Title: **A Phase II Study of the Aurora A Kinase Inhibitor Alisertib in Combination with 7+3 Induction Chemotherapy in Patients with High-risk Acute Myeloid Leukemia**

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TITLE: A phase II study of the aurora A kinase inhibitor alisertib in combination with 7+3 induction chemotherapy in patients with high-risk acute myeloid leukemia

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NCI-Supplied Agent(s): N/A

Other Agent(s): Alisertib, MLN8237, Takeda Pharmaceuticals
Cytarabine, commercial
Idarubicin hydrochloride, commercial
Daunorubicin, commercial

IND #: 117653

IND Sponsor: Massachusetts General Hospital Cancer Center

SYNOPSIS AND SCHEMA

Patient Population: Adults, over age 18, with newly diagnosed, pathologically-confirmed acute myeloid leukemia (AML), eligible for induction chemotherapy, and with higher-risk disease, as defined by at least one of the following; a) age ≥ 65, b) association with poor-risk karyotype, c) secondary AML arising out of antecedent myeloid malignancy, d) or due to prior radiation or chemotherapy.

Performance Status: ECOG 0-2

Primary Objective:

- To determine the rates of complete remission (CR) and complete remission with incomplete count recovery (CRi) in higher risk patients receiving alisertib in combination 7+3 induction chemotherapy, and to assess whether this is higher than the historical rates seen in this population with 7+3 induction alone.

Secondary Objectives:

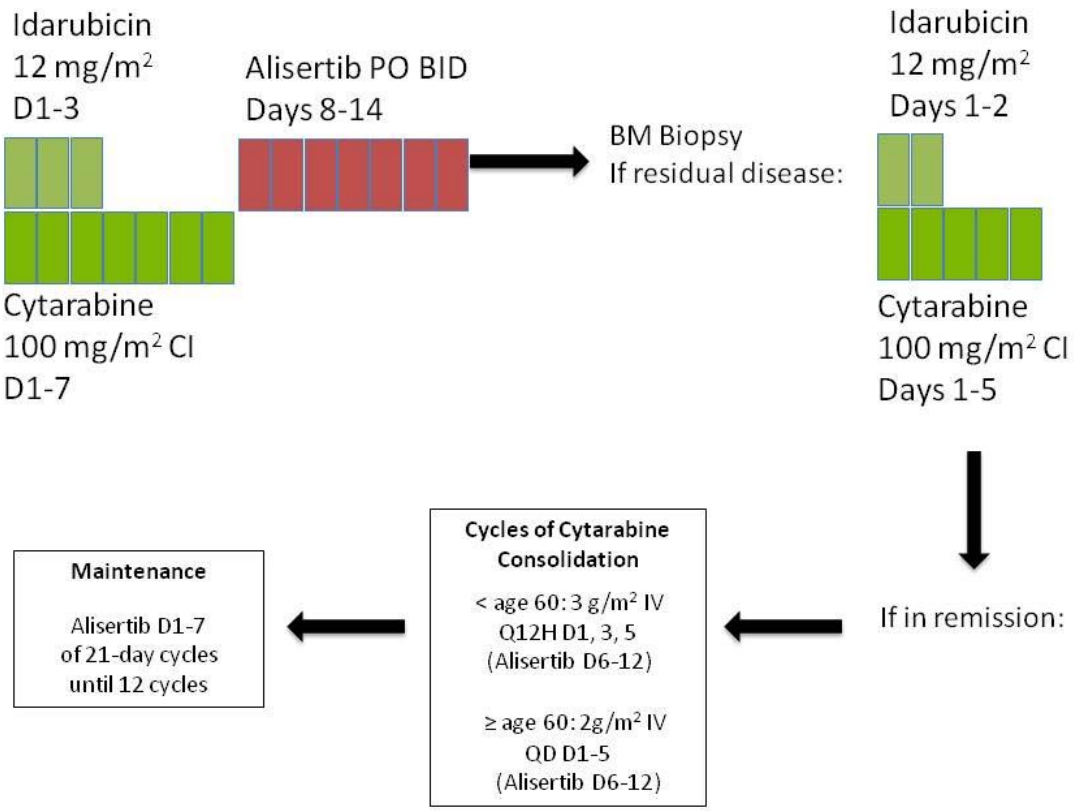
- To determine if the 1-year overall survival (OS) rate, relapse free survival (RFS), remission duration of higher risk patients receiving alisertib in combination with conventional intensive cytotoxic chemotherapy, and to assess whether this is higher than the historical rates seen in this population with conventional intensive chemotherapy alone.
- To describe the frequency and severity of adverse events for patients treated on this study.
- To describe the interaction of pretreatment disease and patient characteristics including morphology, cytogenetics, immunophenotype, molecular/genetic features, WBC count and hemogram, and performance status on clinical outcomes.
- To describe the pharmacodynamic effects of alisertib during treatment with this induction combination

Phase of Study: II

Treatment Plan:
This is a phase II clinical study trial to assess the efficacy of the aurora A kinase alisertib in combination with 7+3 induction chemotherapy in patients with higher-risk newly diagnosed AML. On day 1 of the trial, all enrolled participants will be initiated on induction chemotherapy with conventional AML induction chemotherapy using the "7+3" induction regimen, consisting of cyatarabine continuous intravenous infusion for seven days and concurrent idarubicin (or daunorubicin if appropriate) as an intravenous bolus for first three days of induction. Oral administration with alisertib, at a dose of 30mg twice daily, will begin on day 8, and will continue for 7 days after conclusion of IV infusion of induction chemotherapy. The exception will be if 7+3

therapy begins in the afternoon or evening, and finishes on Day 8, in which case alisertib may be first given in the morning of Day 9. During induction, patients who have residual disease at day 14 may have re-induction with "5+2" chemotherapy, but they will not receive additional dosing of alisertib at that time. Toxicity will be assessed throughout the course of the study, until the final study visit. Pharmacodynamic evaluation will be performed prior to treatment, at day 14 of induction, at count recovery following induction, and at any time of relapse or suspected relapse. Following count recovery after induction, if patients proceed to consolidative chemotherapy with cytarabine, they will receive alisertib during consolidation at day 6 following conclusion of cytarabine administration, and will continue for 7 days. Upon count recovery, following consolidation cycles of therapy, maintenance cycles of alisertib will be started for 7 days, followed by 14 days off, and will be continued in this manner as 21-day cycles, for up to 12 cycles, until disease progression, or until onset of unacceptable toxicity. We will plan to follow patients for 12 months after the last patient enrollment.

Study flowchart



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1. OBJECTIVES

1.1 Study Design

This study is a Phase II study which seeks to assess the efficacy of the aurora A kinase inhibitor, alisertib, combined with standard "7+3" induction chemotherapy for patients with newly-diagnosed, higher risk acute myeloid leukemia (AML). Oral administration with alisertib, at a dose of 30mg twice daily, will begin following conclusion of cytarabine continuous infusion, and will continue for 7 days during induction chemotherapy. Toxicity will be assessed throughout the course of the study, until the final study visit. Pharmacodynamic evaluation will be performed prior to treatment, at day 14 of induction, at count recovery following induction, and at the time of relapse or suspected relapse. Following count recovery after induction, if patients proceed to consolidative chemotherapy with cytarabine, they will receive alisertib during consolidation at day 6 following conclusion of cytarabine administration, and will continue for 7 days. Upon count recovery, following consolidation cycles of therapy, maintenance cycles of alisertib will be started for 7 days, followed by 14 days off, and will be continued in this manner as 21-day cycles, for up to 12 cycles, until disease progression, or until onset of unacceptable toxicity.

1.2 Primary Objectives

- To determine the rates of complete remission (CR), complete remission with incomplete count recovery (CRi) in higher risk patients receiving alisertib in combination 7+3 induction chemotherapy, and to assess whether this is higher than the historical rates seen in this population with 7+3 induction alone.

1.3 Secondary Objectives

- To determine if the 1-year overall survival (OS) rate, relapse free survival (RFS), remission duration of higher risk patients receiving alisertib in combination with chemotherapy, and to assess whether this is higher than the historical rates seen in this population with conventional intensive chemotherapy alone.
- To describe the frequency and severity of adverse events for patients treated on this study.
- To describe the interaction of pretreatment disease and patient characteristics including morphology, cytogenetics, immunophenotype, molecular/genetic features, WBC count and hemogram, and performance status on clinical outcomes.
- To describe the pharmacodynamic effects of alisertib during treatment with this induction combination

2. BACKGROUND

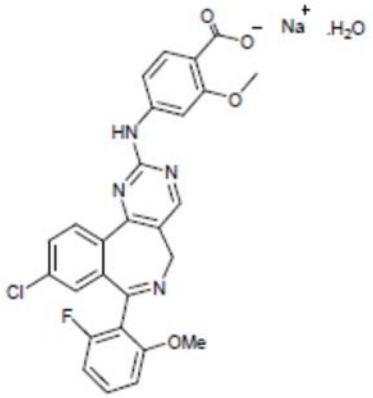
2.1 Alisertib (MLN8237)

2.1.1 Alisertib: structure and chemical properties

Alisertib is a selective small molecule inhibitor of Aurora A kinase that is being developed for the treatment of advanced malignancies. Information regarding the chemical name, structure, and properties of MLN8237 is listed below, as is the structure of MLN8237.

Research Name	MLN8237-004 ML00653668
Chemical Name	sodium 4- -2- methoxybenzoate hydrate
Common Name	sodium 4-{[9-chloro-7-(2-fluoro-6 methoxyphenyl) 5*H* pyrimido[5,4-d][2]benzazepin-2-yl]amino}-2-methoxybenzoate hydrate
Generic Name	Not available
Proprietary Name	Not available
USAN Name	alisertib (free acid), alisertib sodium
pINN Name	alisertib
CAS Registry Number	1208255-63-3
Classification	Aurora A kinase inhibitor
Molecular Formula	C27H19ClFN4NaO4.H2O
Molecular Weight	558.92 (sodium salt) 518.92 (free acid)

Structure of MLN8237-004



Physical, Chemical, and Pharmaceutical Properties

In its solid state, alisertib (MLN8237-004) drug substance is an off-white to yellow solid with an assay value of 96.0% to 104.0% (w/w) on an anhydrous basis, as determined by reverse phase high performance liquid chromatography (HPLC). The solubility of the drug substance in water (pH = 8.4) is 8.5mg/mL. The drug substance is a stable monohydrate with a pKa of 4.53 for the free acid.

Formulation

MLN8237-004 Enteric-Coated Tablets will be used.

- The MLN8237-004Tablet: An enteric coated tablet dosage form of MLN8237-004 has been developed for use in clinical studies. Three different enteric coated tablet dosage strengths, 10, 50, and 100 mg, expressed as MLN8237 free acid, were formulated. The key formulation excipients that aid in the in vivo absorption of the drug are sodium bicarbonate, sodium lauryl sulfate, and enteric coating. The other formulation excipients such as povidone, microcrystalline cellulose, croscarmellose sodium, and sodium stearyl fumarate serve as manufacturing aids.

Storage

- Tablet: MLN8237-004 enteric-coated tablets are packaged in HDPE bottles with rayon coil, induction seal, desiccant packs, and polypropylene child-resistant cap and stored at USP-controlled room temperature (20°C to 25ºC) with excursions permitted from 15°C to 30°C.

Handling

Alisertib (MLN8237-004) drug product is a cytotoxic anticancer drug. As with other potentially toxic compounds, caution should be exercised when handling the 3 dosage forms of MLN8237-004 (capsules, tablets, and oral solution). Refer to published guidelines regarding the proper handling and disposal of anticancer agents.[1, 2]

2.1.2 Alisertib (MLN8237): Pharmacology

2.1.2.1 *In Vitro* Pharmacology

The data from both enzymatic and cell-based assays demonstrated that MLN8237 is a selective and potent inhibitor of Aurora A kinase.[3-7] Nonclinical studies show that MLN8237 is an adenosine triphosphate (ATP)-competitive and reversible inhibitor of Aurora A kinase with an inhibition constant (Ki) of 0.43 nM.[3] In HCT-116 human colon cancer cells, MLN8237 inhibited Aurora A kinase activity with a half-maximal inhibitory concentration (IC50) of 6.7 nM, and was approximately 200-fold more selective for Aurora A than for Aurora B (IC50 = 1534 nM).[4] Moreover, MLN8237 was at least 250-fold more selective for Aurora A when compared to other kinases tested in vitro.[5] Consistent with the outcomes associated with Aurora kinase inhibition, an antimitotic agent, MLN8237 administration in vitro resulted in formation of abnormal mitotic spindles, an accumulation of mitotic cells, and a decrease in the proliferation of a broad range of tumor cell lines grown in culture.[4, 8-14]

2.1.2.2 *In Vivo* Pharmacology

Treatment of nude mice bearing human colon HCT-116 tumor xenografts with MLN8237 resulted in increased mitotic cells in the tumor, a phenotype that recapitulates the results in cultured cells.[15-17] In orally dosed mice bearing HCT-116 tumor xenografts, the duration of increased mitotic index was longer lasting in the more efficacious dose groups (10 and 30mg/kg) than the less efficacious dose group (3mg/kg). MLN8237 was efficacious against a variety of subcutaneous solid tumor xenograft models (breast, colon, lung, prostate and neuroblastoma) in mice when dosed orally for approximately 21 days.[18-23] Moreover, MLN8237 demonstrated robust antitumor activity in a number of hematologic malignancies, including diffuse large B cell lymphomas and acute lymphoblastic leukemias.[21, 22] The oral dose and schedule that was maximally efficacious varied for each model from 10 to 30 mg/kg QD in some models or 20 mg/kg BID in other models.[20, 22] Studies in the HCT-116 colon tumor model demonstrated that less frequent dosing (eg, 5 days-on followed by 5 days-off) was also efficacious.[20] Osmotic mini-pump studies in HCT-116 tumors showed that the pharmacodynamic effect and efficacy are saturated when the plasma concentration reaches approximately 1μM.[15, 17] The relationship between PK and pharmacodynamics in orally dosed mice was consistent with this observation. The maximally efficacious oral doses of MLN8237 in the HCT-116 model (10 and 30 mg/kg QD) resulted in plasma concentrations approximating 1 μM for 8 to 12 hours post-dose. As detailed in the discussion of the phase 1 clinical studies, these exposure levels were readily achieved in patients with advanced malignancies treated with MLN8237.

2.1.2.3 Safety Pharmacology

In exploratory safety pharmacology studies, MLN8237 exhibited minimal activity against the rapidly activating component of the delayed rectifier potassium current (I_{Kr}) encoded by the human ether-à-go-go related gene (hERG; IC_{50} and K_i > 100 μM).[24] MLN8237 had in vitro activity

against the gamma aminobutyric acid A alpha-1 ($GABA_A\alpha1$) benzodiazepine binding site (Ki = 290 nM).[25] In an exploratory non-GLP-compliant single-dose in vivo CNS safety pharmacology study, effects consistent with the binding of MLN8237 to the $GABA_A\alpha1$ binding site (gait abnormalities) were seen in rats at doses and exposures that exceeded those that were tolerated on a daily oral dosing schedule.[26] In an exploratory non-GLP-compliant single-dose in vivo cardiovascular safety pharmacology study in radio-telemetry-instrumented beagle dogs, effects consistent with the binding of MLN8237 to the $GABA_A\alpha1$ binding site (increases in heart rate and blood pressure, modest increase in body temperature) were seen in dogs at doses and exposures that exceeded those that were tolerated on a daily oral dosing schedule.[27] No MLN8237-related effects on clinical signs or physical examination findings indicative of impaired respiratory function (i.e., labored our shallow breathing), or microscopic changes in the lungs of animals that survived until scheduled termination, were noted at tolerated doses in GLP-compliant repeat-dose toxicology studies.

2.1.2.4 Nonclinical Pharmacokinetics and Product Metabolism
Studies to evaluate the PK of MLN8237 were conducted in nude mice bearing human colon HCT-116 tumor xenografts and in Sprague-Dawley rats, beagle dogs, cynomolgus monkeys, and chimpanzees.[15, 17, 28-31]

Absorption
MLN8237 was highly permeable in the Caco-2 cell assay, with an apparent permeability (P_{app}) of 62.4 and $47.1x10^{-6}$ cm/sec in the apical-to-basolateral and basolateral-to-apical directions, respectively, at a pH of 7.4.[32] MLN8237 demonstrated good oral bioavailability in all animal species tested.[33, 34]

Distribution
MLN8237 had a moderate volume of distribution at steady state (V_{ss}) in all nonclinical species studied, ranging from 0.74 L/kg to 3.5 L/kg.[15, 17, 28-31] MLN8237 is highly bound to plasma proteins in all species (percentages of unbound MLN8237 [f_u]: 1.9% in mouse, 0.70% in rat, 2.2% in dog, 1.7% in chimpanzee, and 2.5% in human plasma).[35]

[^{14}C]MLN8237-derived radioactivity was rapid and extensively distributed to tissues.[36] The highest concentrations for most tissues were reached at 0.25 hours after a single oral administration. Concentrations of >20µg equiv/g at 0.25 hours post dose were found in the small intestine (268.920µg equiv/g), liver (91.860µg equiv/g), adrenal cortex (50.504µg equiv/g), stomach (39.040µg equiv/g), adrenal medulla (24.235µg equiv/g), and kidney cortex (23.135µg equiv/g). The highest concentrations were observed in contents of the alimentary canal at 0.25 to 24 hours post-dose (≤67.596 to 2692.620µg equiv/g), which probably reflected the radioactivity from the dosing formulation and bile. Bile concentrations were also relatively high (≤831.288µg

equiv/g), but concentrations in urine were much lower (0.292µg equiv/g), which suggested that biliary excretion was the major route of elimination after PO administration. The concentrations observed in the central nervous system were the lowest of all tissues (≤0.525 to 0.726µg equiv/g) and were below the quantitation limit (BQL) at 4 hours post-dose. Low concentrations were also observed in the testis (≤0.718µg equiv/g), bone (≤0.411µg equiv/g), and the lens of the eye (0.124µg equiv/g).

Tissue concentrations in most tissues declined rapidly and were approximately an order of magnitude lower at 4 hours post-dose. Concentrations in approximately half of the tissues were BQL at 24 hours post-dose and all, except liver and alimentary canal contents, were BQL at 48 hours post-dose.

Radioactivity was detected by quantitative whole-body autoradiography (QWBA) analysis of cardiac blood through 24 hours post-dose (highest at 0.25 hours, 14.770µg equiv/g) and concentrations became BQL at 48 hours.[36] Blood-to-plasma ratios were approximately 0.64 through 4 hours post-dose and had decreased slightly at 24 hours postdose. When analyzed by liquid scintillation counting (LSC), blood concentrations of radioactivity closely matched concentrations measured by QWBA.

The in vitro blood-to-plasma partitioning of MLN8237 in whole blood from healthy human volunteers was determined.[37] At a concentration of 1µM, the mean blood-to-plasma ratios of MLN8237 in humans (pooled male and female samples) after 10 and 60 minutes of incubation at 37°C were 0.46 and 0.55, respectively. At a concentration of 10µM, the mean blood-to-plasma ratios of MLN8237 in humans after 10 and 60 minutes of incubation at 37°C were 0.45 and 0.59, respectively. MLN8237 (1 to 10µM) was preferentially distributed into human plasma, with a mean blood-to-plasma ratio of 0.57 when equilibrium was reached at 60 minutes.

Metabolism

MLN8237 is metabolized by both glucuronidation and oxidation pathways with no marked species differences in metabolism observed from in vitro studies. Urinary excretion of MLN8237 was negligible in the rat and chimpanzee (< 1% of the dose).[38] An in vivo metabolic profiling study in rats using ^{14}C-MLN8237 showed that unchanged MLN8237 was the predominant circulating component in plasma, accounting for approximately 93.0% of the total radioactivity in plasma over a 24-hour period.[39] In addition, the major metabolic route of 14C-MLN8237 in rats is acyl glucuronidation, with >77.9% of the dose metabolized by this pathway.[40]

The oxidative metabolites (M2 and M3) were formed by CYP3A4, 1A2, 2C9, and 2C19, while UGT1A1, 1A3, and 1A8 mediated M1 formation.[38] Using human liver microsomes with the appropriate cofactors, the percent contribution of CYP and UGT was calculated to be 13.1% and 86.9%, respectively, showing that CYP isozymes play a minor role in the metabolism of

MLN8237.[41]

Excretion
After oral administration of 14C-MLN8237 to intact rats, radioactivity was eliminated primarily in the feces, while radioactivity was eliminated primarily in the bile of bile-duct cannulated rats.[39] After IV administration of MLN8237 in rats and chimpanzees, less than 1% of the dose was found as the parent form in urine collected over 24 hours post-dose.[29, 31]

Pharmacokinetic Drug Interactions
The potential of MLN8237 (when administered at the MTD of 50 mg BID) to inhibit the major CYP isozymes 1A2, 2C9, 2C19, 2D6, and 3A4/5 is unlikely (IC_{50}>100μM, estimated K_i>50μM).[42, 43] Based on the 50-mg BID regimen, with a mean Cmax of ~2.7μM, MLN8237 is unlikely to produce drug-drug interactions (DDIs) by inhibiting major CYP isozymes. Because MLN8237 is metabolized by multiple CYP isozymes and uridine diphosphate-glucuronosyltransferase (UGT) isozymes, CYP-based metabolism plays a minor role in vitro, and major UGT-based interactions are less common, it is anticipated that CYP or UGT inhibitors would not substantially affect the PK of MLN8237.

In a Caco-2 cell assay, MLN8237 showed excellent permeability and no efflux. MLN8237 is not a human P-gp substrate, but inhibited the P-gp-mediated efflux of paclitaxel (Taxol®) in Caco-2 cells with an IC_{50} of 4.0μM.[32] MLN8237 is unlikely to produce inhibition of P-gp at systemic sites of transport because of the substantially lower steady-state unbound C_{max} at the clinical dose of 50 mg BID (67nM). However, the potential for inhibition of intestinal P-gp efflux by higher intestinal luminal concentrations of MLN8237 cannot be ruled out; therefore, orally administered narrow therapeutic range substrates of P-gp (eg, digoxin) should be administered with caution. MLN8237 is a weak inhibitor of BCRP, OATP, and MRP2.[32] The potential for co-administered MLN8237 to result in drug interactions with chemotherapeutic agents that are known P-gp substrates has not been established at the dose and schedules employed in clinical studies.

Other Pharmacokinetics Studies
On the basis of the GLP-compliant toxicokinetic (TK) evaluations of MLN8237 after repeated daily oral dose administration, exposure increased with dose in an approximately dose-proportional manner from 1 to 50mg/kg in rats and from 0.15 to 1.5mg/kg in dogs. There was no obvious accumulation or auto-induction observed throughout the dosing period.[44-46]

2.1.3 MLN8237: Toxicology.
A series of nonclinical safety assessment studies has been conducted to support the clinical development of MLN8237.

Rat Two-Cycle Repeat-Dose Toxicology Study

A GLP-compliant study was conducted to evaluate the potential toxicity and toxico-kinetic profile of MLN8237 when administered to Sprague Dawley rats once daily via oral gavage for a total of 2 cycles (each cycle consisting of daily dosing for 7 consecutive days; cycles were separated by a 14-day non-dosing period), as well as to evaluate the recovery from, persistence, or progression of any effects during a minimum 14-day recovery period.[46] Microscopically, the principal change observed in all affected tissues was increased mitotic figures (mitotic arrest)/apoptosis, which is consistent with the outcomes associated with Aurora A kinase inhibition. This finding was seen in tissues/organs of rats dosed with ≥ 5 mg/kg/day, with incidence and severity correlating to basal cellular replication rate and increasing with increasing dose. In this study 50 mg/kg/day was considered to be the severely toxic dose in 10% of animals administered (STD10) MLN8237. Lethality occurred as a result of infections that arose secondary to pharmacologically mediated effects on dividing cells in bone marrow and lymphoid tissues. A dosage of 15 mg/kg/day generated pharmacologic effects in rats and was clearly tolerated. A dosage of 5 mg/kg/day was well tolerated. A dosage of 1 mg/kg/day was the NOEL in both male and female rats.

Rat Six-Cycle Repeat-Dose Toxicology Study

The objectives of this GLP-compliant study were to evaluate the potential toxicity and toxicokinetic profile of MLN8237 when administered to Sprague Dawley rats QD via oral gavage for a total of 6 cycles (each cycle consisting of daily dosing for 21 consecutive days; cycles were separated by a 7-day non-dosing period), as well as to evaluate the recovery from, persistence, or progression of any effects during a minimum 28-day recovery period.[47] Administration of MLN8237 orally (by gavage) to Sprague Dawley rats for 6 cycles at dosages of 2, 5, or 15 mg/kg resulted in the death of 1 male rat from the 5-mg/kg group, as well as 1 male and 1 female rat from the 15 mg/kg group. In rats that survived to the scheduled interim and primary ecropsies, administration of MLN8237 at 5 mg/kg (primary necropsy only) and 15 mg/kg (interim and primary necropsies) resulted in microscopic changes consisting of single-cell necrosis and/or lymphoid depletion predominating in tissues/organs with cell populations having a high basal cellular replication rate similar to what was noted in the 2-cycle repeat-dose toxicology study. In addition, administration of MLN8237 at 5 and 15 mg/kg resulted in hematologic alterations secondary to bone marrow suppression of erythrocyte and leukocyte precursors. These test article-related hematologic and microscopic findings were consistent with outcomes associated with Aurora A kinase inhibition.[48] Concomitant test article--related lower body weight gains and lower food consumption were noted over the 6 dosing cycles in the 15-mg/kg group. After a 28-day recovery period, all microscopic findings had completely resolved and hematology alterations indicated continuing resolution.

Dog Two-Cycle Repeat-Dose Toxicology Study

A GLP-compliant study was conducted to evaluate the potential toxicity and toxico-kinetic profile of MLN8237 when administered to beagle dogs once daily via oral lavage for a total of 2 cycles (each cycle consisting of daily dosing for 7 consecutive days; cycles were separated by a 14-day

non-dosing period), as well as to evaluate the recovery from, persistence, or progression of any effects during a minimum 16-day recovery period.[45] In the repeat-dose oral toxicology study in dogs, the DLT associated with MLN8237 administration occurred as a result of target-mediated effects of mitotic arrest and apoptosis (single-cell necrosis) of replicating cells in the intestine, bone marrow, and lymphoid tissues. A dosage of 1.5 mg/kg/day for 7 consecutive days was tolerated in 4 of 6 males. This dosage was adjusted to 1.0 mg/kg/day for the second 7-day dosing cycle. Administration of 1.0 mg/kg/day for 7 consecutive days was tolerated by all of the dogs (5 males and 6 females). Therefore, 1.0 mg/kg/day was the highest non-severely toxic dose (HNSTD). A dosage of 0.45 mg/kg/day generated pharmacologic effects and was well-tolerated. A dosage of 0.15 mg/kg/day was the NOEL.

Dog Six-Cycle Repeat-Dose Toxicology Study
The objectives of this GLP-compliant study were to evaluate the potential toxicity and toxico-kinetic profile of MLN8237 when administered to beagle dogs QD via oral gavage for a total of 6 cycles (each cycle consisting of daily dosing for 21 consecutive days; cycles were separated by a 7-day non-dosing period), as well as to evaluate the recovery, persistence, or progression of any effects during a minimum 28-day recovery period.[49] Administration of MLN8237 to beagle dogs QD via oral gavage at a dosage of 1.0 mg/kg for up to 6 days was not tolerated and resulted in lethality and dose reduction to 0.75 mg/kg for the surviving 4 animals for the remainder of the dosing period. Cyclical administration of MLN8237 to beagle dogs QD via oral gavage for the following 5 complete cycles, was well tolerated at a dosage of 0.75 mg/kg. Cyclical administration of MLN8237 to beagle dogs QD via oral gavage for a total of 6 cycles (each cycle consisting of daily dosing for 21 consecutive days with cycles separated by a 7-day nondosing period) was well-tolerated at dosages of 0.45 and 0.15 mg/kg. Test article-related effects were limited to hematology and microscopic findings, all of which were consistent with outcomes associated with Aurora A kinase inhibition and were similar to what was noted in the 2-cycle repeat-dose toxicology study. The microscopic findings in the 0.45- and 1.0/0.75-mg/kg groups had completely resolved at the recovery necropsy, and the hematology findings had partially resolved in the 1.0/0.75-mg/kg group at the recovery necropsy.

2.1.4 Clinical Experience

There have been multiple company-sponsored clinical studies of alisertib (MLN8237) that are either ongoing or have completed enrollment. The solubility of MLN8237 molecules is low in acidic pH; hence, the drug was originally developed as the buffered PIC formulation for the initial phase 1 clinical studies. Subsequently, the ECT formulation was developed and has been evaluated in multiple studies.

Initial studies enrolled patients with advanced solid tumors. The major goals of each study were to evaluate safety, pharmacokinetics, and pharmacodynamics, and to determine the DLT(s) and MTD of alisertib (MLN8237) when administered for 7 consecutive days, followed by a 14-day

recovery period. The studies also included cohorts to evaluate longer dosing periods up to 14 or 21 days. In the first-in-human study phase I study conducted in the United States, 65 patients received MLN8237 across dose cohorts evaluating various dose levels and schedules with the PIC formulation. 65 pts received 5-150 mg QD for 7 days or 25-70 mg QD for 14 or 21 days. In addition, to minimize C_{max}-associated somnolence risk, divided treatments were given up to maximum administered doses 60 mg BID for 7 days or 40 mg BID for 14 days. Tumors were colorectal (14), lung (13), ovary (6), head and neck (6), prostate (4), and others (22). Dose limiting toxicities included grades 3-4 neutropenic fever (n=4), thrombopenia (n=3), somnolence (n=2), and mucositis (n=1), seen at doses ≥ 110 mg/d x 7 days, ≥ 40 mg BID x 14 days, and in 1 of 11 pts at 50 mg BID x 7 days. Most common adverse events included nausea (57%), diarrhea (52%), fatigue (48%), and alopecia (35%). The recommended phase 2 dose was 50 mg BID for 7 days. Durable partials response was documented in one patient with platinum-refractory ovarian cancer and treatment was for over 1.5 years (> 30 cycles). Eight patients continued treatment for ≥ 6 cycles with stable disease. Alisertib **(**MLN8237) was deemed to be generally well tolerated.[50]

Concurrent with the above study, data was presented on pharmacokinetic and pharmacodynamic results from two phase 1 studies of alisertib (MLN8237) in patients with advanced solid tumors. In 117 patients who took MLN8237, the drug was absorbed rapidly (median T_{max}, 2h), steady-state exposures were achieved by 7 days and were dose-proportional over dosing ranges of 5-200mg per day. The recommended phase II dose 50mg BID × 7 days produced steady-state average concentrations (geometric mean ~1.7μM), exceeding the efficacious concentration estimated preclinically (1μM). Mean $t_{1/2}$ was ~21h. At steady-state exposures, skin mitotic and apoptotic indices and tumor mitotic index increased, and tumor mitotic cell chromosome alignment and spindle bipolarity decreased, all demonstrating AAK inhibition. In evaluable pts with paired tumor biopsies, exposure-related changes were observed by D7, including decreases in chromosome alignment and spindle bipolarity.[51]

Another phase I trial of enteric coated (ECT) alisertib in advanced solid tumors was presented in the last year. The agent was administered orally BID for 7 days, followed by 14 days of rest, in 21-day cycles. 17 pts were treated, with frequent tumor types including sarcoma (n=4), and ovarian (n=2). The MTD was identified as 50 mg BID, with a DLT of Grade 4 febrile neutropenia was reported at 50 mg BID. The most common adverse events were neutropenia (47%) and fatigue (47%), and the most common grade≥3 adverse event was neutropenia (41%), which was reversible. Following oral administration, MLN8237 ECT was absorbed at a moderate rate with an overall median T_{max} of 3 h and with dose-related increase in exposure over ranges of 10–50 mg BID. Mean $t_{½}$ was ~20 h following multiple dosing, consistent with previous results.[52]

Alisertib has also been studied in solid tumor phase II trials as single agent and in combination.[54, 55] A study in combination with paclitaxel in a phase I/II trial of advanced solid tumor malignancies reported grade 4 diarrhea, grade 3 stomatitis (both at paclitaxel 80 mg/m^2 with

alisertib 20 mg BID) and grade 4 neutropenic fever (at paclitaxel 60 mg/m^2 with alisertib 30 mg BID). A MTD was not reached. Most common adverse events were neutropenia (n=18), anemia (n=13) and nausea (n=12). 21 pts had grade ≥3 drug-related adverse events, with the most common being neutropenia (n=15). Eight patients experienced partial responses by RECIST and/or CA125 levels, and three patients had stable disease for ≥ 6 months.[55]

2.1.4.1 Clinical Studies in Hematologic Malignancies

Alisertib has also been studied in hematologic malignancies, with promising results. A phase 1 study of advanced hematologic malignancies was conducted across 6 dose cohorts, evaluating the following dose levels and schedules, initially using the PIC formulation of alisertib (initial cohort size n = 2-7): 25mg, 35 mg QD x 21 days; and 35, 45, 65, and 90 mg QD x14 days. With the 14-day schedule, the MTD of alisertib (PIC formulation) was determined to be 45 mg QD in adult patients with advanced hematologic malignancies, established following occurrence of DLTs in four patients at higher dose levels: grade 4 thrombocytopenia, grade 4 neutropenia (65 mg QD); grade 3 febrile neutropenia, grade 4 thrombocytopenia (90 mg QD). The study was amended and expanded to evaluate the ECT formulation of alisertib administered BID with a 7-day schedule, specifically to characterize safety and pharmacokinetics with this formulation. The MTD for the ECT formulation was established as 50 mg BID, with four DLTs observed in three patients: at 40 mg QD for 14 days, one patient had grade 4 febrile neutropenia, and one patient had grade 4 bullous dermatitis and grade 4 neutropenia; at 50 mg BID for 7 days, one of nine patients had grade 4 neutropenia. In total, 55 patients were included (PIC n=28, ECT n=27). 60% of patients had non-Hodgkin's lymphoma, 35% multiple myeloma, and 5% chronic lymphocytic leukemia/small lymphocytic lymphoma. The most common drug-related adverse events (AEs) were neutropenia (PIC 46%, ECT 63%), thrombocytopenia (39%, 41%), and diarrhea (29%, 41%). 24 patients (PIC n=12, ECT n=12) had serious adverse events; the most common was febrile neutropenia (PIC n=1, ECT n=4). Tolerability appeared similar between the PIC and ECT formulations. Six patients (PIC n=3, ECT n=3) died during the study, but no deaths were considered related to alisertib. In the 42 evaluable patients, five (PIC n=3, ECT n=2) had partial responses, and 11 had a best response of stable disease (PIC n=6, ECT n=5). 26 patients had progressive disease (PIC n=14, ECT n=12).[56]

In a phase II trial of single agent alisertib in aggressive B-cell and T-cell non-Hodgkin lymphomas, patients were treated at a dose of 50mg twice daily for 7 days on 21 day cycles. 48 patients were enrolled, of whom 41 were evaluable for response. Most common Grade 3/4 adverse events were neutropenia (63%), thrombocytopenia (31%), stomatitis (15%), febrile neutropenia (13%) and fatigue (6%). The overall response rate (ORR) was 32%, with response by histology being 20% for DLBCL, 23% for MCL 23% and 57% for T- cell NHL. The investigators concluded that the generally manageable toxicity profile suggested an opportunity to combine alisertib with other agents [57]

2.1.4.2 Clinical Studies in Myeloid Malignancies

Relevant to the current study, a phase II, single-arm, multicenter study was performed in patients with advanced AML and high-grade myelodysplastic syndrome (MDS). Patients were treated at 50 mg BID for 7 consecutive days in a 21-day cycle. In all, 57 patients were enrolled, with 46 having AML, 21 of whom had secondary AML. 49 patients (86%) patients had received prior therapies, and 9 (16%) had received 3 prior therapies. Treatment-related grade 3/4 adverse events were seen in 24 (42%) patients and included febrile neutropenia (11%), thrombocytopenia (9%), anemia (9%), fatigue (7%), neutropenia (7%), and somnolence (4%). 22 on-study deaths were reported, caused by events unrelated to alisertib. Overall, 6 (13%) responses were observed, all in AML patients, with 5 patients experiencing a PR, and one documented CR in a 79-year old female with no prior therapy, who remained in CR through 16 cycles. Also, 17 patients with AML (49%) and 2 with MDS (20%) achieved stable disease.[58]

At our institution, we have just finished accrual on a phase I, 3+3 cohort dose-escalation study of alisertib combined with standard 7+3 induction for newly diagnosed AML. We recently presented our data at the oral session during the 2014 American Society of Hematology Conference. For this study, those with acute promyelocytic leukemia or with AML and core-binding factor alterations were excluded. All patients received 7+3 induction (continuous infusion cytarabine 100mg/m^2 x 7 days and intravenous idarubicin 12mg/m^2 x 3 days), after which on day 8, alisertib was administered twice daily (BID) for 7 days. Dose escalation occurred via three cohorts (10mg BID, 20mg BID, 30mg BID). All underwent a mid-induction bone marrow biopsy, following course of alisertib, to assess for residual disease, which if present, was to be treated with 5+2 re-induction (cytarabine 100mg/m^2 x 5 days, idarubicin 12mg/m^2 x 2 days) without alisertib. Following induction, up to four cycles of consolidation were allowed, using cytarabine (3g/m^2 BID days 1,3,5 for those aged <60 and 2g/m^2 daily days 1-5 for those aged ≥60) followed by alisertib, according to dose cohort, on days 6-12. After consolidation, alisertib maintenance was initiated, at cohort dose level, for days 1-7 of 21 day cycles, to be continued for 12 months or until disease progression. Those eligible for stem cell transplant (SCT), following induction and/or consolidation courses, were removed from study for this purpose.

Data on 16 patients were presented (n=3, 10mg BID; n=7, 20mg BID; n=6, 30mg BID). One patient (6%) had therapy-related AML and six (38%) had antecedent MDS or myeloproliferative neoplasm. Eleven of the 16 patients (69%) had high risk disease, as per older age (age over 65), high risk cytogenetics, and /or derived from preceding myeloid malignancy. *FLT3* mutations were detected in 3 (19%), *NPM1* mutations in 3 (19%), *CEBPA* mutations in 2 (13%), and *IDH1/IDH2* mutations in 6 patients (38%). One patient in cohort 2 died of sepsis, deemed unrelated to study drug, prior to completion of the toxicity assessment period and was replaced. Six (38%) patients have gone on to SCT. All patients received induction, and 9 patients received at least one cycle of consolidation, and 3 patients started maintenance therapy. Alisertib was well tolerated. All patients experienced expected grade 4 toxicities of anemia, thrombocytopenia, and febrile

neutropenia seen during the induction phase of treatment. One DLT was encountered, of delayed thrombocytopenia (grade 4 at day 40), was seen in the 20mg BID cohort. No other DLTs were detected, and toxicities which could be possibly or probably be attributed to alisertib were grade 1/2 rash, nausea, and oral mucositis. [59]

Doses of up to 30mg twice daily were well-tolerated, and this therefore was determined to be the recommended phase II dose (RP2D). 14 of 16 (88%) patients achieved remission following induction (12 cases of CR and 2 of CRp). During the course of the study, three patients experienced relapse, and three have died (two due to disease). Thirteen of 14 evaluable patients had an ablated mid-induction marrow biopsy, with the remaining patient showing 6% blasts in a 20% cellular marrow at mid-induction. None of the patients required re-induction with 5+2 at mid-induction. Based on this data obtained during dose-escalation, [59] it was deemed that alisertib was well tolerated in conjunction with standard induction chemotherapy in newly diagnosed AML, and held promise as a therapeutic agent in AML, when combined with induction chemotherapy. Since the presentation of this data in abstract form, patient numbers can be updated. In total, 19 of 22 patients (86%) have achieved remission. Of these 19 patients, 7 have relapsed during the course of the study, and of these, 5 have died of relapsed AML.

2.2 Study Disease: Acute Myeloid Leukemia (AML)

AML is characterized by an arrest in differentiation and uncontrolled proliferation of myeloid precursors in the bone marrow. This underlying process frequently leads to hematopoietic insufficiency, and when undifferentiated cells escape the marrow, to significant leukocytosis, with often devastating and life-threatening sequelae. Unfortunately, successful treatment of AML remains a difficult challenge. Although the majority of adults under age 60 with de novo AML achieve a complete remission (CR) with traditional anthracycline and cytarabine based induction regimens, the long-term survival rates continue to be poor at approximately 30-40%. [60, 61] Remission rates and prognosis are significantly poorer for patients with high-risk AML, which includes older patients, those with poor-risk cytogenetic features, and those whose disease has arisen from myelodysplastic syndromes (MDS), myeloproliferative disorders (MPD), as well as those with secondary AML from environmental exposures or prior chemotherapy. In such cases, a remission is achieved in less than 40% of cases, with very poor long-term survival rates of less than 10%.[61, 62]

Anthracyclines and cytarabine have been the primary agents used for AML induction therapy for more than two decades. Early Cancer and Leukemia Group B (CALG-B) studies established the utility of these agents in induction regimens.[63] CALGB-7421 later demonstrated that induction with 7 days of cytarabine and 3 days of anthracycline (7+3) was superior to 5 days of cytarabine and 2 days of anthracycline (5+2).[64] Increasing the intensity of the regimen by extending the infusion of cytarabine to 10 days was also investigated, and was shown to have no impact in complete remission rate. Since that time, 7+3 has been extensively used as first line induction

regimens in AML, although outcomes continue be frequently inadequate and poor, especially in older patients and those with high risk AML.[61]

Therefore, there is an urgent need for the development of novel approaches in the treatment of AML to improve outcomes for patients, especially those with high-risk disease. In the last few years, a variety of targeted and novel therapies have been combined with standard induction therapy, with some promising initial results[65-67], although the majority of advanced phase studies to date have failed to show an advantage in outcomes with combination therapies.

2.3 Rationale

As mentioned above, there is a pressing need to develop novel approaches to the treatment of AML. Aurora kinases are a family of oncogenic serine-threonine kinases that regulate multiple phases of the mitotic signaling cascade. Inhibition of aurora A kinase (AAK) leads to mitotic errors, followed by aneuploidy, apoptosis, and senescence, which maybe especially applicable to cells recently or concurrently exposed to cytotoxic chemotherapy. Alisertib, also known as MLN8237, is an ATP-competitive, orally available inhibitor of AAK, which has been evaluated for safety and efficacy in Phase I and II trials of hematological malignancies including AML. [58] Recently, we have concluded a phase I study of alisertib in combination with "7+3" conventional induction chemotherapy, and found this combination to be safe and well-tolerated. In addition, there was a strong suggestion of efficacy with a significantly less than expected need for "5+2" re-induction and a significantly higher than expected rate of remission. Of note, the majority of patients on this study harbored high risk disease, as defined by age, preceding marrow disease, and cytogenetic risk. [59] The next logical step in establishing the efficacy of alisertib in combination with standard induction chemotherapy in patients with higher risk AML, who have particularly poor outcomes with standard therapies.

This protocol therefore describes a phase II clinical trial of alisertib, at a dose of 30 mg twice daily, combined with induction chemotherapy (7+3 cytarabine- and idarubicin
-based therapy) for patients with higher risk AML. 30mg twice daily is the RP2D based on a recently completed dose-escalation study of this combination. This will be a highly translational project, encouraged by laboratory investigation of AAK activity in myeloid malignancies and promising recent clinical data mentioned above, and will be informed by correlative studies assessing in vivo pharmacodynamic parameters.

2.4 Correlative Studies

We will perform gene panel sequencing on bone marrow aspirate and/or blood samples collected at baseline , day 14 of induction (range 13-16), count recovery following induction (range 35-45, 55-65 if re-induction given), and at any time of relapse or suspicion of relapse. All samples will be processed in a CLIA certified lab. De-identified samples obtained from blood, bone marrow, and skin biopsies from selected cases will be subjected to broader sequence analysis (whole exome

or genome). These exploratory analyses will be performed with several hypotheses in mind. First, we will evaluate whether a specific mutational profile is associated with treatment response. Second, we will determine whether there is selective sensitivity to therapy among specific founding clones and/or subclones. Third, we will investigate somatic mutation patterns that are associated with disease progression. Additional laboratory studies will be performed to characterize the functional consequences of aurora kinase inhibition on leukemic blasts during treatment.

2.5 Hypothesis

The addition of alisertib to "7+3" induction chemotherapy will enhance the rate of remission and other clinical outcomes in patients with high-risk acute myeloid leukemia.

3. PARTICIPANT SELECTION

3.1 Eligibility Criteria

3.1.1 Participants must have pathologically confirmed, newly diagnosed high-risk acute myeloid leukemia, as defined by at least one of the following criteria

A. Age ≥65 years
B. Age ≥ 18 years with adverse risk karyotype, as per European Leukemia Net Guidelines (See Addendum B)
C. Age ≥ 18 years with antecedent or underlying myelodysplastic syndrome, CMML, or myeloproliferative neoplasm
D. Age ≥ 18 years with AML with MDS-related changes

**Note:
For patients in category A, a sample to evaluate patient cytogenetics will be sent at the time of diagnosis per standard clinical care and the absence of favorable risk cytogenetics must be confirmed by Day 8. If the cytogenetic analysis reveals that the patient harbors favorable risk cytogenetics, or if the cytogenetic results are not received prior to Day 8, the participant will be removed from the study. Patients removed due to presence of favorable cytogenetics would be considered to be inevaluable and will be replaced. In addition, patients who receive less than half of their total alisertib dose during induction would be considered to be inevaluable and will be replaced.

3.1.2 Adults, age 18 years or older at the time of diagnosis, eligible for standard induction chemotherapy according to their treating physician, who fulfill criteria in section 3.1.1.

3.1.3 ECOG performance status 0-2 (Karnofsky ≥60%, see Appendix A)

3.1.4 Left ventricular ejection fraction > 50% as measured by echocardiogram or MUGA scan

3.1.5 Must not have received systemic antineoplastic therapy including radiation therapy within 14 days of the study enrollment, except hydroxyurea or 6-mercaptopurine for the purposes of cytoreduction. Patients may also have received *all-trans* retinoic acid (ATRA) if there is an early suspicion of acute promyelocytic leukemia (APL, M3-AML), although if confirmed to have APL these patients will be excluded from the study.

3.1.6 Adequate renal function as defined by: calculated creatinine clearance ≥ 40 mL/min (Cockcroft-Gualt Forumla)

3.1.7 Direct bilirubin < 2.0 x upper limit of nomal (ULN), SGOT (AST) and SGPT (ALT)< 2.5 x ULN. AST and/or ALT may be up to 5X ULN if thought to be secondary to leukemia.

3.1.8 The effects of alisertib on the developing human fetus are unknown. For this reason and because other chemotherapeutic 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. 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 for the duration of study participation, and 6 months after completion of therapy.

3.1.9 Subject must be able to take oral medication and to maintain a fast as required for 2 hours before and 1 hour after alisertib administration.

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

3.2 Exclusion Criteria

3.2.1 Patients will be excluded from this study if they are found to harbor "favorable" risk cytogenetics:

3.2.2 Patients with acute bilineal/biphenotypic leukemia

3.2.3 Participants who have had systemic chemotherapy or radiotherapy within 14 days prior to entering the study, except for hydroxyurea or 6-MP as noted. Empiric intrathecal chemotherapy during a diagnostic lumbar pucture is allowed, as long as CNS disease is not suspected.

3.2.4 Participants who are receiving or have received any other investigational agents within 14 days of enrollment.

3.2.5 Immunotherapy or therapy with monoclonal antibodies or small tyrosine kinase inhibitors within the past 4 weeks prior to treatment with the trial drug

3.2.6 Persistence of clinically relevant therapy related toxicity from previous anti-cancer therapy

3.2.7 Prior allogeneic bone marrow or organ transplantation

3.2.8 Individuals with a history of a different malignancy are ineligible except for the following circumstances. Individuals with a history of other malignancies are eligible if they have been disease-free for at least 3 years and are deemed by the investigator to be at low risk for recurrence of that malignancy. Individuals with the following cancers are eligible if diagnosed and treated within the past 3 years: cervical cancer in situ, and basal cell or squamous cell carcinoma of the skin.

3.2.9 Current clinical central nervous system (CNS) symptoms deemed by the investigator to be related to leukemic CNS involvement (no lumbar puncture required, clinical assessment per investigator's judgment is sufficient).

3.2.10 If applicable, patient with ≥ Grade 2 peripheral neuropathy within 14 days before enrollment

3.2.11 Prior treatment with alisertib

3.2.12 Known history of hepatitis C infection or suspected currently active hepatitis C infection. Known or suspected history of hepatitis B infection will be excluded when any of the following conditions are met:

- Received hematopoietic stem cell transplantation (either allogenic or autologous), or
- Received any rituximab-containing treatment regimen in the last 12 months before entering the study, or Active hepatitis B as deemed by serology or viral load

3.2.13 Current or history of congestive heart failure New York Heart Association (NYHA) class 3 or 4, or any history of documented diastolic or systolic dysfunction (LVEF <50%, as measured by MUGA scan or echocardiogram). Prior to study entry, any ECG abnormality at screening has to be documented by the investigator as not medically relevant

3.2.14 Known hypersensitivity to the trial drugs or other contraindication to standard "7+3" induction chemotherapy.

3.2.15 Known history of uncontrolled sleep apnea syndrome, or sleep apnea requiring supplemental oxygen, and other conditions that could result in excessive daytime sleepiness.

3.2.16 A medical condition requiring use of proton pump inhibitors (PPIs); or histamine 2 (H2) receptor antagonists. Patients who intermittently use these medications, must meet the following criteria:

- No use of PPIs within 5 days before the first dose of alisertib
- No use of H2 antagonist or pancreatic enzymes within 24 hours before the first dose of alisertib

3.2.17 Patients with mental deficits or psychiatric conditions that preclude them from giving informed consent or following protocol.

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

3.2.19 Known GI disease or GI procedures that could interfere with the oral absorption or tolerance of alisertib. Examples include, but are not limited to partial gastrectomy, history of small intestine surgery, and celiac disease.

3.2.20 Pregnant women are excluded from this study because alisertib, along with standard induction chemotherapy, carries 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 alisertib as well as cytarabine and idarubicin, breastfeeding should be avoided. Confirmation that the subject is not pregnant must be established by a negative serum β-human chorionic gonadotropin (β-hCG) pregnancy test result obtained during screening. Pregnancy testing is not required for post-menopausal or surgically sterilized women.

3.2.21 Although not absolute exclusion criteria, because of known drug-drug interactions, below are issues that should be considered during enrollment:

Treatment with clinically significant enzyme-inducing drugs, including known P-glycoprotein inducers (including St John's wort and rifampicin) should be used only if absolutely necessary and considered to be the best available choice for the patient. If possible, it is recommended that alternatives to known substrates, inhibitors or inducers of P-glycoprotein be considered. Cases should be discussed with the principal investigator, and may be allowed as per his/her discretion.

3.2.22 Patients with psychological, familial, social, or geographic factors that otherwise preclude them from giving informed consent, following the protocol, or potentially hamper compliance with study treatment and follow-up.

3.2.23 Patients who are otherwise felt unable to comply with the protocol, in the opinion of the investigator.

3.3 Inclusion of Women and Minorities

Both men and women of all races and ethnic groups, and any member of an underrepresented population, are eligible for this trial.

4. REGISTRATION PROCEDURES

4.1 General Guidelines for DF/HCC and DF/PCC Institutions

Institutions will register eligible participants with the DF/HCC Quality Assurance Office for Clinical Trials (QACT) central registration system. Registrations must occur prior to the initiation of protocol therapy. Any participant not registered to the protocol before protocol therapy begins will be considered ineligible and registration will be denied.

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

Following registration, participants may begin protocol therapy. Issues that would cause treatment delays should be discussed with the Overall Principal Investigator (PI). If a participant does not receive protocol therapy following registration, the participant's registration on the study must be canceled. Notify the QACT Registrar of registration cancellations as soon as possible.

4.2 Registration Process for DF/HCC and DF/PCC Institutions

The QACT registration staff is accessible on Monday through Friday, from 8:00 AM to 5:00 PM Eastern Standard Time. In emergency situations when a participant must begin protocol therapy during off-hours or holidays, call the QACT registration line at [redacted] and follow the instructions for registering participants after hours.

The registration procedures are as follows:

- Obtain written informed consent from the participant prior to the performance of any protocol specific procedures or assessments.

- Complete the QACT protocol-specific eligibility checklist using the eligibility assessment documented in the participant's medical record and/or research chart. **To be eligible for registration to the protocol, the participant must meet all inclusion and exclusion criterion as described in the protocol and reflected on the eligibility checklist.**

  Reminder: Confirm eligibility for ancillary studies at the same time as eligibility for a treatment protocol. Registration to both treatment and ancillary protocols will not be completed if eligibility requirements are not met for all studies.

- Fax the eligibility checklist(s) and all pages of the consent form(s) to the QACT at [redacted]. For Phase I protocols, attach participant dose level assignment confirmation from the sponsor.

- The QACT Registrar will (a) review the eligibility checklist, (b) register the participant on the protocol, and (c) randomize the participant when applicable.

- An email confirmation of the registration and/or randomization will be sent to the Overall PI, study coordinator(s) from the Lead Site, treating investigator and registering person immediately following the registration and/or randomization.

4.3 General Guidelines for Other Investigative Sites

N/A

4.4 Registration Process for Other Investigative Sites

N/A

5. TREATMENT PLAN

5.1 Treatment Regimen

All subjects who meet the inclusion/exclusion criteria will be considered for enrollment. Subjects will only be approached after obtaining permission from their physician and healthcare team at that time. The healthcare team will introduce the study to the patient and obtain permission from the patient to be contacted by the study staff. After permission is obtained, one of the co-investigators of this study will explain the study to potential subjects in detail. It will be emphasized that the study is entirely optional. All details of the study will be explained to the subject with any questions answered by a co-investigator or a member of the healthcare team. A copy of the study's consent form will be provided to all participants. Written informed consent will be obtained at that time. The patient will be ensured that his or her decision to participate in the study will not affect his or her individual care. As stated above, all subjects who sign consent will be registered via QACT as per standard institutional guidelines.

Enrolled patients will provide blood and urine for assays that are part of standard of care for their treatment as well as tests which are required by the study. Those required by the study are as follows, and should occur within 14 days prior to enrollment: complete blood count and differential, complete metabolic panel consisting of electrolytes panel, renal function, and hepatic function assays, coagulation parameters, LDH, uric acid, hepatitis B/C serologies, and urinalysis. EKG and an echocardiogram will also be required prior to initiation of treatment. A MUGA scan can be used in place of an echocardiogram, if necessary. Standard institutional procedures can be followed for imaging and obtaining blood and urine cultures if there is suspicion of infection. A bone marrow biopsy and aspirate will also be performed within 14 days of enrollment.

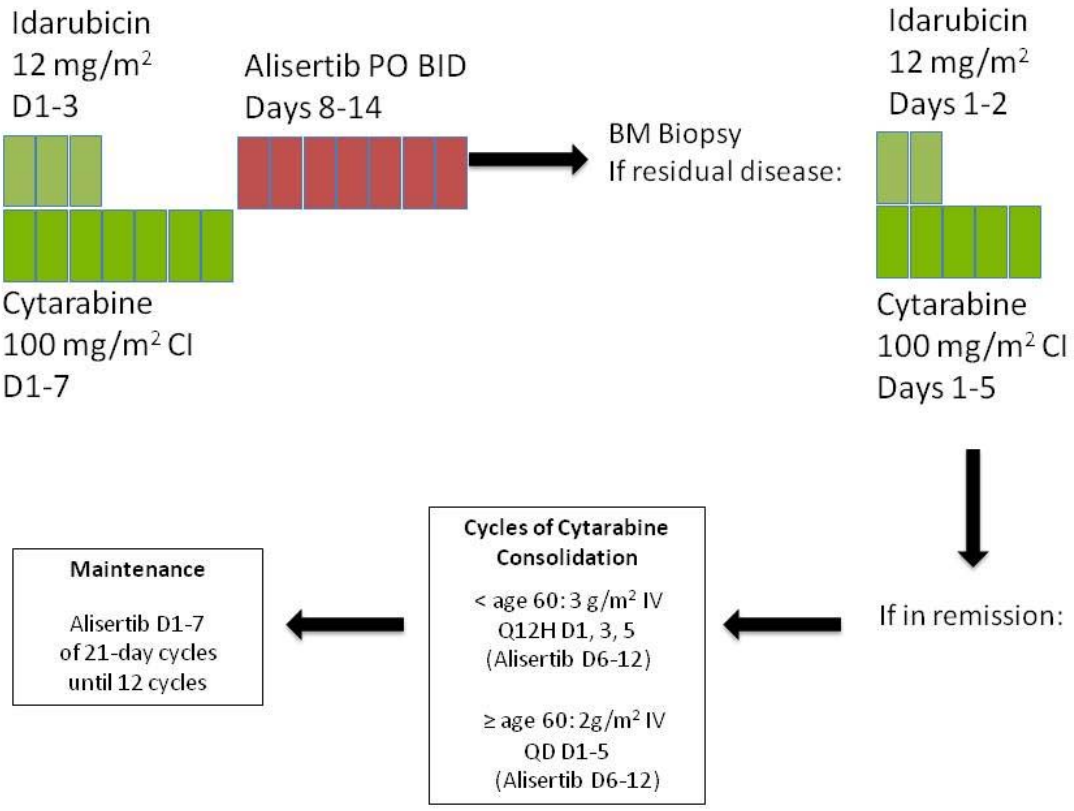
Induction treatment and cytarabine consolidation will be administered on an inpatient basis. During consolidation and maintenance, alisertib may be given on an outpatient basis. . All patients

will have a central venous catheter line placed (a peripherally inserted central catheter (PICC) will also be allowed) during induction. Patient are not required to have a central line during consolidation or maintenance. Reported adverse events and potential risks for alisertib, cytarabine, and idarubicin, as well as appropriate dose modifications for these agents are described in **Section 6**. No investigational or commercial agents or therapies other than those described below may be administered with the intent to treat the patient's malignancy.

Prior to onset of induction therapy, for patients with significant leukocyotosis, hydroxyurea can be administered to lower the WBC, as per discretion of treating physician. Leukapheresis can also be used for reduction in the WBC count. It is recommended that patients start allopurinol prior to receiving chemotherapy and this be continued until at least day 5.

5.2 Agent Administration

Treatment on study will be according to the schema outlined below:



5.2.1. Remission Induction Therapy:
On day 1 of the study, all patients will be initiated on the "7+3" induction regimen, which consists of the following:

- Cyatarabine 100mg/m^2/day intravenous continuous infusion days 1-7 of induction cycle
- Idarubicin 12mg/m^2/day intravenous bolus days 1-3 of induction cycle. If idarubicin is not available, daunorubicin 60mg/m^2/day intravenous bolus on days 1-3 will also be allowed as an alternative. The dose of idarubicin (or daunorubicin) should be adjusted based on bilirubin level as per **Section 6.2**.

Treatment with alisertib at a dose of 30mg twice daily (every 12 hours) will begin on day 8 (or day 9), after conclusion of "7+3" induction course. **Section 5.2.3**

5.2.2. Day 14 Bone Marrow Aspiration and Possible Re-Induction Therapy

A bone marrow biopsy will be performed on day 14 (range day 13-16) of induction.

- If bone marrow cellularity is ≥ 20% and there are greater than 5% myeloblasts present, the treating physician is to administer "5+2" re-induction, consisting of the following:
  -Cyatarabine 100mg/m^2/day continuous infusion days 1-5 of re-induction
  -Idarubicin 12mg/m^2/day bolus days 1-2 of re-induction. If idarubicin is not available, daunorubicin 60mg/m^2/day intravenous bolus on days 1-2 will also be allowed as an alternative. The dose of idarubicin or daunorubicin should be adjusted based on bilirubin level as per **Section 6.2**.
  -Alisertib will <u>not</u> be given during re-induction

- If regardless of bone marrow cellularity, there are ≤ 5% myeloblasts present, no re-induction is necessary.

- If bone marrow cellularity is < 20% and there are greater than 5% myeloblasts present, re-induction will be at the discretion of the treating physician.

5.2.3 Treatment with Alisertib

During Induction, treatment with alisertib will be as follows:
-Alisertib 30mg twice daily (every 12 hours) will be given on days 8-14 or 9-15

Alisertib will be begin on day 8, after the conclusion of "7+3" induction course. The exception would be if 7+3 therapy begins in the afternoon or evening, and therefore finishes late on day 8, in which case the first dose of Alisertib may be given on Day 9,

If "5+2" re-induction is administered, alisertib will not be given during this re-induction.

During Consolidation, treatment with Alisertib will be as follows: -Alisertib at 30mg PO twice daily (every 12 hours) will be administered for days 6-12 at after the conclusion of Cytarabine. Please note . if dose reduction took place for alisertib-related toxicity during induction, that dose-reduced dose will also be used during consolidation and subsequent maintenance

Following count recovery after induction and/or consolidation course(s), maintenance cycles of alisertib will be started.and continued for 12 cycles, or until disease progression or unacceptable toxicity. Alisertib delays and dose modification are detailed in Section 6.

During Maintenance, treatment with Alisertib will be as follows:

- Alisertib 30mg PO twice daily (every 12 hours) will be administered for days 1-7 followed

by 14 days off, in 21 day cycles.

Subjects must be fasting for 2 hours before and 1 hour after Alisertib (MLN8237) administration. Only water is allowed during the fasting period. Subjects should take Alisertib within 3 hours of a missed dose. If more than 3 hours have passed, then that dose should be omitted, and the subject should resume treatment with the next scheduled dose. Vomited or missed doses of Alisertib will not be made up or re-dosed on this study.

5.2.4 Remission Bone Marrow Aspiration and Biopsy
Upon a peripheral blood count recovery of ANC of at least 1000/μL and a platelet count of at least 100,000/μL, following course of induction (and re-induction, if necessary) chemotherapy, a bone marrow aspirate and/or biopsy will be performed to assess and document response to therapy. A bone marrow aspirate and/or biopsy will also be performed if hematologic recovery has not occurred by day 42 (Range 35-45) (or day 63 if 5+2 re-induction was given based on day 14 bone marrow biopsy, range 55-65), or at any time that AML regeneration is suspected.

Response criteria will be as defined by modified International Working Group (IWG) as per Cheson et al [68]. If patient is found to have persistent AML, he/she will be discontinued from the study and proceed with additional management off-study per discretion of the treating oncologist.

5.2.5 Remission Consolidation Therapy
Patients who are eligible for hematopoietic stem cell transplantation (HSCT), per their treating physician, or other post-remission strategies, after discussion with and per the discretion of the principal investigator, can come off study for this purpose.

Patients in CR or CRi who are unable or unwilling to undergo HSCT can receive up to four cycles of remission consolidation therapy, at the discretion of their treating physician. Each consolidation cycle is typically four weeks (28 days) in duration, and should begin within two to four weeks following hematologic recovery (ANC ≥ 1000/mcL and platelet count ≥ 100,000/mcL), but not sooner than four weeks from the beginning of the previous cycle.

Patients who have CRI (ANC ≥ 1000/mcL and platelet count ≤ 100,000/mcL) may undergo remission consolidation therapy after discussion with the principal investigator.

Remission consolidation therapy will consist of the following for patients younger than age 60:

- Cytarabine 3 g/m^2 by IV infusion over 3 hours (or institutional standard) given every 12 hours on Days 1, 3, and 5. (6 doses)
- Alisertib 30 mg PO given twice daily (every 12 hours) on Days 6-12

Remission consolidation therapy will consist of the following for patients equal to or greater than age 60:

- Cytarabine 2 g/m^2 per day by IV infusion over 3 hours (or institutional standard) on Days 1-5. (5 doses)
- Alisertib 30 mg PO given twice daily (every 12 hours) on Days 6-12

Prior to cytarabine infusion, mandatory corticosteroid eye drops will be given per institutional standards. Patients should be monitored closely and a serial fashion for onset of cerebellar deficits or central nervous system symptomatology, as per institutional guidelines.

In those patients receiving cycles of consolidation therapy, alisertib (MLN8237) will be started on day 6, and given by mouth at a dose of 30mg twice daily, for a period of seven days. If dose reduction took place for alisertib-related toxicity during induction, that dose-reduced dose will also be used during consolidation and subsequent maintenance. Dose reductions for toxicity related to alisertib will be as outlined in **Section 6.1**.

5.2.6 Maintenance Therapy

Upon count recovery, (ANC ≥ 1000/mcL and/or platelet count ≥ 100,000/mcL), following at least 2 cycles of consolidation, maintenance therapy with alisertib will be initiated. The first cycle of maintenance should begin within two to four weeks following hematologic recovery (ANC ≥ 1000/mcL and/or platelet count ≥ 100,000/mcL), but not sooner than four weeks from the beginning of the previous cycle. If the first cycle of maintenance cannot be started within four weeks following hematologic recovery (as mentioned above), the patient would be considered off study treatment. See **section 6.1.1** for further information regarding delay and dose modifications.

Alisertib will be given at 30mg BID for days 1 through 7 of 21 day cycles. Maintenance therapy will continue for a total of 12 cycles or until disease progression. A window of up to +/- 3 days is allowed to start the successive cycles of maintenance to accommodate patient/provider availability and holidays.

5.3 General Concomitant Medication and Supportive Care Guidelines

Antiemetics will be used according to standard practices. Patients will receive prophylaxis directed against candidiasis, and/or herpes simplex virus (HSV), and bacterial infection, as needed, according to individual institutional practices.

Hematopoietic growth factors may be used with PI approval.

Caution should be used o with the use of proton pump inhibitors, H2 antagonists, antacids, or pancreatic enzymes, that could potentially impact the absorption of Alisertib. .

The following guidelines should be used if using these medications:

- No use of PPI within 5 days prior to Alisertib and while on Alisertib. PPI may be restarted after 24hrs after the last dose of Alisertib.
- No use of H2 antagonist or pancreatic enzymes within 24 hours before the first dose of alisertib and may be restarted 24 hrs after the last dose of Alisertib.
- Antacids should not be taken 2hrs prior to Alisertib and for 1 hour after the dose.
- Carafate should not be taken 6 hrs prior to Alisertib and for 1 hour after the dose

Treatment with clinically significant enzyme-inducing drugs, including known P-glycoprotein inducers (including St John's wort and rifampicin) should be used only if absolutely necessary and considered to be the best available choice for the patient. If possible, it is recommended that alternatives to known substrates, inhibitors or inducers of P-glycoprotein be considered. Cases should be discussed with the principal investigator, and may be allowed as per their discretion.

5.4 Duration of Therapy

In the absence of disease progression or unacceptable adverse event, alisertib will be continued for a period of up to 12 cycles of maintenance, based on the schedule above.

In the absence of treatment delays due to adverse events, treatment may continue as above until 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, or
- General or specific changes in the patient's condition render the patient unacceptable for further treatment in the judgment of the investigator.
- Patient becomes pregnant or begins breast-feeding
- Failure to return for follow-up

5.5 Duration of Follow Up

Alisertib will be administered as above, until the conclusion of the study, unless relapse, death, or intolerable toxicity takes place, at which point treatment will cease. Patients discontinued from therapy for unacceptable adverse events will be followed until resolution or stabilization of the adverse event. Also, regardless, all participants will be followed for 30 days after removal from study or until death, whichever occurs first.
Patients will be contacted at least every 3 months for a period of up to 12 months for disease and survival status and collection of subsequent anticancer treatment information until death or study

closure, whichever occurs first. Results from routine labs will be collected as needed to follow patients for peripheral blood count recovery.

5.6 Criteria for Removal from Study

Patients will be removed from study when any of the criteria listed in **Section 5.4** applies, or if the study physician determines they cannot comply with the protocol. The reason for study removal and the date the patient was removed will be documented in the Case Report Form (CRF). Alternative care options will be discussed with the participant. In the event of unusual or life-threatening complications, participating investigators must immediately notify the Principal Investigator, Amir T. Fathi MD, Partners pager 12042.

A QACT Treatment Ended/Off Study Form will be filled out when a participant comes off study. This form can be found on the QACT website or obtained from the QACT registration staff.

6. DOSING DELAYS/DOSE MODIFICATIONS

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

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

6.1 Alisertib, Dose Modifications/Delays

All adverse events experienced by participants will be collected from the time of the first dose of study treatment, through the study and until the final study visit. Participants continuing to experience toxicity at the off study visit may be contacted for additional assessments until the toxicity has resolved or is deemed irreversible.

6.1.1 Management of Toxicity

It is anticipated that all patients will experience grade 3 or 4 myelosuppression during induction and consolidation. There are no dose modifications planned for abnormal blood counts during these phases of treatment.

However, during maintenance phase, when concurrent chemotherapy is not administered with alisertib, if a patient has grade 4 neutropenia or thrombocytopenia on day 1 the next cycle should be delayed until that cytopenia is grade 2 or less, and patients should be followed weekly. Patients will be allowed to delay subsequent cycles for up to 2 weeks. If the patient is delayed more than 2 weeks they will be removed from study. If during a cycle a patient experiences grade 4 neutropenia

or thrombocytopenia, doses should be held until grade 2 or less. Once the patient's neutropenia or thrombocytopenia is at a grade 2 or less, dosing may be resumed at the next lower dose level as per Table 1. If the cytopenia does not resolve to grade 2 within 2 weeks, the patient will be removed from study. If the dose is held during maintenance for this purpose, doses will not be made up. Patients should continue at the lower dose of Alisertib for all subsequent maintenance cycles.

If at any time during treatment, patients with CTCAE 4.0 Grade 3 non-hematologic toxicity that is possibly, probably or definitely related to alisertib (MLN8237) and persisting for >48 hours without resolution to ≤ Grade 2 will have dosing of alisertib interrupted (or sooner at the discretion of the investigator if considered necessary for patient safety) with the exception of those Grade 3 or 4 non-hematologic toxicities that are associated with AML and its treatment with induction or consolidation chemotherapy.

If the alisertib-related Grade 3 non-hematologic toxicity reverses to ≤ Grade 1 or baseline within 7 days of stopping the study drug, then the patient may resume their dosing schedule at one dose level lower, according to the dose de-escalation schedule below (See **Table 1** below). For example, as a scenario, if a patient had been receiving a dose of alisertib 30 mg by mouth twice daily, they would then be de-escalated to dose of 20mg by mouth twice daily, and so on per dosing levels detailed below. If the patient develops grade 3 non-hematologic toxicity at a dose of 10mg once daily, they will discontinue alisertib.

Doses will not be made up if days of treatment are missed due to toxicity. If the Grade 3 non-hematologic toxicity does not resolve within the window provided above, patients will not be re-dosed with alisertib and will discontinue treatment.

Table 1: Dose de-escalation for toxicity	
Dose Level	**Alisertib Dose**
Level 0	30 mg by mouth twice daily
Level -1	20 mg by mouth twice daily
Level -2	10 mg by mouth twice daily
Level -3	10 mg by mouth once daily

Patients with non-hematological study drug-related Grade 4 will not be re-dosed with alisertib and will discontinue treatment. In addition, dose reduction will be allowed with persistent non-hematologic grade 2 toxicities, including GI toxicities, at the discretion of the investigator. Dose reductions will be allowed during the induction, consolidation, and maintenance phases of therapy. Once a patient's dose is reduced they should continue at that dose level for the rest of the treatment program.

Should unanticipated circumstances arise that might require minor variances from the prescribed dosing and schedule of the protocol therapy or recommended supportive care, the principal investigator should be contacted in advance for discussion and approval in order to ensure safety and determine/allow patients to continue to receive treatment on study.

6.1.1.1 Drug-Drug Interaction Risks

Preliminary results from the recently completed alisertib human ADME Study C14014 indicate that oxidative metabolism is the predominant route of elimination for alisertib. Preliminary results of subsequent in vitro drug metabolism studies suggest that the oxidative metabolism of alisertib is mainly mediated via CYP3A4. These findings suggest that alisertib systemic exposures may be potentially increased when co-administered with clinically significant CYP3A inhibitors. The clinical effect of co-administration of a CYP3A inhibitor on the pharmacokinetics of alisertib is unknown at this time and a DDI study with itraconazole (a strong CYP3A inhibitor) is planned. Until these results become available, it is recommended that the concomitant use of moderate or strong CYP3A inhibitors or grapefruit juice/grapefruit-containing products be avoided. Selection of an alternate concomitant medication with no or minimal CYP3A inhibition potential is recommended. If co-administration of moderate or strong CYP3A inhibitors is medically necessary, investigators should monitor for toxicities and follow the dose modifications for toxicity per study protocol.

6.1.2 Specific Toxicities and Management

Nausea and Vomiting
Specifically for in relation to alisertib, prophylactic antiemetic therapy will not be used in this study unless it becomes clear that alisertib is causing acute nausea and vomiting. If prophylactic antiemetic therapy is needed, 5-HT_3 receptor antagonists (without corticosteroids) should be tried first. Prophylactic anti-emetics can be otherwise used as per institutional guidelines for other chemotherapy administered during the study.

Although this study will not initially employ prophylactic antiemetics, there is no prohibition against antiemetic use in the management of a patient who develops nausea or vomiting, or both.

Diarrhea
Specifically for use of alisertib, antidiarrheal medications will not be used prophylactically; however, patients can be instructed to take loperamide, 4 mg, at the occurrence of the first loose stool and then 2 mg every 2 hours until they are diarrhea-free for at least 12 hours if thought to be alisertib-related diarrhea. Fluid intake should be maintained to avoid dehydration.

Central Nervous System Effects
If a patient experiences excessive sedation believed to be related to alisertib, treatment with alisertib should be interrupted. Patients whose sedation is not considered immediately life-threatening should be carefully monitored and given appropriate supportive care. If the patient's level of consciousness is considered to be life-threatening, necessary measures should be instituted to secure the airway, ventilation, and intravenous access

6.1.3 Management of cardiac toxicity
Given the rare reports of cardiac toxicities in patients receiving alisertib, extra caution will be exercised to monitor and manage patients in this regard, as necessary. Alisertib will be discontinued if:
A. There is persistent clinically significant arrhythmia thought to be related to Alisertib.
B. Onset of congestive heart failure with related symptoms, and which does not respond promptly to standard management.

6.2 Dose Modifications of Idarubicin (or Daunorubicin) for Related Hepatotoxicity

Initial and subsequent idarubicin (or daunorubicin) doses should be modified as follows for hepatotoxicity:

Total Bilirubin (mg/dL)	**% Dose to Give**
≤ 2	100%
> 2 – ≤ 3.0	75% (25% dose reduction)
> 3.0	50% (50% dose reduction)

Assess total bilirubin daily when administering idarubicin (or daunorubicin) during remission induction or re-induction treatment.

6.3 Dose Modifications for Treatment with cytarabine consolidation therapy

- Contributions of concomitant medications (i.e. psychotropic drugs) to neurotoxicity should be assessed and other medications discontinued if possible.

-For neurotoxicity ≥ grade 2 due to cytarabine during consolidation therapy, discontinue high-dose cytarabine for the remainder of the cycle.

-For patients younger than 60 years, cytarabine may be considered at the subsequent consolidation therapy cycle with a dose modification from 3 g/m^2 to 2 g/m^2 every 12 hours on days 1, 3 and 5 if the toxicity has resolved to ≤ grade 1.

-For patients equal to or greater than 60 years, cytarabine may be considered at the next consolidation therapy cycle with a dose modification from 2 g/m^2 to 1 g/m^2 daily on days 1 through 5 of consolidation if the toxicity has resolved to ≤ grade 1.

-For a second occurrence of neurotoxicity ≥ grade 2, cytarabine for consolidation should be permanently discontinued.

Should unanticipated circumstances arise that might require minor variances from the prescribed dosing of cytarabine or idarubicin (or daunorubicin if indicated) and schedule of the protocol therapy, the principal investigator should be contacted in advance for discussion and approval in order to ensure safety and determine/allow patients to continue to receive treatment on study. Dose reductions for idarubicin (or daunorubicin if indicated) or cytarabine may be implemented if clinically indicated, and after discussion with principal investigator of study, for treatment-related adverse events that do not meet above dose reduction criteria.

7. ADVERSE EVENTS: LIST AND REPORTING REQUIREMENTS

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

7.1 Adverse Event Characteristics

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

  An adverse event (AE) is defined as any untoward medical occurrence, including an exacerbation of a pre-existing condition, in a patient in a clinical investigation who received a pharmaceutical product. The event does not necessarily have to have a causal relationship with this treatment.

  All adverse events occurring during the course of the clinical trial (i.e., from signing the informed consent onwards through the observational phase) will be collected, documented and reported to the sponsor by the investigator according to the specific definitions and instructions detailed in the 'Adverse Event Reporting' section of the

Investigator Site File. The observational phase for AE reporting is defined from the first dose of study treatment until the End of Treatment visit, or 30 days after the last administration of study medication, whichever is the latest.

A serious adverse event (SAE) is defined as any AE which results in death, is immediately life-threatening, results in persistent or significant disability / incapacity, requires or prolongs patient hospitalization, is a congenital anomaly / birth defect, or is to be deemed serious for any other reason if it is an important medical event when based upon appropriate medical judgment which may jeopardize the patient and may require medical or surgical intervention to prevent one of the other outcomes listed in the above definitions.

Patients may be hospitalized for administrative or social reasons during the trial (e.g. days on which infusion takes place, long distance from home to site). These and other hospitalizations planned at the beginning of the trial do not need to be reported as an SAE.

All adverse events, serious and non-serious, will be fully documented on the appropriate CRF(s) / eCRFs. For each adverse event, the investigator will provide the onset, end, intensity, treatment required, outcome, seriousness and action taken with the investigational drug. The investigator will determine the relationship of the investigational drug to all AEs as defined in the 'Adverse Event Reporting' Section of the Investigator Site File.

The basis for judging the intensity of the AE as well as the causal relationship between the investigational product and the AE is described below.

- **For expedited reporting purposes only:**
  - AEs for the agent(s) that are listed above should be reported only if the adverse event varies in nature, intensity or frequency from the expected toxicity information which is provided.
  - The attribution of any AE to the study drug as “definite,” “probable,” or “possible” will be considered as related to the therapy (“yes”), while unlikely and unrelated are considered as not related (“no”) for regulatory reporting purposes.
  - Other AEs for the protocol that do not require expedited reporting are outlined in the next section (Expedited Adverse Event Reporting) under the sub-heading of Protocol-Specific Expedited Adverse Event Reporting Exclusions.

- **Attribution** of the AE:

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

7.1 Adverse Events Associated with Alisertib

Toxicities have been noted in Phase I and II studies involving alisertib, the majority of grade 3 or 4 AEs being hematologic in nature.

Experience with alisertib has been in the single-agent and in the combination (with cytotoxic chemotherapy) setting. Additionally, 3 alisertib formulations (PIC, ECT, and oral solution) have entered clinical studies. The adverse events (AEs) were classified according to the Medical Dictionary for Regulatory Activities (MedDRA).

Treatment-emergent adverse events, regardless of causality, occurring in ≥10% of patients are displayed, as coded by MedDRA System Organ Class (SOC) and Preferred Term in **Table 2** for events noted in single agent studies to date, and in **Table 3** for events noted specifically in single agent studies of hematologic malignancies.

Table 2:

MedDRA System Organ Class MedDRA Preferred Term	Alisertib 50 mg BID 7D Q21 C14001–C14007 + C14012 Incidence N = 530 n (%)
Subjects with at Least 1 Adverse Event	515 (97)
Gastrointestinal disorders	397 (75)
Diarrhoea	222 (42)
Nausea	167 (32)
Stomatitis	164 (31)
Vomiting	120 (23)
Constipation	68 (13)
Abdominal Pain	65 (12)
Glossodynia	
Blood and lymphatic system disorders	380 (72)
Neutropenia	251 (47)
Anaemia	237 (45)
Thrombocytopenia	147 (28)
Leukopenia	146 (28)
Febrile neutropenia	64 (12)
General disorders and administration site conditions	377 (71)
Fatigue	246 (46)
Pyrexia	108 (20)
Asthenia	72 (14)
Oedema peripheral	70 (13)
Chills	
Skin and subcutaneous tissue disorders	299 (56)
Alopecia	186 (35)
Rash	
Nervous system disorders	236 (45)
Somnolence	104 (20)
Headache	57 (11)
Dizziness	56 (11)
Metabolism and nutrition disorders	241 (45)
Decreased appetite	114 (22)
Dehydration	56 (11)
Hypokalaemia	
Respiratory, thoracic and mediastinal disorders	217 (41)
Dyspnoea	86 (16)
Cough	66 (12)
Respiratory tract congestion	
Dyspnoea exertional	
Oropharyngeal pain	
Sinus congestion	
Infections and infestations	203 (38)
Oral candidiasis	
Musculoskeletal and connective tissue disorders	173 (33)
Pain in Extremity	
Back pain	
Musculoskeletal	
Musculoskeletal chest pain	
Investigations	175 (33)
Weight decreased	
White blood cell count decreased	
Neutrophil count decreased	
Blood magnesium decreased	
Psychiatric disorders	107 (20)
Confusional state	
Vascular disorders	74 (14)
Injury, poisoning and procedural complications	57 (11)
Ligament sprain	
Fall	
Eye disorders	
Renal and urinary disorders	53 (10)
Reproductive system and breast disorders	
Scrotal swelling	

Table 3

MedDRA System Organ Class MedDRA Preferred Term	Incidence N = 155 n (%)
Subjects with at Least 1 Adverse Event	**155 (100)**
Gastrointestinal disorders	**120 (77)**
Diarrhoea	70 (45)
Nausea	51 (33)
Stomatitis	40 (26)
Vomiting	32 (21)
Abdominal pain	26 (17)
Constipation	20 (13)
Blood and lymphatic system disorders	**115 (74)**
Neutropenia	69 (45)
Anaemia	65 (42)
Thrombocytopenia	54 (35)
Leukopenia	44 (28)
Febrile neutropenia	31 (20)
General disorders and administration site conditions	**110 (71)**
Fatigue	69 (45)
Asthenia	26 (17)
Pyrexia	25 (16)
Oedema peripheral	21 (14)
Infections and infestations	**80 (52)**
Skin and subcutaneous tissue disorders	**80 (52)**
Alopecia	46 (30)
Nervous system disorders	**78 (50)**
Somnolence	40 (26)
Dizziness	17 (11)
Respiratory, thoracic and mediastinal disorders	**75 (48)**
Dyspnoea	28 (18)
Cough	25 (16)
Metabolism and nutrition disorders	**60 (39)**
Anorexia	23 (15)
Dehydration	18 (12)
Investigations	**51 (33)**
Musculoskeletal and connective tissue disorders	**50 (32)**
Psychiatric disorders	**35 (23)**
Vascular disorders	**29 (19)**
Hypotension	19 (12)
Cardiac disorders	**23 (15)**
Injury, poisoning and procedural complications	**23 (15)**
Renal and urinary disorders	**19 (12)**

The most common treatment-emergent adverse events of ≥ grade 3 severity are displayed by causality, as coded by MedDRA SOC and Preferred Term, in **Table 4** for single-agent alisertib in

studies performed in Europe and US, and in **Table 5** for single-agent alisertib in studies in hematologic malignancies.

Table 4

		Alisertib 50 mg BID 7D Q21 C14001–C14007 and C14012
MedDRA System Organ Class MedDRA Preferred Term		**N = 530 n (%)**
Subjects with at Least 1 Grade ≥ 3 Adverse Event	Incidence	420 (79)
	Related	332 (63)
Blood and lymphatic system disorders	Incidence	306 (58)
	Related	271 (51)
Neutropenia	Incidence	209 (39)
	Related	202 (38)
Leukopenia	Incidence	109 (21)
	Related	97 (18)
Anaemia	Incidence	103 (19)
	Related	70 (13)
Thrombocytopenia	Incidence	82 (15)
	Related	69 (13)
Febrile neutropenia	Incidence	59 (11)
	Related	44 (8)
Gastrointestinal disorders	Incidence	111 (21)
	Related	76 (14)
Abdominal pain	Incidence	
	Related	
General disorders and administration site conditions	Incidence	87 (16)
	Related	50 (9)
Investigations	Incidence	89 (17)
	Related	64 (12)
Neutrophil count decreased	Incidence	
	Related	
Infections and infestations	Incidence	72 (14)
	Related	21 (4)
Metabolism and nutrition disorders	Incidence	60 (11)
	Related	22 (4)

Table 5

MedDRA System Organ Class MedDRA Preferred Term	Incidence N = 155 n (%)	Causality Related	 Not Related
Subjects with Any Grade ≥ 3 Treatment-Emergent AE	**134 (86)**	**97 (63)**	**37 (24)**
Blood and lymphatic system disorders	**94 (61)**	**78 (50)**	**16 (10)**
Neutropenia	58 (37)	55 (35)	3 (2)
Thrombocytopenia	40 (26)	34 (22)	6 (4)
Anaemia	39 (25)	29 (19)	10 (6)
Leukopenia	38 (25)	34 (22)	4 (3)
Febrile neutropenia	26 (17)	15 (10)	11 (7)
Infections and infestations	**36 (23)**	**7 (5)**	**29 (19)**
General disorders and administration site conditions	**27 (17)**	**14 (9)**	**13 (8)**
Gastrointestinal disorders	**26 (17)**	**18 (12)**	**8 (5)**
Investigations	**21 (14)**	**17 (11)**	**4 (3)**
Metabolism and nutrition disorders	**18 (12)**	**6 (4)**	**12 (8)**
Nervous system disorders	**17 (11)**	**10 (6)**	**7 (5)**

7.2.1 Treatment-Related Adverse Events, Grade 3 or Greater

The data from series of trials of single agent alisertib in US and Europe demonstrates a 39% incidence of grade 3 or greater treatment-related neutropenia, while the incidence of infection and febrile neutropenia was 14% and 11%, respectively (**Table 4**). In single-agent studies, the most common (at least 10% of patients) adverse events, regardless of causality and severity, were blood and lymphatic system disorders (72%) and gastrointestinal disorders (75%), although fatigue (46%) and skin manifestations (56%) were also common. Within the blood and lymphatic category, leukopenia, anemia, and thrombocytopenia were reported as a grade ≥ 3 events in 21%, 19%, and 15% of the patients, respectively, in single-agent studies. Gastrointestinal Disorders had an incidence of 21%.

For studies conducted in patients with hematologic malignancies (**Table 5**), ≥ grade 3 neutropenia and thrombocytopenia had an incidence of 37% and 26%, respectively. ≥ grade 3 anemia and febrile neutropenia occurred in 25% and 17% of patients, respectively. Gastrointenstinal disorderswere seen in 17% of patients. Results from the phase 1 and phase 2 studies indicated that the more frequent toxicities can be monitored by routine clinical evaluations, and represent mechanistic effects in proliferating tissues (bone marrow, GI epithelium, hair follicles). While the reversibility of alopecia has not been established, other prevalent toxicities (myelotoxicities and GI disorders) are largely reversible and manageable by dose reduction, interruption of the planned dosing schedule, or supportive care.

Although the majority of treatment-related SAEs reflect mechanistic toxicities of alisertib in proliferative tissues (bone marrow, GI epithelium, hair follicles), some SAEs have been reported that appear to be possible off-target effects unrelated to already well established Aurora A kinase functions. For example, two patients in two separate studies developed cardiac abnormalities including left ventricular dysfunction and ejection fraction decrease. In both patients, a significant decrease in LVEF was observed during or after the first treatment cycle and prior anthracycline therapies were noted in addition to confounding histories and comorbidities. These events were confounded by pre-existing morbidities including prior radiation to the heart, possible tumor involvement of the pericardium, and prior treatment-resistant tachycardia. Cardiac toxicity was not seen in a phase I study of alisertib in combination 7+3 induction.

In addition, skin rash has been described as an identified risk for treatment with alisertib. Palmar-plantar erythrodysaesthesia syndrome has been observed predominantly in a pediatric patient population, with 2 such serious cases occurring in an adult patient population. One patient experienced a serious case of bullous dermatitis. There was no mucous membrane involvement and the event improved with antibiotics, methylprednisolone, and local wound care.

7.3 Adverse Events Associated with Idarubicin and Cytarabine

Please refer to the appropriate package inserts for the comprehensive list of adverse events for each agent.

Idarubicin: Toxicities includes myelosuppression and cardiotoxicity. Cumulative dose beyond approximately 150 mg/m^2 (and equivalent cumulative doses with other anthracyclines) results in increased risk for myocardial injury, and congestive heart failure. Radiation therapy involving the heart and previous anthracycline administration increases the risk for cardiomyopathy. Hepatic and renal dysfunction may occur. Other reactions include reversible alopecia, nausea and vomiting, anorexia, diarrhea and mucositis. If extravasation occurs during administration, tissue necrosis can result. Rarely, chills, fever, skin rash and anaphylactoid reactions can occur. The occurrence of secondary acute leukemia has been reported rarely in patients treated with anthracycline chemotherapy.

Cytarabine: Toxicities include myelosuppression, nausea, vomiting, diarrhea, anorexia, anal ulceration, stomatitis, rash, headache, fever, myalgia, malaise, bone pain, chest pain, hepatic and renal dysfunction, and alopecia. Central nervous system toxicity, i.e., significant cerebral and cerebellar, dysfunction, progression to coma, has been seen with high doses, such as those given in consolidation. Severe cardiomyopathy has been reported with high dose Ara-C in combination with cyclophosphamide. Progressive ascending paralysis has occurred in two patients receiving IV and intrathecal ara-C. Marked keratoconjunctivitis has also occurred with high doses. Ara-C

can cause fetal harm when administered to a pregnant woman; however, there are no adequate and well-controlled studies in pregnant women.

7.4 Adverse Event Reporting Definitions

7.4.1 Adverse Event (AE):

An adverse event (AE) is any undesirable sign, symptom or medical condition or experience that develops or worsens in severity after starting the first dose of study treatment or any procedure specified in the protocol, even if the event is not considered to be related to the study.

Abnormal laboratory values or diagnostic test results constitute adverse events only if they induce clinical signs or symptoms or require treatment or further diagnostic tests.

7.4.2 Serious adverse event (SAE):

A serious adverse event (SAE) is any adverse event, occurring at any dose and regardless of causality that:

- Results in death
- Is life-threatening. Life-threatening means that the person was at immediate risk of death from the reaction as it occurred, i.e., it does not include a reaction which hypothetically might have caused death had it occurred in a more severe form.
- Requires or prolongs inpatient hospitalization (i.e., the event required at least a 24-hour hospitalization or prolonged a hospitalization beyond the expected length of stay). Hospitalization admissions and/or surgical operations scheduled to occur during the study period, but planned prior to study entry are not considered SAEs if the illness or disease existed before the person was enrolled in the trial, provided that it did not deteriorate in an unexpected manner during the trial (e.g., surgery performed earlier than planned).
- Results in persistent or significant disability/incapacity. Disability is defined as a substantial disruption of a person's ability to conduct normal life functions.
- Is a congenital anomaly or birth defect; or
- Is an important medical event when, based upon appropriate medical judgment, it may jeopardize the participant and require medical or surgical intervention to prevent one of the outcomes listed above. Examples of such medical events include allergic bronchospasm requiring intensive

treatment in an emergency room or at home; blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse.

Events **not** considered to be serious adverse events are hospitalizations for:

- routine treatment or monitoring of the studied indication, not associated with any deterioration in condition, or for elective procedures
- elective or pre-planned treatment for a pre-existing condition that did not worsen
- emergency outpatient treatment for an event not fulfilling the serious criteria outlined above and not resulting in inpatient admission
- respite care

7.4.3 Expectedness - Adverse events can be 'Expected' or 'Unexpected.'

Expected adverse event
Expected adverse events are those that have been previously identified as resulting from administration of the agent. For the purposes of this study, an adverse event is considered expected when it appears in the current adverse event list, the Investigator's Brochure, the package insert or is included in the informed consent document as a potential risk.

Unexpected adverse event
For the purposes of this study, an adverse event is considered unexpected when it varies in nature, intensity or frequency from information provided in the current adverse event list, the Investigator's Brochure, the package insert or when it is not included in the informed consent document as a potential risk.

7.4.4 Attribution

Attribution is the relationship between an adverse event or serious adverse event and the study treatment. Attribution will be assigned as follows:

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

7.5 Procedures for AE and SAE Recording and Reporting

Participating investigators will assess the occurrence of AEs and SAEs at all participant evaluation time points during the study.

All AEs and SAEs whether reported by the participant, discovered during questioning, directly observed, or detected by physical examination, laboratory test or other means, will be recorded in the participant's medical record and on the appropriate study-specific case report forms.

The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 4.0 will be utilized for AE reporting. All appropriate treatment areas should have access to a copy of the CTCAE version 4.0.

A copy of the CTCAE version 4.0 can be downloaded from the CTEP website at: http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm.

7.5.1 Reporting to the Study Sponsor

7.5.1.1 Serious Adverse Event Reporting:

All serious adverse events that occur after the initial dose of study treatment, during treatment, or within 30 days of the last dose of treatment must be reported to the DF/HCC Overall Principal Investigator on the local institutional SAE form. This includes events meeting the criteria outlined in Section 7.4.2, as well as the following:

- Grade 2 (moderate) and Grade 3 (severe) Events – Only events that are unexpected and possibly, probably or definitely related/associated with the intervention.

- All Grade 4 (life-threatening or disabling) Events – Unless expected AND specifically listed in the protocol as not requiring reporting.

- All Grade 5 (fatal) Events – When the participant is enrolled and actively participating in the trial OR when the event occurs within 30 days of the last study intervention.

  Note: If the participant is in long term follow up, report the death at the time of continuing review.

Participating investigators must report each serious adverse event to the DF/HCC Overall Principal Investigator within 24 hours of learning of the occurrence. In the event that the participating investigator does not become aware of the serious adverse event immediately (e.g., participant sought treatment elsewhere), the participating investigator is to report the event within 24 hours after learning of it and document the time of his or her first awareness of the adverse event. Report

serious adverse events by telephone, email or facsimile to:

Overall Principal Investigator: Amir T. Fathi MD
[redacted]

Within the following 24-48 hours, the participating investigator must provide follow-up information on the serious adverse event. Follow-up information should describe whether the event has resolved or continues, if and how the event was treated, and whether the participant will continue or discontinue study participation.

AEs which are serious must be reported to Millennium Pharmacovigilance's SAE Reporting designee, Cognizant Contacts, from first dose of alisertib up to and including 30 days after administration of the last dose. When possible, signs and symptoms indicating a common underlying pathology should be noted as one comprehensive event. Any SAE that occurs at any time after completion of alisertib treatment or after the designated follow-up period that the investigator and/or sub-investigator considers to be related to any study drug must be reported to Cognizant Contacts. Planned hospital admissions or surgical procedures for an illness or disease that existed *before the patient was enrolled in the trial* are not to be considered AEs unless the condition deteriorated in an unexpected manner during the trial (e.g., surgery was performed earlier or later than planned). All SAEs should be monitored until they are resolved or are clearly determined to be due to a patient's stable or chronic condition or intercurrent illness(es).

This is an investigator-initiated study. The principal investigator, Amir T. Fathi, MD (who may also sometimes be referred to as the sponsor-investigator), is conducting the study and acting as the sponsor. Therefore, the regulatory obligations of the principal investigator include both those of a sponsor and those of an investigator.

Sponsor-investigator must report all SAEs, regardless of expectedness or relationship with any study drug, to Cognizant Contacts as soon as possible, but no later than 5 calendar days of the sponsor-investigator's observation or awareness of the event. In the event that this is a multisite study, the sponsor-investigator is responsible to ensure that the SAE reports are sent to Cognizant Contacts from all sites participating in the study. Subinvestigators must report all SAEs to the sponsor-investigator so that the sponsor-investigator can meet his/her foregoing reporting obligations to Cognizant Contacts, unless otherwise agreed between the sponsor-investigator and subinvestigator(s). Cognizant Contacts may request follow-up information to a reported SAE, which the sponsor-investigator will be responsible for providing to Cognizant Contacts.

Millennium will provide a sample SAE Report Form representative of the information Millennium Pharmacovigilance/Cognizant Contacts may request in follow-up.

The SAE report must include event term(s), serious criteria, and the investigator's or sub-investigator's determination of both the intensity of the event(s) and the relationship of the event(s) to study drug administration.

Intensity for each SAE, including any lab abnormality, will be determined by using the NCI CTCAE, version used at your institution, as a guideline, whenever possible.The criteria are available online at http://ctep.cancer.gov/reporting/ctc.html.

Relationship to all study drugs for each SAE will be determined by the investigator or sub-investigator by responding yes or no to the question: Is there a reasonable possibility that the AE is associated with the study drug(s)?

Sponsor-investigator must also provide Cognizant Contacts with a copy of all communications with applicable regulatory authorities related to the study or study drug(s), including, but not limited to, telephone conversation logs, as soon as possible but no later than 5 calendar days of such communication.

Sponsor-investigator will be responsible for forwarding such reports to any subinvestigator(s).

Millennium Pharmacovigilance Designee
SAE and Pregnancy Reporting Contact Information:
[redacted]

Suggested Reporting Form: Millennium Serious Adverse Events Form (provided by Millennium)

7.5.1.2 Non-Severe Adverse Event Reporting:

Non-serious adverse events will be reported to the DF/HCC Overall Principal Investigator on the toxicity Case Report Forms.

7.5.1.3 Procedures for Reporting Drug Exposure during Pregnancy and Birth Events

If a woman becomes pregnant or suspects that she is pregnant while participating in this study, she must inform the investigator immediately and must permanently discontinue study drug(s). All pregnancies and suspected pregnancies must be reported to Cognizant Contacts (see Section 4.2 for contact information) immediately. The pregnancy must be followed for the final pregnancy outcome (ie, delivery, still birth, miscarriage) and Cognizant Contacts will request this information from the investigator.

If a female partner of a male patient becomes pregnant during the male patient's

participation in this study, this must be reported to Cognizant Contacts immediately (see Section **Error! Reference source not found.** for contact information). Every effort should be made to follow the pregnancy for the final pregnancy outcome.

Suggested Pregnancy Reporting Form:

Pregnancy Report Form (Provided by Millennium)

7.5.2 Reporting to the Institutional Review Board (IRB)

Investigative sites within DF/HCC will report all serious adverse events directly to the DFCI Office for Human Research Studies (OHRS).

Other investigative sites should report serious adverse events to their respective IRB according to the local IRB's policies and procedures in reporting adverse events. A copy of the submitted institutional SAE form should be forwarded to:

Overall Principal Investigator: Amir T. Fathi MD

[redacted]

The DF/HCC Principal Investigator will submit SAE reports from outside institutions to the DFCI Office for Human Research Studies (OHRS) according to DFCI IRB policies and procedures in reporting adverse events.

7.5.3 Reporting to Hospital Risk Management

Participating investigators will report to their local Risk Management office any subject safety reports or sentinel events that require reporting according to institutional policy.

7.6 Monitoring of Adverse Events and Period of Observation

All adverse events, both serious and non-serious, and deaths that are encountered from initiation of study intervention, throughout the study, and within 30 days of the last study intervention should be followed to their resolution, or until the participating investigator assesses them as stable, or the participating investigator determines the event to be irreversible, or the participant is lost to follow-up. The presence and resolution of AEs and SAEs (with dates) should be documented on the appropriate case report form and recorded in the participant's medical record to facilitate source

data verification.

For some SAEs, the study sponsor or designee may follow-up by telephone, fax, and/or monitoring visit to obtain additional case details deemed necessary to appropriately evaluate the SAE report (e.g., hospital discharge summary, consultant report, or autopsy report).

Participants should be instructed to report any serious post-study event(s) that might reasonably be related to participation in this study. Participating investigators should notify the DF/HCC Overall Principal Investigator and their respective IRB of any unanticipated death or adverse event occurring after a participant has discontinued or terminated study participation that may reasonably be related to the study.

7.7 Product Complaints

A product complaint is a verbal, written, or electronic expression that implies dissatisfaction regarding the identity, strength, purity, quality, or stability of a drug product. Individuals who identify a potential product complaint situation should immediately contact MedComm Solutions (see below) and report the event. Whenever possible, the associated product should be maintained in accordance with the label instructions pending further guidance from a Millennium Quality representative.

For Product Complaints,
call MedComm Solutions at
[redacted]
(US and International)

Product complaints in and of themselves are not AEs. If a product complaint results in an SAE, an SAE form should be completed and sent to Millennium Pharmacovigilance (refer to Section 8.2).

A product complaint is a verbal, written, or electronic expression that implies dissatisfaction regarding the identity, strength, purity, quality, or stability of a drug product. Individuals who identify a potential product complaint situation should immediately contact MedComm Solutions (see below) and report the event. Whenever possible, the associated product should be maintained in accordance with the label instructions pending further guidance from a Millennium Quality representative.

A medication error is a preventable event that involves an identifiable patient and that leads to inappropriate medication use, which may result in patient harm. While overdoses and underdoses constitute medication errors, doses missed inadvertently by a patient do not. Individuals who identify a potential medication error situation should immediately contact MedComm Solutions (see below) and report the event.

8. DATA AND SAFETY MONITORING

8.1 Data Reporting

8.1.1 **Method**

The QACT will collect, manage, and monitor data for this study.

8.1.2 **Data Submission**

The schedule for completion and submission of case report forms (paper or electronic) to the QACT is as follows:

Form	**Submission Timeline**
Eligibility Checklist	Complete prior to registration with QACT
On Study Form	Within 14 days of registration
Baseline Assessment Form	Within 14 days of registration
Treatment Form	Within 10 days of the last day of the cycle
Adverse Event Report Form	Within 10 days of the last day of the cycle
Response Assessment Form	Within 10 days of the completion of the cycle required for response evaluation
Off Treatment/Off Study Form	Within 14 days of completing treatment or being taken off study for any reason
Follow up/Survival Form	Within 14 days of the protocol defined follow up visit date or call

8.2 Safety Meetings

The DF/HCC Data and Safety Monitoring Committee (DSMC) will review and monitor toxicity and accrual data from this trial. The committee is composed of clinical specialists with experience in oncology and who have no direct relationship with the study. Information that raises any questions about participant safety will be addressed with the Principal Investigator and study team.

The DSMC will meet quarterly and/or more often if required to review toxicity and accrual data. Information to be provided to the committee may include: up-to-date participant accrual; current dose level information; DLT information; all grade 2 or higher unexpected adverse events that have been reported; summary of all deaths occurring within 30 days for Phase I or II protocols; for gene transfer protocols, summary of all deaths while being treated and during active follow-up; any response information; audit results, and a summary provided by the study team. Other information (e.g. scans, laboratory values) will be provided upon request.

8.3 Monitoring

Involvement in this study as a participating investigator implies acceptance of potential audits or inspections, including source data verification, by representatives designated by the DF/HCC Overall Principal Investigator (or Protocol Chair) or DF/HCC. The purpose of these audits or inspections is to examine study-related activities and documents to determine whether these activities were conducted and data were recorded, analyzed, and accurately reported in accordance with the protocol, institutional policy, Good Clinical Practice (GCP), and any applicable regulatory requirements.

All data will be monitored for timeliness of submission, completeness, and adherence to protocol requirements. Monitoring will begin at the time of participant registration and will continue during protocol performance and completion.

9. REGULATORY CONSIDERATIONS

9.1 Protocol Review and Amendments

This protocol, the proposed informed consent and all forms of participant information related to the study (e.g., advertisements used to recruit participants) and any other necessary documents must be submitted, reviewed and approved by a properly constituted IRB governing each study location.

Any changes made to the protocol must be submitted as amendments and must be approved by the IRB prior to implementation. Any changes in study conduct must be reported to the IRB. The DF/HCC Overall Principal Investigator (or Protocol Chair) will disseminate protocol amendment information to all participating investigators.

All decisions of the IRB concerning the conduct of the study must be made in writing.

9.2 Informed Consent

All participants must be provided a consent form describing this study and providing sufficient information for participants to make an informed decision

about their participation in this study. The formal consent of a participant, using the IRB approved consent form, must be obtained before the participant is involved in any study-related procedure. The consent form must be signed and dated by the participant or the participant's legally authorized representative, and by the person obtaining the consent. The participant must be given a copy of the signed and dated consent document. The original signed copy of the consent document must be retained in the medical record or research file.

All patient confidentiality will be respected. All study members will comply fully with all HIPPA regulations and policies in the administration of this study. All research team members are to follow the institutional HIPAA policies and procedures, including the 'minimal use' of protected health information when conducting recruitment activities, determining eligibility and retaining research charts. We will document these discussions and file the documentation in the regulatory file.

9.3 Ethics and Good Clinical Practice (GCP)

This study is to be conducted according to the following considerations, which represent good and sound research practice:

- E6 Good Clinical Practice: Consolidated Guidance
  www.fda.gov/downloads/RegulatoryInformation/Guidances/UCM129515.pdf

- US Code of Federal Regulations (CFR) governing clinical study conduct and ethical principles that have their origin in the Declaration of Helsinki

    - Title 21 Part 11 – Electronic Records; Electronic Signatures
      www.access.gpo.gov/nara/cfr/waisidx_02/21cfr11_02.html

    - Title 21 Part 50 – Protection of Human Subjects
      www.access.gpo.gov/nara/cfr/waisidx_02/21cfr50_02.html

    - Title 21 Part 54 – Financial Disclosure by Clinical Investigators
      www.access.gpo.gov/nara/cfr/waisidx_02/21cfr54_02.html

    - Title 21 Part 56 – Institutional Review Boards
      www.access.gpo.gov/nara/cfr/waisidx_02/21cfr56_02.html

    - Title 21 Part 312 – Investigational New Drug Application
      www.access.gpo.gov/nara/cfr/waisidx_02/21cfr312_02.html

- State laws

- DF/HCC research policies and procedures
  http://www.dfhcc.harvard.edu/clinical-research-support/clinical-research-unit-cru/policies-and-procedures/

It is understood that deviations from the protocol should be avoided, except when necessary to eliminate an immediate hazard to a research participant. In such case, the deviation must be reported to the IRB according to the local reporting policy.

9.4 Study Documentation

The investigator must prepare and maintain adequate and accurate case histories designed to record all observations and other data pertinent to the study for each research participant. This information enables the study to be fully documented and the study data to be subsequently verified.

Original source documents supporting entries in the case report forms include but are not limited to hospital records, clinical charts, laboratory and pharmacy records, recorded data from automated instruments, microfiches, photographic negatives, microfilm or magnetic media, and/or x-rays.

9.5 Records Retention

All study-related documents must be retained for the maximum period required by applicable federal regulations and guidelines or institutional policies.

10. PHARMACEUTICAL INFORMATION

10.1 Alisertib (MLN8237)

As stated above, in its solid state, alisertib (MLN8237-004) drug substance is an off-white to yellow solid with an assay value of 96.0% to 104.0% (w/w) on an anhydrous basis, as determined by reverse phase high performance liquid chromatography (HPLC). The solubility of the drug substance in water (pH = 8.4) is 8.5mg/mL. The drug substance is a stable monohydrate with a pKa of 4.53 for the free acid.

Formulation

An enteric coated tablet dosage form of MLN8237-004 has been developed for use in clinical studies. Three different enteric coated tablet dosage strengths, 10, 50, and 100 mg, expressed as MLN8237 free acid, were formulated. The key formulation excipients that aid in the in vivo absorption of the drug are sodium bicarbonate, sodium lauryl sulfate, and enteric coating. The

other formulation excipients such as povidone, microcrystalline cellulose, croscarmellose sodium, and sodium stearyl fumarate serve as manufacturing aids.

The study drug, as provided by Millennium, will be labeled and handled at the investigative site as open-label material; packaging labels will fulfill all requirements specified by governing regulations. MLN8237 will be supplied as ECT in 10 mg strength. The 60-cc HDPE bottles will have a child-resistant cap and be labeled for take-home use. Patients will receive instructions for home use of MLN8237, including the requirement that MLN8237 be administered as intact tablets

Storage
Alisertib (MLN8237-004) enteric-coated tablets are packaged in HDPE bottles with rayon coil, induction seal, desiccant packs, and polypropylene child-resistant cap and stored at USP-controlled room temperature (20°C to 25ºC) with excursions permitted from 15°C to 30°C.

Handling
Alisertib (MLN8237-004) drug product is a cytotoxic anticancer drug. As with other potentially toxic compounds, caution should be exercised when handling the 3 dosage forms of MLN8237-004 (capsules, tablets, and oral solution). Refer to published guidelines regarding the proper handling and disposal of anticancer agents.[1, 2]

10.2 Idarubicin

Desription: (Idarubicin® package insert): Idarubicin HCl Injection contains idarubicin hydrochloride and is a sterile, semi-synthetic, preservative-free solution antineoplastic anthracycline for intravenous use. Chemically, idarubicin hydrochloride is (1S,3S)-3-Acetyl-1,2,3,4,6,11-hexahydro-3,5,12-trihydroxy-6,11-dioxo-1-naphthacenyl-3-amino-2,3,6-trideoxy-α-L-lyxo-hexopyranoside, hydrochloride.
Molecular weight: 533.95
Molecular formula: C26H27NO9•HCl

Idarubicin HCl Injection is a sterile, red-orange, isotonic parenteral preservative-free solution, available in 5 mL (5 mg), 10 mL (10 mg) and 20 mL (20 mg) single-use-only vials. Each mL contains idarubicin HCI 1 mg and the following inactive ingredients: glycerin 25 mg and water for injection q.s. Hydrochloric acid is used to adjust the pH to a target of 3.5.

Warnings: Idarubicin is intended for administration under the supervision of a physician who is experienced in leukemia chemotherapy. Idarubicin is a potent bone marrow suppressant. Idarubicin should not be given to patients with pre-existing bone marrow suppression induced by previous drug therapy or radiotherapy unless the benefit warrants the risk.

Severe myelosuppression will occur in all patients given a therapeutic dose of this agent for induction, consolidation or maintenance. Careful hematologic monitoring is required. Deaths due to infection and/or bleeding have been reported during the period of severe myelosuppression. Facilities with laboratory and supportive resources adequate to monitor drug tolerability and protect and maintain a patient compromised by drug toxicity should be available. It must be possible to treat rapidly and completely a severe hemorrhagic condition and/or a severe infection.

Pre-existing heart disease and previous therapy with anthracyclines at high cumulative doses or other potentially cardiotoxic agents are co-factors for increased risk of idarubicin-induced cardiac toxicity and the benefit to risk ratio of idarubicin therapy in such patients should be weighed before starting treatment with idarubicin. Myocardial toxicity as manifested by potentially fatal congestive heart failure, acute lifethreatening arrhythmias or other cardiomyopathies may occur following therapy with idarubicin. Appropriate therapeutic measures for the management of congestive heart failure and/or arrhythmias are indicated. Cardiac function should be carefully monitored during treatment in order to minimize the risk of cardiac toxicity of the type described for other anthracycline compounds. The risk of such myocardial toxicity may be higher following concomitant or previous radiation to the mediastinal-pericardial area or in patients with anemia, bone marrow depression, infections, leukemic pericarditis and/or myocarditis. While there are no reliable means for predicting congestive heart failure, cardiomyopathy induced by anthracyclines is usually associated with a decrease of the left ventricular ejection fraction (LVEF) from pretreatment baseline values.

Since hepatic and/or renal function impairment can affect the disposition of idarubicin, liver and kidney function should be evaluated with conventional clinical laboratory tests (using serum bilirubin and serum creatinine as indicators) prior to and during treatment.

Please refer to the product's package insert for full prescribing and toxicity information.

10.3 Cytarabine (Cytosine Arabinoside) (Ara-C)

Description: (Cytarabine® package insert) Cytarabine for Injection, commonly known as ara-C, an antineoplastic, is a sterile lyophilized material for reconstitution and intravenous, intrathecal or subcutaneous administration. It is available in multi-dose vials containing 100 mg, 500 mg, 1 g or 2 g sterile cytarabine. The pH of Cytarabine for Injection, is adjusted, when necessary, with hydrochloric acid and/or sodium hydroxide. Cytarabine is chemically 4-amino-1-ß-D-arabinofuranosyl-2(1H)-pyrimidinone.
Molecular weight: 243.22
Molecular formula: C9H13N3O5

Cytarabine is an odorless, white to off-white, crystalline powder which is freely soluble in water and slightly soluble in alcohol and in chloroform.

Warnings: For induction therapy, patients should be treated in a facility with laboratory and supportive resources sufficient to monitor drug tolerance and protect and maintain a patient compromised by drug toxicity. The main toxic effect of Cytarabine at doses used during induction is bone marrow suppression with leukopenia, thrombocytopenia and anemia. Less serious toxicity includes nausea, vomiting, diarrhea and abdominal pain, oral ulceration, and hepatic dysfunction. The physician must judge possible benefit to the patient against known toxic effects of this drug in considering the advisability of therapy with Cytarabine for Injection.

Please refer to the product's package insert for full prescribing and toxicity information.
The investigator, or a responsible party designated by the investigator, must maintain a careful record of the inventory and disposition of the agent using the NCI Drug Accountability Record or another comparable drug accountability form. (See the CTEP website at http://ctep.cancer.gov/protocolDevelopment for the "Policy and Guidelines for Accountability and Storage of Investigational Agents" or to obtain a copy of the drug accountability form.)

11. BIOMARKER, CORRELATIVE, AND SPECIAL STUDIES

11.1 Laboratory Correlative Studies

Bone marrow aspirate samples will be obtained prior to treatment, at mid-treatment assessment (range day 13-16), and at count recovery (day 42 or day 63 if 5+2 re-induction was given), or according to the investigator's discretion, whichever occurs first, , as well as at any time of suspected relapse. Additionally, although not mandated, if patients agree to a skin biopsy, a single 6mm punch biopsy will be performed concurrently during the bone marrow aspiration procedure on day 14 (window day 13-16). The punch biopsy will be collected in a specimen cup containing saline (NOT formalin).

The study will require the following volumes of aspirated bone marrow in purple top tubes (if inaspirable, peripheral blood in purple top tubes may be substituted):

- Pre-treatment: Bone marrow aspirate (5-10 mL) [or Blood (20 mL)]
- Day 14 (Range days 13-16): Bone marrow aspirate (5-10 mL) [or Blood (20 mL)]; skin biopsy (6mm punch)- optional
- Count recovery or at Day 42 (Range days 35-45) – or day 63 if 5+2 given (Range days 55-65)): Bone marrow aspirate (5-10) mL [or Blood (20 mL)]
- In the event of suspected/confirmed relapse: Bone marrow aspirate (5-10 mL) [or Blood (20 mL)]

All specimens should be maintained at room temperature and delivered within 1 day to the processing laboratories. Specimens may be shipped via FedEx priority overnight Monday through

Thursday (excluding holidays). For after hours/evening collections, specimens may be maintained at room temperature and shipped within 4 days of collection. Specimens should be labeled with study 15-334, pt initials, pt study ID number, date and time collection, specimen type: blood, marrow, skin, and must be accompanied by the appropriate requisition form. Bone marrow and/or blood specimens should be delivered to:

Skin biopsies should be delivered to:

We will perform gene panel sequencing ("Rapid Heme Panel") on bone marrow aspirate samples (or peripheral blood) collected at study enrollment, mid-treatment, count recovery, and at relapse/suspicion of relapse. In selected cases, remaining DNA from bone marrow (or peripheral blood) and skin biopsies will be subjected to broader sequence analysis (whole exome or genome). These exploratory analyses will be performed with several hypotheses in mind. First, we will evaluate whether a specific mutational profile is associated with treatment response. Second, we will determine whether there is selective sensitivity to therapy among specific founding clones and/or subclones. Third, we will investigate somatic mutation patterns that are associated with disease progression. Additional laboratory studies will be performed to characterize the functional consequences of aurora kinase inhibition on leukemic blasts during treatment.

12. STUDY CALENDAR

Baseline evaluations are to be conducted within 14 days prior to start of protocol therapy.

Assessments must be performed prior to administration of any study agent.

Of note, the cycle 1, Day 1 labs in Calendar below do not need to be re-assessed for purposes for study enrollment eligibility.	**Baseline** (within 14 days of enrollment)	**Day 1**	**Day 8 (Range 6-10)**	**Day 14 (Range 13-16)**	**Count recovery or Day 43 (Range 35-45), or Day 63 (if 5+2 given, range 55-65)**	**Day 1 of each Consolidation cycle (+/- 3 days)**	**Day 1 of each Maintenance cycle (+/- 3 days)**	**Relapse**
ASSESSMENTS								
Written consent	X							
Medical history	X							
Review of prior therapy	X							
Physical exam	X	X	X	X	X	X	X	X
Vital signs	X	X	X	X	X	X	X	X
Weight	X	X	X	X	X	X	X	X
Height	X							
Concomitant meds	X	X	X	X	X	X	X	X
Performance status	X					X	X	X
Symptom and Toxicity Notation	X	X	X	X	X	X	X	X
LABORATORY EVALUATIONS								
CBC w/ diff	X	X	X	X	X	X	X	X
Complete Chemistry Panel (including magnesium and phosphate)	X	X	X	X	X	X	X	X
ALT, AST, total bili, direct bili	X	X	X	X	X	X	X	X
LDH	X	X	X	X	X	X	X	X
Uric acid	X	X	X	X	X	X	X	X
PT, PTT	X							
D-Dimer, fibrinogen	X							
Hepatitis B S Ag and serology	X							
Hepatitis C serology	X							

Serum/urine pregnancy test for females of child-bearing potential	X							
EKG	X							
MUGA scan or echocardiogram	X							
DISEASE STATUS								
Bone marrow biopsy/aspirate	X			X	X			X
Skin Biopsy (Optional)				X				
TREATMENT **Alisertib** to be administered twice daily starting on day 8 (or 9) for 7 days during induction, given days 6-12 following cytarabine during consolidation cycles (up to 4 cycles), and then resumed on count recovery for 7 days, followed by 14 days off, and continued in this manner as 21-day cycles for up to 12 cycles.								

13. MEASUREMENT OF EFFECT

13.1 Response Criteria

Complete remission (CR): Bone marrow showing less than 5% myeloblasts with normal maturation of all cell lines, an ANC of at least 1000/µL and a platelet count of 100,000/µL, absence of blast in peripheral blood, absence of identifiable leukemic cells in the bone marrow, existing extramedullary disease. If possible, at least one bone marrow biopsy should be performed to confirm CR.

Complete Remission with Incomplete Blood Count Recovery (CRi): Same as for CR but without achievement of ANC at least 1000/uL (CRi) and/or platelet count of 100,000/uL (CRp).

Partial Remission: All hematologic criteria of CR are fulfilled, and a decrease of bone marrow blast percentage to 5 to 25 percent; and decrease of pretreatment bone marrow blast percentage by at least 50 Percent.

Morphologic Leukemia Free State: Bone marrow blasts <5 percent; absence of blasts with Auer rods; absence of extramedullary disease; no hematologic recovery required

Resistant Disease: Failure to achieve CR or CRi.

Recurrence/Morphologic Relapse: Reappearance of leukemic blasts in the peripheral blood or > 5% blasts in the bone marrow not attributable to any other cause (e.g. bone marrow regeneration

after consolidation therapy). The appearance of new dysplastic changes should be considered relapse. The reappearance or development of cytologically proven extramedullary disease indicates relapse. Molecular and/or genetic relapse is characterized by reappearance of a cytogenetic or molecular abnormality.

14. STATISTICAL CONSIDERATIONS

This is a phase II, two-stage, single center, to assess the efficacy of alisertib in combination 7+3 induction chemotherapy in the adult higher risk AML population. Higher risk AML patients are those that (a) are at least 18 years of age with a poor risk karyotype and/or secondary AML, or (b) at least 65 years of age with AML.

The primary endpoint of this study is the rate of complete remission (CR) or complete remission with incomplete count recovery (CRi) in higher risk patients receiving alisertib in combination with 7+3 induction chemotherapy. The CR+CRi rate will be compared to the historical rates seen in this population with 7+3 induction alone. Secondary endpoints include: 1-year overall survival (OS), relapse free survival (RFS), remission duration, and toxicity for patients receiving alisertib in combination with conventional cytotoxic chemotherapy.

14.1 Sample Size, Accrual Rate, and Study Duration

A total of 39 evaluable patients will be enrolled based on the Simon two-stage design assuming a null CR rate of 0.45, a CR+CRi rate of 0.70 for alisertib in combination 7+3 induction chemotherapy, a 0.05 significance level, and 90% power. The first stage will accrue 13 patients; if more than 6 CR+CRi's are observed during the first stage, the trial will continue onto the second stage accruing 26 more patients. If more than 22 CR+CRi's are observed out of 39 then alisertib in combination 7+3 induction chemotherapy will be considered effective. Under this design, the probability of stopping after the first stage is 0.64. Enrollment for the first stage assessment will be held before enrolling the first patient to the second stage.

Patients removed due to presence of favorable cytogenetics prior to day 8 would be considered to be inevaluable and will be replaced. In addition, patients who receive less than half of their total alisertib dose during induction would be considered to be inevaluable and will be replaced. We anticipate that approximately 3 eligible adults per month will be accrued to this MGH/DFCI for an approximate total accrual of 39 evaluable adult high risk AML patients over 13 months.

14.2 Statistical Design/Endpoints

Primary Endpoint

The primary endpoint of this study is the proportion of patients who achieve a CR or a CRi (defined in section 13.1) compared to the historical rate of 0.45. It is hypothesized that the CR+CRi rate for alisertib in combination 7+3 induction chemotherapy will be 0.70. A one-sided single proportion test will be used to test the hypothesis that the remission rate of alisertib in 7+3 chemotherapy is greater than the 7+3 historical rate.

Secondary Endpoints

The one year overall survival and relapse free survival after treatment will be estimated using the Kaplan-Meier method, and presented as a 90% confidence interval using Greenwood's formula for standard errors of the estimate. Overall survival (OS) is defined as the time from study entry to death from any cause. Relapse free survival (RFS) is defined as the time from achieving a CR to the first of disease recurrence or death. RFS applies only to the subset of patients who achieve a CR+CRi at the end of induction therapy. Remission duration will also be estimated using the method of Kaplan-Meier. All toxicities encountered during the study will be evaluated according to the NCI criteria assigned to the protocol (CTCAE version 4.0).

15. PUBLICATION PLAN

Dr. Amir T. Fathi holds the primary responsibility for publications related to this study. The results should be made public within 24 months of reaching the end of the study. The end of the study is the time point at which the last data items are to be reported, or after the outcome data are sufficiently mature for analysis, as defined in the section on Sample Size, Accrual Rate and Study Duration. If a report is planned to be published in a peer-reviewed journal, then that initial release may be an abstract that meets the requirements of the International Committee of Medical Journal Editors. A full report of the outcomes should be made public no later than three (3) years after the end of the study.

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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.

ADDENDUM B

Adverse Risk Karyotype Groups per European Leukemia Net (ELN) Guidelines Döhner H et al, Blood 2010;115(3):453-474
• Inv(3)(q21q26.2) or t(3;3)(q21;q26.2) • t(6;9)(p23;q34) • t(v;11)(v;q23), [not including t(9;11)(p22;q23)]* • Monosomy 5 or del(5q) • Monosomy 7 • Abnormal 17p • Complex karyotype (≥3 abnormalities)