DATE: July 24, 2020

TO: CTEP Protocol and Information Office

FROM: Kamal Menghrajani, MD

SUBJECT: Amendment to update the participating organizations and other administrative changes.

SUMMARY OF CHANGES – Protocol:

I. PIO Recommendations:

#	Section	Comments
1.	4.1	**Please delete the information within this subsection and replace with the following language.** **4.1 Investigator and Research Associate Registration with CTEP** 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 at 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 Oncology Patient Enrollment Network (OPEN), Rave, or acting as a primary site contact) must complete their annual registration using CTEP's web-based Registration and Credential Repository (RCR) at https://ctepcore.nci.nih.gov/rcr. RCR utilizes five person registration types. • IVR: MD, DO, or international equivalent, • NPIVR: advanced practice providers (*e.g.*, NP or PA) or graduate level researchers (*e.g.*, PhD), • AP: clinical site staff (*e.g.*, RN or CRA) with data entry access to CTSU applications (*e.g.*, Roster Update Management System [RUMS], OPEN, Rave,), • Associate (A): other clinical site staff involved in the conduct of NCI-sponsored trials, and • Associate Basic (AB): individuals (*e.g.*, pharmaceutical company employees) with limited access to NCI-supported systems. RCR requires the following registration documents:

<table>
<tr><th>#</th><th>Section</th><th>Comments</th></tr>
<tr><td></td><td></td><td>
<table>
<tr><th>Documentation Required</th><th>IVR</th><th>NPIVR</th><th>AP</th><th>A</th><th>AB</th></tr>
<tr><td>FDA Form 1572</td><td>✓</td><td>✓</td><td></td><td></td><td></td></tr>
<tr><td>Financial Disclosure Form</td><td>✓</td><td>✓</td><td>✓</td><td></td><td></td></tr>
<tr><td>NCI Biosketch (education, training, employment, license, and certification)</td><td>✓</td><td>✓</td><td>✓</td><td></td><td></td></tr>
<tr><td>GCP training</td><td>✓</td><td>✓</td><td>✓</td><td></td><td></td></tr>
<tr><td>Agent Shipment Form (if applicable)</td><td>✓</td><td></td><td></td><td></td><td></td></tr>
<tr><td>CV (optional)</td><td>✓</td><td>✓</td><td>✓</td><td></td><td></td></tr>
</table>

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

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

In addition, all investigators act as the Site-Protocol PI, consenting/treating/drug shipment, or as the CI on the DTL must be rostered at the enrolling site with a participating organization (*i.e.*, Alliance).

Additional information is located 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.

RESPONSE: This has been updated.</td></tr>
<tr><td>2.</td><td>4.2</td><td>**Please delete the information within this subsection and replace with the following language.**

4.2 Site Registration
This study is supported by the NCI Cancer Trials Support Unit (CTSU).

Sites participating with the NCI Central Institutional Review Board (NCI CIRB) must submit the Study Specific Worksheet for Local Context (SSW) to the CIRB using IRBManager to indicate their intent to open the study locally. The NCI</td></tr>
</table>

#	Section	Comments
		CIRB's approval of the SSW is automatically communicated to the CTSU Regulatory Office, but sites are required to contact the CTSU Regulatory Office at CTSURegPref@ctsu.coccg.org to establish site preferences for applying NCI CIRB approvals across their Signatory Network. Site preferences can be set at the network or protocol level. Questions about establishing site preferences can be addressed to the CTSU Regulatory Office by emailing the email address above or calling 1-888-651-CTSU (2878). In addition, the Site-Protocol PI (*i.e.*, the investigator on the IRB/REB approval) must meet the following five criteria to complete processing of the IRB/REB approval record: • Holds an Active CTEP status, • Rostered at the site on the IRB/REB approval (*applies to US and Canadian sites only*) and on at least one participating roster, • If using NCI CIRB, rostered on the NCI CIRB Signatory record, • Includes the IRB number of the IRB providing approval in the Form FDA 1572 in the RCR profile, and • Holds the appropriate CTEP registration type for the protocol. **Additional Requirements** Additional requirements to obtain an approved site registration status include: • An active Federalwide Assurance (FWA) number, • An active roster affiliation with the Lead Protocol Organization (LPO) or a Participating Organization, and • Compliance with all protocol-specific requirements (PSRs). **RESPONSE: This has been updated.**
3.	4.2.2	**Please delete the following bullet point.** • 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.) **RESPONSE: This has been updated.**
4.	4.2.3	**Please delete the information within this subsection and replace with the following language.** 4.2.3 Submitting Regulatory Documents

#	Section	Comments
		Submit required forms and documents to the CTSU Regulatory Office via the Regulatory Submission Portal on the CTSU website. To access the Regulatory Submission Portal, log on to the CTSU members' website → Regulatory → Regulatory Submission. Institutions with patients waiting that are unable to use the Regulatory Submission Portal should alert the CTSU Regulatory Office immediately at 1-866-651-2878 in order to receive further instruction and support. **RESPONSE: This has been updated.**
5.	4.2.4	**Please delete the information within this subsection and replace with the following language.** 4.2.4 Checking Site Registration Status You can verify your site's registration status on the members' side of the CTSU website. • Log on to the CTSU members' website • Click on *Regulatory* at the top of your screen • Click on *Site Registration* • Enter your 5-character CTEP Institution Code and click on Go Note: The status shown only reflects institutional compliance with site registration requirements as outlined above. 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. **RESPONSE: This has been updated.**
6.	4.3.1, ~~4.3.2~~	**Please delete the information within these 2 subsections and replace with the following language.** 4.3.1 OPEN / IWRS The Oncology Patient Enrollment Network (OPEN) is a web-based registration system available on a 24/7 basis. OPEN is integrated with CTSU regulatory and roster data and with the Lead Protocol Organization (LPOs) registration/randomization systems or Theradex Interactive Web Response System (IWRS) for retrieval of patient registration/randomization assignment. OPEN will populate the patient enrollment data in NCI's clinical data management system, Medidata Rave.

#	Section	Comments
		Requirements for OPEN access: • A valid CTEP-IAM account. • To perform enrollments or request slot reservations: Be on an LPO roster, ETCTN Corresponding roster, or Participating Organization roster with the role of Registrar. Registrars must hold a minimum of an AP registration type. • If a DTL is required for the study, the registrar(s) must hold the OPEN Registrar task on the DTL for the site. • Have an approved site registration for a protocol prior to patient enrollment. To assign an Investigator (IVR) or Non-Physician Investigator (NPIVR) as the treating, crediting, consenting, drug shipment (IVR only), or receiving investigator for a patient transfer in OPEN, the IVR or NPIVR must list the IRB number used on the site's IRB approval on their Form FDA 1572 in RCR. If a DTL is required for the study, the IVR or NPIVR must be assigned the appropriate OPEN-related tasks on the DTL. Prior to accessing OPEN, site staff should verify the following: • Patient has met all eligibility criteria within the protocol stated timeframes, and • 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. Access OPEN at https://open.ctsu.org or from the OPEN link on the CTSU members' website. Further instructional information is in the OPEN section of the CTSU website at https://www.ctsu.org or https://open.ctsu.org. For any additional questions, contact the CTSU Help Desk at 1-888-823-5923 or ctsucontact@westat.com. **RESPONSE: This has been updated. Subsection 4.3.2 was deleted and subsequent sections were renumbered.**
7.	4.2.2	**Please add this information to this section.** This Study will use the ETCTN Specimen Tracking System (STS). • All biospecimens collected for this trial must be submitted using the

#	Section	Comments
		ETCTN Specimen Tracking System (STS) unless otherwise noted. • The system is accessed through special Rave user roles: "CRA Specimen Tracking" for data entry at the treating institutions and "Biorepository" for users receiving the specimens for processing and storage at reference labs and the Biorepository. • Please refer to the Medidata Account Activation and Study Invitation Acceptance link on the CTSU website under the Rave/DQP tab. • **Important: Failure to complete required fields in STS may result in a delay in sample processing.** Any case reimbursements associated with sample submissions will not be credited if samples requiring STS submission are not logged into STS. **RESPONSE: This has been added.**
8.	10.3.1	**If this study will use Rave Cancer Therapy Evaluation Program Adverse Event Reporting System (CTEP-AERS) Integration, include this section.** **1.1.1 Rave-CTEP-AERS Integration** The Cancer Therapy Evaluation Program Adverse Event Reporting System (CTEP-AERS) integration enables evaluation of post-baseline AEs entered in Rave to determine whether they require expedited reporting, and facilitates entry in CTEP-AERS for those AEs requiring expedited reporting. All AEs that occur after baseline are collected in Medidata Rave using the Adverse Event form, which is available for entry at each treatment or reporting period, and used to collect AEs that start during the period or persist from the previous reporting period. The Clinical Research Associate (CRA) will enter AEs that occur prior to the start of treatment on a baseline form that is not included in the Rave-CTEP-AERS integration. AEs that occur prior to enrollment must begin and end on the baseline Adverse Event form and should not be included on the standard Adverse Events form that is available at treatment unless there has been an increase in grade. Prior to sending AEs through the rules evaluation process, site staff should verify the following on the Adverse Event form in Rave: • The reporting period (course/cycle) is correct, and • AEs are recorded and complete (no missing fields) and the form is query-free (fields added to the form during study build do not need to be query-free for the integration call with CTEP-AERS to be a success).

#	Section	Comments
		The CRA reports AEs in Rave at the time the Investigator learns of the event. If the CRA modifies an AE, it must be re-submitted for rules evaluation. Upon completion of AE entry in Medidata Rave, the CRA submits the AE for rules evaluation by completing the Expedited Reporting Evaluation form. Both NCI and protocol-specific reporting rules evaluate the AEs submitted for expedited reporting. A report is initiated in CTEP-AERS using information entered in Medidata Rave for AEs that meet reporting requirements. The CRA completes the report by accessing CTEP-AERS via a direct link on the Medidata Rave Expedited Reporting Evaluation form. In the rare occurrence that Internet connectivity is lost, a 24-hour notification is to be made to CTEP by telephone at 301-897-7497. Once Internet connectivity is restored, the 24-hour notification that was phoned in must be entered immediately into CTEP-AERS using the deep link from Medidata Rave. Additional information about the CTEP-AERS integration is available on the CTSU website: • Study specific documents: Protocols > Documents > Education and Promotion, and • Expedited Safety Reporting Rules Evaluation user guide: Resources > CTSU Operations Information > User Guides. NCI requirements for SAE reporting are available on the CTEP website: • NCI Guidelines for Investigators: Adverse Event Reporting Requirements is available at https://ctep.cancer.gov/protocolDevelopment/electronic_applications/docs/aeguidelines.pdf. **RESPONSE: This has been updated.**
9.	13.2	**Please delete the information within this subsection and replace with the following language.** **13.2 Data Reporting** Medidata Rave is a clinical data management system being used for data collection for this trial/study. Access to the trial in Rave is controlled through the CTEP-IAM system and role assignments. To access Rave via iMedidata:

#	Section	Comments

		• Site staff will need to be registered with CTEP and have a valid and active CTEP-IAM account, and
		• Assigned one of the following Rave roles on the relevant Lead Protocol Organization (LPO) or Participating Organization roster at the enrolling site: Rave CRA, Rave Read Only, Rave CRA (LabAdmin), Rave SLA, or Rave Investigator. Refer to https://ctep.cancer.gov/investigatorResources/default.htm for registration types and documentation required.
		  ○ To hold Rave CRA or Rave CRA (Lab Admin) role, site staff must hold a minimum of an AP registration type,
		  ○ To hold Rave Investigator role, the individual must be registered as an NPIVR or IVR, and
		  ○ To hold Rave Read Only role, site staff must hold an Associates (A) registration type.
		
		Upon initial site registration approval for the study in Regulatory Support System (RSS), all persons with Rave roles assigned on the appropriate roster will be sent a study invitation e-mail from iMedidata. To accept the invitation, site staff must log in to the Select Login (https://login.imedidata.com/selectlogin) using their CTEP-IAM username and password, and click on the *accept* link in the upper right-corner of the iMedidata page. Site staff 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 iMedidata screen. If an eLearning is required and has not yet been taken, the link to the eLearning will appear under the study name in iMedidata instead of the *Rave EDC* link; once the successful completion of the eLearning has been recorded, access to the study in Rave will be granted, and a *Rave EDC* link will display under the study name.
		
		Site staff that have not previously activated their iMedidata/Rave account at the time of initial site registration approval for the study in RSS will also receive a separate invitation from iMedidata to activate their account. Account activation instructions are located on the CTSU website in the Rave section under the Rave resource materials (Medidata Account Activation and Study Invitation Acceptance). Additional information on iMedidata/Rave is available on the CTSU members' website in the Data Management > Rave section 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.
		
		RESPONSE: This has been updated.

10.	13.3	**Please add this subsection.**

#	Section	Comments
		Data Quality Portal The Data Quality Portal (DQP) provides a central location for site staff to manage unanswered queries and form delinquencies, monitor data quality and timeliness, generate reports, and review metrics. The DQP is located on the CTSU members' website under Data Management. The Rave Home section displays a table providing summary counts of Total Delinquencies and Total Queries. DQP Queries, DQP Delinquent Forms, and the DQP Reports modules are available to access details and reports of unanswered queries, delinquent forms, and timeliness reports. Review the DQP modules on a regular basis to manage specified queries and delinquent forms. The DQP is accessible by site staff that are rostered to a site and have access to the CTSU website. Staff that have Rave study access can access the Rave study data using a direct link on the DQP. To learn more about DQP use and access, click on the Help icon displayed on the Rave Home, DQP Queries, and DQP Delinquent Forms modules. Note: Some Rave protocols may not have delinquent form details or reports specified on the DQP. A protocol must have the Calendar functionality implemented in Rave by the Lead Protocol Organization (LPO) for delinquent form details and reports to be available on the DQP. Site staff should contact the LPO Data Manager for their protocol regarding questions about Rave Calendaring functionality. **RESPONSE: This has been added and subsequent section renumbered.**

II. Changes Requested by the PI:

#	Section	Comments
11.	Header, Title Page	Updated the protocol version date.
12.	General	General formatting throughout the document.
13.	Title Page	Added the ClinicalTrials.gov number. Removed LAOs MN026, NC010, and NJ066 from the Participating Organizations.

#	Section	Comments
		Removed the Non-Member Collaborators, as requested by CTEP. Changed the Study Contact individual.
14.	3.1.3	Clarified the targetable mutations.
15.	4.2.2	Clarified who to contact to schedule the SIV.
16.	6.1, 6.4	Clarified that hydroxyurea may be given concurrently.

SUMMARY OF CHANGES – Consent Form:

#	Section	Comments
17.	Header	Updated the version date.

NCI Protocol #: 10200

Local Protocol #: N/A

ClinicalTrials.gov Identifier: NCT03701295

TITLE: A Phase Ib/II Study of the Histone Methyltransferase Inhibitor Pinometostat in Combination with Azacitidine in Patients with 11q23-Rearranged Acute Myeloid Leukemia

Corresponding Organization: **LAO-MD017** JHU Sidney Kimmel Comprehensive Cancer Center LAO

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Participating Organizations:

LAO-11030 / University Health Network Princess Margaret Cancer Center LAO
LAO-CA043 / City of Hope Comprehensive Cancer Center LAO
LAO-CT018 / Yale University Cancer Center LAO
LAO-MA036 / Dana-Farber - Harvard Cancer Center LAO
LAO-MD017 / JHU Sidney Kimmel Comprehensive Cancer Center LAO
LAO-OH007 / Ohio State University Comprehensive Cancer Center LAO
LAO-PA015 / University of Pittsburgh Cancer Institute LAO
LAO-TX035 / University of Texas MD Anderson Cancer Center LAO
LAO-NCI / National Cancer Institute LAO

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Baltimore, MD 21287
Telephone: (410) 955-4044
jmurra33@jhmi.edu

NCI-Supplied Agent(s): Pinometostat NSC# 795144
Other Agent(s): Azacitidine, NSC#102816 Supplier: Commercial
IND Sponsor: *DCTD, NCI*

Protocol Type / Version # / Version Date:

Original / v 1.0 / 04/18/2018
Original / v.2.0 / 06/25/2018
Original / v.3.0 / 07/20/2018
Original / v.4.0 / 08/15/2018
Original / v.5.0 / 09/13/2018
Original / v.6.0 / 10/17/2018
Original / v.7.0 / 11/27/2018
Original / v.8.0 / 02/27/2019
Original / v. 9.0 / 04/10/2019
Original / v.10.0 / 05/03/2019
Original / v.11.0 / 09/10/2019
Original / v.12.0 / 10/25/2019
Original / v.13.0 / 07/24/2020

SCHEMA

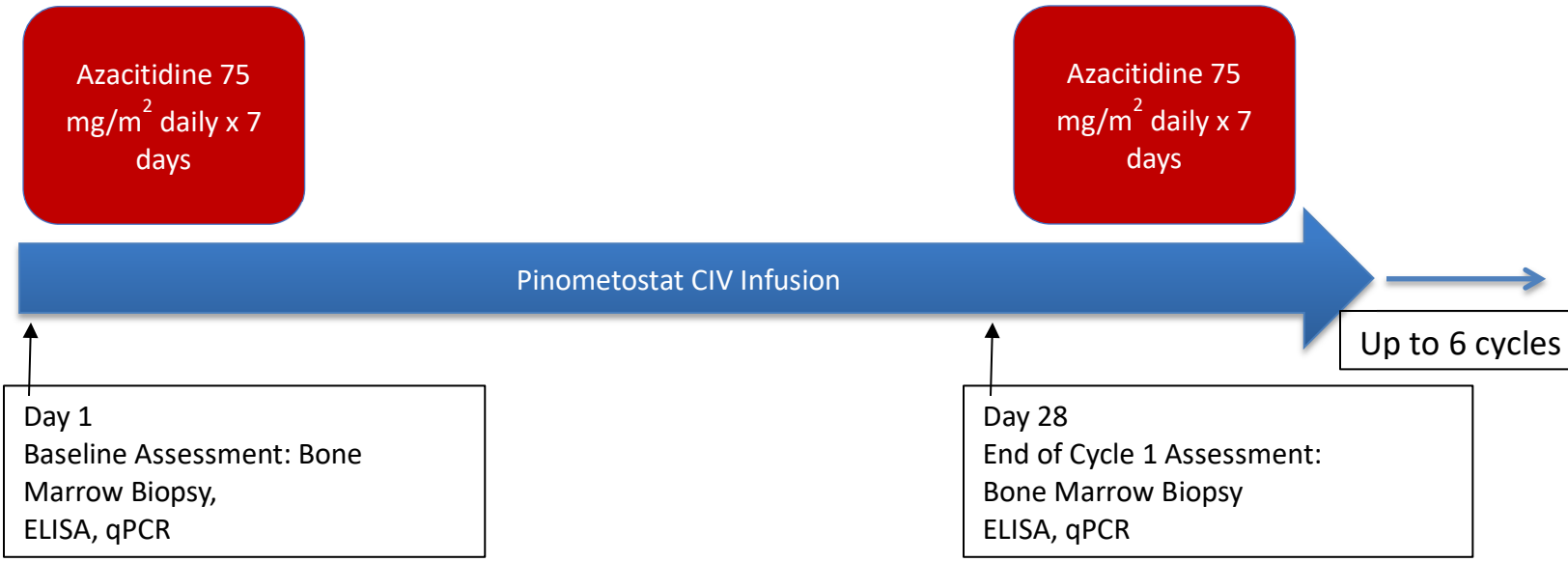

Azacitidine 75 mg/m2 daily x 7 days
Azacitidine 75 mg/m2 daily x 7 days
Pinometostat CIV Infusion
Up to 6 cycles
Day 1
Baseline Assessment: Bone Marrow Biopsy,
ELISA, qPCR
Day 28
End of Cycle 1 Assessment:
Bone Marrow Biopsy
ELISA, qPCR

TABLE OF CONTENTS

1. OBJECTIVES

1.1 Primary Objectives

1.1.1 Phase Ib Safety / Tolerability Run In - Primary Objective:

- To determine if the combination of pinometostat, at a dose of 54 or 90mg/m2/day, and azacitidine, at a dose of 75 mg/m2 daily for 7 days, is safe and tolerable in patients with MLL-rearranged acute myeloid leukemia, either in the relapsed / refractory setting or in those who choose not to undergo standard induction therapy in the previously untreated setting

1.1.2 Phase II Primary Objective:

- To determine the preliminary efficacy, as determined by overall response rate (CR, CRi, PR, and MLFS), of pinometostat administered at the maximum tolerated dose from the phase 1b, combined with azacitidine administered at 75 mg/m^2 daily for 7 days, in patients with MLL-rearranged acute myeloid leukemia, either in the relapsed / refractory setting or in those who choose not to undergo standard induction therapy in the previously untreated setting. Patients will be treated with 6 cycles of therapy on trial, with the option to continue if they are deriving significant clinical benefit. As only two dose levels are being studied, the MTD may not be reached. In this case, a recommended phase 2 dose (RP2D) of either 54 or 90 mg/m2 pinometostat will be evaluated in the Phase 2 portion of the study, based on the dosing used in the previous phase 1 trial of single-agent pinometostat[1].

1.2 Secondary Objectives

1.2.1 Phase Ib Safety / Tolerability Run In - Secondary Objectives:

- Perform correlative studies to evaluate for on-target effects, cellular differentiation, and decreased leukemia cell proliferation in these patients
- Correlative studies will include assessment of patterns of histone methylation at H3K79 by ELISA; evaluation of expression of downstream targets of DOT1L-mediated hypermethylation (HOXA9 and Meis1) by qPCR; measurement of myeloid cell differentiation by differential and flow cytometry on bone marrow biopsy samples and peripheral blood samples
- To observe and record anti-tumor activity. Although the clinical benefit of these drug(s) has not yet been established, the intent of offering this treatment is to provide a possible therapeutic benefit, and thus the patient will be carefully monitored for tumor response and symptom relief in addition to safety and tolerability.

1.2.2 Phase II Secondary Objectives:

- Perform correlative studies to evaluate for on-target effects, cellular differentiation, and decreased leukemia cell proliferation in these patients.
- Correlative studies will include assessment of patterns of histone methylation at H3K79 by ELISA; evaluation of expression of downstream targets of DOT1L-mediated hypermethylation (HOXA9 and Meis1) by qPCR; measurement of

myeloid cell differentiation by differential and flow cytometry on bone marrow biopsy samples and peripheral blood samples

2. BACKGROUND

2.1 Study Disease(s)

Acute myeloid leukemia (AML) is a cancer of proliferative, clonal, abnormally differentiated cells of the hematopoietic system.[1] Genomic profiling and cytogenetics have significantly improved our ability to discern prognosis and likelihood of response to therapy for patients with AML.[2] Patients at high risk for poor response to standard induction therapy or early relapse include those with unfavorable abnormalities seen on routine cytogenetic testing, including partial tandem duplications and rearrangements in the 11q23 locus, also known as the Mixed Lineage Leukemia (*MLL*) locus[2]. *MLL* rearrangements are not uncommon and are found in about 10% of adult AML[3–5].

In *MLL*-rearranged AML, aberrant recruitment of the disruptor of telomeric silencing 1-like (DOT1L), a histone H3 lysine 79 (H3K79) methyltransferase, and subsequent hypermethylation of downstream targets *HOXA9* and *Meis1* is central to leukemogenesis[3,6] (Figure 1).

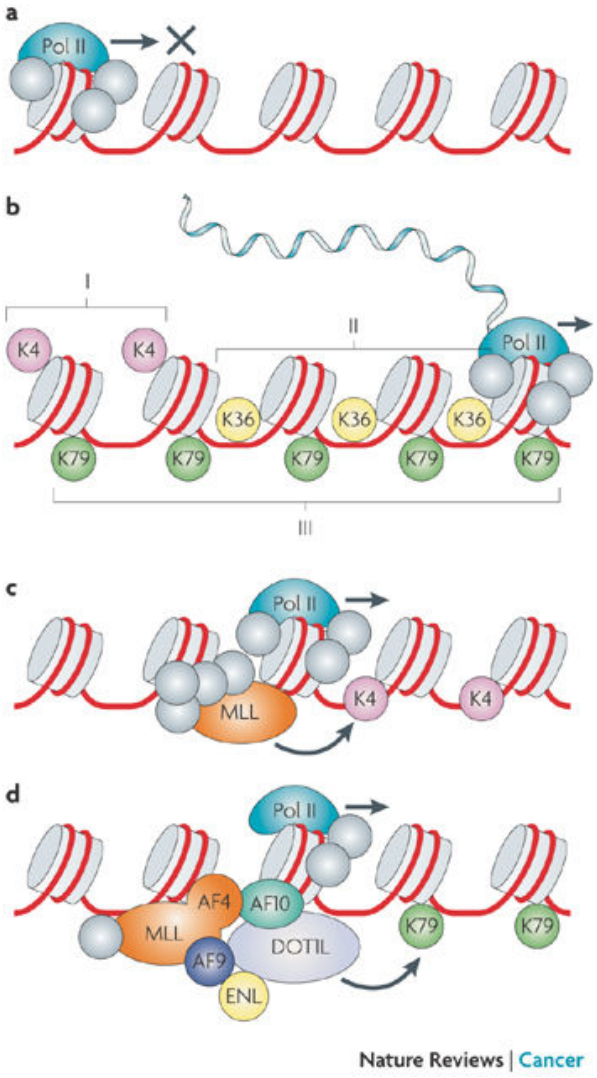


Figure 1. Role of DOT1L and histone methylation in leukemogenesis[3]. A. RNA polymerase II (Pol II) can bind the promoter region of a gene but cannot proceed with transcription without specific methylation marks on the histone core. B. Unique histone marks can be found on different regions of a gene, and may impart unique activities. RNA transcription may be initiated when a promoter region (I) carries histone H3 lysine 4 (H3K4) methylation marks, and extend when an open-reading frame region (II) carries a histone H3 lysine 36 (H3K36). Histone H3 lysine 79 (H3K79) methylation marks have a broad distribution across promoter and open-reading frame regions (III). C. Mixed lineage leukemia (MLL) is a member of a multiprotein complex that mediates methylation of H3K4 within the promoter region of genes occupied by RNA polymerase II. D. A hypothetical function for MLL fusion proteins is presented. MLL fusion(s) that lack the SET (Su(var)3-9, Enhancer-of-zeste, Trithorax) domain H3K4 methyltransferase activity may recruit the H3K79 methyltransferase DOT1L. MLL fusion-mediated recruitment of DOT1L to promoters normally occupied by MLL, (such as the HoxA cluster) allows H3K79 methylation of the HoxA cluster, which may lead to aberrant expression of HoxA cluster genes.

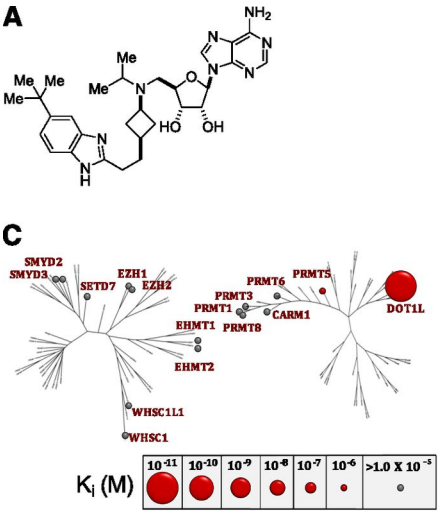
DOT1L has been previously shown to be necessary for the development and maintenance of *MLL*-rearranged leukemia[7]. In one pre-clinical study, a conditional DOT1L deletion model was used to show that DOT1L-deficient cells are depleted of the global H3K79 methylation mark. Loss of DOT1L activity in *MLL*-rearranged cells attenuated cell viability and colony formation potential. The effect was found to contribute to the viability of cells immortalized by multiple MLL oncoproteins, and not only the fusion products that are known to directly recruit DOT1L[8].

While MLL-rearranged AML is the most well-studied instance of DOT1L-mediated leukemogenesis, recent studies have shown that this pathway also drives other types of AML. Several researchers have shown that DOT1L-mediated aberrant methylation of downstream targets (HOXA9 and Meis1) is also seen in AML with *DNMT3A*, *NPM1*, and *IDH1/2* mutations, with the strongest signal seen in *NPM1*-mutated disease[9–11]. Not only are similar patterns of DOT1L-mediated changes seen, but these types of AML have also been shown to be responsive to small-molecule inhibition of DOT1L. As such, patients with these mutations could be considered candidates for further studies of combination DOT1L inhibitor and hypomethylating agent therapy.

2.2 CTEP IND Agent

A small molecule inhibitor, pinometostat, discovered by Epizyme and now under the purview of CTEP, was found to be a potent and selective inhibitor of DOT1L histone methyltransferase activity. Use of this agent in a continuous IV infusion in a rat xenograft model of MLL-rearranged leukemia showed complete, sustained tumor regressions without significant appreciable toxicities[12].

Pinometostat was then moved into the clinic for first-in-human studies. This agent directly targets DOT1L with sub-nanomolar affinity and >37,000 fold selectivity as compared to other histone methyltransferases[12].



Property	EPZ-5676	EPZ004777
DOT1L inh. K_i (nM)	0.08 ± 0.03	0.3 ± 0.02
Residence time (hr)	> 24	1
Fold selectivity	> 37,000	> 1,000
MV4-11 prolif. IC_{50} (nM)	3.5 ± 0.7	151 ± 42
H3K79me2 inh. IC_{50} (nM)	2.7 ± 0.9	11 ± 4
HOXA9 inh. IC_{50} (nM)	67 ± 27	691 ± 145
MEIS1 inh. IC_{50} (nM)	53 ± 17	984 ± 234

Figure 2. Structure, binding, and inhibitory activity of Pinometostat (EPZ-5676)[12]. A. Chemical structure of EPZ-5676, now called pinometostat. B. Superposition of DOT1L-EPZ-5676 (green) and DOT1L-SAM (gray; Protein Data Base accession number 3QOW). To accommodate the extended hydrophobic tail of the inhibitor, significant rearrangement of the protein is required, including the loop between β-strands 10 and 11 (L10-11). C. Selectivity profile of

pinometostat inhibitory activity against representative members of the lysine (left) and arginine (right) enzyme families. The diameter of the sphere for each enzyme is directly related to the magnitude of inhibition by pinometostat. Larger circles correlate to increased potency; gray circles indicate no measurable inhibition up to 10 μM of pinometostat. D. Comparison of pinometostat and EPZ004777 potency, selectivity, and cell-based activity. The enzyme inhibition Ki values for EPZ004777 and pinometostat in DOT1L enzymatic assays are listed (n = 3; mean values ± SD are shown). Residence time for each compound is listed and was calculated as the reciprocal of the enzyme-ligand dissociation rate as determined by surface plasmon resonance. Also listed are inhibitory activities for both compounds in MV4-11 proliferation assays (n = 3 [pinometostat] or n = 2 [EPZ004777]; mean values ± SD are shown), MV4-11 cell H3K79me2 ELISA assays (n = 2; mean values ± SD are shown) and MV4-11 cell *HOXA9* and *MEIS1*qRT-PCR assays (n = 2; mean values ± SD are shown).

A Phase I trial of pinometostat in patients with advanced hematologic malignancies, including acute leukemia with MLL rearrangements, showed some promise. Eight of 34 patients with MLL-rearranged leukemia showed a response to the drug, with one patient in a morphologic CR, one in a cytogenetic CR, two patients with resolution of leukemia cutis, and the remaining with signs of differentiation or leukocytosis (Table 1). Correlative studies showed that pinometostat plasma concentrations in these patients were correlated with the inhibition of global H3K79 methylation, as well as with reductions in methylation of target genes HOXA9 and Meis1.[13]

Table 1. Dose of pinometostat, number of patients enrolled at each dose, and corresponding response from the phase I trial of this drug as a single agent in relapsed / refractory hematologic malignancies.

Dose mg/m²/day	Number of patients (n=51)	Marrow Response (n=3)	Leukemia cutis resolved (n=3)	Leukocytosis/ Differentiation (n=9)
12	1	-	-	-
24	5	-	-	1
36	4	-	1	2
54	6	2 CR	1	1
54 (28 day CIV)	8	-	1	1
80	3	-	-	2
90 (28 day CIV)	24	1 PR	-	2

As the dose of 54 mg/m2/day showed two CRs in the single-agent trial and the dose of 90 mg/m2/day showed one PR in the single-agent trial, these are the two doses planned for use in this combination study. Plasma pinometostat concentrations and H3K79 methyltransferase inhibition were found to decline rapidly between days 21 and 28 when the infusion was discontinued at day 21, and thus this trial will use a 28-day continuous IV infusion[1].

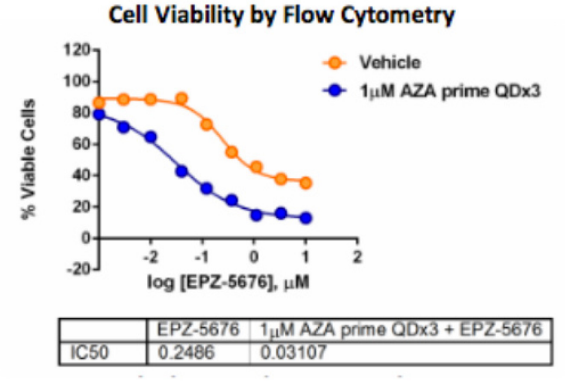
2.3 Other Agent: Azacitidine

Azacitidine is a nucleoside analogue of the cytosine which functions as an antineoplastic agent by incorporating into DNA and RNA, leading to chain termination, and by acting as a DNA methyltransferase inhibitor. In recent years, azacitidine has come into practice as a promising treatment for adult AML patients who are not candidates for induction chemotherapy, and is listed as a therapeutic option by the NCCN. Azacitidine was previously shown to prolong overall survival in comparison to best supportive care in the phase 3 AZA-001 trial for treatment-naïve older adults with 20-30% bone marrow blasts[14]. Newer data from the international phase 3 study of azacitidine compared to conventional care regimens in treatment-naive older adults with > 30% blasts shows that the response rate to this hypomethylating agent is around 30% (27.8% for CR / CRi and 1.2% for PR) with a 4-month overall survival benefit[15]. Similarly, a 2011 Italian study showed an overall response rate of 32% to single-agent azacitidine in adults with AML[16]. Azacitidine is administered IV or subcutaneously at a dose of 75mg/m^2 per day for 7 days. Therapy may be administered over seven sequential days, but is often administered with a two-day break in clinical practice.

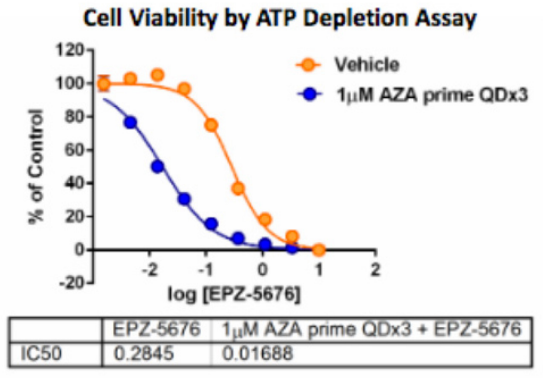
2.4 Rationale

Given that DOT1L-mediated leukemogenesis is mediated by both histone and DNA hypermethylation, a combination approach employing both pinometostat and a DNA hypomethylating agent would be a rational next step in developing up-front therapies for adults with AML with aberrant DOT1L signaling who are unable to tolerate or unwilling to undergo treatment with induction chemotherapy. A previous study by Epizyme showed that the combination of pinometostat with the hypomethylating agent azacitidine led to synergistic antiproliferative effects in vitro in MOLM-13 and MV4-11 MLL cell lines. Their previously published data showed that AML cell lines with a translocation t(9;11) or t(4;11) treated with both azacitidine and pinometostat had increased rates of cytotoxicity when compared to treatment with either drug alone[17].

Figure 3. Synergistic effects of EPZ-5676 (Pinometostat) and azacitidine on cell viability as measured by flow cytometry and ATP depletion[17].



	EPZ-5676	1μM AZA prime QDx3 + EPZ-5676
IC50	0.2486	0.03107



	EPZ-5676	1μM AZA prime QDx3 + EPZ-5676
IC50	0.2845	0.01688

2.5 Correlative Studies Background

The objective of the correlative studies is to better understand whether this combination is having the anticipated on-target effects and, if so, what the sequelae of those on-target effects may be.

Correlative studies will include morphology, flow cytometry, cytogenetics, and a myeloid genetics panel, all of which are standard of care. Integrated biomarkers for identification of 11q23 / MLL rearrangements would be performed as part of these assessments.

Additionally, we seek to study changes in methylation patterns with the use of this combination. This is important for understanding on-target effects as MLL-rearranged leukemia is driven by hypermethylation, and pinometostat works to inhibit DOT1L methyltransferase activity and 5-azacytidine is a DNA hypomethylating agent. The assay of choice would be an ELISA assay to quantitate the extent of H3K79 methylation in leukemia cells.

As the mechanism of leukemogenesis in MLL-rearranged leukemia relies on hypermethylation of H3K79 by DOT1L, leading to increased transcription of HOXA9 and Meis1, we anticipate that expression levels of these two genes will decrease with treatment. As such, we plan to use qPCR to evaluate gene expression level prior to and while on treatment.

We anticipate that pinometostat will induce differentiation of myeloid blasts into more mature myeloid forms, based on previous research on pinometostat. We anticipate that MLL rearrangements will be detectable in more mature myeloid cells, e.g. neutrophils, allowing us to evaluate the extent of differentiation. We will perform these assays on selected samples.

We considered running ChIP-seq for H3K27 methylation (as these histones may be mono-, di-, or trimethylated, and each of these is mediated by DOT1L). Pinometostat's inhibiton of DOT1L may lead to an accumulation of non-methylated or hypomethylated forms of H3K79 and ChIP-seq can help measure the valence of that histone methylation, as the amount of methylation has previously been described to correlate to the robustness of gene expression. However, the use of this assay would be limited by cost.

Determining azacitidine incorporation and DNA methylation would be an exploratory biomarker, also performed by the Sidney Kimmel Comprehensive Cancer Center Analytical Pharmacology Core. This would allow for the assessment of 5-aza-2`-deoxycytidine incorporation into genomic DNA and the extent of global DNA methylation. This will be one of the first assessments of 5-aza-2`-deoxycytidine incorporation relative to the extent of global DNA methylation in human samples and may give further insight into the mechanism of action of azacitidine.

3. PATIENT SELECTION

3.1 Eligibility Criteria

3.1.1 Patients must have histologically or cytologically confirmed acute myeloid leukemia.

3.1.2 Patients must have an 11q23 translocation or partial tandem duplication, confirmed by cytogenetics, FISH, or myeloid panel. Both de novo and therapy-related AML with an 11q23 rearrangement or PTD are considered eligible.

3.1.3 Patients may not have any other targetable mutations (such as FLT3, IDH1, and IDH2) identified on myeloid mutational panel testing or must refuse treatment with a targeted agent if such a mutation is detected.

3.1.4 Age ≥18 years.

3.1.5 ECOG performance status < 3 (Karnofsky >60%; see Appendix A).

3.1.6 Patients must have adequate organ function as defined below, unless thought to be related to the underlying disease:

- PT and aPTT	≤ 1.5x ULN
- total bilirubin	< 2 times the upper limit of institutional normal (ULN) unless due to Gilbert's disease
- AST(SGOT)/ALT(SGPT)	≤2.5 X institutional upper limit of normal
- creatinine	≤ 2 times the upper limit of institutional normal (ULN)
OR	
- creatinine clearance	GFR ≥30 mL/min/1.73 m^2

3.1.7 Patients treated in the up-front setting must decline standard-of-care therapy.

3.1.8 Ability to understand and the willingness to sign a written informed consent document. Participants with impaired decision-making capacity with a close legal guardian / caregiver may be considered.

3.1.9 Patients must have measurable disease, defined as abnormal blasts detectable in the peripheral blood or bone marrow or the presence of extramedullary disease, including leukemia cutis. Patients with extramedullary disease but no bone marrow disease are still considered eligible. See Section 12 (Measurement of Effect) for the evaluation of measurable disease.

3.1.10 Patients may have had previous treatment with standard-of-care or experimental agents. Patients who have previously undergone bone marrow transplantation may also be included.

3.1.11 Patients who are HIV+, HBV+, and / or HCV+ may be eligible as follows:

- Human Immunodeficiency Virus (HIV)-infected patients on effective antiretroviral therapy with undetectable viral load within 6 months are eligible for this trial. The antiretroviral therapy should not strongly induce or inhibit CYP3A4.
- If evidence of chronic Hepatitis B virus (HBV) infection, HBV viral load must be undetectable on suppressive therapy if indicated.
- If history of Hepatitis C virus (HCV) infection, must be treated with undetectable HCV viral load.

3.1.12 The effects of pinometostat on the developing human fetus are unknown. For this reason and because histone methyltransferase inhibitors as well as hypomethylating agents are known to be teratogenic, women of child-bearing potential and men must agree to use adequate contraception (hormonal or barrier method of birth control; abstinence) prior to study entry, and for the duration of study participation and for 4 weeks after the last dose of study treatment. Should a woman become pregnant or suspect she is pregnant while she or her partner is participating in this study, she should inform her treating physician immediately. Men treated or enrolled on this protocol must also agree to use adequate contraception prior to the study, for the duration of study participation, and 90 days after completion of pinometostat and azacitidine administration.

3.2 Exclusion Criteria

3.2.1 Patients who are receiving any other investigational agents.

3.2.2 Patients with active CNS disease are excluded from this clinical trial because they may develop progressive neurologic dysfunction that would confound the evaluation of neurologic and other adverse events. Patients with a prior history of CNS disease will not be excluded.

3.2.3 History of allergic reactions attributed to compounds of similar chemical or biologic composition to pinometostat or azacitidine

3.2.4 Patients receiving medications or substances that are inhibitors or inducers of the CYP3A4 or CYP450 system should have their medications reviewed and adjusted for interactions as appropriate for local institutional practice. Because the lists of these agents are constantly changing, it is important to regularly consult a frequently-updated medical reference. As part of the enrollment/informed consent procedures, the patient will be counseled on the risk of interactions with other agents, and what to do if new medications need to be prescribed or if the patient is considering a new over-the-counter medicine or herbal product.

3.2.5 Patients receiving any medications or substances that are inhibitors or inducers of MATE1 and MATE2-K transporters should have their medications reviewed and adjusted for interactions as appropriate for local institutional practice. Pinometostat has been demonstrated to be an inhibitor of MATE1 and MATE2-K transporters in vitro, although the clinical significance of this is unclear. Drug interactions may occur between Pinometostat and other therapies that are MATE substrates, including metformin.

Consultation with a frequently updated medical reference and / or pharmacist should be sought to guide necessary changes in the patient's other medications.

3.2.6 Uncontrolled intercurrent illness including, but not limited to, ongoing or active infection, symptomatic congestive heart failure, unstable angina pectoris, uncontrolled or clinically significant cardiac arrhythmia, or psychiatric illness/social situations that would limit compliance with study requirements. Patients with these conditions that are medically well controlled may be considered for enrollment.

3.2.7 Pregnant women are excluded from this study because pinometostat is an agent with the potential for teratogenic or abortifacient effects. Because there is an unknown but potential risk for adverse events in nursing infants secondary to treatment of the mother with pinometostat, breastfeeding should be discontinued if the mother is treated with pinometostat. These potential risks may also apply to other agents used in this study.

3.2.8 HIV-positive patients on combination antiretroviral therapy should have their regimen reviewed for potential pharmacokinetic interactions with pinometostat and azacitidine. In the event of a potential interaction, alternative therapies may be considered in consultation with the patient's primary HIV physician.

3.2.9 Absence of an 11q23 rearrangement or absence of an 11q23 partial tandem duplication

3.2.10 Patients with an active bleeding diathesis.

3.2.11 Patients at increased risk of QT prolongation (e.g. from known long-QT syndrome) or who have a corrected QT interval that is persistently longer than 450ms despite adjustments to other medications

3.2.12 Patients who are eligible for or willing to receive intensive induction therapy for de novo AML

3.3 Inclusion of Women and Minorities

NIH policy requires that women and members of minority groups and their subpopulations be included in all NIH-supported biomedical and behavioral research projects involving NIH-defined clinical research unless a clear and compelling rationale and justification establishes to the satisfaction of the funding Institute & Center (IC) Director that inclusion is inappropriate with respect to the health of the subjects or the purpose of the research. Exclusion under other circumstances must be designated by the Director, NIH, upon the recommendation of an IC Director based on a compelling rationale and justification. Cost is not an acceptable reason for exclusion except when the study would duplicate data from other sources. Women of childbearing potential should not be routinely excluded from participation in clinical research. Please see http://grants.nih.gov/grants/funding/phs398/phs398.pdf.

See section 9.2 for the planned distribution of subjects by sex/gender, race, and ethnicity in the Planned Enrollment Report table.

4. REGISTRATION PROCEDURES

4.1 Investigator and Research Associate Registration with CTEP

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 at 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 Oncology Patient Enrollment Network (OPEN), Rave, or acting as a primary site contact) must complete their annual registration using CTEP's web-based Registration and Credential Repository (RCR) at https://ctepcore.nci.nih.gov/rcr.

RCR utilizes five person registration types.

- IVR: MD, DO, or international equivalent,
- NPIVR: advanced practice providers (*e.g.*, NP or PA) or graduate level researchers (*e.g.*, PhD),
- AP: clinical site staff (*e.g.*, RN or CRA) with data entry access to CTSU applications (*e.g.*, Roster Update Management System [RUMS], OPEN, Rave,),
- Associate (A): other clinical site staff involved in the conduct of NCI-sponsored trials, and
- Associate Basic (AB): individuals (*e.g.*, pharmaceutical company employees) with limited access to NCI-supported systems.

RCR requires the following registration documents:

Documentation Required	IVR	NPIVR	AP	A	AB
FDA Form 1572	✓	✓			
Financial Disclosure Form	✓	✓	✓		
NCI Biosketch (education, training, employment, license, and certification)	✓	✓	✓		
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 Cancer Trials Support Unit (CTSU) websites and applications. In addition, IVRs and NPIVRs must list all clinical practice sites and Institutional Review Boards (IRBs) covering their practice sites on the FDA Form 1572 in RCR to allow the following:

- Addition to a site roster,
- Assign the treating, credit, consenting, or drug shipment (IVR only) tasks in OPEN,
- Act as the site-protocol Principal Investigator (PI) on the IRB approval, and

- Assign the Clinical Investigator (CI) role on the Delegation of Tasks Log (DTL).

In addition, all investigators act as the Site-Protocol PI, consenting/treating/drug shipment, or as the CI on the DTL must be rostered at the enrolling site with a participating organization (*i.e.*, Alliance).

Additional information is located 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.

4.2 Site Registration

This study is supported by the NCI Cancer Trials Support Unit (CTSU).

Sites participating with the NCI Central Institutional Review Board (NCI CIRB) must submit the Study Specific Worksheet for Local Context (SSW) to the CIRB using IRBManager to indicate their intent to open the study locally. The NCI CIRB's approval of the SSW is automatically communicated to the CTSU Regulatory Office, but sites are required to contact the CTSU Regulatory Office at CTSURegPref@ctsu.coccg.org to establish site preferences for applying NCI CIRB approvals across their Signatory Network. Site preferences can be set at the network or protocol level. Questions about establishing site preferences can be addressed to the CTSU Regulatory Office by emailing the email address above or calling 1-888-651-CTSU (2878).

In addition, the Site-Protocol PI (*i.e.*, the investigator on the IRB/REB approval) must meet the following five criteria to complete processing of the IRB/REB approval record:

- Holds an Active CTEP status,
- Rostered at the site on the IRB/REB approval (*applies to US and Canadian sites only*) and on at least one participating roster,
- If using NCI CIRB, rostered on the NCI CIRB Signatory record,
- Includes the IRB number of the IRB providing approval in the Form FDA 1572 in the RCR profile, and
- Holds the appropriate CTEP registration type for the protocol.

Additional Requirements

Additional requirements to obtain an approved site registration status include:

- An active Federalwide Assurance (FWA) number,
- An active roster affiliation with the Lead Protocol Organization (LPO) or a Participating Organization, and
- Compliance with all protocol-specific requirements (PSRs).

4.2.1 Downloading Regulatory Documents

Site registration forms may be downloaded from the *10020* protocol page located on the CTSU Web site. Permission to view and download this protocol is restricted and is based

on person and site roster data housed in the CTSU RSS. To participate, Investigators and Associates must be associated with the Corresponding or Participating protocol organization in the RSS.

- Go to https://www.ctsu.org and log in 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, then select *LAO-MD017*, and protocol *#10200*
- Click on LPO Documents, select the Site Registration documents link, and download and complete the forms provided. (Note: For sites under the CIRB initiative, IRB data will load to RSS as described above.)

4.2.2 Requirements For 10200 Site Registration

- A Site initiation visit (SIV) is required for each participating site prior to activation. The local site PI must participate on the call as well as their research nurse, study coordinator, and pharmacist. To schedule a SIV, please email the Protocol Contact and reference the protocol in the subject line of the email.

- Training Requirement:
  - **All data entry users** (Clinical Research Associate role) at each participating site will need to complete the Theradex-led training.
  - Theradex will provide a certificate of completion, which will need to be submitted to the CTSU through the Regulatory Submission Portal.
  - The training is a one-time only requirement per individual. If an individual has previously completed the training for another ETCTN study, the training does not need to be completed again nor does the certificate of completion need to be resubmitted to the CTSU. However, new versions of the Specimen Tracking system may require new training.
  - This training will need to be completed before the first patient enrollment at a given site.
  - Peter Clark and Diana Vulih are the main points of contact at Theradex for the training (PClark@theradex.com and DVulih@theradex.com, Theradex phone: 609-799-7580).

This Study will use the ETCTN Specimen Tracking System (STS).

- All biospecimens collected for this trial must be submitted using the ETCTN Specimen Tracking System (STS) unless otherwise noted.
- The system is accessed through special Rave user roles: "CRA Specimen Tracking" for data entry at the treating institutions and "Biorepository" for users receiving the specimens for processing and storage at reference labs and the Biorepository.
- Please refer to the Medidata Account Activation and Study Invitation Acceptance link

on the CTSU website under the Rave/DQP tab.

- **Important: Failure to complete required fields in STS may result in a delay in sample processing.** Any case reimbursements associated with sample submissions will not be credited if samples requiring STS submission are not logged into STS.

4.2.3 Submitting Regulatory Documents

Submit required forms and documents to the CTSU Regulatory Office via the Regulatory Submission Portal on the CTSU website.

To access the Regulatory Submission Portal, log on to the CTSU members' website → Regulatory → Regulatory Submission.

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

4.2.4 Checking **Site** Registration Status

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

- Log on to the CTSU members' website
- Click on *Regulatory* at the top of your screen
- Click on *Site Registration*
- Enter your 5-character CTEP Institution Code and click on Go

Note: The status shown only reflects institutional compliance with site registration requirements as outlined above. 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.

4.3 Patient Registration

4.3.1 OPEN / IWRS

The Oncology Patient Enrollment Network (OPEN) is a web-based registration system available on a 24/7 basis. OPEN is integrated with CTSU regulatory and roster data and with the Lead Protocol Organization (LPOs) registration/randomization systems or Theradex Interactive Web Response System (IWRS) for retrieval of patient registration/randomization assignment. OPEN will populate the patient enrollment data in NCI's clinical data management system, Medidata Rave.

Requirements for OPEN access:

- A valid CTEP-IAM account.
- To perform enrollments or request slot reservations: Be on an LPO roster, ETCTN Corresponding roster, or Participating Organization roster with the role of Registrar. Registrars must hold a minimum of an AP registration type.
- If a DTL is required for the study, the registrar(s) must hold the OPEN Registrar task on the DTL for the site.
- Have an approved site registration for a protocol prior to patient enrollment.

To assign an Investigator (IVR) or Non-Physician Investigator (NPIVR) as the treating, crediting, consenting, drug shipment (IVR only), or receiving investigator for a patient transfer in OPEN, the IVR or NPIVR must list the IRB number used on the site's IRB approval on their Form FDA 1572 in RCR. If a DTL is required for the study, the IVR or NPIVR must be assigned the appropriate OPEN-related tasks on the DTL.

Prior to accessing OPEN, site staff should verify the following:

- Patient has met all eligibility criteria within the protocol stated timeframes, and
- 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.

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

4.3.2 OPEN/IWRS Questions?

Further instructional information on OPEN is provided on the OPEN tab 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.
Theradex has developed a Slot Reservations and Cohort Management User Guide, which is available on the Theradex website:
http://www.theradex.com/clinicalTechnologies/?National-Cancer-Institute-NCI-11.

This link to the Theradex website is also on the CTSU website OPEN tab.

For questions about the use of IWRS for slot reservations, contact the Theradex Helpdesk at 609-619-7862 or Theradex main number 609-799-7580;
CTMSSupport@theradex.com.

4.4 General Guidelines

Following registration, patients should begin protocol treatment as soon as possible. Issues that would cause treatment delays should be discussed with the Principal Investigator. If a patient does not receive protocol therapy following registration, the patient's registration on the study may be canceled. The Study Coordinator should be notified of cancellations as soon as possible.

5. BIOMARKER, CORRELATIVE, AND SPECIAL STUDIES

5.1 Biomarker Plan

Biomarker Name AND Lab PI and Site	**Assay (CLIA: Y/N)**	**Use (Integral, Integrated, or Exploratory) AND Purpose**	**Tissue/Body Fluid Tested and Timing of Sample Collection**	**M/O**	**Use of NCI Resources (No / Pending Approval / Approved)**	**Funding Source(s)**
11q23 rearrangement or partial tandem duplication Eytan Stein / Memorial Sloan-Kettering Cancer Center (MSKCC) Lab PI email:	Cytogenetics and FISH CLIA: Y **BRC Review: No**	Integral Evaluate eligibility	Bone marrow aspirate, peripheral blood Prior to enrollment and therapy cessation	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	CTEP CRADA
Myeloid mutational panel for NPM1 and no other targetable mutations Eytan Stein / Memorial Sloan-Kettering Cancer Center (MSKCC) Lab PI email:	Myeloid Mutations CLIA: Y **BRC Review: No**	Integral Evaluate eligibility	Bone marrow aspirate Prior to enrollment and therapy cessation	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	CTEP CRADA
Myeloid differentiation by Flow/FISH Eytan Stein / Memorial Sloan-Kettering Cancer Center (MSKCC) Lab PI email:	FACS, Flow cytometry, FISH CLIA: N **BRC Review: Yes** **Submission Deadline:**	Integrated Evaluate differentiation of cells with treatment	Bone marrow aspirate, peripheral blood End of cycle 1, end of cycle 3, and therapy cessation	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	CTEP CRADA
H3K79 ELISA Scott Armstrong / Dana-Farber Cancer Institute (DFCI) Lab PI email:	H3K79 ELISA CLIA: N **BRC Review: No**	Integrated Quantitate methylation	Bone marrow aspirate Prior to enrollment, end of cycle 1, end of cycle 3, and therapy cessation	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	CTEP CRADA
qPCR for HOXA9 and Meis1 Expression Scott Armstrong / Dana-Farber Cancer Institute (DFCI) Lab PI email:	qPCR CLIA: N **BRC Review: No**	Integrated Quantitate gene expression	Bone marrow aspirate Prior to enrollment, end of cycle 1, end of cycle 3, and therapy cessation	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	CTEP CRADA
MRD Negativity by Flow Cytometry Eytan Stein / Memorial Sloan-Kettering Cancer Center (MSKCC) Lab PI email:	Flow cytometry CLIA: Y **BRC Review: No**	Integrated Evaluate response to therapy	Bone marrow aspirate End of cycle 1, end of cycle 3, and therapy cessation	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	CTEP CRADA
Azacitidine PK profile Michelle A. Rudek / Johns Hopkins Lab PI email:	LC-MS/MS CLIA: N GLP: Y **BRC Review: No**	Integrated Pharmacokinetic profile of azacitidine alone	Blood with THU to stabilize azacitidine C1D1 pre, and 0.25, 0.5, and 1 hr post	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	UM1 for sample processing; CTEP / NIH / foundation grants for PK assessment
Cell storage / DNA storage Eytan Stein / Memorial Sloan-Kettering Cancer Center (MSKCC) Lab PI email:	Cell storage / DNA storage CLIA: N **BRC Review: No**	Exploratory Evaluate valence of methylation in future	Bone marrow aspirate, nail clippings Prior to enrollment, end of cycle 1, end of cycle 3, and therapy cessation; nail clippings only prior to enrollment	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	CTEP CRADA

Biomarker Name AND Lab PI and Site	**Assay (CLIA: Y/N)**	**Use (Integral, Integrated, or Exploratory) AND Purpose**	**Tissue/Body Fluid Tested and Timing of Sample Collection**	**M/O**	**Use of NCI Resources (No / Pending Approval / Approved)**	**Funding Source(s)**
5-aza-2'-deoxycytidine Genomic Incorporation (from azacitidine) and DNA demethylation Michelle A. Rudek / Johns Hopkins Lab PI email:	LC-MS/MS CLIA: N GLP: Y **BRC Review: No**	Exploratory Pharmacodynamic profile of azacitidine conversion to 5-aza-2'-deoxycytidine triphosphate and incorporation into DNA and its effect on DNA demethylation	PBMC and bone marrow aspirate PBMC: C1D8; Bone marrow aspirate: End of cycle 1, end of cycle 3, and therapy cessation	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	CTEP CRADA

The objective of the correlative studies is to better understand whether this combination is having the anticipated on-target effects and, if so, what the sequelae of those on-target effects may be.

Correlative studies will include morphology, flow cytometry, cytogenetics, and a myeloid genetics panel, all of which are standard of care. These will be performed on bone marrow biopsy samples taken at each time point - prior to therapy initiation and at the end of cycle 1 for the phase Ib, and prior to therapy initiation, end of cycle 1, end of cycle 3, end of cycle 6 and / or after therapy cessation for the phase II. Specimen collection for these assays will be mandatory. Integral biomarkers for identification of 11q23 / MLL rearrangements would be performed as part of these assessments. The assays will be performed at MSKCC or local CLIA-certified pathology laboratories, which have experience processing samples from patients with leukemia.

Additionally, we seek to study changes in methylation patterns with the use of this combination. This is important for understanding on-target effects as MLL-rearranged leukemia is driven by hypermethylation, and pinometostat works to inhibit DOT1L methyltransferase activity and 5-azacytidine is a DNA hypomethylating agent. The assay of choice would be an ELISA assay to quantitate the extent of H3K79 methylation in leukemia cells, performed on mandatory samples from bone marrow samples identified above. The ELISA would be performed in Dr. Scott Armstrong's lab at DFCI, which has extensive experience with this assay.

The ELISA would not be a CLIA-certified test and would be solely performed for research purposes as part of an integrated biomarker. Samples would be viably cryopreserved at each institution and shipped to the study coordinator MSKCC for storage. At the end of the study, the samples would be shipped to DFCI, care of Dr. Armstrong's laboratory, and the assay would be batch run on all samples.

As the mechanism of leukemogenesis in MLL-rearranged leukemia relies on hypermethylation of H3K79 by DOT1L, leading to increased transcription of HOXA9 and Meis1, we anticipate that expression levels of these two genes will decrease with treatment. As such, we plan to use qPCR to evaluate gene expression level prior to treatment on mandatory bone marrow samples at the time points identified above. The qPCR assays will be run in the Armstrong lab at DFCI, which has extensive experience running these qPCR assays. This would not be a CLIA-certified

test and would be solely performed for research purposes as an integrated biomarker. Samples would be viably cryopreserved at each institution and shipped to the study coordinator MSKCC for storage. At the end of the study, the samples would be shipped to DFCI, care of Dr. Armstrong's laboratory, and the assay would be batch run on all samples.

We anticipate that pinometostat will induce differentiation of myeloid blasts into more mature myeloid forms, based on previous research on pinometostat. As such, we will prospectively viably cryopreserve bone marrow aspirates in anticipation of flow-sorting blasts and myeloid cells. We will perform flow cytometry to assess for the upregulation of myeloid differentiation markers (such as CD11b) on each myeloid subtype, as well as perform FISH to track the compartmentalization of the MLL rearrangement. We anticipate that MLL rearrangements will be detectable in more mature myeloid cells, e.g. neutrophils, allowing us to evaluate the extent of differentiation.

We will perform these assays on selected samples. CD11B is part of the standard myeloid panel run by the CLIA-certified flow cytometry lab at MSKCC. Bone marrow samples from the above delineated timepoints will be collected and will be a mandatory part of the trial. Bone marrow aspirate samples will be centrifuged and the buffy coat will be viably cryopreserved in DMSO.

Bone marrow will be used to ensure that more blasts can be recovered and myeloid forms at different phases of maturation can be included. While we could consider assays to evaluate for synergy of these two agents, this falls outside the scope of this phase I study – we would need reference evaluations of patient treatment on each agent alone, which is not the goal of this trial. Additionally, in vitro assays of synergy have previously been performed in pre-clinical work. Western blots for H3K79 methylation were considered, however we would perform the ELISA first as it provides more throughput with a single assay.

We also considered running ChIP-seq for H3K27 methylation (as these histones may be mono-, di-, or trimethylated, and each of these is mediated by DOT1L). Pinometostat's inhibiton of DOT1L may lead to an accumulation of non-methylated or hypomethylated forms of H3K79 and ChIP-seq can help measure the valence of that histone methylation, as the amount of methylation has previously been described to correlate to the robustness of gene expression. However, the use of this assay would be limited by cost. As such, we could consider storing chromatin cross-linked in formaldehyde or freezing down viable cells. If we were to choose to run the assay in future, we would then shear the DNA to obtain nucleosomal DNA and run the assay at that time.

Cell storage / DNA storage will be performed on both bone marrow samples and nail clippings, the latter to be used for comparison to germline DNA so that relevant acquired mutations will be easier to detect.

The analysis of azacitidine drug concentration in plasma would be performed by Dr. Michelle A. Rudek, PhD, PharmD, an Associate Professor of Oncology and the director of the Sidney Kimmel Comprehensive Cancer Center at Johns Hopkins (SKCCC) Analytical Pharmacology Core. She has expertise in bioanalysis, drug metabolism, and pharmacokinetic and pharmacodynamic analysis. The core operates according to Good Laboratory Practices (GLP) and would analyze the samples from cycle 1, day 1 taken prior to therapy administration and at

15 min, 30 min, and 60 min after administration for both the phase Ib and phase II portions of the trial. This analysis would involve liquid chromatography coupled with tandem mass spectrometry (LC-MS/MS) and would be performed on mandatory peripheral blood samples which will be cryopreserved at the local site and shipped to Dr. Rudek's lab at SKCCC.

Determining azacitidine incorporation and DNA methylation would be an exploratory biomarker, also performed by the SKCCC Analytical Pharmacology Core. This would allow for the assessment of 5-aza-2`-deoxycytidine incorporation into genomic DNA and the extent of global DNA methylation. Decitabine, 2'-deoxycytidine, and 5-methyl-2'-deoxycytidine levels from digested DNA will be measured by LC-MS/MS in the DNA from bone marrow samples taken at the end of cycle 1, the end of cycle 3, and at the end of treatment for both the phase Ib and phase II portions of the trial. The analytical assay has been developed and validated by the SKCCC Analytical Pharmacology Core.

This will be one of the first assessments of 5-aza-2`-deoxycytidine incorporation relative to the extent of global DNA methylation in human samples and may give further insight into the mechanism of action of azacitidine.

Correlative studies will be performed at the intervals outlined in the table so as to produce meaningful and interpretable results. As sample collection at these intervals is commonly performed as standard of care, these timepoints should be feasible for assessment.

Specimen Collection Schedule

Specimen Type	**Baseline (Pre-treatment)**	***C1D1 prior to azacitidine, and 0.25hr, 0.5hr, and 1 hr post azacytidine treatment***	***C1D8***	***End of cycle 1***	***End of cycle 3***	***End of cycle 6 and / after cessation of therapy, whichever is first***
Bone marrow	X			X	X	X
Peripheral Blood	X	X	X			

5.2 Integral Laboratory or Imaging Studies

5.2.1.1 *11q23 rearrangement or partial tandem duplication*
5.2.1.2 Collection of Specimen(s): Locally performed
5.2.1.3 Handling of Specimens(s): Locally performed
5.2.1.4 Shipping of Specimen(s): N/A
5.2.1.5 Site(s) Performing Correlative Study: Local laboratory or per local practice of using a commercial laboratory

5.2.1.6 *Myeloid mutational panel for NPM1 and no other targetable mutations*
5.2.1.7 Collection of Specimen(s): Locally performed
5.2.1.8 Handling of Specimens(s): Locally performed
5.2.1.9 Shipping of Specimen(s): N/A
5.2.1.10 Site(s) Performing Correlative Study: Local laboratory or per local practice of using a commercial laboratory

5.3 Integrated Correlative Studies

5.3.1.1 *H3K79 ELISA*
5.3.1.2 Collection of Specimen(s): Locally performed
5.3.1.3 Handling of Specimens(s): Local study coordinator
5.3.1.4 Shipping of Specimen(s): To Armstrong Lab at DFCI
5.3.1.5 Site(s) Performing Correlative Study: Armstrong Lab at DFCI

5.3.1.6 *qPCR for HOXA9 and Meis1 Expression*
5.3.1.7 Collection of Specimen(s): Locally performed
5.3.1.8 Handling of Specimens(s): Local study coordinator
5.3.1.9 Shipping of Specimen(s): To Armstrong Lab at DFCI
5.3.1.10 Site(s) Performing Correlative Study: Armstrong Lab at DFCI

5.3.1.11 *Myeloid differentiation by Flow / FISH*
5.3.1.12 Collection of Specimen(s): Locally performed
5.3.1.13 Handling of Specimens(s): Local study coordinator
5.3.1.14 Shipping of Specimen(s): To MSKCC
5.3.1.15 Site(s) Performing Correlative Study: MSKCC

5.3.1.16 *MRD Negativity by Flow Cytometry*
5.3.1.17 Collection of Specimen(s): Locally performed
5.3.1.18 Handling of Specimens(s): Local study coordinator
5.3.1.19 Shipping of Specimen(s): To MSKCC
5.3.1.20 Site(s) Performing Correlative Study: MSKCC

5.3.1.21 *Azacitidine PK profile*
5.3.1.22 Collection of Specimen(s): Locally performed
5.3.1.23 Handling of Specimens(s): Local study coordinator
5.3.1.24 Shipping of Specimen(s): To Rudek lab at SKCCC

5.3.1.25 Site(s) Performing Correlative Study: Rudek lab at SKCCC

5.4 Exploratory/Ancillary Correlative Studies

5.4.1.1 *Cell storage / DNA storage*
5.4.1.2 Collection of Specimen(s): Locally performed
5.4.1.3 Handling of Specimens(s): Local study coordinator
5.4.1.4 Shipping of Specimen(s): To MSKCC
5.4.1.5 Site(s) Performing Correlative Study: MSKCC

5.4.1.6 *5-aza-2'-deoxycytidine Genomic Incorporation (from Azacitidine) and DNA Demethylation*
5.4.1.7 Collection of Specimen(s): Locally performed
5.4.1.8 Handling of Specimens(s): Local study coordinator
5.4.1.9 Shipping of Specimen(s): To Rudek lab at SKCCC
5.4.1.10 Site(s) Performing Correlative Study: Rudek lab at SKCCC

5.5 Special Studies

5.5.1.1.1 N/A

6. TREATMENT PLAN

6.1 Agent Administration

Treatment will be administered on an outpatient basis. Reported adverse events and potential risks are described in Section 10. Appropriate dose modifications are described in Section 7. Hydroxyurea may be given concurrently for cytoreduction. No other investigational or commercial agents or therapies other than those described below may be administered with the intent to treat the patient's malignancy.

Dose Escalation Schedule		
Dose Level	**Dose**	
	Pinometostat	**Azacitidine**
Level -1	36 mg/m^2/day CIV infusion	75 mg/m^2/day for 7 days / cycle
Level 1*	54 mg/m^2/day CIV infusion	75 mg/m^2/day for 7 days / cycle
Level 2	90 mg/m^2/day CIV infusion	75 mg/m^2/day for 7 days / cycle
**Starting dose levels*		

Regimen Description					
Agent	***Premedications; Precautions***	***Dose***	***Route***	***Schedule***	***Cycle Length***
Pinometostat	*None*	*** in NS to a final concentration between 0.54 mg/ml and 5.63 mg/ml*	*Continuous IV infusion*	*Days 1-28*	*28 days (4 weeks)*
Azacitidine	*Consider antiemetic premedication as per local institutional practicess, e.g. 8mg ondansetron 60min prior to administration*	*75 mg/m^2/day*	*IV or SC, as per local institutional preference; given daily for 7 of the first 10 days of each cycle, e.g. given for 5 days, 2-day break, and given for an additional 2 days*	*Given for 7 of the first 10 days of each cycle*	
***Doses as appropriate for assigned dose level.*					

6.1.1 CTEP IND Agent: Pinometostat

Pinometostat is given as IV continuous infusion administered at a constant flow rate. Pinometostat must be diluted in 0.9% sodium chloride to a final concentration between 0.54 mg/ml and 5.63 mg/ml prior to administration.

Patients should have a peripherally inserted central catheter (PICC) or similar central venous access device with a dedicated lumen for pinometostat alone.

Pinometostat will be administered as a continuous IV infusion through the dedicated lumen of the central venous access device. The patient will return to the study center approximately every 72 hours to have the infusion medication refilled. The use of pumps to administer the medication should follow manufacturer's and institutional guidelines. The patient or caregiver should be trained on how to use the pump or other device used to administer the medication. The patient or caregiver should also be trained on signs that may indicate that there is an issue with drug delivery.

Refer to section 8.1 for compatible IV infusion bags and sets.
The starting dose of pinometostat in the phase 1b will be 54 mg/m^2/day (actual body surface area (BSA)). BSA is calculated as per institutional policy; alternatively, the Du Bois Method (Arch Intern Med. 1916; 17:863-871) may be used.

BSA should be calculated prior to each cycle. The administered dose should be adjusted if the patient's weight changes $\geq$ 10% from baseline and may be rounded to the nearest mg.

6.1.2 Other Agent(s)

Prophylactic antiemetics can be given prior to azacitidine administration as per local institutional practices. One example of this would be the administration of 8mg ondansetron 60 min prior to azacitidine administration.

6.1.3 Other Modality(ies) or Procedures

N/A

6.2 Definition of Dose-Limiting Toxicity

Definition of dose-limiting toxicities, as well as guidelines for DLT management and dose modifications are outlined in Section 7.

Dose escalation will proceed within each cohort according to the following scheme.

Number of Patients with DLT at a Given Dose Level	Escalation Decision Rule
0 out of 3	Enter 3 patients at the next dose level.
≥2	Dose escalation will be stopped. This dose level will be declared the maximally administered dose (highest dose administered). Three (3) additional patients will be entered at the next lowest dose level if only 3 patients were treated previously at that dose.
1 out of 3	Enter at least 3 more patients at this dose level. • If 0 of these 3 patients experience DLT, proceed to the next dose level. • If 1 or more of this group suffer DLT, then dose escalation is stopped, and this dose is declared the maximally administered dose. Three (3) additional patients will be entered at the next lowest dose level if only 3 patients were treated previously at that dose.
≤1 out of 6 at highest dose level below the maximally administered dose	This is generally the recommended phase 2 dose. At least 6 patients must be entered at the recommended phase 2 dose.

6.3 Dose Expansion Cohorts:

This study will be performed as a phase Ib / II. No expansion cohort will be studied; instead, patients will be added to the phase II study and treated at the RP2D. Monitoring of all safety and toxicity data is done by the Principal Investigator and the Corresponding Organization on a real-time basis as data are entered into Medidata Rave using the Web Reporting Module. All participating sites are expected to notify the Principal Investigator when a DLT has occurred.

6.4 General Concomitant Medication and Supportive Care Guidelines

Patients who are neutropenic should be given appropriate prophylaxis as per the local institutional practice.

Pinometostat is metabolized by CYP3A and is a mild, time-dependent inhibitor of CYP3A. Those patients receiving medications metabolized by CYP3A with narrow therapeutic windows should be monitored carefully while receiving pinometostat. All strong CYP3A4 inhibitors or inducers, including azole antifungals that are strong CYP3A4 inhibitors, are excluded from use in these patients.

Pinometostat is metabolized by CYP450 and patients receiving any medications or substances that are inhibitors or inducers of the CYP450 system should have their medications reviewed and adjusted for interactions as appropriate for local institutional practice.

Medications metabolized by CYP3A with narrow therapeutic windows should be used with caution, as EPZ-5676 is potentially a mild, time-dependent inhibitor of CYP3A. The use or initiation of strong CYP3A4 substrates, inhibitors or inducers will be considered on a case-by-

case basis (based on risk-benefit) only after discussion with the Medical Monitor, and only when no alternative medication is possible.

An updated listing of CYP substrates, inhibitors, and inducers can be found at http://medicine.iupui.edu/clinpharm/ddis/table.aspx.

Pinometostat is also metabolized to a minor extent by CYP2C19. It is a substrate of MATE2K and likely a weak substrate for OATP1B3, MATE1, and P-gp. It is also an inhibitor of MATE1 and MATE2K. Use caution when administered with strong inhibitors and inducers of CYP2C19, OATP1B3, P-gp, MATE1, and MATE2K. Use caution with concomitant use of other CYP3A and MATE1 and MATE2-K substrates, especially those with a narrow therapeutic window.

The use of hydroxyurea in patients with acute myeloid leukemia, for control of circulating blast count, is allowed to be given concurrently.

Supportive care measures and symptomatic treatment for any drug-related toxicity may be instituted if clinically indicated. Intermittent use of dexamethasone (10 mg) is permitted as an antiemetic, though chronic steroid use should be discouraged as steroids may be regarded as anti-cancer treatment.

The prophylactic use of hematopoietic colony stimulating factors is prohibited. The therapeutic use of hematopoietic colony stimulating factors should be conducted according to the 2015 ASCO Guideline for use of white blood cell growth factors[19]. Blood transfusions may also be administered as needed per the judgment of the Investigator.

Because there is a potential for interaction of pinometostat with other concomitantly administered drugs, the case report form must capture the concurrent use of all other drugs, over-the-counter medications, or alternative therapies. The Principal Investigator should be alerted if the patient is taking any agent known to affect or with the potential for drug interactions. The study team should check a frequently-updated medical reference for a list of drugs to avoid or minimize use of. Appendix C (Patient Drug Information Handout and Wallet Card) should be provided to patients if available.

6.5 Duration of Therapy

In the absence of treatment delays due to adverse event(s), treatment may continue for 6 cycles. Patients may continue on the study combination if they are deriving clinical benefit. Patients will be taken off the study if one of the following criteria applies:

- Disease progression
- Intercurrent illness that prevents further administration of treatment
- Unacceptable adverse event(s)
- Patient decides to withdraw from the study

- General or specific changes in the patient's condition render the patient unacceptable for further treatment in the judgment of the investigator
- Clinical progression
- Patient non-compliance
- Pregnancy
    - All women of child bearing potential should be instructed to contact the investigator immediately if they suspect they might be pregnant (*e.g.*, missed or late menstrual period) at any time during study participation.
    - The investigator must immediately notify CTEP in the event of a confirmed pregnancy in a patient participating in the study.
- Termination of the study by sponsor
- The drug manufacturer can no longer provide the study agent

The reason(s) for protocol therapy discontinuation, the reason(s) for study removal, and the corresponding dates must be documented in the Case Report Form (CRF).

6.6 Duration of Follow Up

Patients will be followed for 1 month after removal from study or until death, whichever occurs first. Patients removed from study for unacceptable adverse event(s) will be followed until resolution or stabilization of the adverse event.

7. DOSING DELAYS/DOSE MODIFICATIONS

The period of assessment for a dose-limiting toxicity (DLT) will be the first cycle of the phase 1B study.

The starting dose of pinometostat in the phase 1b will be 54 mg/m^2/day (actual BSA). The dose will be escalated in two successive cohorts of patients who will be entered sequentially to each dose level (54 mg/m^2/day or 90 mg/m^2/day). Dose escalation will be performed until ≥Grade 2 (National Cancer Institute (NCI) Common Terminology Criteria for Adverse Events (CTCAE) version 5.0) drug-related (defined as: any AE which cannot be clearly attributed by the Investigator to another cause, such as intercurrent illness or concomitant medication) toxicity in any patient occurs. Dose escalation will stop when the dose reaches 90 mg/m^2/day and no further doses will be tested. If the maximum tolerated dose (MTD) is not reached, a recommended phase 2 dose (RP2D) may be used in its place.

Three patients will be assigned to the cohort at which any ≥Grade 2 drug-related toxicity occurred and dose escalation will proceed according to the common 3+3 design. If none of the first 3 patients at the 54 mg/m^2/day dose experience first cycle dose limiting toxicity (DLT; see DLT definition below), new patients may be entered at the next higher dose level. If 1 of 3 patients experience a first cycle DLT, up to 3 more patients will be started at that same dose level. If 2 or more experience first cycle DLT, no further patients are started at that dose and a lower dose level of 36 mg/m2/day may be tried.

The MTD is defined as the dose level below which >1 patient out of 3 or ≥2 patients out of 6 experience a DLT. The second dose level may begin accrual only if all patients at the first dose level have been observed for a minimum of 28 days, i.e. through the first 28-day cycle. However, if at Day 28 the patient has Grade 3 or 4 neutropenia or thrombocytopenia, (not attributable to persistent leukemia; see DLT definition below), the patient will not begin Cycle 2 and must be observed until Day 42 in order to determine whether a DLT has occurred. In this instance, dose escalation cannot proceed until at least Day 42 so that resolution may be assessed. If patients do not fulfill the definition of severe myelosuppression at Day 28, or if they recover between Days 28 and 42, no hematological DLT will be deemed to have occurred. Patients may then proceed to cycle 2 for further treatment of persistent disease or in the setting of a response.

Once the MTD is reached, the cohort at the MTD will be expanded in the phase II portion of the trial.

A DLT will be defined as a significant suspected adverse reaction or clinically significant abnormal laboratory value assessed as unrelated to disease progression, intercurrent illness, or concomitant medications (only AEs that are incontrovertibly due to an extraneous cause will be excluded as a DLT) and of onset within the first 28 days on study that meets any of the following criteria:

- Any Grade 5 toxicity
- Grade 4 neutropenia lasting ≥ 42 days from start of cycle in absence of evidence of active leukemia

- Any Grade ≥ 3 non-hematologic toxicity not clearly resulting from the underlying leukemia EXCEPT:
    - Alopecia
    - Grade 3 fatigue, asthenia, fever, anorexia, or constipation
    - Grade 3 nausea, vomiting, or diarrhea not requiring tube feeding, total parenteral nutrition, or requiring or prolonging hospitalization
    - Infection, bleeding, or other expected direct complication of cytopenias due to active underlying leukemia
    - Grade 3 infusion reaction including cytokine release syndrome, if successfully managed and which resolves within 72 hours
    - Grade 3 or 4 tumor lysis syndrome if it is successfully managed clinically and resolves within 7 days without end-organ damage
    - Grade 3 or 4 isolated electrolyte abnormalities (i.e., those occurring without clinical consequence) that resolve, with or without intervention, to less than Grade 2 in less than 72 hours will not be considered DLT

The above criteria should also be used for AEs for the phase 2 part of the trial. Safety will be assessed by AEs, SAEs, vital signs, physical examinations, and review of biochemistry and hematology laboratory values.

After completion of the first cycle / DLT-assessment period, further toxicities will be managed as for patients treated on the phase 2 portion of the study. Patients who do not achieve a CR or PR after the first cycle of therapy may continue therapy, with the duration of therapy determined as outlined in section 6.5.

During the phase 2, guidance on how to manage common non-hematologic toxicities is given below. If a patient tolerates pinometostat at the MTD and azacitidine at 75 mg/m^2 but has a hematologic toxicity, the next cycle of azacitidine administration may be delayed out to day 42 while the pinometostat is continued. If the most recent bone marrow on treatment shows persistent leukemia, the patient may continue to be treated with combination therapy with supportive care given for cytopenias.

After discussion with the medical monitor, a lower dose of pinometostat therapy may be considered after the DLT period in the event of toxicities. The dose of azacitidine should remain constant at 75mg/m^2 daily for 7 days given over a 10-day period at the start of each cycle.

If recurrent hematologic toxicity occurs in someone who has achieved a remission, alternative dosing schedules of pinometostat could be explored as follows:

If the recommended phase II dose is 90 mg/m^2/day

Dose Level	Pinometostat Dose
-1	54 mg/m^2/day CIV infusion
0	90 mg/m^2/day CIV infusion

If the recommended phase II dose is 54 mg/m^2/day

Dose Level	Pinometostat Dose
-1	36 mg/m^2/day CIV infusion
0	54 mg/m^2/day CIV infusion

Azacitidine administration may be held until up to day 42 of the cycle, but the dosage should not be reduced.

Indications for dose modifications of pinometostat are as follows. For specific concerns not outlined here, please contact the medical monitor.

Nausea	**Management/Next Dose for Pinometostat**
≤ Grade 1	Continue administration, provide supportive care, monitor
Grade 2	Continue administration, provide supportive care, monitor
Grade 3	Provide supportive care. If tube feeding or TPN are required due to nausea and there is no improvement to ≤ Grade 2 after three days of supportive treatment as per local institutional practice, hold pinometostat until nausea has resolved to ≤ Grade 2. Pinometostat should restarted at one dose level lower.
Grade 4	N/A
*Patients requiring a delay of >2 weeks should go off protocol therapy. **Patients requiring > one dose reductions should go off protocol therapy.	
Recommended management: antiemetics.	

Vomiting	**Management/Next Dose for Pinometostat**
≤ Grade 1	Continue administration, provide supportive care, monitor
Grade 2	Continue administration, provide supportive care, monitor
Grade 3	Provide supportive care. If tube feeding or TPN are required due to vomiting and there is no improvement to ≤ Grade 2 after three days of supportive treatment as per local institutional practice, hold pinometostat until vomiting has resolved to ≤ Grade 2. Pinometostat should restarted at one dose level lower.
Grade 4	Hold therapy until vomiting has resolved to ≤ Grade 2, restart at lower dose if appropriate**
*Patients requiring a delay of >2 weeks should go off protocol therapy. **Patients requiring > 1 dose reductions should go off protocol therapy.	
Recommended management: antiemetics.	

Diarrhea	**Management/Next Dose for Pinometostat**
≤ Grade 1	Continue administration, provide supportive care, monitor
Grade 2	Continue administration, provide supportive care, monitor
Grade 3	If ongoing x 48h, hold therapy until ≤ Grade 2, restart at same dose.

Diarrhea	**Management/Next Dose for Pinometostat**
	Pinometostat should restarted at one dose level lower*.
Grade 4	Hold therapy until ≤ Grade 2, restart at lower dose if appropriate**
Recommended management: Loperamide antidiarrheal therapy Dosage schedule: Adults- 4 mg at first onset, followed by 2 mg with each loose motion until diarrhea-free for 12 hours (maximum dosage: 16 mg/24 hours) Children – 2mg TID dosing may be used for children who are small for their chronological age but are 30kg or greater Adjunct anti-diarrheal therapy is permitted and should be recorded when used. *Patients requiring a delay of >2 weeks should go off protocol therapy. **Patients requiring > one dose reduction should go off protocol therapy.	

Fever	**Management/Next Dose for Pinometostat**
≤ Grade 1	Continue administration, monitor
Grade 2	Continue administration, monitor
Grade 3	Hold pinometostat until fever has resolved. Pinometostat should restarted at one dose level lower.
Grade 4	Hold pinometostat until fever has resolved. Pinometostat should restarted at one dose level lower.
Recommended management: antimicrobial or supportive therapy as appropriate based on clinical evaluation	

Anemia	**Management/Next Dose for Pinometostat**
≤ Grade 1	Continue administration, monitor
Grade 2	Continue administration, monitor
Grade 3	Continue administration, monitor
Grade 4	If anemia thought to be related to underlying leukemia, continue administration. If anemia thought to be related to drug effect and lasts ≥ 14 days, hold pinometostat until anemia has resolved to ≤ Grade 1 and restart at dose level -1*
Anemia is anticipated with treatment. *Patients requiring > one dose reduction should go off protocol therapy.	

Neutropenia	**Management/Next Dose for Pinometostat**
≤ Grade 1	Continue administration, monitor
Grade 2	Continue administration, monitor
Grade 3	Continue administration, monitor
Grade 4	If neutropenia is thought to be related to underlying leukemia, continue administration. If neutropenia is thought to be related to drug effect and lasts ≥ 14 days, hold pinometostat until neutropenia has resolved to ≤ Grade 1 and restart at dose level -1*
Neutropenia is anticipated with treatment. Persistent grade 4 neutropenia (lasting ≥ 14 days) would necessitate dose de-escalation. Fever with neutropenia necessitating ICU admission should be discussed with the medical monitor. *Patients requiring > one dose reduction should go off protocol therapy.	

Thrombocytopenia	**Management/Next Dose for Pinometostat**
≤ Grade 1	Continue administration, monitor
Grade 2	Continue administration, monitor
Grade 3	Continue administration, monitor
Grade 4	If thrombocytopenia is thought to be related to underlying leukemia, continue administration. If thrombocytopenia is thought to be related to drug effect and lasts ≥ 14 days, hold pinometostat until thrombocytopenia has resolved to ≤ Grade 1 and restart at dose level -1*
Thrombocytopenia is anticipated with treatment. *Patients requiring > one dose reduction should go off protocol therapy.	

Bone Marrow Hypocellular	**Management/Next Dose for Pinometostat**	**Management/Next Dose for Azacitidine**
≤ Grade 1	Continue administration, monitor	Continue administration, monitor
Grade 2	Continue administration, monitor	Continue administration, monitor
Grade 3	Continue administration, monitor	If hypocellular with no morphologic evidence of leukemia, then hold azacitidine for 2 weeks and repeat a bone marrow biopsy
Grade 4	If persistent aplasia with no morphologic evidence of leukemia after two week azacitidine hold, hold pinometostat for 2 weeks and repeat a bone marrow biopsy	
Inform medical monitor of grade 4 toxicity.		

7.1 Guidelines for Management of QT Prolongation

The discussion of the emergency management of torsade de pointes and its hemodynamic consequences is beyond the scope of this guideline.

Both nonclinical and clinical data provided suggest a risk of QT prolongation with pinometostat, and the risk may be additive in combination with azacitidine. As such, a baseline EKG should be performed to evaluate for QT prolongation. A weekly EKG should be performed during cycle 1 to monitor for QT prolongation.

Subjects may be at increased risk for the development of QT prolongation when treated with pinometostat and azacitidine in combination with fluoroquinolones, azole antifungal agents or serotonin (5-HT3) antagonists. Investigators need to be vigilant; refrain from administering concomitant medications associated with QT prolongation and if no other therapeutic options are available, monitor subjects receiving pinometostat and azacitidine with the combination of these drugs, and evaluate EKG and electrolytes (including potassium, magnesium, and calcium) particularly in subjects presenting with nausea, vomiting or diarrhea.

Subjects who experience prolongation of the QTc interval using Fridericia's equation (QTcF) interval to > 480 msec (CTCAE Grade ≥ 2) while treated with pinometostat and azacitidine should be promptly evaluated for causality of the QTcF prolongation and managed according to the following guidelines:

- Levels of electrolytes (potassium, calcium and magnesium) should be checked and supplementation given to correct any values outside the normal range.

- Concomitant therapies should be reviewed and adjusted as appropriate for medication with known QT prolonging effects.

- If no other cause is identified and the Investigator believes it is appropriate, particularly if QTcF remains elevated (after above measures have been implemented, or as determined by the Investigator), study drug may be interrupted, and an EKG should be rechecked in approximately 1 week after the QTcF prolongation was first observed or more frequently as clinically indicated. If QTcF has recovered or improved and the Investigator believes it is safe to do so, re-challenge with pinometostat should be considered if held.

- EKGs should be conducted at least weekly (e.g., at every scheduled visit) for 2 weeks following QTcF reduction ≤ 480 msec.

Grade 2 (QTcF > 480 and ≤ 500 msec)

- The dose of pinometostat may be reduced to a dose approved by the Medical Monitor without interruption of dosing. The pinometostat dose may be re-escalated to the prior dose in ≥ 14 days after QT prolongation has decreased to ≤ Grade 1.

Grade 3 (QTcF > 500 msec)

- Hospitalization for continuous cardiac monitoring and evaluation by a cardiologist should both be considered.

- Dosing with pinometostat will be interrupted. If QTcF returns to within 30 msec of baseline or < 450 msec within 14 days, treatment may be resumed at a reduced dose

- The pinometostat dose cannot be re-escalated following dose reduction for Grade 3 QTcF prolongation unless the prolongation was associated with an electrolyte abnormality or concomitant medication

Grade 4 (QTcF > 500 msec or > 60 msec change from baseline with torsade de pointes or polymorphic ventricular tachycardia or signs/symptoms of serious arrhythmia)

- Subjects should be admitted to hospital for continuous cardiac monitoring and discharged only after review by a cardiologist.

- Dosing with pinometostat and azacitidine should be permanently discontinued.

7.2 Tumor Lysis Syndrome

Patients should be monitored for tumor lysis syndrome (TLS), which may be most likely to occur at the time of therapy initiation in patients with high tumor burden. TLS should be defined by the Howard criteria, with laboratory TLS defined by uric acid > 8.0mg/dL, phosphorus > 4.5 mg/dL, potassium > 6.0 mmol/L, and corrected calcium < 7.0 mg/dL with the presence of at least two abnormalities present during the same 24-hour period within 3 days prior to the start of therapy or 7 days afterward. Clinical tumor lysis syndrome is defined by car[20]diac dysrhythmia, sudden death, seizure, neuromuscular irritability, hypotension, heart failure, increasing creatinine of 0.3 mg/dL or a single value of 1.5 times the ULN, or oliguria (< 0.5 ml/kg/hr over 6 hours on average). See reference for further details.

Patients may receive prophylaxis and treatment for tumor lysis syndrome, including hydration, allopurinol, or rasburicase, as is appropriate per local institutional guidelines.

7.3 Differentiation Syndrome

Symptoms of differentiation syndrome may include fever, weight gain or edema, respiratory symptoms with or without infiltrates, pleural or pericardial effusions, hypotension, and acute renal failure. If differentiation syndrome is clinically suspected, initiate prompt administration of corticosteroids at a suggested dose of 10 mg dexamethasone IV or PO every 12 hours until resolution of symptoms and signs as well as hemodynamic monitoring, for a minimum of 3 days is recommended, until improvement. Hydroxyurea at a suggested dose of 1–4 g PO twice or three times per day may be used for subjects with leukocytosis in the setting of differentiation syndrome. Additionally, initiation of furosemide and/or prompt initiation of leukapheresis is recommended if clinically required. For all cases of suspected differentiation syndrome, pinometostat should be immediately held.

8. PHARMACEUTICAL INFORMATION

A list of the adverse events and potential risks associated with the investigational or commercial agents administered in this study can be found in Section 10.1.

8.1 CTEP IND Agent(s)

8.1.1 CTEP IND Agent #1 - Pinometostat (NSC #795144)

Chemical Name or Amino Acid Sequence: (2R,3R,4S,5R)-2-(6-amino-9H-purin-9-yl)-5-((((1r,3S)-3-(2-(5-(tert-butyl)-1H-benzo[d]imidazol-2-yl)ethyl)cyclobutyl)(isopropyl) amino)methyl)tetrahydrofuran-3,4-diol

Other Names: EPZ-5676, EPZ005676

Classification: DOT1L inhibitor

CAS Registry Number: 1380288-87-8

Molecular Formula: $C_{30}H_{42}N_8O_3$ **M.W.:** 562.71 g/mol

Mode of Action: Pinometostat is a selective human DOT1-like, histone H3 methyltransferase (DOT1L) inhibitor. Pinometostat exhibits concentration and time depending inhibition of global methylation of lysine 79 of histone H3 (H3K79) in cultured cells. Pinometostat induces apoptosis in cells that accumulate in the G1/G0 phase. It also inhibits the expression of the key MLL fusion target genes HOXA9 and MEIS1.

Description: A clear to yellow liquid adjusted to a pH of 5.2-6.2 with 1N sodium hydroxide or 1N hydrochloric acid as needed.

How Supplied: Pinometostat is supplied by Epizyme, Inc. and distributed by the Pharmaceutical Management Branch, CTEP/DCTD/NCI. Pinometostat injection is provided as 100 mg/10 mL (10 mg/ml) solution in type 1 borosilicate glass serum vials closed with butyl rubber stoppers and aluminum overseals. The other components of the vial include hydroxypropyl betadex, citric acid, anhydrous, sodium hydroxide, hydrochloric acid, and water for injection.

Preparation: Pinometostat must be diluted with 0.9% sodium chloride injection, USP prior to administration to a final concentration between 0.54 mg/ml and 5.63 mg/ml. Add the calculated dose of pinometostat to the appropriate volume of 0.9% sodium chloride in a non-DEHP PVC, polyolefin, or EVA IV infusion bag. ***Note: mixing of different drug lots in the same infusion bag should be avoided.*** Then gently agitate the infusion bag to ensure mixing. Up to a 90-hour dose may be prepared. An overfill volume of 10% or per institution policy can be prepared to allow for priming of the infusion set tubing. Sites should prepare the appropriate final infusion volume according to standard practice based on a constant flow rate.

Storage: Store at room temperature between 15 °C -30 °C.

If a storage temperature excursion is identified, promptly return pinometostat to 15 °C -30 °C and quarantine the supplies. Provide a detailed report of the excursion (including documentation of temperature monitoring and duration of the excursion) to PMBAfterHours@mail.nih.gov for determination of suitability.

Stability: Stability studies are ongoing. The intravenous infusion is stable at room temperature and can be infused for a maximum of 90 hours. The infusion bag can be stored at room temperature prior to infusion; the total time in bag, including both preparation and infusion time, should not exceed 114 hours total. The infusion must be completed within 114 hours after infusion bag preparation.

Route and Method of Administration: Continuous, intravenous infusion via central venous catheter with a pump for up to 90 hours at a constant flow rate. Compatible IV infusion sets are non-DEHP PVC, polyolefin, or EVA with or without a 0.2-micron filter.

Potential Drug Interactions: Pinometostat is metabolized in vitro predominantly by CYP3A with a minor contribution from CYP2C19. It is also a substrate for MATE2-K and likely a weak substrate for OATP1B3, MATE1, and P-gp. Pinometostat is not a substrate for OCT1, OCT2, OATP1B1 or BCRP. Use of strong CYP3A inhibitors and inducers should be avoided. Use caution when administered with strong inhibitors and inducers of CYP2C19, OATP1B3, P-gp, MATE1, and MATE2-K.

Pinometostat is a weak to moderate inhibitor of CYP3A4/5 in vitro. Pinometostat is an inhibitor of MATE1 and MATE2-K in vitro but not CYP1A2, 2B6, 2C8, 2C9, 2C19, 2D6, P-gp, BCRP, OAT1, OAT3, OCT1, OCT2, OATP1B1, OATP1B3 or BSEP. Use caution with concomitant use of other CYP3A and MATE1 and MATE2-K substrates, especially those with a narrow therapeutic window.

Pinometostat is not an inducer of CYP1A2, 2B6, or 3A4/5 activity or mRNA levels.

Patient Care Implications: Women of child-bearing potential (WOCP) must use acceptable contraceptives while on pinometostat and for 4 weeks after the last dose of pinometostat. Males with partners that are WOCP must use acceptable methods of contraception while on pinometostat and for 90 days after the last dose of pinometostat. It is unknown if pinometostat is excreted in breast milk so should not be administered to nursing mothers.

Availability

Pinometostat is an investigational agent supplied to investigators by the Division of Cancer Treatment and Diagnosis (DCTD), NCI.

Pinometostat is provided to the NCI under a Collaborative Agreement between the

Pharmaceutical Collaborator and the DCTD, NCI (see Section 13.4).

8.1.2 Agent Ordering and Agent Accountability

8.1.2.1 NCI-supplied agents may be requested by eligible participating Investigators (or their authorized designee) at each participating institution. The CTEP-assigned protocol number must be used for ordering all CTEP-supplied investigational agents. The eligible participating investigators at each participating institution must be registered with CTEP, DCTD through an annual submission of FDA Form 1572 (Statement of Investigator), NCI Biosketch, Agent Shipment Form, and Financial Disclosure Form (FDF). If there are several participating investigators at one institution, CTEP-supplied investigational agents for the study should be ordered under the name of one lead participating investigator at that institution.

In general, sites may order initial agent supplies when a subject is being screened for enrollment onto the study.

Submit agent requests through the PMB Online Agent Order Processing (OAOP) application. Access to OAOP requires the establishment of a CTEP Identity and Access Management (IAM) account and the maintenance of an "active" account status, a "current" password, and active person registration status. For questions about drug orders, transfers, returns, or accountability, call or email PMB any time. Refer to the PMB's website for specific policies and guidelines related to agent management.

8.1.2.2 Agent Inventory Records – The investigator, or a responsible party designated by the investigator, must maintain a careful record of the receipt, dispensing and final disposition of all agents received from the PMB using the appropriate NCI Investigational Agent (Drug) Accountability Record (DARF) available on the CTEP forms page. Store and maintain separate NCI Investigational Agent Accountability Records for each agent, strength, formulation and ordering investigator on this protocol.

8.1.3 Investigator Brochure Availability

The current versions of the IBs for the agents will be accessible to site investigators and research staff through the PMB OAOP application. Access to OAOP requires the establishment of a CTEP IAM account and the maintenance of an "active" account status, a "current" password and active person registration status. Questions about IB access may be directed to the PMB IB Coordinator via email.

8.1.4 Useful Links and Contacts

- CTEP Forms, Templates, Documents: http://ctep.cancer.gov/forms/
- NCI CTEP Investigator Registration: RCRHelpDesk@nih.gov
- PMB policies and guidelines: http://ctep.cancer.gov/branches/pmb/agent_management.htm
- PMB Online Agent Order Processing (OAOP) application:

https://ctepcore.nci.nih.gov/OAOP
- CTEP Identity and Access Management (IAM) account: https://ctepcore.nci.nih.gov/iam/
- CTEP IAM account help: ctepreghelp@ctep.nci.nih.gov
- IB Coordinator: IBCoordinator@mail.nih.gov
- PMB email: PMBAfterHours@mail.nih.gov
- PMB phone and hours of service: (240) 276-6575 Monday through Friday between 8:30 am and 4:30 pm (ET)

8.2 Commercial Agent: Azacitidine (NSC # 102816)

Product description:

Chemical Name: 4-amino-1-β-D-ribofuranosyl-s-triazin-2(1*H*)-one

Other Names: 5-azacitidine, 5-aza, azacitidine, Vidaza®

Classification: antimetabolite, DNA hypomethylating agent

CAS Registry Number: 320-67-2

Molecular Formula: $C_8H_{12}N_4O_5$ **M.W.:** 244.2

Approximate Solubility: 89 g/L at 25° C in water

Mode of Action: Azacitidine is a pyrimidine nucleoside analog of cytidine. Azacitidine is incorporated into DNA and RNA at high doses and causes direct cytotoxicity as a result of inhibiting DNA and RNA functions. It also inhibits DNA methyltransferase at low doses, therefore causing DNA hypomethylation and subsequent alterations in gene expression.

How Supplied: Azacitidine for injection is supplied as a lyophilized powder in 100 mg single-use vials packaged in cartons of 1 vial (NDC 67211-102-01). It is commercially available. Each vial contains 100 mg of azacitidine and 100 mg of mannitol as a freeze-dried cake or powder.

Solution preparation: As per package insert

Route of administration: Azacitidine may be administered either subcutaneously or intravenously, depending on local institutional practice.

Subcutaneous: The manufacturer recommends equally dividing volumes >4 mL into 2 syringes and injecting into 2 separate sites; however, policies for maximum SubQ administration volume may vary by institution; interpatient variations may also apply. Rotate sites for each injection (thigh, abdomen, or upper arm). Administer subsequent injections at least 1 inch from previous injection sites; do not inject into tender, bruised, red, or hard areas. Allow refrigerated

suspensions to come to room temperature (up to 30 minutes) prior to administration. Resuspend by inverting the syringe 2 to 3 times and then rolling the syringe between the palms for 30 seconds.

IV: Infuse over 10 to 40 minutes; infusion must be completed within 1 hour of (vial) reconstitution.

Agent Ordering: The agent is commercially available and is considered a standard-of-care drug in this setting.

9. STATISTICAL CONSIDERATIONS

9.1 Study Design/Endpoints

Phase Ib Safety / Tolerability Run In - Primary Endpoint:

- Safety and tolerability will be assessed by evaluating the number of patients out of 6 who experience a dose-limiting toxicity by cycle 1, day 42, using a standard 3+3 design. Two out of 6 events will lead to a dose-reduction or trial discontinuation as described in the analytic plan.

Phase Ib Safety / Tolerability Run In - Primary Endpoint Analytic Plan:

- An incidence of one in six (or fewer) patients experiencing a DLT, as described in section 7, will be considered a safe and tolerable dose.
- Patients who receive at least a full course of therapy will be considered to be evaluable for toxicity/DLT and will not be replaced unless the PI determines that further additional patients at that dose level are required for safety purposes. Patients who experience a DLT prior to completing a full course of therapy will be accounted for in the safety assessment.
- A standard 3+3 design will be used to study patients on an initial pinometostat dose of 54 mg/m2/day continuous IV infusion, in combination with azacitidine at a dose of 75mg/m2 daily for 7 days starting on cycle 1, day 1. If two of six experience a DLT within the first 42 days as defined above, the dose of pinometostat will be decreased to 36 mg/m2/day, and it will be combined with azacitidine at a dose of 75mg/m2 daily for 7 days in a separate cohort of patients. If two of six experience a DLT within the first 42 days as defined above, the combination will be deemed intolerable and the study will be terminated. If three patients tolerate the combination of standard-dose azacitidine with pinometostat at 54 mg/m2/day, the pinometostat dose may be escalated to 90 mg/m2/day and tested as per standard 3+3 design. Patients who complete cycle 1 would be allowed to stay on single-agent pinometostat through their day 42 evaluation. Patients who achieve count recovery prior to day 42 may be considered for additional combination therapy with azacitidine and may complete up to 6 cycles of combination therapy as described for patients directly enrolled on the phase II protocol.

Phase II Primary Endpoint:

- Response to combination therapy defined as CR, CRi, PR, or MLFS, with or without minimal residual disease (MRD), on bone marrow biopsy as defined by the 2017 European Leukemia Network guidelines will be assessed. Baseline bone marrow biopsy will be performed within 14 days prior to starting therapy, at the end of cycle 1 (day 28 +/- 2 days or time of count recovery), at the end of cycle 3 (+/- 2 days or at the time of count recovery after three cycles of azacitidine administration), at the end of cycle 6 (+/- 2 days or at the time of count recovery after six cycles of azacitidine administration), and / or at the time of therapy discontinuation +/- 3 days. Patients who have stable disease after 4 cycles of combination therapy or progressive disease at any time should be taken off the trial.

Phase II Primary Endpoint Analytic Plan:

- Response rate will be defined as the number of patients who achieve a CR, CRi, PR, or morphologic leukemia-free state (MLFS), with or without MRD, at any time point. Patients will be evaluated through the time of count recovery after 6 cycles of combination therapy.
- A Simon two-stage minimax design will be employed whereby a 20% response rate is considered not promising, a 40% response rate is considered promising, and the probabilities of a type I error (falsely accepting a non-promising therapy) and type II error (falsely rejecting a promising therapy) are set at 0.10 and 0.10, respectively. In the first stage of this design, 19 patients will be accrued. All patients will be followed for a minimum of 24 weeks. If at least 4 patients respond then an additional 17 patients will be accrued to the second stage. At the end of the trial, if 11 or more patients respond out of a total of 36 patients, then this combination will be considered worthy of further investigation. This design yields at least a 0.90 probability of a positive result if the true response rate is at least 40% and yields a 0.90 probability of a negative result if the true response rate is 20%.

9.2 Sample Size/Accrual Rate

Maximum planned sample size is 48 patients, with estimated monthly accrual rate of 3-6 patients among all sites per month.

PLANNED ENROLLMENT REPORT

Racial Categories	Ethnic Categories				Total
	Not Hispanic or Latino		Hispanic or Latino		
	Female	Male	Female	Male	
American Indian/ Alaska Native	0	0	0	0	**0**
Asian	2	2	0	0	**4**
Native Hawaiian or Other Pacific Islander	0	0	0	0	**0**
Black or African American	3	4	0	0	**7**
White	8	9	2	2	**21**
More Than One Race	2	2	0	0	**4**
Total	**15**	**17**	**2**	**2**	**36**

9.3 Stratification Factors

N/A

9.4 Analysis of Secondary Endpoints

Phase Ib Safety / Tolerability Run In - Secondary Endpoints:

- Assess safety and tolerability by evaluating the number of patients who experience a dose-limiting toxicity at any time during therapy as defined above.
- H3K79 methylation (assessed by ELISA) on the bone marrow biopsy specimen from prior to therapy initiation and on bone marrow biopsy from day 28 (or time of count recovery)
- Expression levels of HOXA9 and Meis1 (assessed by qPCR), Absolute neutrophil and absolute monocyte count at baseline and day 28
- Evaluate fraction of cells with 11q23 rearrangements before treatment and at day 28
- Evaluate for a rise in absolute neutrophil / absolute monocyte count from baseline

Phase Ib Safety / Tolerability Run In - Secondary Endpoint Analytic Plans:

- Compare quantitative methylation and methylation valence (e.g. if H3K27 has been methylated once, twice, or three times) from baseline and subsequent bone marrow samples using descriptive statistics and graphical displays. A Wilcoxon signed rank test will be used to assess post-treatment differences as compared to baseline. The variability in the assay will be evaluated by collecting a sample at the time of enrollment and at the time of combination therapy initiation, and by running the same sample on the assay multiple times. This variability will be taken into account in determining what percentage change in methylation will be considered significant.
- Compare expression level by qPCR of HOXA9 and Meis1 levels from baseline and subsequent bone marrow samples using descriptive statistics and graphical displays. A Wilcoxon signed rank test will be used to assess post treatment differences as compared to baseline. The variability in the assay will be evaluated by collecting a sample at the time of enrollment and at the time of combination therapy initiation, and by running the same sample on the assay multiple times. This variability will be taken into account in determining what percentage change in methylation will be considered significant.
- Evaluate differentiation by performing a differential on the bone marrow biopsy and peripheral blood samples at baseline and from subsequent time points and assessing percentage of monocytes / neutrophils, bands, and myeloid forms present. Samples will be banked and cytogenetic and FISH analysis of these cells will be performed to evaluate for the presence of MLL-rearrangement in patients who respond to therapy, given funding limitations on performing such an assessment on all patients.

Phase Ib Safety / Tolerability Run In - Exploratory Endpoints:

- Correlative studies will include assessment of incorporation of 5-aza-2`-deoxycytidine incorporation into genomic DNA and the extent of global DNA

methylation and correlation with systemic azacitidine exposure and pharmacodynamics effects.

Phase Ib Exploratory Endpoint Analytic Plans:

- Expect global DNA demethylation / correlation between decitabine systemic PK and decitabine incorporation into genomic DNA; ratio of DAC content/1000 2dC or 5mC content/1000 2dC to express DAC incorporation or global DNA methylation, respectively & methylation changes calculated relative to control 5mC content/1000 2dC ratio. Correlation between systemic azacitidine PK and decitabine incorporation into genomic DNA and other PD measures.

Phase II Secondary Endpoints:

- Assess safety and tolerability by evaluating the number of patients who experience a dose-limiting toxicity at any time during therapy as defined above.
- Assess patient response based on relapsed / refractory or previously untreated status
- H3K79 methylation (assessed by ELISA) on the bone marrow biopsy specimen from prior to therapy initiation and on subsequent bone marrow biopsy samples
- Expression levels of HOXA9 and Meis1 (assessed by qPCR), Absolute neutrophil and absolute monocyte at baseline and on subsequent bone marrow biopsy samples
- Evaluate fraction of cells with 11q23 rearrangements before treatment and on subsequent bone marrow biopsy samples
- Evaluate for a rise in absolute neutrophil / absolute monocyte count at the end of cycle 1 as compared to baseline

Phase II Secondary Endpoint Analytic Plans:

- Patients will be followed for the development of DLTs as described above. All DLTs will be tracked to detect potential toxicities with combination therapy
- Patients will be stratified based on treatment for relapsed / refractory or previously untreated disease. The response of each stratum to combination therapy, defined as CR, CRi, MLFS, or PR, with or without MRD, on bone marrow biopsy as defined by the 2017 European Leukemia Network guidelines, will be described. Kaplan-Meier estimates will be calculated and compared for each stratum, however the small number of patients likely to be enrolled in each stratum will limit the power of this study to detect a difference among them. A report will be generated to look at all patients, all eligible patients who were enrolled, and all evaluable patients.
- Compare quantitative methylation and methylation valence (e.g. if H3K27 has been methylated once, twice, or three times) from baseline and subsequent bone marrow samples using descriptive statistics and graphical displays. A Wilcoxon signed rank test will be used to assess post treatment differences as compared to baseline. The variability in the assay from the phase Ib studies will be taken into account in determining what percentage change in methylation will be considered significant.
- Compare expression level by qPCR of HOXA9 and Meis1 levels from baseline and subsequent bone marrow samples using descriptive statistics and graphical displays. A Wilcoxon signed rank test will be used to assess post treatment differences as compared to baseline. The variability in the assay from the phase Ib studies will be

taken into account in determining what percentage change in methylation will be considered significant.

- Compare 11q23 translocation level as determined by FISH from baseline and subsequent bone marrow samples using descriptive statistics and graphical displays. A Wilcoxon signed rank test will be used to assess post treatment differences as compared to baseline. The variability in the assay from the phase Ib studies will be taken into account in determining what percentage change in translocation detection will be considered significant.
- Evaluate differentiation by performing a differential on the bone marrow biopsy sample and peripheral blood samples at baseline and subsequent bone marrows and assessing percentage of monocytes / neutrophils, bands, and myeloid forms present. Samples will be banked and cytogenetic and FISH analysis of these cells will be performed to evaluate for the presence of MLL-rearrangement in patients who respond to therapy, given funding limitations on performing such an assessment on all patients.

Phase II Exploratory Endpoints:

- Correlative studies will include assessment of incorporation of 5-aza-2`-deoxycytidine incorporation into genomic DNA and the extent of global DNA methylation and correlation with systemic azacitidine exposure and pharmacodynamics effects.

Phase II Exploratory Endpoint Analytic Plans:

- Expect global DNA demethylation / correlation between decitabine systemic PK and decitabine incorporation into genomic DNA; ratio of DAC content/1000 2dC or 5mC content/1000 2dC to express DAC incorporation or global DNA methylation, respectively & methylation changes calculated relative to control 5mC content/1000 2dC ratio. Correlation between systemic azacitidine PK and decitabine incorporation into genomic DNA and other PD measures.

9.5 For phase 2 protocols only: Reporting and Exclusions

9.5.1 Evaluation of Toxicity

All patients will be evaluable for toxicity from the time of their first treatment with pinometostat.

9.5.2 Evaluation of Response

All patients included in the study must be assessed for response to treatment, even if there are major protocol treatment deviations or if they are ineligible. Each patient will be assigned one of the following categories: 1) complete response, 2) partial response, 3) stable disease, 4) morphologic leukemia-free state, 5) early death from malignant disease, 6) early death from toxicity, 7) early death because of other cause, 8) progressive disease, or 9) unknown (not assessable, insufficient data). [Note: By arbitrary convention, category 9 usually designates the "unknown" status of any type of data in a clinical

database.]

All of the patients who met the eligibility criteria (with the possible exception of those who received no study medication) should be included in the main analysis of the response rate. Patients in response categories 5-9 should be considered to have a treatment failure (disease progression). Thus, an incorrect treatment schedule or drug administration does not result in exclusion from the analysis of the response rate. Precise definitions for categories 4-9 will be protocol specific.

All conclusions should be based on all eligible patients. Subanalyses may then be performed on the basis of a subset of patients, excluding those for whom major protocol deviations have been identified (*e.g.*, early death due to other reasons, early discontinuation of treatment, major protocol violations, etc.). However, these subanalyses may not serve as the basis for drawing conclusions concerning treatment efficacy, and the reasons for excluding patients from the analysis should be clearly reported. The 95% confidence intervals should also be provided.

9.6 Early Stopping Criteria

The study includes sequential stopping rules to monitor for safety in the phase II study after safety has been confirmed in the first six patients. These rules are based on the 30 patients who will accrue after the safety run-in. The toxicity definitions for these rules mirror the DLT definition. In the event that three in the first six patients, five in the first 12 patients, six in the first 18 patients, seven in the first 24 patients, or eight patients at any point among the remaining 30 patients fulfill the toxicity criteria, accrual to the study will stop due to a lack of safety. If the true risk of toxicity is 15%, the probability the intervention will be ruled unsafe is 10%. If the true risk increases to 40%, the probability increases to 95%.

10. ADVERSE EVENTS: LIST AND REPORTING REQUIREMENTS

Adverse event (AE) monitoring and reporting is a routine part of every clinical trial. The following list of AEs (Section 10.1) and the characteristics of an observed AE (Sections 10.2 and 10.3) will determine whether the event requires expedited reporting via the CTEP Adverse Event Reporting System (CTEP-AERS) **in addition** to routine reporting.

10.1 Comprehensive Adverse Events and Potential Risks List(s) (CAEPRs)

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

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

10.1.1 CAEPRs for CTEP IND Agent

10.1.1.1 CAEPR for Pinometostat

Comprehensive Adverse Events and Potential Risks list (CAEPR)
for
Pinometostat (EPZ-5676, NSC 795144)

The Comprehensive Adverse Events and Potential Risks list (CAEPR) provides a single list of reported and/or potential adverse events (AE) associated with an agent using a uniform presentation of events by body system. In addition to the comprehensive list, a subset, the Specific Protocol Exceptions to Expedited Reporting (SPEER), appears in a separate column and is identified with bold and italicized text. This subset of AEs (SPEER) is a list of events that are protocol specific exceptions to expedited reporting to NCI (except as noted below). Refer to the 'CTEP, NCI Guidelines: Adverse Event Reporting Requirements' http://ctep.cancer.gov/protocolDevelopment/electronic_applications/docs/aeguidelines.pdf for further clarification. The CAEPR does not provide frequency data; refer to the Investigator's Brochure for this information. Below is the CAEPR for Pinometostat (EPZ-5676).

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

Version 1.1, January 7, 2019[1]

Adverse Events with Possible Relationship to Pinometostat (EPZ-5676) (CTCAE 5.0 Term)	Specific Protocol Exceptions to Expedited Reporting (SPEER)
BLOOD AND LYMPHATIC SYSTEM DISORDERS	
Anemia	
Febrile neutropenia	***Febrile neutropenia (Gr 2)***
Leukocytosis	
CARDIAC DISORDERS	
Heart failure	
GASTROINTESTINAL DISORDERS	
Diarrhea	
Nausea	
Vomiting	
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS	
Fatigue	
INVESTIGATIONS	
Alanine aminotransferase increased	
Aspartate aminotransferase increased	
Blood bilirubin increased	
Electrocardiogram QT corrected interval prolonged	
Lymphocyte count decreased	
Neutrophil count decreased	
Platelet count decreased	
White blood cell decreased	
METABOLISM AND NUTRITION DISORDERS	
Hypocalcemia	
Hypophosphatemia	
SKIN AND SUBCUTANEOUS TISSUE DISORDERS	
Rash maculo-papular	

[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.

Adverse events reported on pinometostat (EPZ-5676) trials, but for which there is insufficient evidence to suggest that there was a reasonable possibility that pinometostat (EPZ-5676) caused the adverse event:

CARDIAC DISORDERS - Cardiac arrest
EYE DISORDERS - Periorbital edema
GASTROINTESTINAL DISORDERS - Colitis; Constipation; Mucositis oral
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS - Edema face; Generalized edema; Localized edema; Malaise; Multi-organ failure
INFECTIONS AND INFESTATIONS - Device related infection; Folliculitis; Fungemia; Lung infection; Sepsis; Thrush
INVESTIGATIONS - Alkaline phosphatase increased; Creatinine increased; Ejection fraction decreased; INR increased; Investigations - Other (ECG PR prolongation); Investigations - Other (ejection fraction increased); Investigations - Other (increase in polymorphonuclear neutrophils and/or monocytes)
METABOLISM AND NUTRITION DISORDERS - Anorexia; Hyperkalemia; Hypermagnesemia; Hypernatremia; Hyperuricemia; Hypoalbuminemia; Hypokalemia; Hypomagnesemia; Tumor lysis

syndrome
MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS - Pain in extremity
NERVOUS SYSTEM DISORDERS - Dysgeusia; Headache; Intracranial hemorrhage; Ischemia cerebrovascular; Lethargy; Presyncope; Tremor
PSYCHIATRIC DISORDERS - Irritability
RENAL AND URINARY DISORDERS - Acute kidney injury
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS - Apnea; Cough; Hypoxia; Pleural effusion; Respiratory failure; Respiratory, thoracic and mediastinal disorders - Other (tachypnea); Sore throat
SKIN AND SUBCUTANEOUS TISSUE DISORDERS - Alopecia; Dry skin; Pruritus
VASCULAR DISORDERS - Hematoma; Hypertension

Note: Pinometostat (EPZ-5676) 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.

10.1.2 Adverse Event List(s) for Commercial Agent(s)

Comprehensive Adverse Events and Potential Risks list (CAEPR)
for
Azacitidine (NSC 102816)

The Comprehensive Adverse Events and Potential Risks list (CAEPR) provides a single list of reported and/or potential adverse events (AE) associated with an agent using a uniform presentation of events by body system. In addition to the comprehensive list, a subset, the Specific Protocol Exceptions to Expedited Reporting (SPEER), appears in a separate column and is identified with bold and italicized text. This subset of AEs (SPEER) is a list of events that are protocol specific exceptions to expedited reporting to NCI (except as noted below). Refer to the 'CTEP, NCI Guidelines: Adverse Event Reporting Requirements' http://ctep.cancer.gov/protocolDevelopment/electronic_applications/docs/aeguidelines.pdf for further clarification. *Frequency is provided based on 1800 patients.* Below is the CAEPR for Azacitidine.

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

Version 2.7, July 30, 2019[1]

Adverse Events with Possible Relationship to Azacitidine (CTCAE 5.0 Term) [n= 1800]			**Specific Protocol Exceptions to Expedited Reporting (SPEER)**
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
BLOOD AND LYMPHATIC SYSTEM DISORDERS			
Anemia			***Anemia (Gr 3)***
	Febrile neutropenia		***Febrile neutropenia (Gr 3)***
CARDIAC DISORDERS			
	Heart failure		***Heart failure (Gr 2)***
	Pericardial effusion		***Pericardial effusion (Gr 2)***
	Sinus tachycardia		***Sinus tachycardia (Gr 2)***
	Supraventricular tachycardia		***Supraventricular tachycardia (Gr 2)***
GASTROINTESTINAL DISORDERS			
	Abdominal pain		***Abdominal pain (Gr 3)***

Adverse Events with Possible Relationship to Azacitidine (CTCAE 5.0 Term) [n= 1800]			Specific Protocol Exceptions to Expedited Reporting (SPEER)
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
	Colitis		***Colitis (Gr 2)***
Constipation			***Constipation (Gr 2)***
Diarrhea			***Diarrhea (Gr 3)***
	Esophagitis		***Esophagitis (Gr 2)***
	Gastrointestinal hemorrhage[2]		
	Mucositis oral		***Mucositis oral (Gr 2)***
Nausea			***Nausea (Gr 3)***
Vomiting			***Vomiting (Gr 3)***
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS			
	Chills		***Chills (Gr 2)***
	Edema limbs		***Edema limbs (Gr 2)***
Fatigue			***Fatigue (Gr 3)***
Fever			***Fever (Gr 3)***
Injection site reaction			***Injection site reaction (Gr 2)***
IMMUNE SYSTEM DISORDERS			
		Allergic reaction	***Allergic reaction (Gr 2)***
		Anaphylaxis	
INFECTIONS AND INFESTATIONS			
Infection[3]			***Infection[3] (Gr 4)***
INJURY, POISONING AND PROCEDURAL COMPLICATIONS			
	Bruising		***Bruising (Gr 2)***
INVESTIGATIONS			
	Alanine aminotransferase increased		***Alanine aminotransferase increased (Gr 4)***
	Alkaline phosphatase increased		***Alkaline phosphatase increased (Gr 2)***
	Aspartate aminotransferase increased		***Aspartate aminotransferase increased (Gr 4)***
	Blood bilirubin increased		***Blood bilirubin increased (Gr 2)***
	GGT increased		***GGT increased (Gr 2)***
	Lymphocyte count decreased		***Lymphocyte count decreased (Gr 4)***
Neutrophil count decreased			***Neutrophil count decreased (Gr 4)***
Platelet count decreased			***Platelet count decreased (Gr 4)***
	Weight loss		***Weight loss (Gr 2)***
	White blood cell decreased		***White blood cell decreased (Gr 4)***
METABOLISM AND NUTRITION DISORDERS			
	Acidosis		***Acidosis (Gr 2)***
	Anorexia		***Anorexia (Gr 3)***
	Hypokalemia		
		Tumor lysis syndrome	
MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS			
	Arthralgia		***Arthralgia (Gr 2)***
	Back pain		***Back pain (Gr 2)***
	Generalized muscle weakness		***Generalized muscle weakness (Gr 2)***
	Myalgia		***Myalgia (Gr 2)***

Adverse Events with Possible Relationship to Azacitidine (CTCAE 5.0 Term) [n= 1800]			Specific Protocol Exceptions to Expedited Reporting (SPEER)
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
	Pain in extremity		***Pain in extremity (Gr 2)***
NERVOUS SYSTEM DISORDERS			
	Dizziness		***Dizziness (Gr 2)***
	Headache		***Headache (Gr 2)***
	Peripheral motor neuropathy		***Peripheral motor neuropathy (Gr 2)***
	Somnolence		***Somnolence (Gr 2)***
PSYCHIATRIC DISORDERS			
	Anxiety		
	Confusion		***Confusion (Gr 2)***
	Insomnia		
RENAL AND URINARY DISORDERS			
		Acute kidney injury	
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS			
		Bronchopulmonary hemorrhage	
	Cough		***Cough (Gr 2)***
	Dyspnea		***Dyspnea (Gr 4)***
	Epistaxis		***Epistaxis (Gr 2)***
	Pharyngolaryngeal pain		
	Postnasal drip		***Postnasal drip (Gr 2)***
	Respiratory, thoracic and mediastinal disorders - Other (abnormal breath sound)[4]		***Respiratory, thoracic and mediastinal disorders - Other (abnormal breath sound)[4] (Gr 2)***
SKIN AND SUBCUTANEOUS TISSUE DISORDERS			
	Alopecia		***Alopecia (Gr 2)***
	Pruritus		***Pruritus (Gr 2)***
	Purpura		***Purpura (Gr 2)***
	Rash maculo-papular		***Rash maculo-papular (Gr 3)***
VASCULAR DISORDERS			
	Hematoma		***Hematoma (Gr 2)***
	Hypotension		***Hypotension (Gr 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]Gastrointestinal hemorrhage includes Anal hemorrhage, Cecal hemorrhage, Colonic hemorrhage, Duodenal hemorrhage, Esophageal hemorrhage, Esophageal varices hemorrhage, Gastric hemorrhage, Hemorrhoidal hemorrhage, Ileal hemorrhage, Intra-abdominal hemorrhage, Jejunal hemorrhage, Lower gastrointestinal hemorrhage, Oral hemorrhage, Pancreatic hemorrhage, Rectal hemorrhage, Retroperitoneal hemorrhage, and Upper gastrointestinal hemorrhage under the GASTROINTESTINAL DISORDERS SOC.

[3]Infection may include any of the 75 infection sites under the INFECTIONS AND INFESTATIONS SOC.

[4]Abnormal breath sounds include rales and rhonchi.

Adverse events reported on azacitidine trials, but for which there is insufficient evidence to suggest that there was a reasonable possibility that azacitidine caused the adverse event:

BLOOD AND LYMPHATIC SYSTEM DISORDERS - Blood and lymphatic system disorders - Other (agranulocytosis); Blood and lymphatic system disorders - Other (lymphadenopathy); Blood and lymphatic system disorders - Other (pancytopenia); Blood and lymphatic system disorders - Other (splenomegaly); Blood and lymphatic system disorders - Other (transfusion: platelets); Bone marrow hypocellular; Leukocytosis
CARDIAC DISORDERS - Atrial fibrillation; Atrial flutter; Atrioventricular block complete; Cardiac arrest; Cardiac disorders - Other (cardiac valve vegetation); Cardiac disorders - Other (Wolff-Parkinson-White syndrome); Chest pain - cardiac; Myocardial infarction; Palpitations; Pericarditis; Restrictive cardiomyopathy; Sinus bradycardia; Ventricular fibrillation
EAR AND LABYRINTH DISORDERS - Hearing impaired; Tinnitus
EYE DISORDERS - Eye disorders - Other (eye/conjunctival hemorrhage); Eye disorders - Other (retina hemorrhage); Papilledema; Uveitis
GASTROINTESTINAL DISORDERS - Abdominal distension; Ascites; Duodenal ulcer; Dyspepsia; Dysphagia; Enterocolitis; Esophageal pain; Esophageal ulcer; Flatulence; Gastritis; Gastrointestinal disorders - Other (enteritis); Gastrointestinal disorders - Other (inguinal hernia, obstructive); Gastrointestinal disorders - Other (intestinal ischemia); Gastrointestinal disorders - Other (intussusception); Gastrointestinal pain; Hemorrhoids; Pancreatitis; Periodontal disease; Small intestinal obstruction; Visceral arterial ischemia
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS - Death NOS; Edema face; Flu like symptoms; Gait disturbance; General disorders and administration site conditions - Other (systemic inflammatory response syndrome); Generalized edema; Malaise; Multi-organ failure; Non-cardiac chest pain; Pain; Sudden death NOS
HEPATOBILIARY DISORDERS - Cholecystitis; Hepatic failure; Hepatobiliary disorders - Other (bile duct stone); Hepatobiliary disorders - Other (hepatic cirrhosis)
IMMUNE SYSTEM DISORDERS - Autoimmune disorder; Immune system disorders - Other (GVHD)
INJURY, POISONING AND PROCEDURAL COMPLICATIONS - Burn; Fall; Fracture; Hip fracture; Injury, poisoning and procedural complications - Other (excoriation); Injury, poisoning and procedural complications - Other (transfusion reaction); Postoperative hemorrhage; Wound dehiscence
INVESTIGATIONS - Activated partial thromboplastin time prolonged; Blood lactate dehydrogenase increased; Creatinine increased; Electrocardiogram QT corrected interval prolonged; INR increased; Investigations - Other (blood urea increased); Investigations - Other (cardiac murmur); Investigations - Other (coagulopathy); Investigations - Other (protein total decreased); Investigations - Other (thrombocytosis); Lipase increased; Lymphocyte count increased; Serum amylase increased
METABOLISM AND NUTRITION DISORDERS - Dehydration; Hyperglycemia; Hyperkalemia; Hyperphosphatemia; Hyperuricemia; Hypoalbuminemia; Hypocalcemia; Hypomagnesemia; Hyponatremia; Hypophosphatemia; Metabolism and nutrition disorders - Other (gout exacerbation); Metabolism and nutrition disorders - Other (hypovolemia); Metabolism and nutrition disorders - Other (low carbon dioxide)
MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS - Arthritis; Bone pain; Chest wall pain; Flank pain; Muscle cramp; Muscle weakness lower limb; Musculoskeletal and connective tissue disorder - Other (chondritis); Musculoskeletal and connective tissue disorder - Other (intervertebral disc protrusion); Musculoskeletal and connective tissue disorder - Other (musculoskeletal stiffness); Neck pain
NEOPLASMS BENIGN, MALIGNANT AND UNSPECIFIED (INCL CYSTS AND POLYPS) - Myelodysplastic syndrome; Neoplasms benign, malignant and unspecified (incl cysts and polyps) - Other (colonic polyp, vaginal polyp); Neoplasms benign, malignant and unspecified (incl cysts and polyps) - Other (metastases to central nervous system); Treatment related secondary malignancy
NERVOUS SYSTEM DISORDERS - Dysesthesia; Dysgeusia; Hydrocephalus; Intracranial hemorrhage; Lethargy; Memory impairment; Nervous system disorders - Other (head injury); Paresthesia; Peripheral sensory neuropathy; Seizure; Stroke; Syncope
PSYCHIATRIC DISORDERS - Delirium; Depression; Hallucinations; Psychiatric disorders - Other (mental status changes)

RENAL AND URINARY DISORDERS - Chronic kidney disease; Dysuria; Hematuria; Proteinuria; Renal and urinary disorders - Other (bladder distention); Renal and urinary disorders - Other (calculus urinary); Renal calculi; Urinary frequency; Urinary retention
REPRODUCTIVE SYSTEM AND BREAST DISORDERS - Erectile dysfunction; Reproductive system and breast disorders - Other (benign prostatic hyperplasia); Uterine hemorrhage; Vaginal hemorrhage
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS - Atelectasis; Hypoxia; Laryngeal hemorrhage; Nasal congestion; Oropharyngeal pain; Pleural effusion; Pleuritic pain; Pneumonitis; Pneumothorax; Productive cough; Pulmonary edema; Respiratory failure; Respiratory, thoracic and mediastinal disorders - Other (chronic obstructive pulmonary disease); Respiratory, thoracic and mediastinal disorders - Other (pharyngeal erythema); Rhinorrhea; Sinus pain; Wheezing
SKIN AND SUBCUTANEOUS TISSUE DISORDERS - Dry skin; Hyperhidrosis; Palmar-plantar erythrodysesthesia syndrome; Skin and subcutaneous tissue disorders - Other (skin laceration); Skin and subcutaneous tissue disorders - Other (skin lesion); Skin and subcutaneous tissue disorders - Other (skin nodule); Skin and subcutaneous tissue disorders - Other (Sweet's Syndrome); Skin induration; Urticaria
VASCULAR DISORDERS - Flushing; Hypertension; Thromboembolic event; Vascular disorders - Other (pallor); Vascular disorders - Other (poor venous access); Vasculitis

Note: Azacitidine 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.

10.2 Adverse Event Characteristics

- **CTCAE term (AE description) and grade:** The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 5.0 will be utilized for AE reporting. All appropriate treatment areas should have access to a copy of the CTCAE version 5.0. A copy of the CTCAE version 5.0 can be downloaded from the CTEP web site http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm.

- **For expedited reporting purposes only:**
  - AEs for the agent that are ***bold and italicized*** in the CAEPR (*i.e.*, those listed in the SPEER column, Section 10.1) should be reported through CTEP-AERS only if the grade is **above** the grade provided in the SPEER.
  - Other AEs for the protocol that do not require expedited reporting are outlined in Section 10.3.4.

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

10.3 Expedited Adverse Event Reporting

10.3.1 Rave-CTEP-AERS Integration

The Cancer Therapy Evaluation Program Adverse Event Reporting System (CTEP-AERS) integration enables evaluation of post-baseline AEs entered in Rave to determine whether they require expedited reporting, and facilitates entry in CTEP-AERS for those AEs requiring expedited reporting.

All AEs that occur after baseline are collected in Medidata Rave using the Adverse Event form, which is available for entry at each treatment or reporting period, and used to collect AEs that start during the period or persist from the previous reporting period. The Clinical Research Associate (CRA) will enter AEs that occur prior to the start of treatment on a baseline form that is not included in the Rave-CTEP-AERS integration. AEs that occur prior to enrollment must begin and end on the baseline Adverse Event form and should not be included on the standard Adverse Events form that is available at treatment unless there has been an increase in grade.

Prior to sending AEs through the rules evaluation process, site staff should verify the following on the Adverse Event form in Rave:

- The reporting period (course/cycle) is correct, and
- AEs are recorded and complete (no missing fields) and the form is query-free (fields added to the form during study build do not need to be query-free for the integration call with CTEP-AERS to be a success).

The CRA reports AEs in Rave at the time the Investigator learns of the event. If the CRA modifies an AE, it must be re-submitted for rules evaluation.

Upon completion of AE entry in Medidata Rave, the CRA submits the AE for rules evaluation by completing the Expedited Reporting Evaluation form. Both NCI and protocol-specific reporting rules evaluate the AEs submitted for expedited reporting. A report is initiated in CTEP-AERS using information entered in Medidata Rave for AEs that meet reporting requirements. The CRA completes the report by accessing CTEP-AERS via a direct link on the Medidata Rave Expedited Reporting Evaluation form.

In the rare occurrence that Internet connectivity is lost, a 24-hour notification is to be made to CTEP by telephone at 301-897-7497. Once Internet connectivity is restored, the 24-hour notification that was phoned in must be entered immediately into CTEP-AERS using the deep link from Medidata Rave.
Additional information about the CTEP-AERS integration is available on the CTSU website:

- Study specific documents: Protocols > Documents > Education and Promotion, and
- Expedited Safety Reporting Rules Evaluation user guide: Resources > CTSU Operations Information > User Guides.

NCI requirements for SAE reporting are available on the CTEP website:

NCI Guidelines for Investigators: Adverse Event Reporting Requirements is available at https://ctep.cancer.gov/protocolDevelopment/electronic_applications/docs/aeguidelines.pdf.

10.3.2 Distribution of Adverse Event Reports

CTEP-AERS is programmed for automatic electronic distribution of reports to the following individuals: Principal Investigator and Adverse Event Coordinator(s) (if applicable) of the Corresponding Organization or Lead Organization, the local treating physician, and the Reporter and Submitter. CTEP-AERS provides a copy feature for other e-mail recipients.

10.3.3 Expedited Reporting Guidelines

Use the NCI protocol number and the protocol-specific patient ID assigned during trial registration on all reports.

Note: A death on study requires both routine and expedited reporting, regardless of causality. Attribution to treatment or other cause must be provided.

Death due to progressive disease should be reported as Grade 5 "Disease Progression" in the system organ class (SOC) "General disorders and administration site conditions. Evidence that the death was a manifestation of underlying disease (*e.g.*, radiological changes suggesting tumor growth or progression: clinical deterioration associated with a disease process) should be submitted.

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

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).		
ALL SERIOUS adverse events that meet the above criteria MUST be immediately reported to the NCI via electronic submission within the timeframes detailed in the table below.		
Hospitalization	**Grade 1 and Grade 2 Timeframes**	**Grade 3-5 Timeframes**

Resulting in Hospitalization ≥ 24 hrs	10 Calendar Days	24-Hour 5 Calendar Days
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 submitted electronically 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 electronically within 10 calendar days of learning of the AE.

[1]Serious adverse events that occur more than 30 days after the last administration of investigational agent/intervention 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 PET or SPECT IND agents, the AE reporting period is limited to 10 radioactive half-lives, rounded UP to the nearest whole day, after the agent/intervention was last administered. Footnote "1" above applies after this reporting period.

Effective Date: May 5, 2011

10.3.4 Additional Protocol-Specific Expedited Adverse Event Reporting Exclusions

For this protocol only, the AEs/grades listed below do not require expedited reporting via CTEP-AERS. However, they still must be reported through the routine reporting mechanism (Section 10.4):

CTCAE SOC	Adverse Event	Grade	≥24h Hospitalization[a]
Blood and lymphatic disorders	Bone marrow hypocellular	4	Regardless
Cardiac Disorders	Atrial fibrillation	2	Regardless
Cardiac Disorders	Atrial flutter	2	Regardless
Ear and labyrinth disorders	Vertigo	2	Regardless
Gastrointestinal disorders	Belching	2	Regardless
Gastrointestinal disorders	Bloating	2	Regardless
Gastrointestinal disorders	Constipation	3	Regardless
Gastrointestinal disorders	Dental caries	3	Regardless
Gastrointestinal disorders	Dry mouth	2	Regardless
Gastrointestinal disorders	Dyspepsia	3	Regardless
Gastrointestinal disorders	Gastroesophageal reflux disease	2	Regardless
Gastrointestinal disorders	Gingival pain	2	Regardless
Gastrointestinal disorders	Hemorrhoids	2	Regardless
Gastrointestinal disorders	Lip pain	3	Regardless

CTCAE SOC	Adverse Event	Grade	≥24h Hospitalization[a]
Gastrointestinal disorders	Toothache	3	Regardless
General disorders and administration site conditions	Infusion site extravasation	2	Regardless
General disorders and administration site conditions	Malaise	3	Regardless
General disorders and administration site conditions	Pain	2	No
Injury, poisoning and procedural complications	Fracture	2	Regardless
Investigations	Activated partial thromboplastin time prolonged	3	No
Investigations	Blood lactate dehydrogenase increased	1	Regardless
Investigations	INR increased	3	No
Metabolism and nutrition disorders	Glucose intolerance	3	Regardless
Metabolism and nutrition disorders	Hypercalcemia	2	No
Metabolism and nutrition disorders	Hyperglycemia	3	No
Metabolism and nutrition disorders	Hyperkalemia	2	No
Metabolism and nutrition disorders	Hypernatremia	2	No
Metabolism and nutrition disorders	Hyperphosphatemia	3	No
Metabolism and nutrition disorders	Hyperuricemia	3	No
Metabolism and nutrition disorders	Hypalbuminemia	3	No
Metabolism and nutrition disorders	Hypocalcemia	2	No
Metabolism and nutrition disorders	Hypoglycemia	2	No
Metabolism and nutrition disorders	Hypokalemia	3	No
Metabolism and nutrition disorders	Hypomagnesemia	2	No

CTCAE SOC	Adverse Event	Grade	≥24h Hospitalization[a]
Metabolism and nutrition disorders	Hyponatremia	3	No
Metabolism and nutrition disorders	Hypophosphatemia	3	No
Metabolism and nutrition disorders	Obesity	4	Regardless
Metabolism and nutrition disorders	Tumor lysis syndrome	3	No
Psychiatric disorders	Agitation	3	Regardless
Psychiatric disorders	Anxiety	3	Regardless
Psychiatric disorders	Delirium	3	Regardless
Psychiatric disorders	Depression	3	Regardless
Psychiatric disorders	Insomnia	3	Regardless
Renal and urinary disorders	Chronic kidney disease	4	Regardless
Renal and urinary disorders	Dysuria	1	Regardless
Renal and urinary disorders	Urinary frequency	2	Regardless
Respiratory, thoracic, and mediastinal disorders	Hiccups	3	No
Respiratory, thoracic, and mediastinal disorders	Nasal congestion	3	Regardless
Respiratory, thoracic, and mediastinal disorders	Sneezing	2	Regardless
Respiratory, thoracic, and mediastinal disorders	Sore throat	3	No
Respiratory, thoracic, and mediastinal disorders	Sleep apnea	3	Regardless
Skin and subcutaneous tissue disorders	Dry skin	3	Regardless
Skin and subcutaneous tissue disorders	Eczema	3	No
Vascular disorders	Hypertension	3	No
Vascular disorders	Thromboembolic event	2	No

[a] Indicates that an adverse event required hospitalization for ≥24 hours or prolongation of hospitalization by ≥24 hours of a patient.

10.4 Routine Adverse Event Reporting

All Adverse Events **must** be reported in routine study data submissions. **AEs reported**

expeditiously through CTEP-AERS must also be reported in routine study data submissions.
Adverse event data collection and reporting, which are required as part of every clinical trial, are done to ensure the safety of patients enrolled in the studies as well as those who will enroll in future studies using similar agents. AEs are reported in a routine manner at scheduled times during the trial using Medidata Rave. For this trial the Adverse Event CRF is used for routine AE reporting in Rave.

10.5 Pregnancy

Although not an adverse event in and of itself, pregnancy as well as its outcome must be documented via **CTEP-AERS**. In addition, the ***Pregnancy Information Form*** included within the NCI Guidelines for Adverse Event Reporting Requirements must be completed and submitted to CTEP. Any pregnancy occurring in a patient or patient's partner from the time of consent to 90 days after the last dose of study drug must be reported and then followed for outcome. Newborn infants should be followed until 30 days old. Please see the "NCI Guidelines for Investigators: Adverse Event Reporting Requirements for DCTD (CTEP and CIP) and DCP INDs and IDEs" (at http://ctep.cancer.gov/protocolDevelopment/adverse_effects.htm) for more details on how to report pregnancy and its outcome to CTEP.

10.6 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 expeditiously 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.

10.7 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 AE reporting unless otherwise specified.

11. STUDY CALENDAR

Baseline evaluations are to be conducted within 14 days prior to start of protocol therapy. Scans and x-rays must be done ≤4 weeks prior to the start of therapy.
In the event that the patient's condition is deteriorating, laboratory evaluations should be repeated within 48 hours prior to initiation of the next cycle of therapy.

	Pre-Study	Wk 1 Cyc 1 Day 1	Cyc 1 Day 8	Wk 3	Wk 4	Wk 5 Cyc 2 Day 1	Wk 6	Wk 7	Wk 8	Wk 9 Cyc 3 Day 1	Wk 10	Wk 11	Wk 12
Pinometostat		A	A	A	A	A	A	A	A	A	A	A	A
Azacitidine		B				B				B			
Informed consent	X												
Demographics	X												
Medical history	X												
Hepatitis B and C testing	X												
Concurrent meds	X	X----------											X
Physical exam	X	X		X		X				X			
Urine Pregnancy Test	X												
Vital signs	X	X		X		X				X			
Height	X												
Weight	X	X		X		X				X			
Performance status	X	X											
CBC w/diff, plts	X	X	X	X	X	X				X			
Serum chemistry[a]	X	X	X	X	X	X				X			
PT / PTT	X	X	X	X	X	X				X			
Uric Acid	X	X											
Serum PK assessments[b]		X											
Serum 5-aza-2'-deoxycytidine genomic incorporation[c]		X	X										
EKG (as indicated)	X	X	X	X	X								
Adverse event evaluation		X----------											X
Nail clippings collection	X												

Bone Marrow Biopsy and Aspirate (with morphology, flow cytometry, cytogenetics, FISH)	X					X‡							
Marrow aspirate myeloid genomic panel	X												
Marrow aspirate for banking samples	X					X‡							
Marrow aspirate for qPCR for HOXA9 & Meis1 and H3K79 ELISA	X					X‡							
Marrow aspirate for 5-aza-2'-deoxycytidine Genomic Incorporation and DNA demethylation[c]	X					X‡							

A: *Pinometostat*: Dose as assigned
B: Azaciridine: Standard dosing of 75mg/m2 given over 7 of the first 10 days of the cycle
a: Albumin, alkaline phosphatase, total bilirubin, bicarbonate, BUN, calcium, chloride, creatinine, glucose, phosphorus, potassium, total protein, SGOT [AST], SGPT [ALT], sodium, magnesium.
b: PK assessments should be done prior to treatment and at 15, 30, and 60 min after azacitidine is started in cycle 1
c: Serum and marrow 5-aza-2'-deoxycytidine genomic incorporation and DNA demethylation samples must be collected in green-top vacutainer containing THU
‡: Bone marrow tests should be performed at the end of cycle 1, end of cycle 3, and end of cycle 6, prior to the start of the next cycle of azacitidine. These can be performed on the first day of the following cycle, prior to the start of azacitidine therapy.

	Wk 13 Cyc 4 Day 1	Wk 14	Wk 15	Wk 16	Wk 17 Cyc 5 Day 1	Wk 18	Wk 19	Wk 20	Wk 21 Cyc 6 Day 1	Wk 22	Wk 23	Wk 24	Off Study[c]	1-mo f/u
Pinometostat	A	A	A	A	A	A	A	A	A	A	A	A	#	
Azacitidine	B				B				B					
Informed consent														
Demographics														
Medical history														
Concurrent meds	X----------X													
Physical exam	X				X				X				X	X
Vital signs	X				X				X				X	X
Height	X													
Weight	X				X				X				X	X

Performance status														
CBC w/diff, plts	X				X				X				X	X
Serum chemistry[a]	X				X				X				X	X
PT/PTT	X				X				X				X	X
EKG (as indicated)														
Adverse event evaluation	X----------X													
Bone Marrow Biopsy and Aspirate (with morphology, flow cytometry, cytogenetics, FISH)	X‡												X*	
Marrow aspirate for banking samples	X‡												X*	
Marrow aspirate for qPCR for HOXA9 & Meis1 and H3K79 ELISA	X‡												X*	
Marrow aspirate for 5-aza-2'-deoxycytidine Genomic Incorporation and DNA demethylation[c]	X‡												X*	

A: *Pinometostat*: Dose as assigned
B: *Azacitidine:* Standard dosing of 75 mg/m2 given over 7 of the first 10 days of the cycle
a: Albumin, alkaline phosphatase, total bilirubin, bicarbonate, BUN, calcium, chloride, creatinine, glucose, phosphorus, potassium, total protein, SGOT [AST], SGPT [ALT], sodium, magnesium.
c: Serum and marrow 5-aza-2'-deoxycytidine genomic incorporation and DNA demethylation samples must be collected in green-top vacutainer containing THU

‡: Bone marrow tests should be performed at the end of cycle 1, end of cycle 3, and end of cycle 6, prior to the start of the next cycle of azacitidine. These tests can be performed on the first day of the following cycle, prior to the start of azacitidine therapy.
*: Bone marrow tests should be done at the end of cycle 6. If a patient receives fewer than 6 cycles or more than 6 cycles, bone marrow tests should be done at the time of therapy cessation as long as at least one month has elapsed since the prior bone marrow test.
#: For patients who continue on pinometostat beyond 6 months, they should follow the same evaluation schedule as outlined for cycle 2.

12. MEASUREMENT OF EFFECT

Although the clinical benefit of these drugs has not yet been established, the intent of offering this treatment is to provide a possible therapeutic benefit, and thus the patient will be carefully monitored for tumor response and symptom relief in addition to safety and tolerability. Patients with measurable disease will be assessed by standard criteria. For the purposes of this study, patients should be re-evaluated at the end of cycle 1, end of cycle 3, and end of cycle 6.

12.1 Antitumor Effect

For the purposes of this study, patients should be re-evaluated for response at the end of cycle 1, end of cycle 3, and end of cycle 6.

Response and progression will be evaluated in this study using the 2017 European Leukemia Net Guidelines[19].

12.1.1 Definitions

Evaluable for toxicity. All patients will be evaluable for toxicity from the time of their first treatment with pinometostat.

Evaluable for objective response. Only those patients who have measurable disease present at baseline, have received at least one cycle of therapy, and have had their disease re-evaluated will be considered evaluable for response. These patients will have their response classified according to the definitions stated below. (Note: Patients who exhibit objective disease progression prior to the end of cycle 1 will also be considered evaluable.)

12.1.2 Disease Parameters

Measurable disease. For previously untreated patients, the presence of at least 20% blasts in the peripheral blood on a complete blood count differential or on a bone marrow aspirate differential, with cytogenetics or FISH studies showing a translocation or cytogenetics or FISH or a myeloid mutational panel showing partial tandem duplication of the 11q23 / MLL locus would qualify as measurable disease. For patients with relapsed / refractory disease, any abnormal disease burden as detected by morphology, flow cytometry, cytogenetics, or molecular testing which has a detectable 11q23 rearrangement or PTD would qualify as having measurable disease. Patients in either setting with a myeloid sarcoma or extramedullary involvement with detectable 11q23 rearrangement / PTD would also qualify as having measurable disease. Diagnosis should be confirmed with a bone marrow biopsy within 14 days prior to the start of therapy.

Measurable extramedullary lesions are defined as those that can be accurately measured in at least one dimension (longest diameter to be recorded) as ≥20 mm (≥2 cm) by chest x-ray or as ≥10 mm (≥1 cm) with CT scan, MRI, or calipers by clinical exam. All tumor measurements must be recorded in millimeters (or decimal fractions of centimeters).

Note: Tumor lesions that are situated in a previously irradiated area might or might not be considered measurable. *If the investigator thinks it appropriate to include them, the conditions under which such lesions should be considered must be defined in the protocol*.

Malignant lymph nodes. To be considered pathologically enlarged and measurable, a lymph node must be ≥15 mm (≥1.5 cm) in short axis when assessed by CT scan (CT scan slice thickness recommended to be no greater than 5 mm [0.5 cm]). At baseline and in follow-up, only the short axis will be measured and followed.

12.1.3 Methods for Evaluation of Measurable Disease

Hematopathology – The percentage of blasts found on the aspirate differential as read by the hematopathologist at the enrolling institution will be used to evaluate response.

Peripheral Blood CBC and Differential – The ANC, platelet count, and differential, including blast percentage from both automated and manual differentials, will be used. If both an automated and manual differential are performed, the latter will be used to evaluate the peripheral blast percentage.

FDG-PET - While FDG-PET response assessments need additional study, it is sometimes reasonable to incorporate the use of FDG-PET scanning to assess diagnosis and progression (particularly possible 'new' disease). This would only be applicable to patients with extramedullary disease. New lesions on the basis of FDG-PET imaging can be identified according to the following algorithm:

a. Negative FDG-PET at baseline, with a positive FDG-PET at follow-up is a sign of PD based on a new lesion.
b. No FDG-PET at baseline and a positive FDG-PET at follow-up: If the positive FDG-PET at follow-up corresponds to a new site of disease, this area should be biopsied to determine if it represents progression of disease. Such cases should be discussed with the medical monitor.
c. FDG-PET may be used to upgrade a response to a CR for a patient with extramedullary disease, no bone marrow involvement, and no other means by which to evaluate their disease involvement (e.g. physical exam) in a manner similar to a biopsy in cases where a residual radiographic abnormality is thought to represent fibrosis or scarring. However, it must be acknowledged that such an approach may lead to false positive CR due to limitations of FDG-PET and biopsy resolution/sensitivity.

Note: A ‘positive’ FDG-PET scan lesion means one which is FDG avid with an uptake greater than twice that of the surrounding tissue on the attenuation corrected image.

12.1.4 Response Criteria

12.1.4.1 Evaluation of Response as per 2017 ELN Guidelines[19]

Complete Remission (CR): Bone marrow blasts < 5%; no circulating blasts, no blasts with Auer rods, no extramedullary disease; ANC > 1000 per microliter, platelets > 100,000 per microliter

Complete Remission with Incomplete Count Recovery(CRi): Bone marrow blasts < 5%; no circulating blasts, no blasts with Auer rods, no extramedullary disease; ANC ≤ 1000 per microliter and / or platelets ≤ 100,000 per microliter

Morphologic Leukemia-Free State (MLFS): Bone marrow blasts < 5%; no blasts with Auer rods, no extramedullary disease; no hematologic recovery required

Partial Remission (PR): All hematologic criteria of CR; decrease in bone marrow blasts to 5-25%; decrease in pre-treatment bone marrow blasts by at least 50%

Progressive Disease (PD): Evidence for an increase in bone marrow blast percentage and/or increase of absolute blast counts in the blood:

- > 50% increase in marrow blasts over baseline (a minimum 15% point increase is required in cases with < 30% blasts at baseline; or persistent marrow blast percentage of >70% over at least 3 mo; without at least a 100% improvement in ANC to an absolute level (>0.5 x 10^9 /L [500/μL], and/or platelet count to >50 x 10^9 /L [50,000/μL] nontransfused); or
- > 50% increase in peripheral blasts (WBC x % blasts) to >25 x 10^9 /L (>25 000/μL) (in the absence of differentiation syndrome); or
- New extramedullary disease

Stable Disease (SD): Absence of CR, CRi, MLFS, PR; criteria for progressive disease not met

12.1.4.2 Evaluation of Non-Target Lesions

N/A

12.1.4.3 Evaluation of Best Overall Response

The best overall response is the best response recorded from the start of the treatment until disease progression/recurrence (taking as reference for progressive disease the smallest measurements recorded since the treatment started). The patient's best response assignment will depend on the achievement of both measurement and confirmation criteria.
The overall response rate includes achievement of CR, CRi, MLFS, and PR.

12.1.5 Duration of Response

Duration of overall response: The duration of overall response is measured from the time measurement criteria are met for CR or PR (whichever is first recorded) until the first date that recurrent or progressive disease is objectively documented (taking as reference

for progressive disease the smallest measurements recorded since the treatment started).

The duration of overall CR is measured from the time measurement criteria are first met for CR until the first date that progressive disease is objectively documented.

Duration of stable disease: Stable disease is measured from the start of the treatment until the criteria for progression are met, taking as reference the smallest measurements recorded since the treatment started, including the baseline measurements.

12.1.6 Progression-Free Survival

Progression-free survival (PFS) will be assessed, defined as the duration of time from start of treatment to time of progression or death, whichever occurs first.

12.1.7 Response Review

All responses will be reviewed by an independent hematopathologist at the study's completion.

12.2 Antitumor Effect – Hematologic Tumors

See above

12.3 Other Response Parameters

N/A

13. STUDY OVERSIGHT AND DATA REPORTING / REGULATORY REQUIREMENTS

Adverse event lists, guidelines, and instructions for AE reporting can be found in Section 10 (Adverse Events: List and Reporting Requirements).

13.1 Study Oversight

This protocol is monitored at several levels, as described in this section. The Protocol Principal Investigator is responsible for monitoring the conduct and progress of the clinical trial, including the ongoing review of accrual, patient-specific clinical and laboratory data, and routine and serious adverse events; reporting of expedited adverse events; and accumulation of reported adverse events from other trials testing the same drug(s). The Protocol Principal Investigator and statistician have access to the data at all times through the CTMS web-based reporting portal. For the Phase 1 portion of this study, all decisions regarding dose escalation/expansion/de-escalation require sign-off by the Protocol Principal Investigator through the CTMS/IWRS. In addition, for the Phase 1 portion, the Protocol Principal Investigator will have at least monthly, or more frequently, conference calls with the Study Investigators and the CTEP Medical Officer(s) to review accrual, progress, and adverse events and unanticipated problems.

For a Phase 1/2 trial, enrollment to the Phase 2 portion of the trial will not begin until a protocol amendment has been submitted which summarizes the Phase 1 results, the recommended Phase 2 dose, and the rationale for selecting it. The amendment must be reviewed and approved by CTEP before enrollment to the Phase 2 portion can begin.

During the Phase 2 portion of the study, the Protocol Principal Investigator will have, at a minimum, quarterly conference calls with the Study Investigators and the CTEP Medical Officer(s) to review accrual, progress, and pharmacovigilance. Decisions to proceed to the second stage of a Phase 2 trial will require sign-off by the Protocol Principal Investigator and the Protocol Statistician.

All Study Investigators at participating sites who register/enroll patients on a given protocol are responsible for timely submission of data via Medidata Rave and timely reporting of adverse events for that particular study. This includes timely review of data collected on the electronic CRFs submitted via Medidata Rave.

All studies are also reviewed in accordance with the enrolling institution's data safety monitoring plan. A central data safety monitoring board consisting of study team members will hold a conference, approximately once per week, to discuss safety issues that arise with the trial.

13.2 Data Reporting

Medidata Rave is a clinical data management system being used for data collection for this trial/study. Access to the trial in Rave is controlled through the CTEP-IAM system and role assignments. To access Rave via iMedidata:

- Site staff will need to be registered with CTEP and have a valid and active CTEP-IAM account, and
- Assigned one of the following Rave roles on the relevant Lead Protocol Organization (LPO) or Participating Organization roster at the enrolling site: Rave CRA, Rave Read Only, Rave CRA (LabAdmin), Rave SLA, or Rave Investigator. Refer to https://ctep.cancer.gov/investigatorResources/default.htm for registration types and documentation required.
    - To hold Rave CRA or Rave CRA (Lab Admin) role, site staff must hold a minimum of an AP registration type,
    - To hold Rave Investigator role, the individual must be registered as an NPIVR or IVR, and
    - To hold Rave Read Only role, site staff must hold an Associates (A) registration type.
- Upon initial site registration approval for the study in Regulatory Support System (RSS), all persons with Rave roles assigned on the appropriate roster will be sent a study invitation e-mail from iMedidata. To accept the invitation, site staff must log in to the Select Login (https://login.imedidata.com/selectlogin) using their CTEP-IAM username and password, and click on the *accept* link in the upper right-corner of the iMedidata page. Site staff 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 iMedidata screen. If an eLearning is required and has not yet been taken, the link to the eLearning will appear under the study name in iMedidata instead of the *Rave EDC* link; once the successful completion of the eLearning has been recorded, access to the study in Rave will be granted, and a *Rave EDC* link will display under the study name.

Site staff that have not previously activated their iMedidata/Rave account at the time of initial site registration approval for the study in RSS will also receive a separate invitation from iMedidata to activate their account. Account activation instructions are located on the CTSU website in the Rave section under the Rave resource materials (Medidata Account Activation and Study Invitation Acceptance). Additional information on iMedidata/Rave is available on the CTSU members' website in the Data Management > Rave section 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.

13.2.1 Method

This study will be monitored by the Clinical Trials Monitoring Service (CTMS). Data will be submitted to CTMS at least once every two weeks via Medidata Rave (or other modality if approved by CTEP). Information on CTMS reporting is available at http://www.theradex.com/clinicalTechnologies/?National-Cancer-Institute-NCI-11. On-site audits will be conducted three times annually (one annual site visit and two data audits). For CTMS monitored studies, after users have activated their accounts, please contact the Theradex Help Desk at (609) 799-7580 or by email at CTMSSupport@theradex.com for additional support with Rave and completion of CRFs.

13.2.2 Responsibility for Data Submission

For ETCTN trials, it is the responsibility of the PI(s) at the site to ensure that all investigators at the ETCTN Sites understand the procedures for data submission for each ETCTN protocol and that protocol specified data are submitted accurately and in a timely manner to the CTMS via the electronic data capture system, Medidata Rave.

Data are to be submitted via Medidata Rave to CTMS on a real-time basis, but no less than once every 2 weeks. The timeliness of data submissions and timeliness in resolving data queries will be tracked by CTMS. Metrics for timeliness will be followed and assessed on a quarterly basis. For the purpose of Institutional Performance Monitoring, data will be considered delinquent if it is greater than 4 weeks past due.

Data from Medidata Rave and CTEP-AERS is reviewed by the CTMS on an ongoing basis as data is received. Queries will be issued by CTMS directly within Rave. The queries will appear on the Task Summary Tab within Rave for the CRA at the ETCTN to resolve. Monthly web-based reports are posted for review by the Drug Monitors in the IDB, CTEP. Onsite audits will be conducted by the CTMS to ensure compliance with regulatory requirements, GCP, and NCI policies and procedures with the overarching goal of ensuring the integrity of data generated from NCI-sponsored clinical trials, as described in the ETCTN Program Guidelines, which may be found on the CTEP (http://ctep.cancer.gov/protocolDevelopment/electronic_applications/adverse_events.htm) and CTSU websites.

An End of Study CRF is to be completed by the PI, and is to include a summary of study endpoints not otherwise captured in the database, such as (for phase 1 trials) the recommended phase 2 dose (RP2D) and a description of any dose-limiting toxicities (DLTs). CTMS will utilize a core set of eCRFs that are Cancer Data Standards Registry and Repository (caDSR) compliant (http://cbiit.nci.nih.gov/ncip/biomedical-informatics-resources/interoperability-and-semantics/metadata-and-models). Customized eCRFs will be included when appropriate to meet unique study requirements. The PI is encouraged to review the eCRFs, working closely with CTMS to ensure prospectively that all required items are appropriately captured in the eCRFs prior to study activation. CTMS will prepare the eCRFs with built-in edit checks to the extent possible to promote data integrity.

CDUS data submissions for ETCTN trials activated after March 1, 2014, will be carried out by the CTMS contractor, Theradex. CDUS submissions are performed by Theradex on a monthly basis. The trial's lead institution is responsible for timely submission to CTMS via Rave, as above.
Further information on data submission procedures can be found in the ETCTN Program Guidelines
(http://ctep.cancer.gov/protocolDevelopment/electronic_applications/adverse_events.htm).

13.3 Data Quality Portal

The Data Quality Portal (DQP) provides a central location for site staff to manage unanswered queries and form delinquencies, monitor data quality and timeliness, generate reports, and review metrics.

The DQP is located on the CTSU members' website under Data Management. The Rave Home section displays a table providing summary counts of Total Delinquencies and Total Queries. DQP Queries, DQP Delinquent Forms, and the DQP Reports modules are available to access details and reports of unanswered queries, delinquent forms, and timeliness reports. Review the DQP modules on a regular basis to manage specified queries and delinquent forms.

The DQP is accessible by site staff that are rostered to a site and have access to the CTSU website. Staff that have Rave study access can access the Rave study data using a direct link on the DQP.

To learn more about DQP use and access, click on the Help icon displayed on the Rave Home, DQP Queries, and DQP Delinquent Forms modules.

Note: Some Rave protocols may not have delinquent form details or reports specified on the DQP. A protocol must have the Calendar functionality implemented in Rave by the Lead Protocol Organization (LPO) for delinquent form details and report to be available on the DQP. Site staff should contact the LPO Data Manager for their protocol regarding questions about Rave Calendering functionality.

13.4 CTEP Multicenter Guidelines

N/A

13.5 Collaborative Agreements Language

The agent(s) supplied by CTEP, DCTD, NCI used in this protocol is/are provided to the NCI under a Collaborative Agreement (CRADA, CTA, CSA) between the Pharmaceutical Company(ies) (hereinafter referred to as "Collaborator(s)") and the NCI Division of Cancer Treatment and Diagnosis. Therefore, the following obligations/guidelines, in addition to the provisions in the "Intellectual Property Option to Collaborator" (http://ctep.cancer.gov/industryCollaborations2/intellectual_property.htm) contained within the terms of award, apply to the use of the Agent(s) in this study:

1. Agent(s) may not be used for any purpose outside the scope of this protocol, nor can Agent(s) be transferred or licensed to any party not participating in the clinical study. Collaborator(s) data for Agent(s) are confidential and proprietary to Collaborator(s) and shall be maintained as such by the investigators. The protocol documents for studies utilizing Agents contain confidential information and should not be shared or distributed without the

permission of the NCI. If a copy of this protocol is requested by a patient or patient's family member participating on the study, the individual should sign a confidentiality agreement. A suitable model agreement can be downloaded from: http://ctep.cancer.gov.

2. For a clinical protocol where there is an investigational Agent used in combination with (an)other Agent(s), each the subject of different Collaborative Agreements, the access to and use of data by each Collaborator shall be as follows (data pertaining to such combination use shall hereinafter be referred to as "Multi-Party Data"):

   a. NCI will provide all Collaborators with prior written notice regarding the existence and nature of any agreements governing their collaboration with NCI, the design of the proposed combination protocol, and the existence of any obligations that would tend to restrict NCI's participation in the proposed combination protocol.

   b. Each Collaborator shall agree to permit use of the Multi-Party Data from the clinical trial by any other Collaborator solely to the extent necessary to allow said other Collaborator to develop, obtain regulatory approval or commercialize its own Agent.

   c. Any Collaborator having the right to use the Multi-Party Data from these trials must agree in writing prior to the commencement of the trials that it will use the Multi-Party Data solely for development, regulatory approval, and commercialization of its own Agent.

3. Clinical Trial Data and Results and Raw Data developed under a Collaborative Agreement will be made available to Collaborator(s), the NCI, and the FDA, as appropriate and unless additional disclosure is required by law or court order as described in the IP Option to Collaborator (http://ctep.cancer.gov/industryCollaborations2/intellectual_property.htm). Additionally, all Clinical Data and Results and Raw Data will be collected, used and disclosed consistent with all applicable federal statutes and regulations for the protection of human subjects, including, if applicable, the *Standards for Privacy of Individually Identifiable Health Information* set forth in 45 C.F.R. Part 164.

4. When a Collaborator wishes to initiate a data request, the request should first be sent to the NCI, who will then notify the appropriate investigators (Group Chair for Cooperative Group studies, or PI for other studies) of Collaborator's wish to contact them.

5. Any data provided to Collaborator(s) for Phase 3 studies must be in accordance with the guidelines and policies of the responsible Data Monitoring Committee (DMC), if there is a DMC for this clinical trial.

6. Any manuscripts reporting the results of this clinical trial must be provided to CTEP by the Group office for Cooperative Group studies or by the principal investigator for non-Cooperative Group studies for immediate delivery to Collaborator(s) for advisory review and comment prior to submission for publication. Collaborator(s) will have 30 days from the date of receipt for review. Collaborator shall have the right to request that publication be delayed for up to an additional 30 days in order to ensure that Collaborator's confidential and

proprietary data, in addition to Collaborator(s)'s intellectual property rights, are protected. Copies of abstracts must be provided to CTEP for forwarding to Collaborator(s) for courtesy review as soon as possible and preferably at least three (3) days prior to submission, but in any case, prior to presentation at the meeting or publication in the proceedings. Press releases and other media presentations must also be forwarded to CTEP prior to release. Copies of any manuscript, abstract and/or press release/ media presentation should be sent to:

Email: ncicteppubs@mail.nih.gov

The Regulatory Affairs Branch will then distribute them to Collaborator(s). No publication, manuscript or other form of public disclosure shall contain any of Collaborator's confidential/ proprietary information.

13.6 Genomic Data Sharing Plan

Genomic data will be generated from samples centrally sequenced at MSKCC. Data pertinent to the interpretation of genomic data, including the minimal phenotype information needed to reproduce the primary analysis —such as associated phenotype data (e.g., clinical information), exposure data, relevant metadata, and descriptive information (e.g., protocols or methodologies used)—will be collected from participating sites and incorporated into the analysis of genomic data. The investigators and statistician and/or bioinformaticians for a study will have access to all data on mutations and variants. This information will be sequestered from access throughout the study until it is analyzed for purposes of reporting and publishing of the study results. As specified in the CRADA for the agents used in the clinical study, the pharmaceutical collaborator will have at least 6 months, longer if needed for a regulatory filing, to review the data and or receive copies of the data once the study is completed and analyzed, or sooner, if specified for purposes of generating Intellectual Property. Once these timeframes have been exceeded, the data will be available through a Data Access Committee (DAC) in the GDC following NCI and Collaborator review of the proposals.

13.7 Incidental/Secondary Findings Disclosure Procedure

This protocol involves genomic testing of the tumor sample, as well as of the patient's normal DNA. A myeloid genomic panel should be performed at the time of diagnosis and results should be relayed to the patient and their family as per local institutional practice. Paired analysis of tumors and patient-matched normal samples will be performed for the sole purpose of unambiguous detection of somatic mutations. Germline DNA will not be annotated for pathogenic mutations. Testing of germline DNA will not be conducted in a CLIA lab but instead as part of the IWG myeloid research panel and, as such, results will not be returned to the patient.

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APPENDIX A PERFORMANCE STATUS CRITERIA

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

APPENDIX B BIOASSAY TEMPLATES

Biomarker Name AND Lab PI and Site	**Assay (CLIA: Y/N)**	**Use (Integral, Integrated, or Exploratory) AND Purpose**	**Tissue/Body Fluid Tested and Timing of Sample Collection**	**M/O**	**Use of NCI Resources (No / Pending Approval / Approved)**	**Funding Source(s)**
11q23 rearrangement or partial tandem duplication Eytan Stein / Memorial Sloan-Kettering Cancer Center (MSKCC) Lab PI email:	Cytogenetics and FISH CLIA: Y **BRC Review: No**	Integral Evaluate eligibility	Bone marrow aspirate, peripheral blood Prior to enrollment and therapy cessation	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	CTEP CRADA
Myeloid mutational panel for no targetable mutations Eytan Stein / Memorial Sloan-Kettering Cancer Center (MSKCC) Lab PI email:	Myeloid Mutations CLIA: Y **BRC Review: No**	Integral Evaluate eligibility	Bone marrow aspirate Prior to enrollment and therapy cessation	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	CTEP CRADA
Myeloid differentiation by Flow/FISH Eytan Stein / Memorial Sloan-Kettering Cancer Center (MSKCC) Lab PI email:	FACS, Flow cytometry, FISH CLIA: N **BRC Review: Yes** **Submission Deadline:**	Integrated Evaluate differentiation of cells with treatment	Bone marrow aspirate, peripheral blood End of cycle 1, end of cycle 3, and therapy cessation	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	CTEP CRADA
H3K79 ELISA Scott Armstrong / Dana-Farber Cancer Institute (DFCI) Lab PI email:	H3K79 ELISA CLIA: N **BRC Review: No**	Integrated Quantitate methylation	Bone marrow aspirate, peripheral blood Prior to enrollment, end of cycle 1, end of cycle 3, and therapy cessation	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	CTEP CRADA
qPCR for HOXA9 and Meis1 Expression Scott Armstrong / Dana-Farber Cancer Institute (DFCI) Lab PI email:	qPCR CLIA: N **BRC Review: No**	Integrated Quantitate gene expression	Bone marrow aspirate, peripheral blood Prior to enrollment, end of cycle 1, end of cycle 3, and therapy cessation	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	CTEP CRADA
MRD Negativity by Flow Cytometry Eytan Stein / Memorial Sloan-Kettering Cancer Center (MSKCC) Lab PI email:	Flow cytometry CLIA: Y **BRC Review: No**	Integrated Evaluate response to therapy	Bone marrow aspirate End of cycle 1, end of cycle 3, and therapy cessation	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	CTEP CRADA
Azacitidine PK profile Michelle A. Rudek / Johns Hopkins Lab PI email:	LC-MS/MS CLIA: N GLP: Y **BRC Review: No**	Integrated Pharmacokinetic profile of azacitidine alone	Blood with THU to stabilize azacitidine C1D1 pre, and 0.25, 0.5, and 1 hr post	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	UM1 for sample processing; CTEP / NIH / foundation grants for PK assessment
Cell storage / DNA storage Eytan Stein / Memorial Sloan-Kettering Cancer Center (MSKCC) Lab PI email:	Cell storage / DNA storage CLIA: N **BRC Review: No**	Exploratory Evaluate valence of methylation in future	Bone marrow aspirate, peripheral blood Prior to enrollment, end of cycle 1, end of cycle 3, and therapy cessation; nail clippings only prior to enrollment	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	CTEP CRADA

Biomarker Name AND Lab PI and Site	**Assay (CLIA: Y/N)**	**Use (Integral, Integrated, or Exploratory) AND Purpose**	**Tissue/Body Fluid Tested and Timing of Sample Collection**	**M/O**	**Use of NCI Resources (No / Pending Approval / Approved)**	**Funding Source(s)**
5-aza-2'-deoxycytidine Genomic Incorporation (from azacitidine) and DNA demethylation Michelle A. Rudek / Johns Hopkins Lab PI email: mrudek2@jhmi.edu	LC-MS/MS CLIA: N GLP: Y **BRC Review: No**	Exploratory Pharmacodynamic profile of azacitidine conversion to 5-aza-2'-deoxycytidine triphosphate and incorporation into DNA and its effect on DNA demethylation	PBMC and bone marrow aspirate PBMC: C1D8; Bone marrow aspirate: End of cycle 1, end of cycle 3, and therapy cessation	M	ETCTN Tissue Bank: No CIMAC: No MoCha: No PADIS: No	UM1 for sample processing and analysis

BRC Document Submission Deadline Key:
A = Documents should be submitted to PIO as soon as possible but **not later than 4 weeks** upon receipt of this consensus review. **BRC approval is required prior to LOI approval for this study.**
B = Documents should be submitted to PIO as soon as possible but **not later than 4 weeks** upon receipt of this consensus review. **BRC approval is required prior to protocol approval** in order to allow sufficient time for BRC review and to comply with OEWG deadlines.
C = Documents should be submitted to PIO as soon as possible but **not later than 4 weeks** upon receipt of this consensus review. **BRC approval is required prior to protocol activation** in order to allow sufficient time for BRC review and to comply with OEWG deadlines.
D = Documents should be submitted to PIO as soon as ready, but no later than the protocol submission.

APPENDIX C: PATIENT DRUG INFORMATION HANDOUT AND WALLET CARD

Information for Patients, Their Caregivers and Non-Study Healthcare Team on Possible Interactions with Other Drugs and Herbal Supplements

The patient _____ is enrolled on a clinical trial using the experimental study drug, **Pinometostat**. This clinical trial is sponsored by the National Cancer Institute. This form is addressed to the patient, but includes important information for others who care for this patient.

These are the things that you as a healthcare provider need to know:

Pinometostat interacts with certain specific enzyme(s) in your liver, certain transport proteins that help move drugs in and out of cells.

- The enzymes in question are **CYP3A4/5 and 2C19**. Pinometostat is broken down by these enzymes and may be affected by other drugs that inhibit or induce these enzymes. Pinometostat also inhibits CYP3A4/5 and must be used carefully with other drugs that are broken down by these enzymes.
- The proteins in question are **OATP1B3, P-gp, MATE1 and MATE2-K**. Pinometostat is moved in and out of cells/organs by OATP1B3, P-gp, MATE1, and MATE2-K and may be affected by other drugs that inhibit or induce these proteins. Pinometostat also inhibits MATE1 and MATE2-K and must be used carefully with other drugs that are moved in and out of cells/organs by these proteins.

To the patient: Take this paper with you to your medical appointments and keep the attached information card in your wallet.

Pinometostat may interact with other drugs which can cause side effects. For this reason, it is very important to tell your study doctors of any medicines you are taking before you enroll onto this clinical trial. It is also very important to tell your doctors if you stop taking any regular medicines, or if you start taking a new medicine while you take part in this study. When you talk about your current medications with your doctors, include medicine you buy without a prescription (over-the-counter remedy), or any herbal supplements such as St. John's Wort. It is helpful to bring your medication bottles or an updated medication list with you.

Many health care providers can write prescriptions. You must tell all of your health care providers (doctors, physician assistants, nurse practitioners, pharmacists) you are taking part in a clinical trial.

These are the things that you and they need to know:

Pinometostat must be used very carefully with other medicines that use certain **liver enzymes or transport proteins to be effective or to be cleared from your system**. Before you enroll onto the clinical trial, your study doctor will work with your regular health care providers to review any medicines and herbal supplements that are considered **strong inducers/inhibitors or substrates of CYP3A4/5, transport proteins MATE1 or MATE2-K.**

- Please be very careful! Over-the-counter drugs (including herbal supplements) may contain ingredients that could interact with your study drug. Speak to your doctors or pharmacist to determine if there could be any side effects.
- Your regular health care provider should check a frequently updated medical reference or call your study doctor before prescribing any new medicine or discontinuing any medicine. Your study doctor's name is _____ and he or she can be contacted at _____.

STUDY DRUG INFORMATION WALLET CARD	
You are enrolled on a clinical trial using the experimental study drug **pinometostat.** This clinical trial is sponsored by the NCI. **Pinometosta** may interact with drugs that are **processed by your liver, or use certain transport proteins in your body**. Because of this, it is very important to: ➢ Tell your doctors if you stop taking any medicines or if you start taking any new medicines. ➢ Tell all of your health care providers (doctors, physician assistants, nurse practitioners, or pharmacists) that you are taking part in a clinical trial. ➢ Check with your doctor or pharmacist whenever you need to use an over-the-counter medicine or herbal supplement.	**Pinometostat** interacts with **specific liver enzymes called CYP3A4/5, transport protein MATE1 and MATE2-K,** and must be used very carefully with other medicines that interact with **these enzymes, transporters, or agent**. ➢ Before you enroll onto the clinical trial, your study doctor will work with your regular health care providers to review any medicines and herbal supplements that are considered **strong inducers/inhibitors or substrates of CYP3A4/5 or transporters MATE1 and MATE2-K.** ➢ Before prescribing new medicines, your regular health care providers should go to a frequently-updated medical reference for a list of drugs to avoid, or contact your study doctor. ➢ Your study doctor's name is _____ and can be contacted at _____.