.1.9 The patient is participating in the trial at an institution which has agreed to perform the imaging research studies, completed the ECOG-ACRIN defined scanner qualification procedures and received ECOG-ACRIN approval as outlined in Section 9.4

_____ 3.1.10 Women must not be pregnant or breast-feeding due to unknown toxic effects of the FLT agent on a fetus

All females of childbearing potential must have a blood test or urine study within 2 weeks prior to registration to rule out pregnancy.

A female of childbearing potential is any woman, regardless of sexual orientation or whether they have undergone tubal ligation, who meets the following criteria: 1) has not undergone a hysterectomy or bilateral oophorectomy; or 2) has not been naturally postmenopausal for at least 24 consecutive months (i.e., has had menses at any time in the preceding 24 consecutive months).

Female? _____ (Yes or No)

Date of blood test or urine study: _____

_____ _____

Physician Signature Date

OPTIONAL: This signature line is provided for use by institutions wishing to use the eligibility checklist as source documentation.

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4. Registration Procedures

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CTEP Registration Procedures

Food and Drug Administration (FDA) regulations and National Cancer Institute (NCI) policy require all individuals contributing to NCI-sponsored trials to register and to renew their registration annually. To register, all individuals must obtain a Cancer Therapy Evaluation Program (CTEP) Identity and Access Management (IAM) account (https://ctepcore.nci.nih.gov/iam). In addition, persons with a registration type of Investigator (IVR), Non-Physician Investigator (NPIVR), or Associate Plus (AP) (i.e., clinical site staff requiring write access to OPEN, RAVE, or TRIAD or acting as a primary site contact) must complete their annual registration using CTEP's web-based Registration and Credential Repository (RCR) (https://ctepcore.nci.nih.gov/rcr). Documentation requirements per registration type are outlined in the table below.

Documentation Required	IVR	NPIVR	AP	A
FDA Form 1572	✔	✔		
Financial Disclosure Form	✔	✔	✔	
NCI Biosketch (education, training, employment, license, and certification)	✔	✔	✔	
HSP/GCP training	✔	✔	✔	
Agent Shipment Form (if applicable)	✔			
CV (optional)	✔	✔	✔	

An active CTEP-IAM user account and appropriate RCR registration is required to access all CTEP and CTSU (Cancer Trials Support Unit) websites and applications. In addition, IVRs and NPIVRs must list all clinical practice sites and IRBs covering their practice sites on the FDA Form 1572 in RCR to allow the following:

- Added to a site roster
- Assigned the treating, credit, consenting, or drug shipment (IVR only) tasks in OPEN
- Act as the site-protocol PI on the IRB approval
- Assigned the Clinical Investigator (CI) role on the Delegation of Tasks Log (DTL).

Additional information can be found on the CTEP website at <https://ctep.cancer.gov/investigatorResources/default.htm>.

For questions, please contact the RCR ***Help Desk*** by email at <RCRHelpDesk@nih.gov>.

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4.1 IRB Approval

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Each investigator or group of investigators at a clinical site must obtain IRB approval for this protocol and submit IRB approval and supporting documentation to the CTSU Regulatory Office before they can be approved to enroll patients. Assignment of site registration status in the CTSU Regulatory Support System (RSS) uses extensive data to make a determination of whether a site has fulfilled all regulatory criteria including but not limited to the following:

- An active Federal Wide Assurance (FWA) number

- An active roster affiliation with the Lead Network or a participating organization
- A valid IRB approval
- Compliance with all protocol specific requirements.

In addition, the site-protocol Principal Investigator (PI) must meet the following criteria:

- Active registration status
- The IRB number of the site IRB of record listed on their Form FDA 1572
- An active status on a participating roster at the registering site.

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Sites participating on the NCI CIRB initiative that are approved by the CIRB for this study are not required to submit IRB approval documentation to the CTSU Regulatory Office. For sites using the CIRB, IRB approval information is received from the CIRB and applied to the RSS in an automated process. Signatory Institutions must submit a Study Specific Worksheet for Local Context (SSW) to the CIRB via IRB Manager to indicate their intent to open the study locally. The CIRB's approval of the SSW is then communicated to the CTSU Regulatory Office. In order for the SSW approval to be processed, the Signatory Institution must inform the CTSU which CIRB-approved institutions aligned with the Signatory Institution are participating in the study.

4.2 Downloading Site Registration Documents

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Site registration forms may be downloaded from the EAI141 protocol page located on the CTSU members' website.

- Go to https://www.ctsu.org and log in to the members' area using your CTEP-IAM username and password
- Click on the Protocols tab in the upper left of your screen
- Either enter the protocol # in the search field at the top of the protocol tree, or
- Click on the By Lead Organization folder to expand
- Click on the ECOG-ACRIN link to expand, then select trial protocol #EAI141
- Click on LPO Documents, select the Site Registration documents link, and download and complete the forms provided.

4.3 Requirements For EAI141 Site Registration

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- IRB approval (For sites not participating via the NCI CIRB; local IRB documentation, an IRB-signed CTSU IRB Certification Form, Protocol of Human Subjects Assurance Identification/IRB Certification/Declaration of Exemption Form, or combination is accepted)
- PET/CT Qualification (through the ACR Imaging Core Laboratory)

4.4 Checking Your Site's Registration Status

You can verify your site registration status on the members' section of the CTSU website.

- Go to https://www.ctsu.org and log in to the members' area using your CTEP-IAM username and password

- Click on the Regulatory tab
- Click on the Site Registration tab
- Enter your 5-character CTEP Institution Code and click on Go

NOTE: The status given only reflects compliance with IRB documentation and institutional compliance with protocol-specific requirements outlined by the Lead Network. It does not reflect compliance with protocol requirements for individuals participating on the protocol or the enrolling investigator's status with the NCI or their affiliated networks.

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4.5 Submitting Regulatory Documents

Submit required forms and documents to the CTSU Regulatory Office via the Regulatory Submission Portal, where they will be entered and tracked in the CTSU RSS.

Regulatory Submission Portal: www.ctsu.org (members' area) → Regulatory Tab → Regulatory Submission

When applicable, original documents should be mailed to:

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CTSU Regulatory Office
1818 Market Street, Suite 3000
Philadelphia, PA 19103

Institutions with patients waiting that are unable to use the Portal should alert the CTSU Regulatory Office immediately at 1-866-651-2878 in order to receive further instruction and support

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4.6 Required Protocol Specific Regulatory Documents

1. Copy of IRB Informed Consent Document.

NOTE: Any deletion or substantive modification of information concerning risks or alternative procedures contained in the sample informed consent document must be justified in writing by the investigator and approved by the IRB.

2. A. CTSU IRB Certification Form.
   Or
   B. Signed HHS OMB No. 0990-0263 (replaces Form 310).
   Or
   C. IRB Approval Letter

NOTE: The above submissions must include the following details:

- Indicate all sites approved for the protocol under an assurance number
- OHRP assurance number of reviewing IRB
- Full protocol title and number
- Version Date
- Type of review (full board vs. expedited)
- Date of review
- Signature of IRB official

4.7 Patient Enrollment

Patients must not start protocol procedures prior to Step 1 registration.

The first, baseline FLT PET/CT scan is optional due to the time restriction, such as the two-day minimum requirement for ordering and delivery of the FLT agent when supplied by commercial distributors. Qualified institutions manufacturing their own FLT may be able to supply FLT for the study and minimize time delays due to FLT ordering (see Section 8.2.9.1). Such centers will complete the FLT PET/CT before treatment initiation when feasible and if it will not delay patient care.

The second, post-treatment FLT PET/CT scan is mandatory and must be completed per study imaging protocol specifications.

Patient enrollment will be facilitated using the Oncology Patient Enrollment Network (OPEN). OPEN is a web-based registration system available on a 24/7 basis. To access OPEN, the site user must have an active CTEP-IAM account (check at <https://ctepcore.nci.nih.gov/iam>) and a 'Registrar' role on either the LPO or participating organization roster. Registrars must hold a minimum of an AP registration type.

All site staff will use OPEN to enroll patients to this study. It is integrated with the CTSU Enterprise System for regulatory and roster data and, upon enrollment, initializes the patient in the Rave database. OPEN can be accessed at https://open.ctsu.org or from the OPEN tab on the CTSU members' side of the website at https://www.ctsu.org. To assign an IVR or NPIVR as the treating, crediting, consenting, drug shipment (IVR only), or investigator receiving a transfer in OPEN, the IVR or NPIVR must list on their Form FDA 1572 in RCR the IRB number used on the site's IRB approval.

Prior to accessing OPEN site staff should verify the following:

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- All eligibility criteria has been met within the protocol stated timeframes
- All patients have signed an appropriate consent form and HIPAA authorization form (if applicable).

NOTE: The OPEN system will provide the site with a printable confirmation of registration and treatment information. Please print this confirmation for your records.

Further instructional information is provided on the OPEN tab of the CTSU members' side of the CTSU website at https://www.ctsu.org or at https://open.ctsu.org. For any additional questions contact the CTSU Help Desk at 1-888-823-5923 or ctsucontact@westat.com.

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4.8 Preregistration (Step 0)

NOTE: Patients must not start protocol procedures prior to Step 1 registration.

4.8.1 The following information will be collected at time of preregistration.

4.8.1.1 Protocol Number

4.8.1.2 Investigator Identification

- Institution and affiliate name (Institution CTEP ID)

- Investigator's name (NCI number)
- Cooperative Group Credit
- Credit Investigator
- Protocol specific contact information

4.8.1.3 Patient Identification

- Patient's initials (first and last)
- Patient's Hospital ID and/or Social Security number
- Patient demographics
  - Gender
  - Birth date
  - Race
  - Ethnicity
  - Nine-digit ZIP code
  - Method of payment
  - Country of residence

4.8.2 Eligibility Verification

Patient must be consented to and be considered a viable candidate for registration to Step 1 on ECOG-ACRIN EAI141.

If bone marrow aspirate has been performed prior to preregistration to Step 0, patient must meet all of the eligibility requirements listed in Section 3.

4.8.3 Additional Requirements

4.8.3.1 Patients must provide a signed and dated, written informed consent form.

NOTE: Copies of the consent are not collected by the ECOG-ACRIN Operations Office. However the ECOG-ACRIN Leukemia Translational Research Laboratory (LTRL) requires institutions to submit a copy of the EAI141 consent and a copy of the HIPAA Authorization form.

4.8.3.2 Bone marrow aspirates or peripheral blood specimens **must be submitted** for defined laboratory research studies. See Section 11.

NOTE: Peripheral blood can be submitted after registration (Step 1) to coincide with clinical assessments if bone marrow aspirates had been obtained prior to consent and will not be submitted at preregistration.

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4.9 Registration (Step 1)

4.9.1 The following information will be collected at time of registration.

4.9.1.1 Protocol Number

4.9.1.2 Investigator Identification

- Institution and affiliate name
- Investigator's name

4.9.1.3 Patient Identification

- Patient's initials (first and last)
- Patient's Hospital ID and/or Social Security number
- Patient demographics
  - Gender
  - Birth date (mm/yyyy)
  - Race
  - Ethnicity
  - Nine-digit ZIP code
  - Method of payment
  - Country of residence

4.9.2 Eligibility Verification

Patients must meet all of the eligibility requirements listed in Section 3.

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4.9.3 Additional Requirements

4.9.3.1 Images are to be submitted as indicated in Section 4.10.

4.9.3.2 Bone marrow smears are to be submitted for future undefined research as outlined in Section 11.

4.9.3.3 Bone marrow aspirates must be submitted for defined laboratory research studies as outlined in Section 11.

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4.9.3.4 Data collection for this study will be done exclusively through the Medidata Rave clinical data management system. Access to the trial in Rave is granted through the Medidata application to all persons with the appropriate roles assigned in Regulatory Support System (RSS). To access Rave via Medidata, the site user must have an active CTEP-IAM account (check at <https://ctepcore.nci.nih.gov/iam>) and the appropriate Rave role (Rave CRA, Read-Only, CRA (Lab Admin, SLA or Site Investigator)) on either the LPO or participating organization roster at the enrolling site. To the hold Rave CRA role or CRA Lab Admin role, the user must hold a minimum of an AP registration type. To hold the Rave Site Investigator role, the individual must be registered as an NPIVR or IVR. Associates can hold read-only roles in Rave.

Upon initial site registration approval for the study in RSS, all persons with Rave roles assigned on the appropriate roster will be sent a study invitation e-mail from Medidata. To accept the invitation, site users must log into the Select

Login (https://login.imedidata.com/selectlogin) using their CTEP-IAM user name and password, and click on the "accept" link in the upper right-corner of the iMedidata page. Please note, site users will not be able to access the study in Rave until all required Medidata and study specific trainings are completed. Trainings will be in the form of electronic learnings (eLearnings), and can be accessed by clicking on the link in the upper right pane of the Medidata screen.

Users that have not previously activated their Medidata/Rave account at the time of initial site registration approval for the study in RSS will also receive a separate invitation from Medidata to activate their account. Account activation instructions are located on the CTSU website, Rave tab under the Rave resource materials (Medidata Account Activation and Study Invitation Acceptance). Additional information on Medidata/Rave is available on the CTSU members' website under the Rave tab at www.ctsu.org/RAVE/ or by contacting the CTSU Help Desk at 1-888-823-5923 or by e-mail at ctsucontact@westat.com.

4.10 Digital Images Data Submission Using TRIAD

TRIAD is the American College of Radiology's (ACR) image exchange application. TRIAD provides sites participating in clinical trials a secure method to transmit DICOM RT and other objects. TRIAD anonymizes and validates the images as they are transferred.

4.10.1 TRIAD Access Requirements

Site radiology staff who will submit images through TRIAD will need to be registered with the Cancer Therapy Evaluation Program (CTEP) and have a valid and active CTEP Identity and Access Management (IAM) account and be registered as an AP, NPIVR or IVR. Please refer to CTEP Registration Procedures of the protocol in Section **Error! Reference source not found.** for instructions on how to request a CTEP-IAM account and complete registration in RCR.

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To submit images, the site user must be on the site's affiliate rosters and be assigned the 'TRIAD site user' role on the CTSU roster. Users should contact the site's CTSU Administrator or Data Administrator to request assignment of the TRIAD site user role. RAs are able to submit standard of care imaging through the same method.

4.10.2 TRIAD Installations

When a user applies for a CTEP-IAM account with the proper user role, he/she will need to have the TRIAD application installed on his/her workstation to be able to submit images. TRIAD installation documentation can be found by following this link https://triadinstall.acr.org/triadclient/

This process can be done in parallel to obtaining your CTEP-IAM account username and password.

If you have any questions regarding this information, please send an e-mail to the TRIAD Support mailbox at TRIAD-Support@acr.org.

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4.11 Instructions for Patients who do not Complete FLT PET/CT per Protocol

The patients, who **NEITHER** complete the optional FLT PET/CT scan **NOR** the required FLT PET/CT scan, will be considered off-study. No further data beyond the post-treatment FLT PET/CT scan time point need to be submitted through Medidata Rave, including data related to the remission bone marrow biopsy (Day 28-35). However, all data related to enrollment, the initial screening visit, and treatment should still be submitted.

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5. Study Design

The overall goal of this study is to evaluate the Negative Predictive Value (NPV) of post-treatment FLT uptake parameters for complete remission (CR) in patients with AML. Patients with AML who are candidates for induction chemotherapy will be enrolled in this prospective imaging trial. Participants will receive an anthracycline (IV days 1-3 x 1-2 cycles) plus infusional cytarabine (100-200 mg/m^2, IV days 1-7 x 1-2 cycles)-based regimen. A total of 83 eligible patients will be accrued in the study. All participants will receive one imaging study according to study protocol: **mandatory** post-treatment FLT PET/CT **within three days before or after the nadir bone marrow biopsy** (between Days 10-17 of treatment). The pre-treatment FLT PET/CT is **optional.** Eligible participants will be actively involved in the trial until remission bone marrow biopsy results are available. Patients will be followed for survival outcomes for at least 1 year (1 year beyond the end of the study accrual period).

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5.1 Imaging Schedule

Optional Baseline Imaging: An optional FLT PET/CT imaging study per protocol should be considered prior to induction chemotherapy, only if it does not interfere with commencement of treatment. Note that institutions working with commercial vendors to receive their FLT should allow for **2 days** between ordering the agent and receiving it for same-day imaging.

Mandatory Post-Treatment Imaging: All participants will receive one imaging study according to study protocol: post-treatment FLT PET/CT within three days before or after the nadir bone marrow biopsy (between Days 10-17 of treatment).

5.2 FLT Administered Dose

The injectable activity of FLT for most studies will be ≤ 0.07 mCi/kg of Fluorine-18, not to exceed 5 mCi with a specific activity greater than 200 Ci/mmol at the time of injection. The amount of injected drug is ≤ 6.1 µg (≤ 25 nmol per dose) of FLT. FLT is administered to subjects by intravenous injection of ≤ 10 mL. The drug solution is stored at room temperature in a gray butyl septum sealed, sterile, pyrogen-free glass vial and has an expiration time of 8 hours. There is no evidence that nonradioactive and radioactive FLT molecules display different biochemical behavior.

5.3 Treatment Plan

Participants will receive an anthracycline (IV days 1-3 x 1-2 cycles) plus infusional cytarabine (100-200 mg/m^2, IV days 1-7 x 1-2 cycles)-based regimen. Patients will be followed for up to 1 year beyond the end of study accrual period. Patients enrolled on this imaging trial also may be enrolled in a cooperative group AML treatment study, with an understanding of data sharing between the groups to ensure the data requirements for the EAI141 project are available.

5.4 Adverse Event Reporting Requirements

5.4.1 Purpose

Adverse event (AE) data collection and reporting, which are required as part of every clinical trial, are done to ensure the safety of the patients enrolled, as well as those who will enroll in future studies using similar agents.

- **Routine reporting:** Adverse events are reported in a routine manner at scheduled times during a trial using Medidata Rave.
- **Expedited reporting:** In addition to routine reporting, certain adverse events must be reported in an expedited manner for timelier monitoring of patient safety and care. The following sections provide information and instructions regarding expedited adverse event reporting.

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5.4.2 Terminology

- **Adverse Event (AE):** Any untoward medical occurrence associated with the use of a drug/ IND imaging agent in humans, whether or not considered related to the IND itself. Therefore, an AE can be **ANY** unfavorable or unintended sign (including an abnormal laboratory finding), symptom, or disease that worsens or is emergent after and *temporally associated* with the use of an IND. Note: events which begin before the reporting period are considered Medical History/Baseline events and events occurring after the reporting period are generally unknown.
- **Serious Adverse Event (SAE):** SAEs are generally Grade 3, 4 or 5 events as outlined in FDA guidance (21 CFR Part 312.32).
- **Attribution:** An assessment of the relationship between the adverse event and the protocol treatment, using the following categories.

ATTRIBUTION	DESCRIPTION
Unrelated	The AE is *clearly NOT related* to treatment.
Unlikely	The AE is *doubtfully related* to treatment.
Possible	The AE *may be related* to treatment.
Probable	The AE is *likely related* to treatment.
Definite	The AE is *clearly related* to treatment.

- **CAEPR (Comprehensive Adverse Events and Potential Risks List):** An NCI generated list of reported and/or potential AEs associated with an agent currently under an NCI IND. Information contained in the CAEPR is compiled from the Investigator's Brochure, the Package Insert, as well as company safety reports.
- **CTCAE:** The NCI Common Terminology Criteria for Adverse Events provides a descriptive terminology that is to be utilized for AE reporting. A grade (severity) is provided for each AE term.
- **Hospitalization (or prolongation of hospitalization):** For AE reporting purposes, a hospitalization is defined as an inpatient hospital stay equal to or greater than 24 hours.
- **Life Threatening Adverse Event:** Any AE that places the subject at immediate risk of death from the AE as it occurred.
- **Investigational New Drug (IND):** Drug/agent whose use is being investigated.
- **Monitoring/Reporting period:** The period for which AEs will be actively monitored and reported. This period is defined on a per

agent basis. For some studies this may be the entire time the patient is on study. For imaging IND agents this is generally from at least 24 hours to up to 30 days after administration depending on the half-life of the agent. Patients always are to be evaluated/contacted at the end of the reporting period to determine if any AEs occurred during the reporting period.

- **Routine reporting:** All adverse events are reported in a routine manner within the monitoring/reporting period during a trial using the approved routine reporting system. Sites must report adverse events into the approved electronic data capture system (Eg., Medidata RAVE). This data will be used by the lead organization to update the patient record in the CDUS.
- **Expedited reporting:** In addition to routine reporting, certain serious adverse events must be reported in an expedited manner directly to NCI (e.g., via CTEP-AERs) for timelier monitoring of patient safety and care.
- **Reportable AEs:** AEs are reportable if they meet the explicit reporting criteria for the protocol. For imaging agent trials, all events within the monitoring period are reportable. In addition, the death of any patient while on-study is reportable regardless of temporal relationship to use of an IND imaging agent.
- **CTEP-AERS (also CAERS):** CTEP Adverse Reporting System – used by sites for expedited reporting of adverse events to the NCI (formerly known as AdEERs).
- **Medidata RAVE (RAVE):** the current electronic data capture (EDC) system for cooperative group studies.
- **Clinical Data Update System (CDUS):** the primary resource of clinical trial data for the Division of Cancer Treatment and Diagnosis (DCTD) and the Division of Cancer Prevention (DCP). The Lead Group or the Lead Institution for the protocol is responsible for compiling and submitting the CDUS data for all participants (data for this report is entered by sites into the group data capture system [RAVE]).
- **Grades:** Grade refers to the severity of the AE. The CTCAE displays Grades 1 through 5 with unique clinical descriptions of severity for each AE based on this general guideline:
- **Grade 1:** Mild; asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated.
- **Grade 2:** Moderate; minimal, local or noninvasive intervention indicated; limiting age-appropriate instrumental ADL.
- **Grade 3:** Severe or medically significant but not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care ADL.
- **Grade 4:** Life-threatening consequences; urgent intervention indicated.
- **Grade 5:** Death related to AE.

- **Activities of Daily Living (ADL)**: Instrumental ADL refer to preparing meals, shopping for groceries or clothes, using the telephone, managing money, etc. Self-care ADL refer to bathing, dressing and undressing, feeding self, using the toilet, taking medications, and not bedridden.

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5.4.3 Reporting Procedure

This study requires that expedited adverse event reporting use the NCI CTEP Adverse Event Reporting System (CTEP-AERS). The CTEP's guidelines for CTEP-AERS can be found at http://ctep.cancer.gov. A CTEP-AERS report must be submitted electronically to ECOG-ACRIN and the appropriate regulatory agencies via the CTEP-AERS Web-based application located at https://eapps-ctep.nci.nih.gov/ctepaers.

In the rare event when Internet connectivity is disrupted a 24-hour notification is to be made by telephone to

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- the AE Team at ECOG-ACRIN (857-504-2900)
- the NCI/CIP (240-276-6510)

An electronic report MUST be submitted immediately upon re-establishment of internet connection.

Supporting and follow up data: Any supporting or follow up documentation must be uploaded to the Supplemental Data Folder in Medidata Rave within 48-72 hours. In addition, supporting or follow up documentation must be faxed to the NCI/CIP (240-276-7890) in the same timeframe.

NCI Technical Help Desk: For any technical questions or system problems regarding the use of the CTEP-AERS application, please contact the NCI Technical Help Desk at ncictephelp@ctep.nci.nih.gov or by phone at 1-888-283-7457.

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

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5.4.4 **Secondary Malignancy:** A secondary malignancy is a cancer caused by treatment for a previous malignancy (e.g., treatment with investigational agent/intervention, radiation or chemotherapy). A secondary malignancy is not considered a metastasis of the initial neoplasm.

CTEP requires all secondary malignancies that occur following treatment with an agent under an NCI IND/IDE be reported via CTEP-AERS. Three options are available to describe the event:

- Leukemia secondary to oncology chemotherapy (e.g., acute myelocytic leukemia [AML])

- Myelodysplastic syndrome (MDS)
- Treatment-related secondary malignancy

Any malignancy possibly related to cancer treatment (including AML/MDS) should also be reported via the routine reporting mechanisms outlined in each protocol.

Second Malignancy: A second malignancy is one unrelated to the treatment of a prior malignancy (and is **NOT** a metastasis from the initial malignancy). Second malignancies require **ONLY** routine reporting via CDUS unless otherwise specified.

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5.4.5 Mechanisms for Adverse Event Reporting

Within the monitoring period:

- **Routine Reporting (RAVE)**
  - **All adverse events** (Grades 1-5) both serious and non-serious *worsening or emergent after* IND administration/intervention, within the monitoring period, regardless of hospitalization or attribution, must be reported using the routine reporting system.
- **Expedited Reporting (CTEP-AERS)**
  - **All serious adverse events** (Grades 1 and 2) *worsening or emergent after* IND administration/intervention, within the monitoring period, and *resulting in hospitalization or prolongation of hospitalization for 24 hours or more*, regardless of attribution, must be entered by the site using expedited reporting.
  - **All serious adverse events** (Grades 3, 4 and 5) *worsening or emergent after* IND administration/intervention, within the monitoring period, *regardless of hospitalization or attribution*, must be entered by the site using expedited reporting.

After the monitoring period:

- **Routine Reporting (RAVE)**
  - All adverse events, regardless of grade and attribution, that are reported through the expedited reporting system also must be reported using the routine reporting system.
  - **All Deaths** (Grade 5) While a patient is on-study, without exception, deaths must be entered via routine reporting.
- **Expedited Reporting (CTEP-AERS)**
  - All serious grade 2 events, resulting in hospitalization or prolongation of hospitalization for 24 hours or more, and having *an attribution of possible, probable, or definite*, must be entered by the site using expedited reporting.
  - All serious adverse events (Grades 3-5) having *an attribution of possible, probable, or definite*, must be entered by the site using expedited reporting.

- All Deaths (Grade 5) on-study, regardless of attribution, must be entered by the site using expedited reporting if it is within 30 days of IND agent administration/intervention. After the 30 day period and through the one year follow-up, only routine reporting is required.

AE monitoring period for F-18 (^{18}F-FLT) PET IND:

The AE monitoring period for ^{18}F-FLT, an ^{18}F based radioisotope imaging agent with a half-life (t ½ of 110 minutes and a 10 half-life equivalent of 18.33 hours) is 24 hours after administration of the IND.

During the 24hours period (the "Monitoring period") after administration (including the infusion of the IND), the patient will be evaluated for Adverse events (AEs) at each imaging session, during any follow up (in person, by phone or by email), and at the end of the monitoring period.

AEs for IND Imaging Agents are defined as any signs of illness or symptoms that have appeared or worsened since the infusion of the IND Imaging Agent. Participants will be queried for potential AEs at multiple time points. These may include:

- At the time of injection;
- Before leaving the PET suite;
- If they call the site as instructed for any concerns at any time during the monitoring period;
- At the end of the monitoring period

The AEs that will be specifically monitored during and after the administration include, but are not limited to:

- localized discomfort at the intravenous (IV) injection site;
- pain;
- respiratory difficulties;
- blood pressure instability;
- flushing;
- dizziness;
- pruritus/rash/hives;
- any other symptoms that could be related to an allergic or anaphylactoid-type reaction;
- Any previously unreported AE which occurred during the monitoring period should be reported using the defined reporting procedures at the time the originating site or PI discovers the unreported AE;

When any AE is reported, concomitant medication taken by the participant in the 2 weeks prior to the event and/or during the time of the AE need to be collected and documented in the patient record and within the approved data collection system. This is currently CAERS for events requiring expedited reporting and Medidata RAVE for

routine reporting so that the patient's record may be updated in the CDUS.

Phase 1 and Phase 2 Studies
Expedited Reporting Requirements for Adverse Events that Occur on Studies under an IND/IDE within 24 Hours of the Last Administration of 18-F [1,2]

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 that 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).

Within the 24 hours Reporting Period

ALL SERIOUS adverse events occurring within the reporting period that meet the above criteria MUST be immediately reported to the NCI via CTEP-AERS within the timeframes detailed in the table below, regardless of attribution.

Hospitalization	Grade 1 and Grade 2 Timeframes	Grade 3-5 Timeframes
Resulting in Hospitalization ≥ 24 hrs	10 Calendar Days	24-Hours: Initial report; 5 Calendar Days: Compete report
Not resulting in Hospitalization ≥ 24 hrs	Not required	

NOTE: Protocol specific exceptions to expedited reporting of serious adverse events are found in the Specific Protocol Exceptions to Expedited Reporting (SPEER) portion of the CAEPR

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 after the reporting period of the investigational agent/intervention *and have an attribution of possible, probable, or definite* require reporting as follows:

Expedited 24-hour notification followed by complete report within 5 calendar days for:

- All Grade 3, 4, and Grade 5 AEs

Expedited 10 calendar day reports for:

- Grade 2 AEs resulting in hospitalization or prolongation of hospitalization

[2] For studies using Isotope based agents, the AE reporting period is limited to 10 radioactive half-lives, rounded UP to the nearest whole day, up to a maximum of 30 days, after the agent/intervention was last administered. MR, CT, US, and Optical Imaging contrast agents will have a 30-day post-IND administration monitoring period. Footnote "1" above applies after this reporting period.

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5.4.6 Additional instructions, requirements and exceptions for FLT IND

Please refer to FLT Adverse Event Reporting Guidance Document in Appendix II.

Additional Instructions

For instructions on how to specifically report events that result in persistent or significant disability/incapacity, congenital anomaly, or birth defect events via CTEP-AERS, please contact the CIP Help Desk at CIPSAEReporting@tech-res.com or 240-276-6510. This will need to be discussed on a case-by-case basis.

FLT specific expedited reporting requirements:

Pregnancies: Pregnancies and suspected pregnancies (including a positive or inconclusive pregnancy test, regardless of age or disease state) occurring while the subject is on FLT, or within 28 days of the subject's last dose of FLT, are considered immediately reportable events. The pregnancy, suspected pregnancy, or positive/ inconclusive pregnancy test must be reported via CTEP-AERS within 24 hours of the Investigator's knowledge.

FLT specific expedited reporting exceptions:

For this protocol, the adverse events listed below (Section 5.5) do not require expedited reporting via CTEP-AERS:

- If an AE meets the reporting requirements of the protocol, and it is listed on the SPEER, it should ONLY be reported via CTEP-AERS if the grade being reported exceeds the grade listed in the parentheses next to the event.

5.4.7 Other recipients of adverse event reports and supplemental data

DCTD/NCI will notify ECOG-ACRIN/pharmaceutical collaborator(s) of all AEs reported to the FDA. Any additional written AE information requested by ECOG-ACRIN MUST be submitted to BOTH the NCI and ECOG-ACRIN.

Adverse events determined to be reportable via CTEP-AERS must also be reported by the institution, according to the local policy and procedures, to the Institutional Review Board responsible for oversight of the patient.

5.5 Comprehensive Adverse Events and Potential Risks for 3'-deoxy-3'-[F-18]fluorothymidine (FLT) (NSC 743144)

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 Agent Specific Adverse Event List (ASAEL), appears in a separate column and is identified with ***bold*** and ***italicized*** text. This subset of AEs (ASAEL) contains events that are considered 'expected' for expedited reporting purposes only. Refer to the 'CTEP, NCI Guidelines: Adverse Event Reporting Requirements' for further clarification. The CAEPR does not provide frequency data; refer to the Investigator's Brochure for this information. Below is the CAEPR for 3'-deoxy-3'-[F-18]fluorothymidine.

Version 1.0, July 1, 2010[1]

Category (Body System)	Adverse Events[2] with Possible Relationship to 3'-deoxy-3'-[F-18]fluorothymidine (CTCAE v4.0 Term)	EXPECTED AEs FOR CTEP-AERS REPORTING Agent Specific Adverse Event List (ASAEL)
	No AEs reported in human studies[2,3].	

[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.

[2]No adverse events have been attributed to Positron-Emission Tomography (PET) imaging/diagnostic administration of [3'-deoxy-3'-[F-18]fluorothymidine at the levels described in the Investigators Brochure. Therefore, no adverse events are expected as a result of the intravenous (IV) administration of 3'-deoxy-3'-[F-18]fluorothymidine for typical PET imaging applications.

[3]As with many intravenously administered agents, 3'-deoxy-3'-[F-18]fluorothymidine could cause an allergic reaction that could potentially pose a threat to life (anaphylaxis). This has not been observed in limited human exposure to date. Reasonable precautions should be taken, consistent with normal radiologic and clinical facility practice. The patient should be monitored until the PET procedure is completed, and trained personnel and emergency equipment should be available per facility standards.

For purposes of informed consent regarding reasonably foreseeable risks to subjects in trials utilizing 3'-deoxy-3'-[F-18]fluorothymidine, the following potential adverse events are considered extremely rare:

- Injection-related risks that may include infection, or accidental extravasation of the dose that may lead to discomfort, localized pain, or infection.
- Risks related to allergic reaction/anaphylaxis that may be life threatening.

NOTE: As with all PET imaging agents, 3'-deoxy-3'-[F-18]fluorothymidine is a radiopharmaceutical that decays with positron emission. As such, it poses an intrinsic radiation exposure risk. However, when administered in accordance with the Investigator's Brochure as a PET imaging agent, this risk is felt to be extremely small. The organ and total body doses associated with FLT PET imaging are comparable to or lower than those associated with other widely used clinical nuclear medicine procedures.

NOTE: 3'-deoxy-3'-[F-18]fluorothymidine 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.

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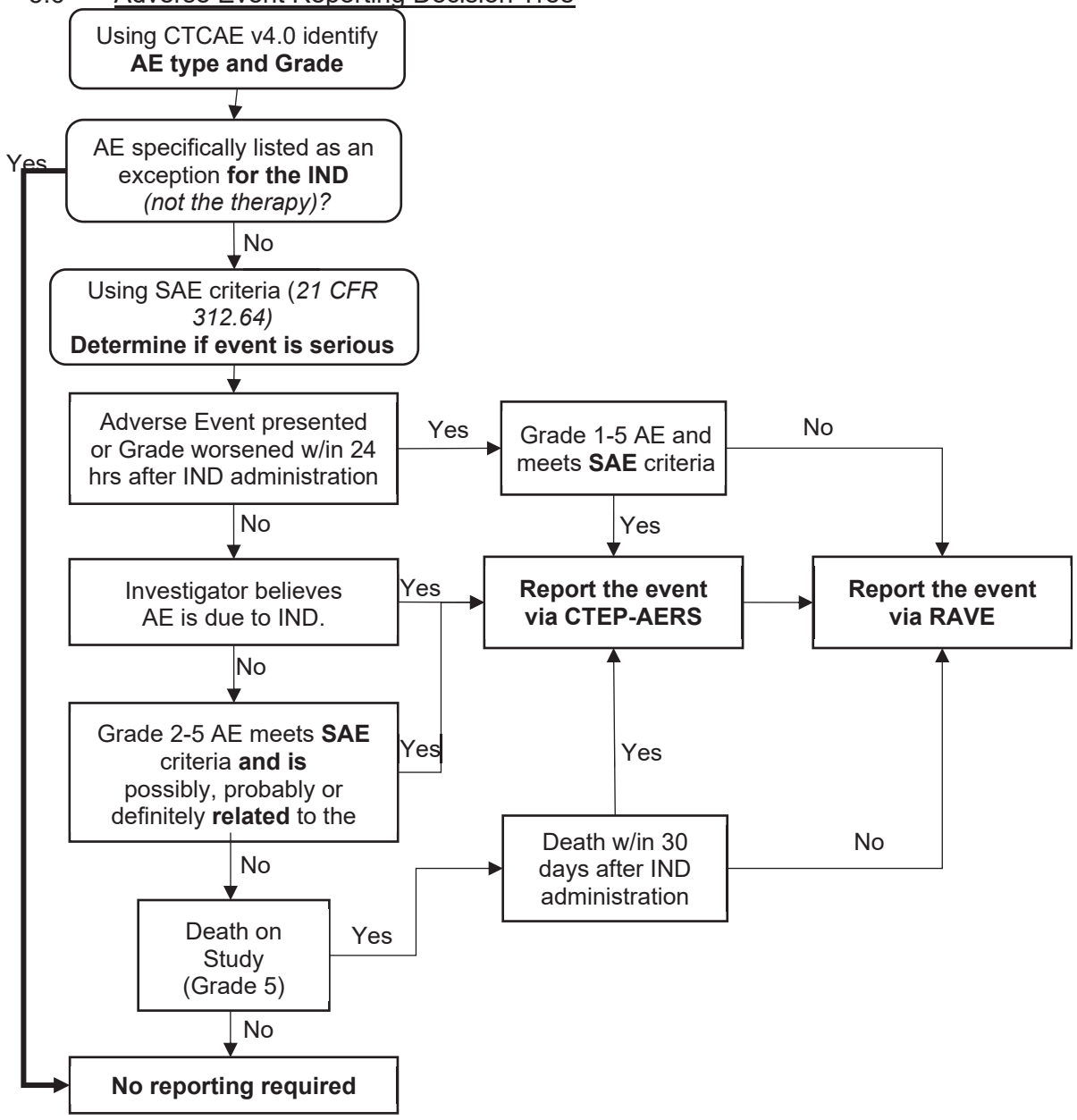
5.6 Adverse Event Reporting Decision Tree



5.7 Duration of Follow-up

Participants will stay on the trial for at least 1 year (1 year beyond the end of the study accrual period) until they complete all trial procedures, specifically the remission bone marrow biopsy (Day 28-35 biopsy), and follow-up collection of relapse free survival and overall survival.

6. Measurement of Effect ACUTE MYELOGENOUS LEUKEMIA (AML) RESPONSE CRITERIA

6.1 Complete Remission (CR)

Requires that all of the following be present for at least 4 weeks.

- Peripheral Blood Counts
  - Neutrophil count ≥ 1.0 x 10^9 /L.
  - Platelet count ≥ 100 x 10^9 /L.
  - Reduced hemoglobin concentration or hematocrit has no bearing on remission status.
  - Leukemic blasts must not be present in the peripheral blood.
- Bone Marrow Aspirate and Biopsy
  - Cellularity of bone marrow biopsy must be > 20% with maturation of all cell lines.
  - ≤ 5% blasts
  - Auer rods must not be detectable.
- Extramedullary leukemia, such as CNS or soft tissue involvement, must not be present.

6.2 Partial Remission (PR)

- Requires that all of the criteria for complete remission be satisfied except that the bone marrow may contain > 5% blasts but < 25% blasts.
- If all other criteria for CR are met, then a value of ≤ 5% blasts with Auer rods or abnormal morphology is considered a partial remission.

6.3 Relapse

Relapse following complete remission is defined as:

- Peripheral Blood Counts
  - Reappearance of blasts in the blood.
- Bone Marrow Aspirate and Biopsy
  - Presence of > 5% blasts, not attributable to another cause (e.g., bone marrow regeneration).
  - If there are no circulating blasts and the bone marrow contains 5% to 20% blasts, then a repeat bone marrow performed ≥ 1 week later documenting more than 5% blasts is necessary to meet the criteria for relapse.

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7. Study Parameters

7.1 Study Parameters

7.1.1 Visit 1: Screening Visit

- A screening assessment will occur to determine eligibility for the study.
- Patients will undergo pre-treatment bone marrow biopsy if one has not already been performed

  NOTE: Pre-treatment bone marrow aspirates are required to be submitted to the ECOG-ACRIN LTRL, see Sections 7.2 and 11 for additional information. If biopsy has already been performed, peripheral blood must be submitted instead at preregistration or after registration.
- A signed consent form will be obtained prior to study trial procedures. Medical history, demographics, height, weight, and physical exam will be obtained. *If patient data are available from clinical records in the appropriate time window, they need not be repeated for the pre-study evaluation.*

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7.1.2 Visit 1A: OPTIONAL FLT PET Imaging Study (FLT-base)

NOTE: Per Eligibility Criteria, any pre-menopausal women or women of childbearing potential MUST not be pregnant as general practice and per institution's standard of care prior to performing any scans.

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- Screening labs will be obtained within the one-week period prior to administration of FLT. Total bilirubin and creatinine should be collected per institutional standard of care. *If patient data are available from clinical records in the appropriate time window, they need not be repeated for the pre-study evaluation.*
- The optional baseline FLT PET (FLT-base) scan will take place within 1 week prior to chemotherapy.
- Specific image acquisition and reconstruction protocols will be provided in a Site Imaging Manual to participating institutions.

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- An adverse event evaluation will be performed at 24 hours. Sites will be required to submit the 'PET Adverse Event Assessment' after the 24 hour monitoring period, even if there are no adverse events to report.

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7.1.3 Visit 2: MANDATORY FLT PET Imaging Study (FLT-post induction)

NOTE: Per Eligibility Criteria, any pre-menopausal women or women of childbearing potential MUST not be pregnant as general practice and per institution's standard of care prior to performing any scans.

Rev. Add3

- Screening labs will be obtained within the one-week period prior to administration of FLT. Total bilirubin and creatinine should be

collected per institutional standard of care. *If patient data are available from clinical records in the appropriate time window, they need not be repeated for the pre-study evaluation.*

- The post treatment FLT PET (FLT-post induction) scan will take place after completion of the induction therapy and within 3 days before or after nadir bone marrow (approximately Days 10-17 after the initiation of the first cycle of the chemotherapy and prior to re-induction if required).

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- An adverse event evaluation will be performed at 24 hours (+/- 4 hours). Sites will be required to submit the 'PET Adverse Event Assessment' after the 24 hour monitoring period, even if there are no adverse events to report.

7.1.4 Visit 3: Nadir Bone Marrow at Day 10 to 17

Following completion of induction chemotherapy, the subject will undergo standard of care nadir bone marrow evaluation (Day 14 bone marrow biopsy) 7-10 days after the completion of induction chemotherapy. Local pathology report data will be collected from this time point as part of follow up for the EAI141 study.

7.1.5 Visit 4: Remission Bone Marrow at Day 28 to 35

Participants will undergo remission bone marrow biopsy at Day 28 to 35 after initial induction chemotherapy, if reinduction chemotherapy is not administered; at Day 28 to 35 after reinduction chemotherapy if reinduction chemotherapy was administered; local pathology report data will be collected from this time point as part of follow up for the EAI141 study.

NOTE: Remission specimens are REQUIRED to be submitted to ECOG-ACRIN LTRL, see sections 7.2 and 11 for additional information.

7.1.6 Sharing of Results

The final results of the study will be shared with the participating centers at the completion of the study. With agreement between the patient and the treating physician, and at the discretion of the participating center, the treating physician may share the experimental results of the FLT PET/CT imaging scans with the participant. The sharing of the informal results should be done with caution, as the FLT PET/CT is still experimental and the significance of these results is unknown. **The results from the FLT PET/CT scans should not be part of the patient's medical record and should not be used to direct or change therapy.** We emphasize that sharing of imaging results with patients remains optional and at the discretion of the participating center, the patient and the treating physician.

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Study Procedures Table

Study Procedures	VISIT 1: SCREENING	VISIT 1A: OPTIONAL Baseline FLT PET/CT (FLT-base)[2]	VISIT 2: Post-Treatment FLT PET/CT (FLT-post)[3]	VISIT 3: Nadir Bone Marrow (Day 10 to 17)[4]	VISIT 4: Remission Bone Marrow (Day 28 to 35)[5]
Informed Consent	X				
Demographics[1]	X				
Medical History[1]	X				
Height[1]	X				
Weight[1]	X	X	X		
Physical Exam[1]	X				
Pre-Scan Pregnancy and Serum Creatinine Evaluations[1]		X	X		
[F-18]FLT PET/CT[6]		X	X		
Bone Marrow Biopsy	X			X	X
Other Clinical Evaluations[7]	X			X	X
Adverse Event Evaluation		X	X		
Biological Sample Submissions	See Sections 7.2 and 11				
Follow-Up Data Collection	Standard Practice Clinical Data Will Be Collected for 1 Year from Enrollment of the Last Participated Accrued to the Study				

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1. If patient data are available from clinical records in the appropriate time window, they will not be repeated for the pre-study evaluation. Total bilirubin and creatinine should be collected per institutional standard of care before FLT PET/CT.
2. Baseline FLT PET/CT imaging (FLT-base) is **optional**, and must be done within 1 week prior to initiation of therapy protocol.
3. Post-treatment FLT PET/CT imaging (FLT-post) must be done after the completion of the induction chemotherapy and within 3 days before or after nadir bone marrow and prior to reinduction if required (Days 10-17 after the initiation of the first cycle of the chemotherapy and prior to reinduction if required)
4. Nadir bone marrow will be performed per standard clinical practice, typically within 7-10 days after the completion of induction therapy.
5. Remission bone marrow will be performed per standard clinical practice, typically at Day 28-35 after treatment began and upon count recovery.
6. **NOTE:** Per Eligibility Criteria, any pre-menopausal women or women of childbearing potential MUST not be pregnant as general practice and per institution's standard of care prior to performing any scans.
7. Other clinical evaluations include: Cytogenetics and molecular studies as mandated by the treatment study in which the patient is enrolled. If the patient is not enrolled in another study, the cytogenetic and molecular studies should be done according to the treating institution's standard of care.

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7.2 Biological Sample Submissions

All specimens submitted must be entered and tracked via the on-line ECOG-ACRIN Sample Tracking System (STS)

	Pre-Treatment [1]	Day 28 to 35 (Remission)	Submit To:
MANDATORY: Bone Marrow Aspirate (Heparin, First Pull)[2,3]	X	X	LTRL
MANDATORY (Only if Bone Marrow Aspirate not submitted): Peripheral Blood (heparin, green or purple [EDTA] top tubes, 30-40mL)[3]	X[4]		LTRL
From patients who answer "Yes" to "I agree to provide additional samples for research."			
Bone Marrow Smears (Wright-Giesma Stained)	X	X	LTRL

1. Signed EAI141 patient consents and HIPAA authorizations must be submitted to the ECOG-ACRIN Leukemia Translational Research Laboratory (LTRL) prior to or at the time of submission of the bone marrow to the LTRL.
2. The laboratory will accept any amount of bone marrow as long as it represents a first pull. Ideally, 2-3mL of aspirate from a separate aspiration site should be submitted.

   For the MRD assessments, aspirate from a separate bone marrow aspiration site must be submitted. ONLY SUBMIT ASPIRATES FROM THE FIRST PULL OF AN ASPIRATION SITE FOR MRD TESTING. DO NOT SUBMIT ASPIRATES FROM THE SECOND OR THIRD PULL OF THE SAME ASPIRATION SITE.

   For patients with an insapirable bone marrow ("dry tap"), or if a bone marrow has been done previously and that patients refuses another aspiration done, call Dr. Paietta's laboratory at (718) 920-4100 to discuss the case and the possibility for submitting peripheral blood only. Be prepared to report the WBC count and the blast count in the peripheral blood at the time of the call.
3. Leftover samples will be stored for future undefined research studies from patients who answer "YES" to "My samples and related information may be kept in a Biobank for use in future health research."
4. Peripheral blood can be submitted after registration to Step 1 to coincide with clinical assessments performed prior to protocol specified imaging. Peripheral blood must be submitted only if bone marrow aspirate will not be submitted at preregistration.

8. Agent Formulation and Procurement

NCI Supplied Agent(s) – General Information

8.1 Supplier of the FLT Agent

8.1.1 Drug Ordering

FLT will be supplied by a commercial vendor of radioisotopes in most cases. The vendor must be authorized within the NCI IND, and the site had been qualified for production by NCI and ECOG-ACRIN. FLT can only be synthesized on site if the chemistry manufacturing and control procedures are filed within the NCI IND and the site has been approved for FLT production under the NCI IND and notified of approval by the NCI Cancer Imaging Program. All other sites will be required to use an approved commercial supplier.

The investigational pharmacist or qualified nuclear medicine technologist at the participating institution will be the responsible party for receiving the FLT, as designated by the site principle investigator.

8.1.2 Drug Returns

If for any reason the study imaging is unable to be completed, sites will allow the radioactivity of the FLT solution to decay and then discard it appropriately per site's policies and procedures. A copy of the policy should be available upon request.

8.1.3 Drug Accountability

The investigator or the investigator-designee must maintain a detailed record of receipt, disposition, and destruction dates of FLT solution, using the Drug Accountability Record form available through CTSU.

In accordance with regulations, the radioisotope vendor conducts several quality control tests on the FLT product prior to release for human administration. Once delivered to the participating institution, doses will be stored in the appropriate storage area in the nuclear medicine facility until they are administered to the patient.

8.2 ^{18}F-FLT

For complete information, please refer to the Investigator's Brochure:

"3'-deoxy-3'-[F-18] fluorothymidine: [F-18]FLT, An Investigational Positron Emission Tomography (PET) Radiopharmaceutical for Injection and intended for use as an *in vivo* diagnostic for imaging active cellular proliferation of malignant tumors", Edition Number 7, Edition date 2009.

8.2.1 Other Names

3'-deoxy-3'-[F-18] fluorothymidine, [F-18]FLT, FLT

8.2.2 Classification

Investigational new drug: Radiopharmaceutical/radiotracer

8.2.3 Mode of Action

The pharmacology of FLT as a therapeutic agent is based on its action as an inhibitor of DNA synthesis. As a diagnostic imaging agent, phosphorylation by thymidine kinase leads to tracer retention in proliferating tissue.

8.2.4 Storage and Stability

In accordance with regulations, the radioisotope vendor conducts several quality control tests on the FLT product prior to release for human administration. Once delivered to the participating institution, doses will be stored in the appropriate storage area in the nuclear medicine facility until they are administered to the patient.

The drug solution is stored at room temperature in a gray butyl septum sealed, sterile, pyrogen-free glass vial and has an expiration time of 8 hours.

8.2.5 Dose Specifics

The administered activity will be 0.07 mCi/kg with a maximum of 5 mCi. The amount of injected drug is ≤ 6.1 µg (≤ 25 nmol per dose) of FLT.

8.2.6 Preparation

The injectable activity of FLT for most studies will be ≤ 0.07 mCi/kg of fluorine-18, not to exceed 5 mCi with a specific activity greater than 200 Ci/mmol at the time of injection. In the dose of FLT, only a small fraction of the FLT molecules are radioactive.

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8.2.7 Route of Administration

FLT is administered to subjects by intravenous injection. There is no evidence that nonradioactive and radioactive FLT molecules display different biochemical behavior.

8.2.8 Incompatibilities

N/A

8.2.9 Availability

8.2.9.1 Drug Ordering

There are two ways to obtain the FLT agent for this study:

1) Commercial vendor supply
2) On-site manufacturing

1) Commercial Vendor Supply. FLT may be supplied by a commercial vendor of radioisotopes if authorized and approved by the NCI CIP. Please see informational document posted on the CTSU regarding FLT dose supply for a listing of the approved commercial vendors and process for set up. The investigator or the investigator-designee will order patient doses of FLT. The investigative

radiopharmaceutical FLT solution will be shipped to the site the same day the participant is to be injected.

2) On-Site Manufacturing. FLT can be manufactured on-site *only* after the site had been qualified and approved for production by NCI Cancer Imaging Program (CIP) and ECOG-ACRIN. For additional information about on-site manufacturing, see "*Regulatory approach to approve manufacturing sites to make IND PET agenda to be used under a Cancer Imaging Program (CIP IND for a specified trial)*".

The investigational pharmacist or qualified nuclear medicine technologist at the participating institution will be the responsible party designated by the investigator.

8.2.9.2 Drug Returns

If for any reason the study imaging is unable to be completed, sites will allow the radioactivity of the FLT solution to decay and then discard it appropriately per site's policies and procedures. A copy of the policy should be available upon request.

8.2.9.3 Drug Accountability

The investigator or the investigator-designee must maintain a detailed record of receipt, disposition, and destruction dates of FLT solution, using the Drug Accountability Record form.

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https://ctep.cancer.gov/forms/docs/agent_accountability.pdf

8.2.10 Side Effects

See Section 5.5 for side effects.

8.2.11 Nursing/Patient Implications

Standard safety precautions required when handling radioactive materials, predominantly during the injection and uptake period should be followed. FLT requirements are similar to those used for other PET tracers, such as FDG.

9. Imaging Protocol

9.1 FLT PET Imaging Studies

Specific image acquisition and reconstruction protocols will be made available through the CTSU. Instructions for images submission is provided in Section 4.10.

9.2 FLT Administration

FLT will be synthesized according to the standard operating procedures provided by the NCI. A summary of the synthesis procedure and associated quality control can be found in the investigator's brochure.

FLT will be administered in the PET imaging suite under physician supervision. The imaging technologist or nurse will administer the FLT by intravenous infusion over one minute, followed by a saline flush. A fully equipped emergency cart and ACLS certified personnel will be available. Reported adverse events and potential risks are described in Section 5.5. The infusion and imaging procedure will be terminated in any patient who exhibits anaphylaxis, chest pain, dyspnea, or grand mal seizure.

9.3 Timing of FLT PET Studies

Two imaging sessions, an *optional* baseline and *mandatory* post-treatment will be performed (see Study Procedures Table).

Optional Baseline: The optional baseline FLT PET/CT imaging must be completed within one week prior to beginning of induction chemotherapy.

Mandatory Post-Treatment: Post-treatment FLT PET/CT must be done after the completion of the induction chemotherapy and **within 3 days before or after nadir bone marrow** (approximately Days 10-17 after the initiation of the first cycle of the chemotherapy and prior to re-induction if required).

9.4 Imaging Quality Assurance (QA)/Quality Control (QC) Procedure

Participant must be scanned on PET/CT scanners that have been qualified by the ACR Imaging Core Laboratory per the protocol-specific instructions made available to participating sites.

Qualification Utility for the Imaging Core Laboratory (QUIC) is the web-based tool for managing the qualification process and communicating with the core lab staff. The QUIC User Guide will be made available to participating sites.

Refer to the instructions about uploading images and completing the required application forms.

A hybrid PET/CT scanner is mandatory. The ability to calculate standardized uptake values (SUVs) is also mandatory. All sequential imaging sessions should be performed on the same PET/CT scanner, whenever possible. Any deviations should be reported to ECOG-ACRIN. QA/QC procedures will include review of DICOM files against study protocols. The PET/CT scanner must be kept calibrated in accordance with the manufacturer's recommendations. The scanner should routinely be assessed for quantitative integrity and stability by being tested using various imaging protocols on a standard phantom. For SUV

measurements, this assessment should include a comparison against a dose calibrator to ensure accuracy; that is, a comparison of the absolute activity measured versus the measured activity injected, should be performed.

A daily QC check must be performed at the beginning of the day, including PET/CT scanner and dose calibrator, in accordance with the manufacturer recommendations. If any of the QC results are outside of the manufacturer's guidelines, the study must be rescheduled and the problem rectified before scanning any patients.

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9.5 Imaging Sessions

The participant will report to the PET suite and undergo FLT injection. Approximately 60 minutes (± 10 min) after injection, a static whole body image will be acquired. If both optional baseline and mandatory post-treatment FLT PET/CT scans are completed, the participant should be scanned on the same scanner for each session. The whole body scan (top of the head [vertex] to below the knees) will be obtained. Details of the imaging procedure are specified in the Site Imaging Manual.

9.6 Supportive Care Guidelines

Any adverse effects, related or non-related to the injection of FLT, will be treated as clinically indicated with no study-related restrictions.

9.7 Dosing Delays/Dose Modifications

9.7.1 FLT Dose

The dose of FLT is based on the radiation dosimetry estimates. Due to the potential of a poor radiosynthetic yield or unavoidable time delays, a lesser amount of radioactivity may be administered at the discretion of the site PI, based on whether clinically acceptable images can be acquired with the dose administered. Any such modifications of the agent infusion will be recorded.

9.8 FLT PET/CT Image Analysis: Central Read

Quantitative Total Marrow Imaging (QTMI) image analysis will be performed at a central analysis laboratory and will include extraction of the whole bone marrow proliferative activity as assessed by analysis of the full FLT PET/CT images as follows [5]:

- The CT images are thresholded to extract the bone;
- These CT bone volumes are expanded to include the marrow resulting in bone + marrow CT masks;
- The masks are applied to the PET images to isolate the PET voxels representing the bone and marrow;
- An SUV threshold of 0.5 is applied to the masked PET images to extract the marrow PET voxels, yielding PET images of the bone marrow.

After the extraction of the spatial bone marrow compartment from the FLT PET/CT imaging, the FLT uptake reflecting the rate of bone marrow proliferation across the bone marrow is analyzed. Preliminary data suggests that image analysis should ideally include analysis of the whole bone marrow to determine a

number of bone marrow proliferation parameters, such as the mean proliferation, maximum proliferation, as well as heterogeneity of proliferation. While each of the proliferation parameters appears predictive of the clinical outcome, each of these parameters likely has different performance characteristics, making them more or less suitable for clinical use.

The extraction analysis will include three imaging parameters (SUV_{mean}, SUV_{max}, SUV_{hetero}).

- Maximum FLT uptake (SUV_{max}), defined as the maximum FLT uptake across the total bone marrow compartment
- Mean FLT uptake (SUV_{mean}), defined as the mean FLT uptake across the total bone marrow compartment
- Heterogeneity of FLT uptake (SUV_{hetero}), defined as the coefficient of variation of FLT uptake across the total bone marrow compartment

While all of these parameters will be extracted and analyzed for each patient, only SUV_{max} will be used as the primary endpoint in the statistical analysis. All other parameters will be used and reported in descriptive manner (see **Statistical Considerations**).

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9.9 Other Comparators

9.9.1 Pathologic Response

This nadir or “day 14 bone marrow” is recommended in the 2012 National Comprehensive Cancer Network (NCCN) Guidelines for the treatment of AML. The ideal response by the NCCN is defined as a hypoplastic marrow with < 10-20% cellularity and < 5-10% residual blasts. This biopsy is a standard measure of induction chemotherapy effectiveness. The results of this analysis will be reported.

9.9.2 Clinical Response

Clinical response will be assessed as below. Criteria are based upon the standard AML response criteria as published by Cheson et al [4]. Remission bone marrow will be performed per standard clinical practice, typically at Day 28-35 after treatment began and upon count recovery. The results of this analysis will be reported.

Table 1. Clinical Response Criteria

Response	ANC (cells/uL)	Platelet Count (cells/uL)	Bone Marrow Blasts (%)
CR	> 1000	> 100,000	< 5%
CRi	> 1000	> 50,000	< 5%
PR	> 1000	> 100,000	> 5% and < 25% and 50% less than pre-therapy
LFS	Any	Any	< 5%
RD			> 5%, not attributable to marrow regeneration

Abbreviations: CR (complete remission), CRi (complete remission with incomplete platelet recovery), PR (partial remission), LFS (leukemia-free state), RD (resistant disease)

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9.10 Data Analysis

The primary goal of our endpoints will be to evaluate the negative predictive value (NPV) of post-treatment FLT PET/CT imaging for complete remission in patients receiving induction chemotherapy for AML. Additional comparisons with imaging, pathologic, clinical and outcome data will also be performed. These comparisons are summarized in Table 2.

Table 2. Planned Comparisons with FLT Parameters

FLT Parameters	Compared To
OPTIONAL Baseline FLT uptake parameters (FLT-base)	CR, RD Nadir bone marrow, relapse-free and overall survival
Post-treatment FLT uptake parameters (FLT-post)	CR, RD Nadir bone marrow, relapse-free and overall survival
Change in FLT uptake parameters (FLT-post vs FLT-base)	CR, RD Nadir bone marrow, relapse-free and overall survival

9.11 Image Analysis

A central review and analysis of all FLT PET/CT imaging will be performed at the ACR Imaging Core Laboratory in coordination with the University of Wisconsin Image Analysis Core (IMAC). The centralized imaging analysis will be used for endpoint calculations. Radiologists will perform local reads of the images to help guide central review. Central review will subsequently be completed; data will be shared at the culmination of the study.

10. Statistical Considerations

The overall objective of this trial is to evaluate the negative predictive value of post-treatment FLT PET/CT imaging for complete remission (CR) in comparison with blast counts from bone marrow biopsy (BMBX) at Day 14.

Definition of complete remission (CR): The CR designation requires that the patient achieve the morphologic leukemia-free state and have an absolute neutrophil count of more than 1,000/uL and platelets of greater than 100,000/uL. Hemoglobin concentration or hematocrit has no bearing on remission status, although the patient must be independent of transfusions. Occasionally, a rare peripheral blood blast may be identified during regeneration; however, if the patient is in CR, the bone marrow would have less than 5% blasts and no Auer rods [4].

Definitions of FLT Positivity and Negativity for CR:

- FLT PET "positive"→ $SUV_{max} \leq 7$
- FLT PET "negative" → $SUV_{max} > 7$

The post-treatment FLT PET/CT needs to be conducted **within three days** before or after the nadir bone marrow biopsy (between Days 10-17 after initiation of first induction cycle and prior to reinduction). After the imaging analysis to collect SUV_{max}, we define a positive testing result as a low uptake value, which corresponds to the lower blast count in the BMBX.

Our primary aim is to evaluate the negative predictive value (NPV) of post-treatment FLT PET/CT imaging for achieving CR in patients receiving induction chemotherapy for AML. The results of the pilot study [5] show that there was a clear separation between responders and non-responders in terms of SUV measurements from FLT PET/CT imaging. For example, the means (standard deviation [SD]) of SUV_{max} for responders and non-responders are 3.6 (0.2) and 11.4 (0.8), respectively. Using the Newton-type algorithm, we found the best cutting point is around $SUV_{max} = 7$. This optimization was done using the *nlm* function in R package *stats*. Thus, our FLT PET data will be dichotomized at $SUV_{max} = 7$ whereas positive FLT PET testing results will be the ones with $SUV_{max} \leq 7$ and the negative FLT PET testing results will be the ones with $SUV_{max} > 7$.

Our sample size calculation is based on hypothesis testing to see if the NPV of post-treatment FLT PET/CT is significantly better than that of the Day 14 bone marrow biopsy. The CR rate is assumed to be around 50% in the study population. In addition, we assume that the FLT PET-positivity rate in the study population is around 50% as well. Under these assumptions, we will need 21 FLT-negative patients to have the power of 80% to test if the NPV of FLT is higher than 0.64 (null hypothesis) given that the true NPV of FLT is 85% or above (alternative hypothesis). The test is one-sided with the target 0.10 significance level. The Exact Binomial test in PASS 2012 was used for the calculations [6]. Since the positivity rate is assumed to be around 50% in the study population, we need to enroll 42 patients. Due to the random nature of the observed numbers of patients meeting the clinical criteria in a prospective study, a sample size correction technique attributed to Pepe (2003) was applied. Our calculation shows that after adding 9 extra patients, we can be 90% sure to yield the required numbers of patients meeting the clinical criteria, which brings the sample size to 51. After an additional 10% inflation to account for potentially incomplete data, we arrive at an overall

sample size of 57 participants. The sample size is 86 evaluable if the true NPV of FLT PET/CT is 80% or above. The sample size is 38 if the true NPV is 90% or above.

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According to our interim monitoring of the early enrollment, the attrition rate is around 38%. The main reason was attributed to the failed remission bone marrow biopsy due to poor health or death. This accounted for ~1/2 of attrition. In addition, there were ~1/3 of attrition due to the failure of completing the post-treatment FLT PET/CT scans (e.g., equipment failure, patients withdrew). We will closely monitor these issues and the enrollment may stop early if we observe a lower attrition rate. Thus, **the overall sample size is up to 83 participants**. The below table shows the required number of participants at various attrition rates.

Attrition rate	Sample size
10%	57
25%	68
38%	**83**

Futility Analysis

A futility analysis will be performed when half of the analyzable participants have completed the trial. The method of conditional power [7] will be used for the futility analysis, which will estimate the probability that NPV with all subjects would be greater than 0.64, conditional on the results accumulated at the time of the futility analysis. If this probability is too small (i.e., < 0.20), implying that it is unlikely that the FLT PET/CT is useful, then a decision may be made to terminate the study early.

Objectives

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10.1 Primary Objectives

10.1.1 To evaluate the negative predictive value (NPV) of post-treatment FLT PET/CT imaging for complete remission (CR) in patients receiving induction chemotherapy for acute myeloid leukemia (AML).

Three imaging parameters (SUV_{mean}, SUV_{max}, SUV_{hetero}) will be measured from an FLT PET/CT scan, and SUV_{max} will be the primary endpoint. The binomial proportion of NPV and the corresponding Exact confidence intervals will be calculated. In addition, the calculated NPV will be tested against the null hypothesis to see if it's significantly larger than 0.64 (NPV of Day-14 BMBX in CR prediction).

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10.2 Secondary Objectives

10.2.1 *To evaluate the positive predictive value (PPV) of post-treatment FLT PET/CT imaging for complete remission;*

The binomial proportion of PPV and the corresponding Exact confidence intervals will be calculated. In addition, the calculated PPV will be tested to see if it's significantly larger than 0.79 (PPV of Day-14 BMBX in CR prediction).

10.2.2 *To estimate the sensitivity and specificity of post-treatment FLT PET/CT for detecting complete remission;*

The binomial proportions and the corresponding Exact confidence intervals will be calculated for sensitivity and specificity estimation.

10.2.3 To correlate FLT PET/CT imaging parameters with biologic correlates (MRD assessment).

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Correlation and regression analysis will be used to assess this aim.

10.2.4 To correlate FLT PET/CT imaging parameters with relapse-free survival and overall survival.

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Kaplan-Meier method and Cox proportional hazard regression will be used to assess this aim

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10.3 Exploratory Objectives

10.3.1 *To evaluate pre-treatment FLT PET/CT activity as a predictor of complete remission;*

No previous studies were available to determine the cutoff for the pre-treatment SUV_{max}. The ROC approach will be used to test the continuous measurement of this variable in predicting CR.

10.3.2 *To evaluate the change between pre-treatment and post-treatment FLT PET/CT activity as a predictor of complete remission;*

No previous studies were available to determine the cutoff for the change from pre-treatment to post-treatment SUV_{max}. The ROC approach will be used to test the continuous measurement of this variable in predicting CR.

Safety Monitoring

Interim analyses of toxicity are performed twice yearly for all ECOG-ACRIN studies. Reports of these analyses are sent to the ECOG-ACRIN Principal Investigator or Senior Investigator at the participating institutions. Expedited reporting of certain adverse events is required, as described in Section **Error! Reference source not found.**.

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10.4 Gender and Ethnicity

Based on previous data from E1900 the anticipated accrual in subgroups defined by gender and race is:

Racial Categories	Ethnic Categories				Total
	Hispanic or Latino		Not Hispanic or Latino		
	Females	Males	Females	Males	
American Indian or Alaskan Native	0	0	0	0	0
Asian	0	0	2	0	2
Native Hawaiian or other Pacific Islander	0	0	0	0	0
Black or African American	0	0	3	2	5
White	4	3	35	34	76
Total	4	3	40	36	83

The accrual targets in individual cells are not large enough for definitive subgroup analyses. Therefore, overall accrual to the study will not be extended to meet individual subgroup accrual targets.

Study Monitoring

This study will be monitored by the ECOG-ACRIN Data Safety Monitoring Committee (DSMC). The DSMC meets twice each year. For each meeting, all monitored studies are reviewed for safety and progress toward completion. When appropriate, the DSMC will also review interim analyses of outcome data. Copies of the toxicity reports prepared for the DSMC meetings are included in the study reports prepared for the ECOG-ACRIN group meeting (except that for double blind studies, the DSMC may review unblinded toxicity data, while only pooled or blinded data will be made public). These group meeting reports are made available to the local investigators, who may provide them to their IRBs. Only the study statistician and the DSMC members will have access to interim analyses of outcome data. Prior to completion of this study, any use of outcome data will require approval of the DSMC. Any DSMC recommendations for changes to this study will be circulated to the local investigators in the form of addenda to this protocol document. A complete copy of the ECOG-ACRIN DSMC Policy can be obtained from the ECOG-ACRIN Coordinating Center.

Evaluation of Toxicity

All subjects will be evaluable for toxicity from the time of injection of FLT until 24 hours after injection.

Evaluation of Preliminary Efficacy

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A futility analysis will be performed when half of the participants (i.e., 34) have completed the trial. The method of conditional power [7] will be used for the futility analysis, which will estimate the probability that NPV with all subjects would be greater than 0.64, conditional on the results accumulated at the time of the futility analysis. If this probability is too small (i.e., < 0.20), implying that it is

unlikely that the FLT PET/CT is useful, then a decision may be made to terminate the study early.

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11. Research Sample Submissions

Bone marrow aspirates and/or peripheral blood must be submitted for defined laboratory research studies being performed at the ECOG-ACRIN LTRL. These laboratory research studies are defined in Section 12.

Bone marrow smears are to be submitted for future undefined research per patient consent.

It is **required** that all samples submitted on this trial be entered and tracked using the ECOG-ACRIN Sample Tracking System (STS) (see Section 11.3). An STS shipping manifest form is to be included with every submission.

All samples must be labeled clearly with the ECOG-ACRIN protocol number (EAI141), ECOG-ACRIN patient sequence number, patient's initials, date of collection and sample type.

11.1 Sample Collection and Submission Schedule

Samples are to be submitted as follows:

11.1.1 Bone marrow and/or peripheral blood are to be submitted to the ECOG-ACRIN LTRL as outlined in Section 11.2. Samples are to be collected at the following time points:

Bone marrow aspirates (or peripheral blood) and bone marrow smears:

- Pre-Treatment

Bone marrow aspirates and bone marrow smears:

- Day 28-35 (Remission)

11.2 Submissions to ECOG-ACRIN Leukemia Translational Research Laboratory (LTRL)

Dr. Paietta's institutional regulations require that she receive a copy of the patient's consent and a copy of the HIPAA authorization at the time of or prior to the submission of the bone marrow samples.

11.2.1 Sample Preparation Guidelines

NOTE: FOR MRD ASSESSMENTS, AN ASPIRATE FROM A SEPARATE BONE MARROW ASPIRATION SITE MUST BE SUBMITTED (THE NEEDLE CAN BE RE-DIRECTED THROUGH THE SAME SKIN PUNCTURE SITE). ONLY SUBMIT ASPIRATES FROM THE FIRST PULL OF AN ASPIRATION SITE FOR MRD TESTING. DO NOT SUBMIT SAMPLES FROM THE SECOND OR THIRD PULL OF THE SAME ASPIRATION SITE.

NOTE: Results of MRD assessments will not be reported to the site.

The following are to be submitted:

1. MANDATORY: Heparinized bone marrow aspirate (the laboratory will accept any amount as long as it represents a first pull). Ideally,

2-3mL of aspirate from a separate aspiration site should be submitted.

For patients with an inaspirable bone marrow ("dry tap"), or if a bone marrow has been done previously and the patient refuses to have another aspiration done, call Dr. Paietta's laboratory at (718) 920-4100 to discuss the case and the possibility for submitting peripheral blood only. Be prepared to report the WBC count and the blast count in the peripheral blood at the time of the call.

ALTERNATIVE TO BONE MARROW ASPIRATE SUBMISSION AT BASELINE: Heparin or EDTA peripheral blood (four [4] green or purple top tubes, 30-40mL).

NOTE: Peripheral blood must be submitted only if bone marrow aspirates will not be submitted at preregistration and can be submitted after registration to Step 1 to coincide with clinical assessments.

2. At least two (2) Wright-Giemsa stained bone marrow smears.

   Submit from patients who answer "Yes" to "I agree to provide additional samples for research."

3. MANDATORY: A copy of the institutional pathology report on the bone marrow must be submitted. The pathology report must include cytogenetic results and any results from fluorescence-in-situ (FISH) hybridization and/or molecular studies done at the submitting institution.

   Please fax to the LTRL at (718) 920-1161

11.2.2 Shipping Procedures

The LTRL must be notified by telephone the day of shipment.

Fax or Email to Dr. Paietta is not acceptable.

Telephone: (718) 920-4100

During off hours, all information regarding the shipment should be left on the answering machine in the LTSL including:

EAI141 patient sequence number

Patient Initials

Type of specimens shipped

Name, telephone number, and institution

For questions regarding the shipment, Dr. Paietta and her staff can be reached at the phone numbers provided on the recorded message, please always try Dr. Paietta first. Questions can also be addressed to Dr. Paietta via e-mail (epaietta@earthlink.net), however, please do not use e-mail for the notification of shipments.

Bone marrow smears should be submitted within one week of collection.

Heparinized bone marrow aspirate and peripheral blood or EDTA samples must be sent fresh (on the day of collection) on cool packs

(do not freeze and do not use ice cubes) by overnight courier (preferably FedEx) to arrive within 24 hours to:

Elisabeth Paietta, Ph.D.
Department of Oncology
Hofheimer 3rd Floor
Leukemia Oncology Laboratory
111 East 210th Street
Bronx, NY 10467
Tel: (718) 920-4100
FAX: (718) 920-1161

The LTRL is open to receive shipments Monday through Saturday. Shipments on Fridays for Saturday delivery must have "Saturday Delivery" marked on the overnight courier slip.

In general, always ship specimens the day before a holiday for delivery the day after the holiday.

An STS shipping manifest form must be generated and shipped with all sample submissions.

Please enter all information into the STS, including time and date of specimen collection and peripheral blood WBC count and blast count.

If samples need to be drawn late at night, on Sunday, or on a holiday when FedEx does not operate, keep the samples in a refrigerator between 10 and 15 degrees Celsius until the next day when it can be shipped.

11.3 ECOG-ACRIN Sample Tracking System

It is **required** that all specimens submitted on this trial be entered and tracked using the ECOG-ACRIN Sample Tracking System (STS). The software will allow the use of either 1) an ECOG-ACRIN user-name and password previously assigned (for those already using STS), or 2) a CTSU username and password.

When you are ready to log the collection and/or shipment of the specimens required for this study, please access the Sample Tracking System software by clicking https://webapps.ecog.org/Tst.

Important: Please note that the STS software creates pop-up windows, so you will need to enable pop-ups within your web browser while using the software. A user manual and interactive demo are available by clicking this link: http://www.ecog.org/general/stsinfo.html

Please take a moment to familiarize yourself with the software prior to using the system.

An STS generated shipping manifest form should be shipped with all specimen submissions.

Please direct your questions or comments pertaining to the STS to ecog-acrin.tst@jimmy.harvard.edu.

11.3.1 Study Specific Notes

Generic Specimen Submission Form (#2981) will be required only if STS is unavailable at time of sample submission. Notify the laboratory of the shipment by faxing a copy of the completed form to the laboratory. Indicate the appropriate laboratory on the submission form:

- ECOG-ACRIN Leukemia Translational Research Laboratory

Retroactively, enter all specimen collection and shipping information when STS is available.

11.4 Use of Specimens in Research

Specimens from patients who consented to allow their specimens to be used for future ECOG-ACRIN approved research studies will be retained in the ECOG-ACRIN Leukemia Tissue Bank (LTB).

Specimens submitted will be processed to maximize their utility for current and future research projects.

If future use is denied or withdrawn by the patient, the specimens will be removed from consideration for use in any future research study.

11.5 Sample Inventory Submission Guidelines

Inventories of all samples submitted from institutions will be tracked via the ECOG-ACRIN STS and receipt and usability verified by the receiving laboratory. Inventories of samples forwarded by the LTRL/LTB and utilized for approved laboratory research studies will be submitted by the LTRL/LTB to the ECOG-ACRIN Operations Office – Boston on a monthly basis in an electronic format.

12. Leukemia Correlative Studies

12.1 Immunophenotype and Molecular Genetics

Immunophenotyping has become an essential part of the diagnostic work-up of all leukemia patients. In fact, the diagnosis of leukemia without immunophenotypic characterization is no longer acceptable. ECOG-ACRIN has, therefore, developed a model system for antigenic data collection that requests specimens from all patients entered on ECOG-ACRIN leukemia trials be studied by the ECOG-ACRIN LTRL. In addition to establishing the leukemia subtype, this centralized testing and data collection has allowed research questions of clinical relevance to be applied to a growing database (e.g., definition of prognostically significant antigen expression levels to eventually yield specific treatment subcategories). In EAI141, an additional significance for central immunophenotyping is provided by the flow cytometric assessment of minimal residual disease (MRD). Sample submissions for MRD determination in bone marrow aspirates will coincide with clinically required bone marrow examinations. The LTRL will use multiparameter 6-color flow cytometry of bone marrow aspirates using leukemia-associated immunophenotypes to detect residual blast cells.

Bone marrow from patients entered on studies of hematologic malignancies are stored in ECOG-ACRIN's LTB for EAI141 embedded and future laboratory studies. The bank provides the scientific community with a source of leukemia specimens that are collected, processed, and maintained following quality control and quality assurance guidelines. The bank will accommodate requests from investigators within and outside ECOG-ACRIN in a timely and efficient manner, with respect to tissue type, tissue preparation, and most importantly, biologic characteristics of specimens.

12.2 Lab Data Transfer Guidelines

The data collected on the above mentioned laboratory research studies will be submitted electronically using a secure data transfer to the ECOG-ACRIN Operations Office – Boston by the investigating laboratories on a quarterly basis or per joint agreement between ECOG-ACRIN and the investigator.

13. Electronic Data Capture

Please refer to the EAI141 Forms Completion Guidelines for the forms submission schedule. Data collection will be performed exclusively in Medidata Rave.

This study will be monitored by the CTEP Data Update System (CDUS) version 3.0. Cumulative CDUS data will be submitted quarterly from the ECOG-ACRIN Coordinating Center to CTEP by electronic means.

14. Patient Consent and Peer Judgment

Current FDA, NCI, state, federal and institutional regulations concerning informed consent will be followed.

15. References

1. Rowe, J.M., *Prognostic factors in adult acute lymphoblastic leukaemia.* Br J Haematol, 2010. **150**(4): p. 389-405.
2. Mattison, R.J., S.M. Luger, and H.M. Lazarus, *New strategies for the evaluation of the nadir bone marrow following induction in acute myeloid leukemia.* Curr Opin Hematol, 2013. **20**(2): p. 93-9.
3. Muzi, M., D.A. Mankoff, J.R. Grierson, J.M. Wells, H. Vesselle, and K.A. Krohn, *Kinetic modeling of 3'-deoxy-3'-fluorothymidine in somatic tumors: mathematical studies.* J Nucl Med, 2005. **46**(2): p. 371-80.
4. Cheson, B.D., et al., *Revised recommendations of the International Working Group for Diagnosis, Standardization of Response Criteria, Treatment Outcomes, and Reporting Standards for Therapeutic Trials in Acute Myeloid Leukemia.* J Clin Oncol, 2003. 21(24): p. 4642-9.
5. Vanderhoek, M., M.B. Juckett, S.B. Perlman, R.J. Nickles, and R. Jeraj, *Early assessment of treatment response in patients with AML using [(18)F]FLT PET imaging.* Leuk Res, 2011. **35**(3): p. 310-6.
6. Hintze, J. 2013. PASS 12. NCSS, LLC. Kaysville, Utah, USA. www.ncss.com
7. Lan KK, Wittes J. *The B-value: a tool for monitoring data.* Biometrics. 1988;44(2):579-85.

Early Assessment of Treatment Response in AML using FLT PET/CT Imaging

Appendix I

Patient Thank You Letter

We ask that the physician use the template contained in this appendix to prepare a letter thanking the patient for enrolling in this trial. The template is intended as a guide and can be downloaded from the web site at http://www.ecog-acrin.org. As this is a personal letter, physicians may elect to further tailor the text to their situation.

This small gesture is a part of a broader program being undertaken by ECOG-ACRIN and the NCI to increase awareness of the importance of clinical trials and improve accrual and follow-through. We appreciate your help in this effort.

[PATIENT NAME] [DATE]

[PATIENT ADDRESS]

Dear [PATIENT SALUTATION],

Thank you for agreeing to take part in this important research study. Many questions remain unanswered in cancer. With the help of people like you who participate in clinical trials, we will achieve our goal of effectively treating and ultimately curing cancer.

We believe you will receive high quality, complete care. I and my research staff will maintain very close contact with you. This will allow me to provide you with the best care while learning as much as possible to help you and other patients.

On behalf of ***[INSTITUTION]*** and ECOG-ACRIN, we thank you again and look forward to helping you.

Sincerely,

[PHYSICIAN NAME]

Early Assessment of Treatment Response in AML using FLT PET/CT Imaging

Appendix II

EAI141 Adverse Event Reporting Guidance Document

Below are scenarios that a site may be faced with when assessing patients prior to and after the Day 14 FLT scan on EAI141. The below scenarios are to provide guidance on how to handle reporting on these issues. This is not an exhaustive list; if there is a situation you are unclear on for reporting please contact the Study Team.

When assessing patients prior to the Day 14 scan, ensure you document the patient was physically and mentally able to undergo the Day 14 scan.

Scenario #1

A subject develops grade 3 febrile neutropenia on hospital day 10 after the beginning of therapy. The grade 3 febrile neutropenia persists despite antibiotics through day 17. FLT is administered and imaging performed on day 14.

Does febrile neutropenia need to be reported as an SAE? No, the SAE is not a new event that occurred after FLT was given. If a fever is documented during the hospitalization prior to the scan, the day 15 fever should not be considered a new AE, therefore no reporting will occur.

Scenario #2

A subject develops grade 3 anemia (hemoglobin < 8 requiring transfusion) on hospital day 7. The patient requires transfusions every 2-3 days through day 20. FLT is administered and imaging performed on day 14. The anemia never becomes life-threatening (grade 4).

Does anemia need to be reported as an SAE? No, anemia was present prior to FLT administration and it has not changed in severity.

Scenario #3

A subject develops grade 3 mucositis. This occurs on day 15, and is attributed to chemotherapy. FLT was administered and imaging conducted on day 14.

Does this need to be reported as an SAE? Yes, though mucositis is not attributed to FLT but instead to chemotherapy, it occurred within the 24 hour monitoring period and must be reported within 24 hours via the AERS.