AflacLL1901
CHOA-AML: A Pilot Study for Newly Diagnosed Pediatric Patients with Acute Myeloid Leukemia (AML)

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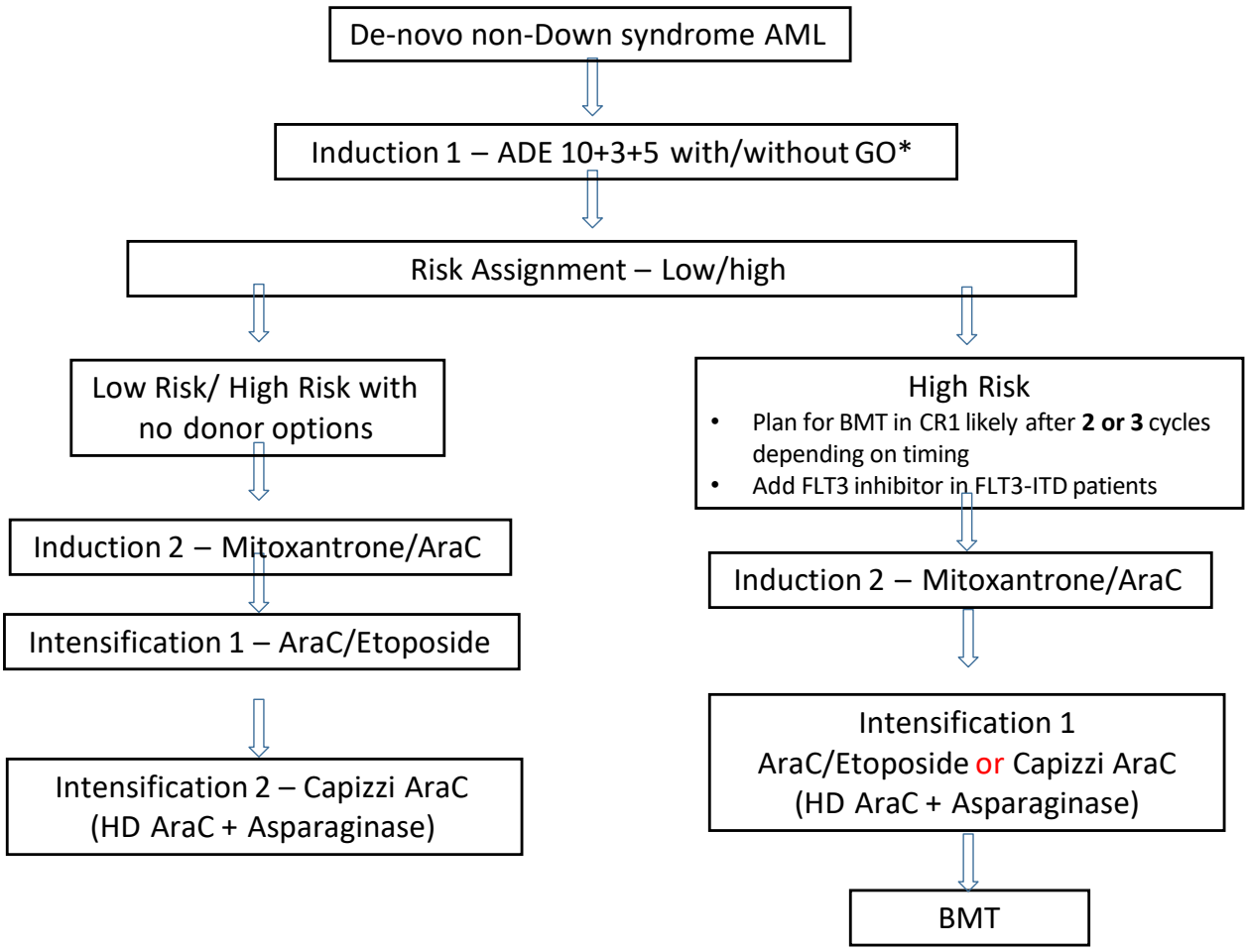
ABSTRACT

Advances in risk stratification and therapy, have improved the event-free survival (EFS) and overall survival (OS) for pediatric acute myeloid leukemia (AML) to approximately 50% and 65% respectively, with current treatment strategies. Patients with good response to induction and/or those who lack high-risk cytogenetic and molecular features [classified as low-risk AML (LR-AML)] have even better outcomes with EFS and OS approaching 70% and 85% respectively; however, treatment-related toxicities remain a major concern. Anthracycline-based therapeutic regimens expose patients to the risk of anthracycline-induced cardiotoxicity. Therefore, strategies that reduce cardiac toxicities using tailored approaches while maintaining and/or improving outcomes are needed for all patients with AML. The Children's Oncology Group (COG) regimens AAML1031 and AAML0531 utilized an anthracycline-intensive backbone for LR-AML with cumulative anthracycline doxorubicin-equivalent doses of up to 492mg/m^2. However, high-risk patients treated with chemotherapy alone received an intensified induction chemotherapy (using mitoxantrone-cytarabine) but with overall reduced doses of anthracycline-equivalent (342mg/m^2). We piloted an institutional practice to treat all LR-AML patients with four cycle regimen (Aflac-AML) with the goal of reducing cumulative anthracycline exposure, thereby reducing the risk of cardiotoxicity, while providing three high-dose cytarabine courses. In this pilot institutional experience with this approach, we were able to maintain excellent outcomes for this low-risk group with 3-year event-free survival (EFS) and OS of 70.0% ± 0.1% and 85.5% ± 0.08% respectively, from end of course 1. Recent evolution in cytogenetic classification has further delineated risk groups in AML. Gemtuzumab ozogamicin (GO), an antibody-drug conjugate was shown to reduce relapse risk in patients with CC genotype with de-novo AML on COG study AAML0531. We propose to study the inclusion of gemtuzumab ozogamicin with our Aflac-AML chemotherapy backbone prospectively to validate its use in all pediatric AML and to further evaluate the approach with a cardiac-sparing goal in an expanded experience for low risk AML.

EXPERIMENTAL DESIGN SCHEMA



* Gemtuzumab ozogamicin (GO) dosing to be determined by SNP testing
Risk Stratification – refer Section 4.0

1.0 GOALS AND OBJECTIVES

1.1 Primary Objective

To estimate the event-free survival (EFS) for newly diagnosed patients with pediatric acute myeloid leukemia using a risk stratified approach, including a modified chemotherapy regimen alone for subjects with low-risk features and allogeneic bone marrow transplantation for those with high risk features.

1.2 Secondary Objectives

1. To estimate the overall survival (OS) for newly diagnosed patients with pediatric acute myeloid leukemia

2. To estimate frequency of minimal residual disease (MRD) negative status after one cycle of induction chemotherapy for AML patients who received GO and those that did not receive GO.

3. To estimate the disease-free survival (DFS) for patients who are MRD negative after induction 1, but lack high risk or lowest risk molecular and cytogenetic features as currently classified

4. To estimate the incidence of early and late cardiotoxicity in patients with de novo AML that receive the four-cycle Aflac-AML regimen with the inclusion of dexrazoxane

5. To estimate the frequency of infection and/or febrile neutropenia by chemotherapy regimen

1.3 Exploratory Objectives

To describe the prevalence of molecular abnormalities in de-novo AML samples using comprehensive genomic testing.

ENDPOINTS

Primary Endpoints include the event-free survival defined as the time from on study to death, failure to achieve remission or relapse

Secondary Endpoints include

- disease-free survival (DFS) from end of first course of therapy, defined as time from end of first course of therapy to death or relapse
- overall survival defined as time from study entry and from end of first course of therapy
- proportion of patients that are in remission (MRD negative) after course 1 in both the GO and non-GO groups
- Disease-free survival for patients who are MRD negative but lack high or low risk molecular and cytogenetic features, defined as time from end of first course of therapy to death or relapse
- Estimate the proportion of patients that develop cardiac ejection fraction <50% (CTCAE V5.0 grade 2 or greater dysfunction) either during therapy (early cardiotoxicity) or after completion of therapy (late cardiotoxicity)
- Estimate the proportion of patients who develop infection and/or febrile neutropenia during each treatment course
- Estimate the effect of infection and/or febrile neutropenia on duration of hospital admission for each treatment course

Exploratory Endpoints include descriptive analysis of molecular abnormalities in de-novo AML.

2.0 BACKGROUND

2.1 Introduction and rationale for development of chemotherapy backbone for de novo AML

Overall survival (OS) rates in pediatric acute myeloid leukemia (AML) approach 65-70% at three years with the current risk adapted chemotherapy and hematopoietic stem cell transplantation (HSCT) approach[1]. The recently concluded Children's Oncology Group (COG) trial in pediatric de-novo AML AAML1031 (2011 – 2017) and its precursor trial AAML0531 (2006 – 2010) used a chemotherapy backbone adapted from the Medical Research Council (MRC) AML trials in order to improve overall outcomes[2-4]. This backbone comprised of an induction course of chemotherapy with cytarabine, daunorubicin and etoposide. Patients were risk stratified into low, intermediate and high risk groups on AAML0531 based on their cytogenetic and molecular features and their response at the end of this first cycle of induction chemotherapy[1]. Patients on AAML0531 received 5 cycles of chemotherapy if they did not meet criteria for consideration of HSCT in first remission (Table 1). In addition, patients were randomized to receiving gemtuzumab ozogamicin (GO) at study entry and received this drug along with chemotherapy in Induction 1 and Intensification 2. Based on results from the MRC12 trial and preliminary analysis of AAML0531, the fifth course of chemotherapy was deleted from the chemotherapy backbone for AAML1031[3]. The risk stratification evolved over the years with the improvement in multi-dimensional flow cytometry (MDFC) and AAML1031 stratified patients into low and high risk categories using response defined by MDFC (measurement of minimal residual disease –MRD) in addition to cytogenetic and molecular features[5].

2.1.1 Rationale for chemotherapy approach in children with low risk AML

Based on the recent COG AAML1031 chemotherapy backbone, patients with low-risk AML received Induction II that is similar to Induction I and two more cycles of consolidation therapy which included two high-dose cytarabine (> 1gm/m^2 per dose) courses in Intensification 1 and Intensification II. Patients with high-risk AML received an intensified second course (Induction II) of chemotherapy comprising of mitoxantrone and high dose cytarabine followed by two more high dose cytarabine containing courses (Intensification I and II) if allogeneic hematopoietic stem cell transplantation was not an option. Thus, these high-risk patients received a total of three courses with high-dose cytarabine as opposed to the low-risk patients who only received two. In addition, in the COG approach, low-risk AML patients received higher doses of anthracycline (492 mg/m^2) and etoposide (1.75 g/m^2) as compared to high-risk patients who did not receive a stem cell transplant[2]. Exposure to high doses of anthracycline and etoposide results high risk of cardiotoxicity and secondary malignancies with the cardiotoxicity risk being highest in patients with acute myeloid leukemia[6,7]. Mertens et al demonstrated that in the Childhood Cancer Survivor Study, cardiac causes and secondary malignancies accounted for excess mortality rates in the childhood cancer survivors as compared to the general population[8]. Cumulative anthracycline dosage >300 mg/m^2 was associated with increased incidence of anthracycline-induced clinical heart failure[9] thereby raising concerns that exposures as high as 492 mg/m^2 in this low-risk cohort on AAML1031 could place them at very high risk of heart failure.

Patients with low-risk AML have favorable outcomes (3-year EFS and OS - 70% and 85% respectively) as compared to patients with high-risk AML (3-year EFS and OS – 30% and 48% respectively)[1]. Regardless of risk, disease relapse still remains the major cause of mortality[1]. Anthracyclines comprise an important component of relapsed AML therapy and higher doses of anthracyclines received in upfront AML therapy preclude their use at the time of relapse[10]. Given the concerns for long term toxicity without clear evidence of benefit to additional anthracyclines and etoposide in low-risk AML patients, we chose to adopt an institutional approach (Aflac-AML regimen) whereby newly diagnosed low-risk AML patients were treated with four cycles of chemotherapy that was identical to the high-risk group per AAML1031 which received chemotherapy alone. The aim of our regimen was to limit anthracycline exposure to 342 mg/m2 and etoposide exposure to 1.25 g/m^2 in low-risk AML patients while maintaining the same favorable outcomes. A comparison of the two regimens is outlined in the table (Table 1) below.

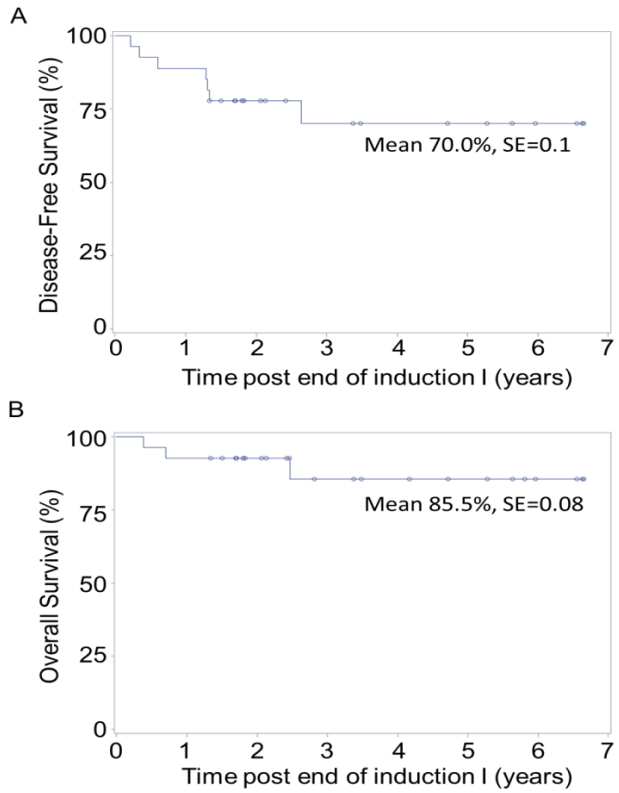
Table 1: Therapy regimen for Aflac-AML, AAML0531 and AAML1031 low-risk patients

Aflac-AML		COG AAML0531		COG AAML1031	
Induction 1					
Cytarabine	100 mg/m²/dose every 12 hours IV Days 1-10	Cytarabine	100 mg/m²/dose every 12 hours IV Days 1-10	Cytarabine	100 mg/m²/dose every 12 hours IV Days 1-10
Daunorubicin	50 mg/m²/dose IV Days 1, 3, 5	Daunorubicin	50 mg/m²/dose IV Days 1, 3, 5	Daunorubicin	50 mg/m²/dose IV Days 1, 3, 5
Etoposide	100 mg/m²/dose IV Days 1-5	Etoposide	100 mg/m²/dose IV Days 1-5	Etoposide	100 mg/m²/dose IV Days 1-5
Induction 2					
Cytarabine	1,000 mg/m²/dose every 12 hours IV Days 1-4	Cytarabine	100 mg/m²/dose every 12 hours IV Days 1-8	Cytarabine	100 mg/m²/dose every 12 hours IV Days 1-8
Mitoxantrone	12 mg/m²/dose IV Days 3-6	Etoposide	100 mg/m²/dose IV Days 1-5	Etoposide	100 mg/m²/dose IV Days 1-5
-----	-----	Daunorubicin	50 mg/m²/dose IV Days 1, 3, 5	Daunorubicin	50 mg/m²/dose IV Days 1, 3, 5
Intensification 1					
Cytarabine	1,000 mg/m²/dose every 12 hours IV Days 1-5	Cytarabine	1,000 mg/m²/dose every 12 hours IV Days 1-5	Cytarabine	1,000 mg/m²/dose every 12 hours IV Days 1-5
Etoposide	150 mg/m²/dose IV Days 1-5	Etoposide	150 mg/m²/dose IV Days 1-5	Etoposide	150 mg/m²/dose IV Days 1-5
Intensification 2					
Cytarabine	3,000 mg/m²/dose every 12 hours IV Days 1, 2, 8, 9	Cytarabine	1,000 mg/m²/dose every 12 hours IV Days 1-4	Cytarabine	1,000 mg/m²/dose every 12 hours IV Days 1-4
Erwinia L-Asparignase	25,000 International Units/m²/dose IM Days 2, 9	Mitoxantrone	12 mg/m²/dose IV Days 3-6	Mitoxantrone	12 mg/m²/dose IV Days 3-6
Intensification 3					
----	----	Cytarabine	3,000 mg/m²/dose every 12 hours IV Days 1, 2, 8, 9	----	----
----	----	*E. Coli* L-Asparignase	6,000 mg/m²/dose IM Days 2, 9	----	----
Abbreviations: COG, Children's Oncology Group; IM, intramuscular; IV, intravenous					

We performed a retrospective review of our institutional cohort of patients with de novo low-risk AML between 0-21 years of age at diagnosis and were treated at the Aflac Cancer and Blood Disorders Center at Children's Healthcare of Atlanta (CHOA) between January 1, 2011 and

December 31, 2016 (manuscript in progress). LR-AML was defined as the presence of NPM1, CEBPA, t(8;21)(q22;q22), inv(16)(p13.1q22), or less than 0.1% blasts (minimal residual disease – MRD negative) at end of Induction I, with the absence of unfavorable cytogenetic or molecular findings. A total of 27 patients with LR-AML treated per Aflac-AML were eligible for analysis with a mean follow up period from time of diagnoses of 3.3 years (SD 2.0). For the study population three-year EFS and OS from end of Induction 1 were 70.0% ± 0.1% and 85.5% ± 0.08% (Figure 1A and B).

Figure 1 A and B: EFS and OS for low-risk patients treated on Aflac-AML regimen



Mean time to relapse for the 7 relapsed patients was 1.11 ± 0.16 years and mean survival time for the 3 patients who died was 1.18 ± 0.65 years (Table 2). Short-term cardiovascular outcomes were determined by echocardiogram measurements taken at diagnosis, between cycles of chemotherapy, and at follow-up. Four of 27 patients (14.8%) experienced grade 2 or higher left ventricular systolic dysfunction (LVSD) during therapy, two of which were associated with bloodstream infection, and 3 of these patients had normal cardiac function at the time of last follow-up. One patient had progressive LVSD which ultimately resulted in death (grade 5). We examined infectious toxicities, ICU admissions, and time to hematologic recovery in each course and compared them to the data available for the precursor AML trial AAML0531 where similar courses of treatment were used and found that they were comparable (Table 3). It is important to note that our institutional practice changed to include regular Levaquin and voriconazole prophylaxis midway through the reporting period for this data.

Table 2: Details of patients who relapsed on Aflac-AML regimen

Pt #	Karyotype/ FISH/molecular	Time to relapse (from EOI)	Current status
1	Inv 16	1.31 yr	Alive, CR2 >4yr
2	Inv 16	1.4 yr	Alive, CR2 >3yr
3	CEPBA mutation	2.8 yr	Alive, CR2
4	t(1;11)(p36,1;q23); complex	0.35yr	Died in relapse
5	t(x;21)(11.2;q13;p13)	1.4 yr (MRD+)	Died in second MRD+ relapse (no HSCT)*
6	t(9;11)(p22;q23)	0.34 yr	Died in relapse
7	t(11;19)(p23;p13.1)	0.72	Alive, relapse post BMT
*developed severe irreversible cardiomyopathy during intensification 1			

Table 3: Comparison of infectious rate between Aflac-AML and AAML0531 regimens

	Induction I		**Induction II**		**Intensification I**		**Intensification II**		**Intensification III**	
	Aflac	0531	Aflac	0531	Aflac	0531	Aflac	0531	Aflac	0531
	ADE	ADE	MA	ADE	AE	AE	Capizzi	MA		Capizzi
Documented infection	**32%**	**35%**	**60%**	37%	**29%**	**49%**	**60%**	**69%**	N/A	**67%**
Neutropenic fever*	**52%**	**31%**	**16%**	22%	**20%**	**23%**	**17%**	**23%**	N/A	**18%**
*in absence of documented infection Comparable regimens represented with like colors										

Given that our rates of treatment associated infection are comparable to published standards along with similar favorable outcomes, we will plan to use our described four cycle cytarabine-intensive regimen piloted in 27 patients as the chemotherapy backbone for this trial. All patients regardless of risk status will receive mitoxantrone-cytarabine as their second course of therapy (Induction II). This will be followed by Intensification I (cytarabine-etoposide) and Intensification II (Capizzi-AraC) for low-risk patients. Patients with high-risk disease will receive an allogeneic HSCT typically after two or three courses of chemotherapy. The third course could be either cytarabine-etoposide or Capizzi-AraC at the treating physician's discretion. High-risk patients unable to receive a HSCT will receive treatment per the Aflac-AML regimen with four cycles of chemotherapy similar to low-risk AML patients.

2.2 Rationale for incorporation of Gemtuzumab Ozogamicin (GO)

Gemtuzumab Ozogamicin (GO, Mylotarg™) is an antibody-drug conjugate (ADC) chemotherapy agent that is composed of a recombinant humanized IgG4, kappa antibody to CD33 conjugated with a cytotoxic antitumor antibiotic, calicheamicin[11]. It binds to CD33 after which the complex is endocytosed and undergoes dissociation in the lysosome. The detached calicheamicin then binds to the minor groove of cellular DNA causing damage and apoptosis[12]. It received accelerated approval in May 2000 by the US FDA for treatment of older adults with CD33 positive AML in first relapse not considered candidates for cytotoxic therapy[13]. CD33 expression is absent on normal

hematopoietic stem cells and is limited to leukemic blasts, thus making it an attractive therapeutic target[14].

Though GO was initially approved for use in patients experiencing first relapse of AML who were unable to tolerate any other chemotherapy, it was subsequently given both as monotherapy and in combination with other chemotherapy agents for patients with *de novo* AML[15-18]. Myelosuppression, infections, hyperbilirubinemia and elevated liver enzymes were the major side effects reported[19]. Hepatic veno-occlusive disease (VOD) was reported in patients who received the drug even in the absence of stem cell transplantation [20]. However, the Southwest Oncology Group (SWOG) study S0106 which prospectively compared the benefit of adding GO to a standard induction regimen of cytarabine and daunorubicin failed to show any survival benefit and reported a 5% induction mortality in the GO group as compared to the non-GO group (1% mortality)[21]. Results of this study led to the withdrawal of the drug from the market in June 2010. Four subsequent studies demonstrated contrasting results showing efficacy of GO in patients with newly diagnosed AML, supporting the consideration for re-approval of GO in AML[22]. In the Children's Oncology Group (COG) randomized phase III trial for pediatric patients with *de novo* AML (AAML0531), GO significantly improved the 3-year event free survival (EFS) by reducing the relapse risk (RR), but had no impact on overall survival[1]. Specifically in patients with FLT3-ITD positive disease, exposure to GO prior to HSCT reduced the risk of relapse significantly compared to those that had not received GO (15% versus 55%, p=0.007) [23]. Following the overwhelming support for the use of GO in AML, the US FDA granted approval for the drug in de-novo and relapsed or refractory disease in September 2017.

Pollard et al divided all patients on AAML0531 based on their CD33 expression into quartiles. Patients with high CD33 expression (Q2-4) had significantly improved EFS and relapse risk [24]. Single nucleotide polymorphisms (SNPs) in CD33 (specifically resulting in increased number of T alleles) showed a positive correlation with poorer response to GO-containing regimens[25]. On the COG AML pilot study AAML 03P1, coding SNPs in CD33 (rs35112940 and rs12459419) were associated with better risk disease and improved outcomes compared to other genotypes[26]. Further characterization and testing in a larger cohort of patients on AAML0531 (n=816) showed that patients with CC genotype (rs12459419) have a significant response to GO as compared to CT or TT genotypes[27]. When genotyping data was correlated with expression, it was found that the presence of T allele (CT/TT) genotype strongly correlates with low expression of CD33. There are 256/816 patients (31%) who had either CT or TT genotype and were in the high expression (Q2-4) group on AAML0531. In this group, addition of GO did not improve relapse risk. Conversely, there were 61/816 individuals (7%) who were CC genotype that showed low quartile (Q1) CD33 expression. The RR even in this small group was still improved significantly with GO through the p-value was just shy of statistical significance (p=0.055)[27]. While determination of CD33 expression is universally possible with routine multi-dimensional flow cytometry, division of the data acquired into quartiles that are validated and can be compared between institutions is challenging. Given that the CD33 genotyping shows strong correlation with relapse risk regardless of CD33 expression, we will be obtaining CD33 genotyping on all patients at the time of diagnosis. We expect the turn-around time for this testing to be approximately 4-5 days, allowing for a single 3 mg/m2 dose of GO to be administered between days 6-10 during Induction I for patients with CC genotype.

2.3 Rationale for use of Dexrazoxane with anthracyclines

Cumulative incidence of heart failure and other cardiac events in patients with acute leukemias ranges from 4-10% at 10-20 years from diagnosis [28,29]. While late cardiotoxicity has been

described well following anthracycline exposure above 250-300 mg/m2 doxorubicin equivalents, early cardiotoxicity is a growing concern[30]. Evaluation of cardiotoxicity (defined as grade 2 or higher left ventricular dysfunction per CTCAE version 5.0) on COG AAML0531 showed an incidence of 12% over a five-year follow-up period with most of the events occurring while on protocol therapy. Black, older patients (>1 year age) and concurrent infection were risk factors associated with worse cardiac outcomes[30].

Dexrazoxane is a bis-dioxopiperazine compound that was introduced in the early 1990's and has been shown to reduce cardiotoxicity related to anthracyclines initially in animal models and later in clinical studies[31,32]. While the exact mechanism is poorly understood, it is believed that anthracyclines form complexes with heavy metals specifically iron and generate free radicals that cause cardiac damage[33]. Dexrazoxane binds to iron and prevents the generation of free radicals thereby reducing cardiotoxicity. Dexrazoxane has been used successfully in combination with various chemotherapy regimens with acceptable toxicity and enhanced cardio-protection[34,35]. While there was concern regarding development of secondary malignancies with exposure to dexrazoxane, recent literature has not supported this side-effect[36]. In this study, all patients will receive dexrazoxane in anthracycline-containing treatment courses. We will closely monitor for infection, cardiac, hepatic and hematologic toxicities as well as development of secondary malignancies. In addition, we will obtain more precise measurements of cardiac function using echocardiogram at the end of each course (Section 8.0) as noted in Table 4.

Table 4: Variables of cardiac function to be determined using echocardiography

Left ventricle (LV) remodeling	LV thickness to dimension ratio
LV myocardial deformation	Strain and Strain rate imaging
LV systolic function	LV Ejection Fraction using biplane Simpson method LV Shortening fraction using 2D and M-mode Mitral and septal S'
LV diastolic function	Early filling peak velocity (E wave) Atrial systole filling peak velocity (A wave) Mitral E/A ratio Isovolumic relaxation time (IVRT) Mitral annular tissue doppler (E' and A') Isovolumic contraction time (IVCT) Aortic ejection time (ET)
LV global function	Spectral Doppler-derived myocardial performance index Tissue Doppler Imaging-derived myocardial performance index

2.4 Use of FLT3-ITD inhibitors in AML

Presence of activating mutations in the FLT3 gene occurs in approximately 15% of pediatric patients with de-novo AML[37]. Patients with FLT-ITD mutations with a high allelic ratio (HAR>0.4) have an extremely poor progression-free survival of 16% compared to those with lower allelic ratios[38] and are therefore categorized in the high risk group post induction. Further analysis of FLT3-ITD mutations has suggested an AR>0.1 to be associated with a poor outcome as well (COG, internal communication). Several FLT3-ITD inhibitors are currently in clinical trials and COG AAML1031 tested the inclusion of one such inhibitor, sorafenib, with the regular chemotherapy backbone in FLT3-ITD positive AML in a non-randomized fashion. The results of this combination are currently under review; however the combination is believed to be safe

(COG, internal communication). In this study, we will use sorafenib as the FLT3 inhibitor of choice in patients with FLT3-ITD HAR (>0.1) AML.

2.5 Genomic testing in de-novo AML

The Therapeutically Applicable Research to Generate Effective Treatments (TARGET) initiative was launched by the National Cancer Institute to support genomic characterization of pediatric cancers with poor outcomes, AML being one of them. As a part of the Target Pediatric AML initiative (TpAML), genomic sequencing will be carried out for clinical purposes through the Foundation One Heme panel. This panel sequences DNA coding regions of 406 genes and selected introns of 31 genes involved in hematologic malignancies. It also performs RNA sequencing of 265 genes commonly rearranged in cancer. All patients on this study will have Foundation One Heme panel sent at diagnosis and we expect results to be available within 4 weeks. We will be able to correlate the data obtained from this panel with disease outcomes. While this panel includes the genes of interest for risk stratification, it is recognized that other investigational sequencing approaches may be options during the course of this trial.

3.0 STUDY DESIGN AND POPULATION

CHOA-AML is a single-center pilot trial. We will enroll up to 30 newly diagnosed pediatric patients with acute myeloid leukemia (AML) at CHOA.

All clinical and laboratory data required for determining eligibility of a patient enrolled must be available in the patient's medical/research record which will serve as the source document for verification.

All clinical and laboratory studies to determine eligibility must be performed within 7 days prior to enrollment unless otherwise indicated. Bone marrow aspiration (BMA)/biopsy must be obtained within 2 weeks prior to start of protocol therapy (repeat the BMA/biopsy if necessary).

3.1 Patient Inclusion Criteria

3.1.1 **Age:** Patients must be less than 21 years of age at the time of study enrollment

3.1.2 **Diagnosis:** Patients must be newly diagnosed with AML

3.1.1.1 Patients with previously untreated primary AML who meet the customary criteria for AML with ≥ 20% bone marrow blasts as set out in the 2016 WHO Myeloid Neoplasm Classification are eligible.

3.1.1.2 Attempts to obtain bone marrow either by aspirate or biopsy must be made unless clinically prohibitive. In cases where it is clinically prohibitive, peripheral blood with an excess of 20% blasts and in which adequate flow cytometric and cytogenetics/FISH testing is feasible can be substituted for the marrow exam at diagnosis.

3.1.1.3 Patients with <20% bone marrow blasts are eligible if they have:

- A karyotypic abnormality characteristic of *de novo* AML *(t(8;21)(q22;q22), inv(16)(p13q22)* or *t(16;16)(p13;q22*) or,
- the unequivocal presence of megakaryoblasts, or
- Biopsy proven isolated myeloid sarcoma (myeloblastoma; chloroma, including leukemia cutis)

3.1.3 **Performance Level:** Patients with acceptable organ function and any performance status are eligible for enrollment

3.2 Patient Exclusion Criteria

3.2.1 Patients with any of the following constitutional conditions are not eligible:

- Fanconi anemia
- Shwachman syndrome
- Any other known bone marrow failure syndrome
- Patients with constitutional trisomy 21 or with constitutional mosaicism of trisomy 21

Note: Enrollment may occur, pending results of clinically indicated studies to exclude these conditions.

3.2.2 Other Excluded Conditions:

- Any concurrent malignancy
- Juvenile myelomonocytic leukemia (JMML)
- Philadelphia chromosome positive AML
- Biphenotypic or bilineal acute leukemia
- Acute promyelocytic leukemia (APL)
- Acute myeloid leukemia arising after a clearly defined preceding myelodysplastic syndrome
- Therapy-related myeloid neoplasms

Note: Enrollment may occur pending results of clinically indicated studies to exclude these conditions.

3.2.3 Sexually Active Patients

Sexually active patients of reproductive potential are not eligible unless they have agreed to use an effective contraceptive method for the duration of their study participation.

3.2.4 Pregnancy and Breast Feeding:

- Female patients of childbearing potential are not eligible unless a negative pregnancy test result has been obtained
- Female patients who are pregnant are ineligible since fetal toxicities and teratogenic effects have been noted for several of the study drugs.
- Lactating females are not eligible unless they have agreed not to breastfeed their infants.

3.3 Definitions

3.3.1 CNS LEUKEMIA AT DIAGNOSIS:

CNS disease at diagnosis is defined as:

- Any number of blasts on a cytospin prep in an atraumatic (< 100 RBCs) lumbar puncture.
- Blasts in a traumatic tap in which the WBC/RBC ratio in the CSF is twice that in the peripheral blood.
- Clinical signs of CNS leukemia (such as facial nerve palsy, brain/eye involvement or hypothalamic syndrome). Extra-ocular orbital masses are not considered CNS leukemia.
- Radiographic evidence of an intracranial, intradural mass consistent with a chloroma.

3.3.2 Method of Evaluation for Traumatic Lumbar Punctures

If the patient has leukemic cells in the peripheral blood and the lumbar puncture is traumatic and contains blasts, the following algorithm should be used to diagnose CNS disease:

$$\frac{\text{CSF WBC}}{\text{CSF RBC}} > 2X \quad \frac{\text{Blood WBC}}{\text{Blood RBC}}$$

A patient with CSF blasts, whose CSF WBC/RBC is 2X greater than the blood WBC/RBC ratio, has CNS disease at diagnosis. Example: CSF WBC = 60/μL; CSF RBC = 1,500/μL; blood WBC = 46,000/μL; blood RBC = 3 X 106/μL:

$$\frac{60}{1{,}500} = \mathbf{0.04} \quad \mathbf{> 2X} \quad \frac{46{,}000}{3 \text{ X } 106} = \mathbf{0.015}$$

3.3 Protection of Human Subjects

This protocol will be reviewed and approved by the Emory University Institutional Review Board (IRB) prior to study initiation. Prospective participants and their families will be informed of the nature of the study and will be given pertinent information as to the intended purpose, possible benefits, and possible adverse events. The procedures and possible hazards to which the patient will be exposed will be explained, as well as alternatives to participation in this trial will be presented in detail to the patient and to the patient's responsible family members. An approved informed consent statement will be read and signed by the patient (or responsible family member if under the age of 18) and an investigator.

Verbal assent will be obtained from children ages 6 to 10 and documented in the patients' medical record, as well as being noted in the case report forms. Written assent will be obtained from children ages 11 to 17, using an approved assent statement, along with use of the informed consent by their parent or guardian. Participants 18 years or older will read/sign the consent form.

3.4 Aflac Office Monitoring Schedule

The trial will be monitored internally according to the following schedule:

- Study monitoring will occur every 6 months until all subjects have completed intervention;

- Once all subjects have completed intervention, a final monitoring visit will occur within 90 days after the last study related procedures and follow-up are completed for all patients enrolled;
- Additional monitoring may be performed in the event of serious adverse event(s), if deemed necessary by the monitoring committee or if requested by the Principal Investigator.

4.0 RISK STRATIFICATION

After completion of first course of chemotherapy (Induction I), patients will be classified into low or high-risk groups.

Low risk

t(8;21)(q21.3;q22) – CBF (*RUNX1-RUNX1T1)*
inv(16)/t(16;16)(p13.1q22.1) – CBF (*CBFB-MYH11)*
NPM1 positive without associated FLT3 mutation
Biallelic CEBPA positive
No High-Risk Prognostic Markers/MRD negative (<0.05%) at end of 1st induction
CBF, CEBPa, NPM positive **and** MRD positive at the end of 1st Induction

High risk

inv(3)(q21q26.3) MECOM-RPN1 fusion
t(3;21)(26.2;q22)
t(3;5)(q25;q34)
t(6;9)(p23;q34.1)(*DEK-NUP214*)
t(8;16)(p11.2;p13.3) KAT6A(8p11.21) Fusion (for patients who are 90 days or older)
t(16;21)(p11;q22) FUS-ERG)
Inv(16) (p13.3q24.3) *CBFA2T3-GLIS2*
Monosomy 7
Monosomy 5/5q-[*EGR1*(5q31) deleted]
KMT2A(MLL) (11q23.3) - t(4;11)(q21;q23) - t(6;11)(q27;q23) - t(10;11)(p11.2;q23) - t(10;11)(p12;q23) - t(11;19)(q23;p13.3)
11p15 rearrangement - *NUP98* (11p15.5)
12p 13.2 rearrangement - *ETV6* – any partner gene
Deletion 12p to include 12p13.2
FLT3 High Allelic Ratio mutations (AR >0.1)
10p12.3 rearrangement - *Non-KMT2A MLLT10 Fusions*
RAM phenotype (high CD56, dim-to-negative expression CD45 & CD38, no HLA-DR)
MRD positive (>0.05%) at end of 1st induction, excluding those with CBF, CEBPa, NPM

5.0 TREATMENT PROGRAM

Dexrazoxane will be administered per institutional standard during all anthracycline-containing courses of this protocol

5.1 Induction 1 – ADE (10+3+5) with GO

The following therapy guidelines are for patients during Induction I. Patients on Induction I will receive gemtuzumab in addition to ADE therapy based on GO genotyping. Induction I lasts a total of 28 days.

Intrathecal Cytarabine: IT
Given at time of diagnostic lumbar puncture or Day 1.

Age-based dosing:

Age (yrs)	Dose
0-0.99	20 mg
1-1.99	30 mg
2-2.99	50 mg
≥ 3	70 mg

For CNS positive patients: add twice weekly IT AraC doses until CNS is clear, plus two additional treatments. A minimum of four intrathecal treatments are to be administered during the period of three weeks following diagnosis.

Cytarabine: Slow IV Push (e.g., over 1-2 minutes)
100 mg/m^2/dose (3.3 mg/kg/dose if BSA < 0.6 m^2) every 12 hours (total of 20 doses) on Days 1-10.

Daunorubicin (DAUN): IV over 15 minutes)
50 mg/m^2/dose (1.67 mg/kg/dose if BSA < 0.6 m^2) every other day (total of 3 doses) on Days 1, 3, 5.
See Section 6 if direct bilirubin is > 2.

Etoposide (ETOP): IV over 60 – 120 minutes
100 mg/m2/dose (3.3 mg/kg/dose if BSA < 0.6 m^2) daily (total of 5 doses) on Days 1-5.
See Section 6 if direct bilirubin is > 2.

Gemtuzumab Ozogamicin (GO): IV over 2 hours
3 mg/m2/dose (0.1 mg/kg/dose if BSA < 0.6 m^2) once (total of 1 dose) on Day 6 through10.

GO to be given once when GO genotyping demonstrates CC genotype. It can be given on any day between days 6-10 of Induction 1.

Pre-medicate with an antihistamine such as diphenhydramine, 1 mg/kg IV (max 50 mg), acetaminophen 10-15 mg/kg PO (max 650 mg) and methylprednisolone (1mg/kg) PO/IV. Repeat every 4 hours as needed to prevent post infusion complications.

Following Induction I, the next course (Induction II) starts on Day 29 or when blood count parameters and clinical condition are acceptable (whichever occurs later). This should not precede the end of course marrow exam.

5.2 Induction 1 for FLT3-ITD patients – ADE (10+3+5) with GO with Sorafenib

The following therapy guidelines are for patients during Induction I with FLT3-ITD mutation. Patients on Induction I will receive gemtuzumab in addition to ADE therapy based on GO genotyping. Induction I lasts a total of 28 days.

Intrathecal Cytarabine: IT
Given at time of diagnostic lumbar puncture or Day 1.

Age-based dosing:

Age (yrs)	Dose
0-0.99	20 mg
1-1.99	30 mg
2-2.99	50 mg
≥ 3	70 mg

For CNS positive patients: add twice weekly IT AraC doses until CNS is clear, plus two additional treatments. A minimum of four intrathecal treatments are to be administered during the period of three weeks following diagnosis.

Cytarabine: Slow IV Push (e.g., over 1-2 minutes)
100 mg/m^2/dose (3.3 mg/kg/dose if BSA < 0.6 m^2) every 12 hours (total of 20 doses) on Days 1-10.

Daunorubicin (DAUN): IV over 15 minutes
50 mg/m^2/dose (1.67 mg/kg/dose if BSA < 0.6 m^2) every other day (total of 3 doses) on Days 1, 3, 5.
See Section 6 if direct bilirubin is > 2.

Etoposide (ETOP): IV over 60 – 120 minutes
100 mg/m2/dose (3.3 mg/kg/dose if BSA < 0.6 m^2) daily (total of 5 doses) on Days 1-5.
See Section 6 if direct bilirubin is > 2.

Gemtuzumab Ozogamicin (GO): IV over 2 hours
3 mg/m2/dose (0.1 mg/kg/dose if BSA < 0.6 m^2) once (total of 1 dose) on Day 6 through10.

GO to be given once when GO genotyping demonstrates CC genotype. It can be given on any day between days 6-10 of Induction 1.

Pre-medicate with an antihistamine such as diphenhydramine, 1 mg/kg IV (max 50 mg), acetaminophen 10-15 mg/kg PO (max 650 mg) and methylprednisolone (1mg/kg) PO/IV. Repeat every 4 hours as needed to prevent post infusion complications.

Sorafenib (SORAF): PO
Days: 11 through 28.
Dose: 200 mg/m^2/dose daily, rounded to accommodate tablet size. The maximum dose will be 400 mg. Dose will be administered per dosing nomogram in Appendix I.

Note: Sorafenib tablets should be taken at least 1 hour before or 2 hours after food. Tablets should be taken with clear liquids (approximately 2 to 4 ounces for children < 12 years and 4 to 8 ounces for ≥ 12 years). If taken with food, sorafenib should be taken with a moderate to low fat meal. Tablets should not be crushed and should be swallowed whole. However, sorafenib tablets can be dispersed in water to facilitate the administration to subjects that cannot swallow tablets

Following Induction I, the next course (Induction II) starts on Day 29 or when blood count parameters and clinical condition are acceptable (whichever occurs later). This should not precede the end of course marrow exam.

5.3 Induction II – MA

The following therapy guidelines are for patients during Induction II. Induction II lasts 28 days or longer.

Progression to Induction II should await disease response determination (MRD and extramedullary disease) in order to correctly assign risk stratum. Patients with high or low risk cytogenetic/molecular features may proceed on to Induction II therapy before disease response results are available. By the beginning of Induction II, patients should have been classified as Low or High risk. **If a patient is classified as High risk at the end of Induction I and no family HLA match is found, initiation of an unrelated donor search should occur.**

Intrathecal Cytarabine (IT ARAC): IT

IT ARAC may be administered on Day 1 of Induction II or with the bone marrow evaluation at the end of Induction I.

Age-based dosing:

Age (yrs)	Dose
0 - 0.99	20 mg
1 - 1.99	30 mg
2 - 2.99	50 mg
≥ 3	70 mg

For newly detected CNS positive patients who were CNS negative at diagnosis: IT cytarabine is to be administered twice weekly until the CSF is clear. A minimum of 4 intrathecal treatments must be given and a maximum of 6 intrathecal treatments may be given. Patients whose CSF is clear at the second spinal tap will receive 3 intrathecal treatments once the CSF is clear and all other patients will receive 2 intrathecal treatments once the CSF is clear. Patients with refractory CNS leukemia following 6 doses of therapy will be taken off protocol therapy.

High Dose Cytarabine (HD ARAC): IV over 1 - 3 hours

Days: 1 through 4.
Dose: 1000 mg/m^2/dose, every 12 hours (i.e., 2000 mg/m^2/day, divided BID) or
33 mg/kg/dose, every 12 hours (i.e., 66 mg/kg/day, divided BID) if BSA < 0.6 m^2.

Suggested premedications and supportive care: Administer steroid eye drops (0.1% dexamethasone or 1% prednisolone ophthalmic solution), 2 drops to each eye every 6 hours, beginning immediately before the first dose and continuing for 24 hours after the last dose. If patient does not tolerate steroid eye drops, may administer artificial tears on an every 2-4 hour schedule.

Patients in whom serum creatinine is > 2 mg/dL or > 2 x normal for age, must have a CrCl performed (Day 1) and doses of cytarabine adjusted as outlined.

MitoXANTRONE (MITOX): IV over 15 - 30 minutes
Days: 3 through 6.
Dose: 12 mg/m^2/dose or
0.4 mg/kg/dose if BSA < 0.6 m^2.

Note: Administer through the tubing of a rapidly infusing solution of D5W or 0.9% NaCl. Avoid extravasation; the use of a central line is suggested. On the Days 3 and 4, mitoXANTRONE should be given 8 hours after the 5th and 7th high dose cytarabine infusions are complete.

Patients with high risk status may proceed to best allogenic donor SCT or alternative donor SCT following Induction II. Depending on timing, these patients may receive a third course of chemotherapy (Intensification I) prior to HSCT. This course could be either AE or HD ARAC/LASP.

Patients with low risk status who had low risk markers and were MRD positive at the end of Induction I and continue to be MRD positive after Induction II will come off protocol.

5.4 Induction II for FLT3-ITD patients – MA with Sorafenib

The following therapy guidelines are for patients during Induction II with FLT3-ITD AML. Induction II lasts 28 days or longer.

Progression to Induction II should await disease response determination (MRD and extramedullary disease) in order to correctly assign risk stratum. Patients with high or low risk cytogenetic/molecular features may proceed on to Induction II therapy before disease response results are available. By the beginning of Induction II, patients should have been classified as Low or High risk. **If a patient is classified as High risk at the end of Induction I and no family HLA match is found, initiation of an unrelated donor search should occur.**

Intrathecal Cytarabine (IT ARAC): IT
IT ARAC may be administered on Day 1 of Induction II or with the bone marrow evaluation at the end of Induction I.

Age-based dosing:

Age (yrs)	Dose
0 - 0.99	20 mg
1 - 1.99	30 mg
2 - 2.99	50 mg
≥ 3	70 mg

For newly detected CNS positive patients who were CNS negative at diagnosis: IT cytarabine is to be administered twice weekly until the CSF is clear. A minimum of 4 intrathecal treatments must be given and a maximum of 6 intrathecal treatments may be given. Patients whose CSF is clear at the second spinal tap will receive 3 intrathecal treatments once the CSF is clear and all other patients will receive 2 intrathecal treatments once the CSF is clear. Patients with refractory CNS leukemia following 6 doses of therapy will be taken off protocol therapy.

High Dose Cytarabine (HD ARAC): IV over 1 - 3 hours
Days: 1 through 4.
Dose: 1000 mg/m^2/dose, every 12 hours (i.e., 2000 mg/m^2/day, divided BID) or
33 mg/kg/dose, every 12 hours (i.e., 66 mg/kg/day, divided BID) if BSA < 0.6 m^2.

Suggested premedications and supportive care: Patients in whom serum creatinine is > 2 mg/dL or > 2 x normal for age, must have a CrCl performed (Day 1) and doses of cytarabine adjusted as outlined.

MitoXANTRONE (MITOX): IV over 15 - 30 minutes
Days: 3 through 6.
Dose: 12 mg/m^2/dose or
0.4 mg/kg/dose if BSA < 0.6 m^2.

Note: Administer through the tubing of a rapidly infusing solution of D5W or 0.9% NaCl. Avoid extravasation; the use of a central line is suggested. On the Days 3 and 4, mitoXANTRONE should be given 8 hours after the 5th and 7th high dose cytarabine infusions are complete.

Sorafenib (SORAF): PO
Days: 7 through 34.
Dose: 200 mg/m^2/dose daily, rounded to accommodate tablet size. The maximum dose will be 400 mg.
Dose to be administered per dosing nomogram in Appendix I.

Note: Sorafenib tablets should be taken at least 1 hour before or 2 hours after food. Tablets should be taken with clear liquids (approximately 2 to 4 ounces for children < 12 years and 4 to 8 ounces for ≥ 12 years). If taken with food, sorafenib should be taken with a moderate to low fat meal. Tablets should not be crushed and should be swallowed whole. However, sorafenib tablets can be dispersed in water to facilitate the administration to subjects that cannot swallow tablets.

Patients with FLT3-ITD AML may proceed to best allogenic donor SCT or alternative donor SCT following Induction II. Depending on timing, these patients may receive a third course of chemotherapy (Intensification I) prior to HSCT. This course could be either AE or HD ARAC/LASP with sorafenib.

5.5 Intensification I – AE

The following Intensification I therapy guidelines are for low risk patients and for patients with high risk disease who do not have an appropriate donor for SCT. Intensification I will last for 28 days or longer. It is suggested, but not required, that patients have an ANC > 1,000/μL and a platelet count > 75,000/μL before proceeding with Intensification I therapy.

Intrathecal Cytarabine (IT ARAC): IT
IT ARAC may be administered on Day 1 of Intensification I or with the bone marrow evaluation at the end of Induction II.

Age-based dosing:

Age (yrs)	Dose

0 - 0.99	20 mg
1 - 1.99	30 mg
2 - 2.99	50 mg
≥ 3	70 mg

High Dose Cytarabine (HD ARAC): IV over 1 - 3 hours
Days: 1 through 5.
Dose: 1000 mg/m^2/dose, every 12 hours, (i.e., 2000 mg/m^2/day, divided BID), or
33 mg/kg/dose, every 12 hours (i.e., 66 mg/kg/day, divided BID) if BSA < 0.6 m^2.

Suggested premedications and supportive care: Administer steroid eye drops (0.1% dexamethasone or 1% prednisolone ophthalmic solution), 2 drops to each eye every 6 hours, beginning immediately before the first dose and continuing for 24 hours after the last dose. If patient does not tolerate steroid eye drops, may administer artificial tears on an every 2-4 hour schedule. Patients in whom serum creatinine is > 2 mg/dL or > 2 x normal for age, must have a CrCl performed (Day 1) and doses of cytarabine adjusted as outlined.

Etoposide (ETOP): IV over 60 - 120 minutes
Days: 1 through 5.
Dose: 150 mg/m^2/dose, or
5 mg/kg/dose if BSA < 0.6 m^2.

Note: Slow rate of administration if hypotension occurs. The use of an in-line filter during the infusion is suggested. Each dose of etoposide should immediately follow the 1st, 3rd, 5th, 7th and 9th doses of cytarabine.

5.6 Intensification I for FLT3-ITD patients – AE with sorafenib

The following Intensification I therapy guidelines are for patients with FLT3-ITD AML who do not have an appropriate donor for SCT. Intensification I will last for 28 days or longer. It is suggested, but not required, that patients have an ANC > 1,000/µL and a platelet count > 75,000/µL before proceeding with Intensification I therapy.

Intrathecal Cytarabine (IT ARAC): IT
IT ARAC may be administered on Day 1 of Intensification I or with the bone marrow evaluation at the end of Induction II.

Age-based dosing:

Age (yrs)	Dose
0 - 0.99	20 mg
1 - 1.99	30 mg
2 - 2.99	50 mg
≥ 3	70 mg

High Dose Cytarabine (HD ARAC): IV over 1 - 3 hours
Days: 1 through 5.
Dose: 1000 mg/m^2/dose, every 12 hours, (i.e., 2000 mg/m^2/day, divided BID), or
33 mg/kg/dose, every 12 hours (i.e., 66 mg/kg/day, divided BID) if BSA < 0.6 m^2.

Suggested premedications and supportive care: Administer steroid eye drops (0.1% dexamethasone or 1% prednisolone ophthalmic solution), 2 drops to each eye every 6 hours, beginning immediately before the first dose and continuing for 24 hours after the last dose. If patient does not tolerate steroid eye drops, may administer artificial tears on an every 2-4 hour schedule. Patients in whom serum creatinine is > 2 mg/dL or > 2 x normal for age, must have a CrCl performed (Day 1) and doses of cytarabine adjusted as outlined.

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Etoposide (ETOP): IV over 60 - 120 minutes

Days: 1 through 5.
Dose: 150 mg/m^2/dose, or
5 mg/kg/dose if BSA < 0.6 m^2.

Note: Slow rate of administration if hypotension occurs. The use of an in-line filter during the infusion is suggested. Each dose of etoposide should immediately follow the 1st, 3rd, 5th, 7th and 9th doses of cytarabine.

Sorafenib (SORAF): PO

Days: 6 through 33
Dose: 200 mg/m^2/dose daily, rounded to accommodate tablet size. The maximum dose will be 400 mg.

Dose to be administered per dosing nomogram in Appendix I.

Note: Sorafenib tablets should be taken at least 1 hour before or 2 hours after food. Tablets should be taken with clear liquids (approximately 2 to 4 ounces for children < 12 years and 4 to 8 ounces for ≥ 12 years). If taken with food, sorafenib should be taken with a moderate to low fat meal. Tablets should not be crushed and should be swallowed whole. However, sorafenib tablets can be dispersed in water to facilitate the administration to subjects that cannot swallow tablets.

5.7 Intensification II – HD ARAC/LASP

The following Intensification II therapy guidelines are for low risk patients and for patients with high risk disease who do not have an appropriate donor for SCT. Intensification II lasts 28 days or longer.

It is suggested, but not required, that patients have an ANC > 1,000/µL and a platelet count > 75,000/µL before proceeding with therapy.

High Dose Cytarabine (HD ARAC): IV over 3 hours

Days: 1, 2 and 8, 9
Dose: 3,000 mg/m^2/dose, every 12 hours (i.e., 6,000 mg/m^2/day, divided BID), or
100 mg/kg/dose, every 12 hours (i.e., 200 mg/kg/day, divided BID) if BSA < 0.6 m^2.

Suggested premedications and supportive care: Administer steroid eye drops (0.1% dexamethasone or 1% prednisolone ophthalmic solution), 2 drops to each eye every 6 hours, beginning immediately before the first dose and continuing for 24 hours after the last dose. If patient does not tolerate steroid eye drops, may administer artificial tears on an every 2-4 hour schedule. Patients in whom serum creatinine is > 2 mg/dL or > 2 x normal for age, must have a CrCl performed (Days 1 and 8) and doses of cytarabine adjusted as outlined.

***Erwinia* L-asparaginase (Erwinaze):** IM (may be given IV over 1 hour)
Days: 2, 9 (one dose per day, 2 doses total for the entire course)
Dose: 25,000 IU/m2 (830 IU/kg/dose if BSA < 0.6 m^2)

Note: Erwinia asparaginase should be given 6 hours after the start of cytarabine doses 4 and 8. If Erwinia asparaginase is not available, pegasparaginase should not be given. Rather, asparaginase should be omitted.

5.8 Intensification II for FLT3-ITD patients – HD ARAC/LASP with sorafenib

The following Intensification II therapy guidelines are for patients with FLT3-ITD AML who do not have an appropriate donor for SCT. Intensification II lasts 28 days or longer.

It is suggested, but not required, that patients have an ANC > 1,000/µL and a platelet count > 75,000/µL before proceeding with therapy.

High Dose Cytarabine (HD ARAC): IV over 3 hours
Days: 1, 2 and 8, 9
Dose: 3,000 mg/m^2/dose, every 12 hours (i.e., 6,000 mg/m^2/day, divided BID), or
100 mg/kg/dose, every 12 hours (i.e., 200 mg/kg/day, divided BID) if BSA < 0.6 m^2.

Suggested premedications and supportive care: Administer steroid eye drops (0.1% dexamethasone or 1% prednisolone ophthalmic solution), 2 drops to each eye every 6 hours, beginning immediately before the first dose and continuing for 24 hours after the last dose. If patient does not tolerate steroid eye drops, may administer artificial tears on an every 2-4 hour schedule. Patients in whom serum creatinine is > 2 mg/dL or > 2 x normal for age, must have a CrCl performed (Days 1 and 8) and doses of cytarabine adjusted as outlined.

***Erwinia* L-asparaginase (Erwinaze):** IM (may be given IV over 1 hour)
Days: 2, 9 (one dose per day, 2 doses total for the entire course)
Dose: 25,000 IU/m2 (830 IU/kg/dose if BSA < 0.6 m^2)

Note: Erwinia asparaginase should be given 6 hours after the start of cytarabine doses 4 and 8. If Erwinia asparaginase is not available, pegasparaginase should not be given. Rather, asparaginase should be omitted.

Sorafenib (SORAF): PO
Days: 11 through 28.
Dose: 200 mg/m^2/dose daily, rounded to accommodate tablet size. The maximum dose will be 400 mg. Dose will be administered per dosing nomogram in Appendix I.

Note: Sorafenib tablets should be taken at least 1 hour before or 2 hours after food. Tablets should be taken with clear liquids (approximately 2 to 4 ounces for children < 12 years and 4 to 8 ounces for ≥ 12 years). If taken with food, sorafenib should be taken with a moderate to low fat meal. Tablets should not be crushed and should be swallowed whole. However, sorafenib tablets can be dispersed in water to facilitate the administration to subjects that cannot swallow tablets.

6.0 DOSE MODIFICATION FOR TOXICITIES

Common Terminology Criteria for Adverse Events (CTCAE)

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

6.1 Allergy to Asparaginase

6.1.1 Local Reactions (Inflammation at Injection Site, Swelling):
Continue asparaginase (*Erwinia*) administration in the presence of Grade 1 allergic reaction (transient flushing or rash; drug fever < 38°C).

Premedication with antihistamines to decrease the risk of overt allergy symptoms is strongly discouraged since antihistamine use may mask the appearance of systemic allergy. Systemic allergy is associated with the presence of asparaginase neutralizing antibodies, which render asparaginase therapy ineffective.

6.1.2 Anaphylaxis/Systemic Allergic Reactions:
Discontinue asparaginase (*Erwinia*) if the patient develops a systemic allergic reaction (urticaria, wheezing, laryngospasm, hypotension, etc.). Should an allergy be diagnosed after the first dose given on Day 2 during Intensification II (for high risk patients), then the dose due on Day 9 should not be administered.

6.2 Allergy to Etoposide

Etoposide allergic reactions may be managed with pre-medications such as diphenhydramine 1 mg/kg IV (maximum dose 50 mg) or an equivalent H-1 receptor antagonist, ranitidine 1 mg/kg IV (maximum dose 50 mg) or an equivalent H-2 receptor antagonist, hydrocortisone 1 - 4 mg/kg IV, and by slowing the rate of the infusion OR etoposide phosphate may be substituted in the same dose and at the same rate. Premedication for etoposide phosphate is recommended.

6.3 Cardiac Toxicity

6.3.1 Left Ventricular Systolic Dysfunction:
Daunorubicin, mitoxantrone, and sorafenib will be held if there is significant evidence of cardiac disease by echocardiogram (shortening fraction < 28% or EF < 55%). Cardiac examination with echocardiogram is required prior to the start of all chemotherapy courses, at the end of protocol therapy, and in follow up. Echocardiogram and cardiac biomarker evaluation (i.e. troponin) are also strongly recommended, particularly in clinical scenarios with potential or suspected cardiac dysfunction.

6.3.2 Daunorubicin and Mitoxantrone:
Do not re-start anthracyclines if held for left ventricular shortening dysfunction that is not associated with a microbiologically proven bacteremia or sepsis. If the left ventricular shortening dysfunction occurred in the setting of microbiologically proven bacteremia or sepsis, then anthracyclines may be reinstituted at the treating clinician's discretion once the shortening fraction has returned to ≥ 28% or EF ≥ 55%.

6.3.3 Sorafenib in Combination with Chemotherapy:
Sorafenib should be held for a shortening fraction of < 28% or EF < 55% regardless of attribution. Sorafenib should be restarted at a reduced dose (100 mg/m^2) when the shortening fraction has returned to ≥ 28% or EF ≥ 55%. If SF decreases below

28% or EF decreases below 55% on 100 mg/m^2, then sorafenib should be discontinued permanently. Sorafenib dose may not be re-escalated in combination with chemotherapy.

6.3.4 Hypertension:
The table below will be used to manage hypertension possibly, probably or definitely related to sorafenib. Management of hypertension not related to sorafenib is at the treating clinician's discretion. Should initiation of anti-hypertensive (anti-HTN) therapy be required, single agent therapy (such as amlodipine or nifedipine) should be started. For patients greater than age 18, rely on CTCAE v.5.0 grading of hypertension for consideration of dose adjustment.

GRADE	MANAGEMENT
For grade 1	No anti-HTN therapy, no change in sorafenib dose
For grade 2	Initiate monotherapy with anti-HTN medication, no change in sorafenib dose
For grade 3	Add additional agent for anti-HTN therapy, hold sorafenib dose. Restart if blood pressure comes back in range for age, gender then dose reduce sorafenib to 100 mg/m^2/day
For grade 4	Permanently discontinue sorafenib

6.4 Coagulopathy

If symptomatic, omit Day 9 asparaginase and consider factor replacement (FFP, cryoprecipitate, factor VIIa). Do not hold asparaginase (*Erwinia*) for abnormal laboratory findings without clinical symptoms.

6.5 Hepatic Toxicity

6.5.1 Transaminases:
If the ALT or AST are > 10 x ULN, attempts should be made to identify the cause and notify the Study Chair. In most cases, the therapy will proceed without modification.

6.5.2 Hyperbilirubinemia:
If the direct bilirubin is > 3 mg/dL, notify the Study Chair. In some cases, it may be necessary to proceed if the bilirubin elevation is a result of the leukemia itself. If the elevated direct bilirubin is not a result of the leukemia, modify the doses of daunorubicin, etoposide and mitoxantrone as follows below. For all cases in which the direct bilirubin is elevated at the point in time that the next course is to begin, consider delaying the course for 1 week to determine whether the direct bilirubin falls to an acceptable level.

6.5.3 Asparaginase:
L-asparaginase has been associated with hepatic toxicity but dosing guidelines for hepatic toxicity are not available. Thus, asparaginase administration in the setting of hepatic toxicity is at the clinician's discretion.

6.5.4 Daunorubicin, Etoposide and Mitoxantrone:
In severe liver dysfunction, the half-life of mitoxantrone is prolonged and the AUC may be more than 3-fold that of patients with normal hepatic function. However,

there are no available dose adjustment guidelines in the literature. Therefore, similar dose reductions of mitoxantrone as outlined below for daunorubicin are utilized on this protocol.

Direct Bilirubin	Daunorubicin	Etoposide	Mitoxantrone
≥ 2 and < 3 mg/dL	50% of the calculated dose	50% of the calculated dose	50% of the calculated dose
≥ 3 and < 5 mg/dL	25% of the calculated dose	25% of the calculated dose	25% of the calculated dose
≥ 5 mg/dL	Hold dose and notify study chair	Hold dose and notify study chair	Hold dose and notify study chair

Full dose of daunorubicin, etoposide and mitoxantrone may resume when the direct bilirubin has fallen to < 1.2 mg/dL.

6.6 Neurologic Toxicity

6.6.1 Cytarabine:

Patients with > Grade 3 CTCAE v 5.0 nervous system disorders from high dose cytarabine should not receive further high dose cytarabine. The most common nervous system disorder is an acute cerebellar syndrome that may manifest itself as ataxia, nystagmus, dysarthria, or dysmetria. However, seizures and encephalopathy have also occurred following therapy with high dose cytarabine.

6.7 Pancreatitis

6.7.1 Asparaginase

Discontinue asparaginase (*Erwinia*) in the presence of hemorrhagic pancreatitis or severe pancreatitis (abdominal pain > 72 hours and > Grade 3 amylase elevation (> 2.0 x ULN). In the case of mild pancreatitis after Day 2, Day 9 asparaginase may be given only if symptoms and signs subside, and amylase levels return to normal. Severe pancreatitis is a contraindication to additional asparaginase administration.

6.8 Renal Toxicity

6.8.1 Cytarabine:

Patients with nephrotoxicity secondary to antibiotics, or antifungals, may have prolonged excretion of cytarabine leading to more severe marrow and extramedullary toxicity. Patients with a serum creatinine > 2 mg/dL or > 2 x normal for age should be hydrated orally or intravenously. Following hydration, the patient must have a creatinine clearance ≥ 60 mL/min/1.73m² as measured preferably by a nuclear GFR Scan or timed urine collection for creatinine clearance before proceeding with high dose cytarabine therapy (doses of 1,000 mg/m2 or greater). If the CrCl is abnormal (< 60 mL/min/1.73m2) then high dose cytarabine should be reduced from twice daily to once daily dosing, at the same previously prescribed doses (e.g., 50% daily dose reduction). With this approach, previous research has shown the prevention of subsequent neurotoxicity in recipients of high dose cytarabine in the face of renal insufficiency.

6.8.2 Etoposide:

In patients with impaired renal function, the following **initial** dose modification of etoposide should be considered based on measured creatinine clearance:

- for CrCl > 60 mL/min, give full dose,
- for CrCl of 15 – 60 mL/min, give 75% of the dose (25% dose reduction).

- for CrCl < 15 mL/min, notify the Study Chair.

(Please note: For dose adjustment in renal dysfunction for children, use the "corrected" value for creatinine clearance measured in mL/min/1.73 m^2**,** which 'corrects' that value for the standardized values of CrCl in adults is measured in mL/min**.** The "corrected" value is loosely interpreted as being equivalent to creatinine clearance measured in an adult patient). **Subsequent doses** should be based on patient tolerance and clinical effect.

6.9 Thrombosis

6.9.1 Asparaginase

Discontinue asparaginase and treat with appropriate antithrombotic therapy, as indicated. Do not hold asparaginase for abnormal laboratory findings without clinical sequelae. For significant thrombosis, not line related, consider evaluation for inherited predisposition to thrombosis.

6.10 Sinusoidal Obstruction Syndrome (SOS, formerly VOD) of the Liver

6.10.1 Gemtuzumab Oozagamicin (GO)

SOS is a known toxicity of GO. No specific predisposing factor is known that may identify the patient at risk.

Criteria for the diagnosis of SOS of the liver

For the purposes of reporting toxicity, the following definition for SOS will be used:
1) Weight gain of more than 10% baseline, and
2) Right upper quadrant pain or tender hepatomegaly, and
3) Total bilirubin 2 mg/dL or greater, and
4) Edema or ascites.

Reversal of portal venous blood flow by ultrasound or pathologic confirmation by liver biopsy are not required for the diagnosis but may be done in support of the clinical diagnosis and/or the evaluation of other diagnostic possibilities. Ultrasound may also be used to assess the severity and course of the patient's SOS.

6.11 Palmar-Plantar Erythrodysesthesia Syndrome, Skin Pain that impacts ADLs or Rash Maculo-Papular

6.11.1 Sorafenib

Grade 1 – Continue sorafenib with topical therapy and supportive care*

Grade 2

- First occurrence – hold sorafenib, if rash improved to <grade 2 in 2 weeks, restart at same dose. If does not improve in 2 weeks, hold till rash improves then start drug at 100 mg/m^2/day dose.
- Second occurrence – hold sorafenib, if rash improved in 2 weeks, restart at 100 mg/m^2/day dose. If does not improve to <grade 2 in 2 weeks, stop sorafenib
- Third occurrence – stop sorafenib

Grade 3
- First occurrence – hold sorafenib, if rash improved in 2 weeks, restart at 100 mg/m^2/day dose. If does not improve to <grade 2 in 2 weeks, stop sorafenib
- Second occurrence – stop sorafenib

*Treatment of sorafenib related Rash/Hand-foot syndrome
- Patients who develop hand-foot syndrome may receive topical emollients (such as Aquaphor) as well as topical or oral steroids or antihistamines if appropriate. Hand foot syndrome may include skin pain without rash that impacts activities of daily living.
- Oral administration of vitamin B 6 (pyridoxine) can also be used for these patients - BSA < 0.5m^2: 50 mg per day; BSA 0.5 - 1.0 m^2: 100 mg per day; BSA 1.1 - 1.5 m^2: 200 mg per day, and BSA > 1.5m^2: 300 mg per day.

7.0 SUPPORTIVE CARE

7.1 General Guidelines

The following guidelines are intended to give general direction for optimal patient care and to encourage uniformity in the treatment of this study population. We will follow our institutional guidelines for management of hospitalization and discharge for these patients.

Hospitalization/Hospital Environment

Hospitalization following each course of chemotherapy is strongly recommended until the absolute phagocyte count (sum of the neutrophils, bands and monocytes) is rising for 2 successive days, and the patient is afebrile and clinically stable.

It is recommended that patients should be assigned to rooms with special air filtration systems such as high efficiency particulate air filters (HEPA) or clean-air rooms with constant positive pressure airflow if at all possible.

Central Venous Access

It is recommended that all patients have a double lumen central venous access line placed prior to the beginning of therapy.

Hyperleukocytosis and Metabolic Derangement

Patients with high peripheral blast counts (> 100,000/μL) may have increased problems related to metabolic abnormalities, bleeding, and hyperviscosity. Transfusion with packed red blood cells should be given very cautiously, since it may increase viscosity and increase the risk of leukostasis. In these patients, the platelet count should be maintained at least greater than 20,000/μL

Continuation of AML Therapy

AML therapy can proceed when there is documented evidence of response to antimicrobial therapy for infections complications, absence of fever or other signs or symptoms of infection, and other starting criteria have been met.

Nutrition

Active measures should be used to prevent weight loss of greater than 10% of pre-illness body weight. If possible, enteral feedings are preferred to parenteral.

Mucosal Evaluation and Care
Mucositis is expected and may be severe; liberal use of pain medications for this condition is encouraged. For patients with poor oral hygiene, consultation by dentistry is recommended prior to initiating therapy.

Suppression of Menstruation
Menstruating females may receive depo-injections or another suppressant during the entire course of this protocol per institutional standards.

Irradiation
Blood products should be irradiated following the current FDA guidelines found at:
http://www.fda.gov/cber/gdlns/gamma.htm

7.2 Infection Prophylaxis

Patients with AML are at a high risk for mortality and morbidity during therapy mainly due to infections. Therefore, patients require an aggressive approach to infection prevention and treatment as outlined below:

- Hospitalization as mentioned in Section 7.1

- Bacterial Prophylaxis
  It is required that all patients except those receiving sorafenib receive broad spectrum gram –positive and gram-negative anti-bacterial prophylaxis at the start of each cycle of therapy and until they have met criteria for discharge as mentioned in section 7.1. Antibiotic can be decided by the treating institution, although levofloxacin is recommended.

- Pneumocystis jiroveci Prophylaxis
  It is required that all patients receive prophylaxis for Pneumocystis jiroveci pneumonia during each cycle. Trimethoprim-sulphamethoxazole, pentamidine isethionate and dapsone are acceptable though trimethoprim-sulphamethoxazole is preferred.

- Fungal prophylaxis
  Prophylactic anti-fungal therapy with voriconazole (<13 yrs), posaconazole (≥13 yrs) or micafungin is required until they have met criteria for discharge as mentioned in section 7.1.

Patients with an ANC < 500/µL (or < 1,000 /µL and falling) and an oral temperature > 38°C twice in 12 hours or ≥ 38.3°C once, should have empiric systemic antibiotics initiated immediately. Institutional guidelines as set forth for patients with AML which include guidelines for fever and neutropenia in AML patients, management of possible and documented fungal infections and pneumocystis prophylaxis will be followed.

8.1 EVALUATIONS/DATA TO BE ACCESSIONED

STUDIES TO BE OBTAINED	Baseline	Each Course of chemotherapy	End of therapy	Relapse/ Refractory
History	X	---	---	---
Physical Exam	X	X	X	X
Vital Signs	X	X	X	X
Height, Weight, BSA	X	X	X	X
Performance status	X	X	X	X
CBC with differential, platelets	X	X	X	X
Liver Function – AST, ALT, Total and Direct Bilirubin, Total Protein, Albumin	X	X	X	X
Kidney Function – BUN, Creatinine	X	X	X	X
Electrolytes (Sodium, Potassium, Chloride, Calcium, Magnesium, Phosphorous) and Glucose	X	X	X	X
Chest X-ray and Pulse oximetry	X	---	---	X
ECHO and EKG	X	X Prior to each course	X	X
Bone marrow aspirate (BMA) and/or biopsy for morphology and flow cytometry to include CD33 expression	X	---	---	X
BMA for FLT3 testing	X	---	---	X
BMA for CD33 Genotype	X	---	---	---
BMA for genomic testing	X	---	---	X
Lumbar Puncture with cell count and cytospin	X	---	---	X
Imaging of chloroma –CT/MRI if applicable	X	---	---	X
BMA for MRD testing	--	X End of each course	X	--
Concomitant Medication Data	X	X	X	X
Adverse Event/Toxicity Monitoring		X	X	

8.2 Data Collection

The REDCap™ system, a secure web application for building and managing online surveys and databases, will be used for data submission. Access to the system will be provided through CHOA, a REDCap™ consortium partner. REDCap will be used to capture data for enrollment, baseline patient, disease and treatment characteristics, and outcomes.

8.3 Data Confidentiality

The confidentiality of each participant record will be rigorously maintained using existing institutional standards. HIPAA and state/federal government regulations for protecting participant privacy and security will be strictly observed. Printing study documents will be discouraged and avoided whenever possible. If it is necessary to print source documentation, it will be maintained and protected by clinical research personnel and kept in a locked file cabinet. All computers used

in the study will be encrypted according to institutional policies. Databases will be stored on the secured servers maintained by Emory University and CHOA

No participant or subject-identifiable information will be given to third parties, including family members (except legal guardians of minors), unless that subject (or legal guardian of minors) has given written or witnessed consent to do so. The results of research studies may be published, but all data will be de-identified prior to publication and individual subjects will not be identified.

If a participant contacts the study's project personnel, he or she will be informed of the status of the research without revealing specific findings.

8.4 Data and Safety Monitoring

This study will be centrally reviewed by the Data Safety and Monitoring Board (DSMB) of the Aflac Clinical Research Office.

The DSMB meets quarterly to review all adverse events and deaths and determine whether any patient safety problems necessitate protocol modifications or discontinuation of the trial.

The DSMB will also meet on an *ad hoc* basis if unexpected safety events occur that may necessitate study suspension or closure. The DSMB will discontinue the review of outcomes when this protocol is closed to accrual.

Before each regularly scheduled DSMB meeting, the study coordinator will submit a report including tabular summaries of all reported SAEs, AEs, and deaths on study to date. The report will also include a brief summary of each previously unreported SAE and death, including an assessment of whether the event was unexpected or related to the study.

If the DSMB recommends protocol or informed consent changes during the study, the recommendations will be reviewed by the Protocol Chair and incorporated into the protocol as deemed appropriate. The protocol with incorporated changes will be distributed to the participating PIs after approval by the Emory University IRB.

8.3.1 Adverse Event Reporting Requirements

The timely reporting of adverse drug reactions (including toxic deaths) is required by the US Food and Drug Administration. The reporting of adverse reactions is in addition to and does not supplant the reporting of toxicities as part of the data reporting for this study. The NCI Common Terminology Criteria for Adverse Events (CTCAE) version 5 will be utilized for adverse event reporting in this trial.

Serious adverse event (SAE) or reaction is any untoward medical occurrence (including an abnormal laboratory finding) associated with the use of a drug in humans that at any dose: results in death, is life-threatening, requires inpatient hospitalization or prolongation of existing hospitalization, results in persistent or significant disability/incapacity, is a congenital anomaly/birth defect, or requires medical or surgical intervention in order to prevent one of the previous listed outcomes. Medical and scientific judgment should be exercised in deciding whether expedited reporting is appropriate in other situations, such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the subject or may require intervention to prevent one of the other outcomes listed in the definition above. These should also usually be

considered serious. Examples of such events are intensive treatment in an emergency room or at home for allergic bronchospasm; or development of drug dependency or drug abuse.

Unexpected adverse drug experience means any adverse drug experience, the specificity or severity of which is not consistent with the current investigator brochure or if an investigator brochure is not required or available, the specificity or severity of which is not consistent with the risk information described in the general investigational plan or elsewhere in the current application, as amended. “Unexpected” as used in this definition, refers to an adverse drug experience that has not been previously observed (e.g., included in the investigator brochure) rather than from the perspective of such experience not being anticipated from the pharmacological properties of the pharmaceutical product.

Attribution: The investigator will be asked to document his/her opinion of the relationship of the event to study medication as follows:

The categories are:
- Definite: The adverse event is clearly related to the study drug(s).
- Probable: The adverse event is most likely related to the study drug(s).
- Possible: The adverse event may be related to the study drug(s).
- Unlikely: The adverse event is doubtfully related to the study drugs(s).
- Unrelated: The adverse event is clearly not related to the study drug(s).

For an SAE, the Primary Study Coordinator should be notified via phone or email within 24 hours of the study personnel becoming aware of the event, and they will notify the DSMB. A copy of the completed Serious Adverse Event form must also be sent to study coordinator through REDCap within 5 calendar days after the incident. The investigator is responsible for reporting serious adverse events and unanticipated problems (UPs) to the IRB according to the guidelines mandated by the institutional IRB. The PI will report to FDA any serious unexpected adverse reactions (SUSARs) according to FDA regulations. Copies of all serious adverse event reports will be kept on file in the Clinical Research Office of the Cancer Center.

9.1 AGENT INFORMATION

9.2 CYTARABINE - ALL ROUTES

(Cytosine arabinoside, Ara-C, Cytosar®) NSC #63878

Source and Pharmacology:
Cytarabine appears to act through the inhibition of DNA polymerase. A limited, but significant, incorporation of cytarabine into both DNA and RNA has also been reported. It exhibits cell phase specificity, primarily killing cells undergoing DNA synthesis (S-phase) and under certain conditions blocking the progression of cells from the G1 phase to the S-phase. Cytarabine is metabolized by deoxycytidine kinase and other nucleotide kinases to the nucleotide triphosphate (Ara-CTP), an effective inhibitor of DNA polymerase. Ara- CTP is inactivated by a pyrimidine nucleoside deaminase, which converts it to the nontoxic uracil derivative (Ara-U). It appears that the balance of kinase and deaminase levels may be an important factor in determining sensitivity or resistance of the cell to cytarabine. It has an initial distributive phase t½ of about 10 minutes, with a secondary elimination phase t½ of about 1 to 3 hours. Peak levels after intramuscular or

subcutaneous administration of cytarabine occur about 20 to 60 minutes after injection and are lower than IV administration. Intrathecally administered doses are metabolized and eliminated more slowly with a t½ of about 2 hours.

Toxicity: (Intravenous, SubQ, IM)

	Common Happens to 21-100 children out of every 100	**Occasional** Happens to 5-20 children out of every 100	**Rare** Happens to < 5 children out of every 100
Immediate: Within 1-2 days of receiving drug	Nausea, vomiting, anorexia *With High Dose*: conjunctivitis	Flu-like symptoms with fever, rash	Ara-C syndrome (fever, myalgia, bone pain, occasionally chest pain, maculopapular rash, malaise, conjunctivitis), anaphylaxis, swelling, pain and redness at the site of the medication injection (SubQ or IM injection) *With High Dose:* cardiomyopathies (vasculitis, and pericarditis), cerebral and cerebellar dysfunction including: encephalopathy, aseptic meningitis, ataxia, dysphasia, nystagmus, a decreased level of consciousness, personality changes, somnolence, seizures
Prompt: Within 2-3 weeks, prior to the next course	Myelosuppression (anemia, thrombocytopenia, leukopenia, megaloblastosis, reticulocytopenia), stomatitis, alopecia	Diarrhea, hypokalemia, hypocalcemia, hyperuricemia *With High Dose*: capillary pulmonary leak syndrome (RDS, pulmonary edema)	Hepatotoxicity, sinusoidal obstruction syndrome (SOS, formerly VOD), urinary retention, renal dysfunction, pain and erythema of the palms and soles
Delayed: Any time later during therapy, excluding the above conditions			Asymptomatic nonoliguric rhabdomyolysis
Unknown Frequency and Timing:	Fetal toxicities and teratogenic effects of cytarabine have been noted in humans. It is unknown whether the drug is excreted in breast milk.		

Toxicity: (Intrathecal)

	Common Happens to 21-100 children out of every 100	**Occasional** Happens to 5-20 children out of every 100	**Rare** Happens to < 5 children out of every 100
Immediate: Within 1-2 days of receiving drug	Nausea, vomiting, fever, headache	Arachnoiditis	Rash, somnolence, meningismus, convulsions, paresis
Prompt: Within 2-3 weeks, prior to the next course			Myelosuppression, ataxia
Delayed: Any time later during therapy, excluding the above condition			Necrotizing leukoencephalopathy, paraplegia, blindness (in combination with XRT & systemic therapy)

Formulation:
Cytarabine for Injection is available in vials of 100 mg, 500 mg, 1 g, and 2 g containing a sterile powder for reconstitution. It is also available at a 20 mg/mL concentration with benzyl alcohol (25 mL per vial) or as a preservative free solution (5 mL, 50 mL per vial), and at a 100 mg/mL concentration with benzyl alcohol (20 mL vial) or as preservative free solution (20 mL vial).

Hydrochloric acid and/or sodium hydroxide may be added to adjust the pH. Store at 25°C (77°F); excursions permitted to 15°-30°C (59°-86°F). Cytarabine solutions should be protected from light.

Guidelines for Administration: See Treatment and Dose Modification sections of the protocol.

IV Infusion:
Reconstitute the lyophilized powder with Bacteriostatic Water for Injection or NS injection. Solution containing bacteriostatic agent should not be used for the preparation of doses > 200 mg/m^2. May be further diluted with dextrose or sodium chloride containing solutions. May give by IV push injection, by IV infusion, or by continuous infusion.

Low Dose (≤ 200 mg/m²/dose): For administration by IV push, reconstitute to a concentration of 20- 100 mg/mL.

High Dose (≥ 1000 mg/m²/dose): Administer steroid eye drops (dexamethasone or prednisolone), 2 drops each eye q6h beginning immediately before the first dose and continuing 24 hours after the last dose. If patient does not tolerate steroid eye drops, administer artificial tears on a q2-4 hour schedule.

Stability: When reconstituted with Bacteriostatic Water for Injection, cytarabine is stable for 48 hours at room temperature. Solutions reconstituted without a preservative should be used immediately. Discard if solution appears hazy. Diluted solutions in D5W or NS are stable for 8 days at room temperature; however, the diluted cytarabine should be used within 24 hours for sterility concerns.

Intrathecal:
For intrathecal administration, dilute with 5-10 mL (or volume per institutional practice) preservative free 0.9% sodium chloride injection, lactated Ringer's injection, Elliot's B solution. The volume of CSF removed should be equal to at least ½ the volume delivered.

Patient Age (years)	Recommended volume	10% CSF volume	CSF Volume *
1 – 1.99	5–10 mL	5 mL	50 ± 10 mL (babies)
2 – 2.99	5-10 mL	8 mL	80 ± 20 mL (younger children)
3 – 8.99	5-10 mL	10 mL	100 ± 20 mL (older children)
9 or greater	5-10 mL	13 mL	130 ± 30 mL (adults)

*Rieselbach, R.E. et.al. Subarachnoid distribution of drugs after lumbar injection; N Engl J Med. 1962 Dec 20; 267:1273-8

Of Note: Larger volumes approximating at least 10% of the CSF volume, isovolumetric delivery, with the patient remaining prone after the procedure may facilitate drug distribution. These procedures have not been validated in clinical trials. They are allowed but not mandated for patients on COG studies.

Stability: Intrathecal cytarabine mixed in NS, lactated Ringer's injection, or Elliot's B solution is stable for 24 hours at 25ºC but contains no preservative and should be administered as soon as possible after preparation.
Supplier: Commercially available from various manufacturers. See package insert for further information.

9.3 DAUNORUBICIN

(Daunomycin, rubidomycin, Cerubidine®) NSC #82151

Source and Pharmacology:

Daunorubicin is an anthracycline antibiotic isolated from cultures of *Streptomyces coeruleorubidus*. Daunorubicin is closely related structurally to doxorubicin only differing in that the side chain of daunorubicin terminates in a methyl group rather than an alcohol. The cytotoxic effect of daunorubicin on malignant cells and its toxic effects on various organs are similar to those of doxorubicin and are thought to be related to nucleotide base intercalation and cell membrane lipid binding activities. Intercalation inhibits nucleotide replication and action of DNA and RNA polymerases. The interaction of daunorubicin with topoisomerase II to form DNA-cleavable complexes appears to be an important mechanism of cytocidal activity. Daunorubicin cellular membrane binding may affect a variety of cellular functions. Enzymatic electron reduction of daunorubicin by a variety of oxidases, reductases, and dehydrogenases generate highly reactive species including the hydroxyl free radical (OH•) which may lead to DNA damage or lipid peroxidation. Daunorubicin is metabolized more rapidly by aldoketoreductases to the active metabolite, daunorubicinol, than is doxorubicin. Daunorubicin hydrochloride is rapidly and widely distributed in tissues, with the highest levels in the spleen, kidneys, liver, lungs, and heart. Daunorubicin serum decay pattern is multiphasic. The initial t½ is approximately 45 minutes followed by a terminal t½ of 18.5 hours. By 1 hour after drug administration, the predominant plasma species is daunorubicinol, which disappears with a half-life of 26.7 hours. Twenty five percent of an administered dose of daunorubicin is eliminated in an active form by urinary excretion and an estimated 40% by biliary excretion.

Toxicity:

	Common Happens to 21-100 children out of every 100	**Occasional** Happens to 5-20 children out of every 100	**Rare** Happens to < 5 children out of every 100
Immediate: Within 1-2 days of receiving drug	Nausea, vomiting, pink or red color to urine, sweat, tears, and saliva	Hyperuricemia, sclerosis of the vein	Diarrhea, anorexia, abdominal pain, extravasation (rare) but if occurs = local ulceration, anaphylaxis, fever, chills, rash, urticaria, acute arrhythmias
Prompt: Within 2-3 weeks, prior to the next course	Myelosuppression (leukopenia, thrombocytopenia, anemia), alopecia	Mucositis (stomatitis and esophagitis), hepatotoxicity	Radiation recall reactions, myocarditis-pericarditis syndrome, conjunctivitis and lacrimation
Delayed: Any time later during therapy			Cardiomyopathy[1] (uncommon at cumulative doses ≤ 550 mg/m², 400 mg/m² with mediastinal radiation, 300 mg/m² in children, or 10 mg/kg in children < 2 yrs or 0.5 m²) (L), hyper-pigmentation of nail beds
Late: Any time after completion of treatment		Subclinical cardiac dysfunction	CHF (on long term follow up in pediatric patients), secondary malignancy (in combination regimens)
Unknown Frequency and Timing:	Fetal toxicities and teratogenic effects of daunorubicin have been noted in animals. It is unknown whether the drug is excreted in breast milk.		

[1] *Risk increases with cardiac irradiation, exposure at a young or advanced age.*
(L) Toxicity may also occur later.

Formulation and Stability:
Daunorubicin is available as red-orange lyophilized powder1 for injection in 20 mg single dose vials and a preservative free 5mg/mL solution2 in 20 mg (4 mL) and 50 mg (10 mL) vials.
¹ Each vial contains 21.4mg of daunorubicin hydrochloride (equivalent to 20 mg of daunorubicin) and 100 mg mannitol.
² Each mL contains 5.3mg daunorubicin hydrochloride (equivalent to 5 mg of daunorubicin), 9 mg of sodium chloride, sodium hydroxide or hydrochloric acid to adjust pH, and Sterile Water for Injection.

Powder for Injection:
Store unreconstituted vial at room temperature, 15°-30°C (59°-86°F). Protect from light. Retain in carton until contents are used. Reconstitute a 20 mg vial with 4 mL SWFI to a final concentration of 5mg/mL. After adding the diluent, the vial should be shaken gently, and the contents allowed to dissolve. The reconstituted solution is stable for 24 hours at room temperature and 48 hours refrigerated. Protect from exposure to sunlight.

Aqueous Solution:
Store refrigerated 2°-8°C, (36°-46°F). Protect from light. Retain in carton until contents are used.

Guidelines for Administration: See Treatment and Dose Modifications sections of the protocol.

Administer by IV side arm into a rapidly flowing infusion solution. Alternately, daunorubicin may be further diluted in saline or dextrose containing solutions and administered by infusion. Protect final preparation from light. To avoid extravasation, the use of a central line is suggested.

Supplier: Commercially available from various manufacturers. See package insert for further information.

9.4 ETOPOSIDE INJECTION
(VePesid®, Etopophos®,VP-16) NSC #141540

Source and Pharmacology:
A semisynthetic derivative of podophyllotoxin that forms a complex with topoisomerase II and DNA which results in single and double strand DNA breaks. Its main effect appears to be in the S and G2 phase of the cell cycle. The initial t½ is 1.5 hours and the mean terminal half-life is 4 to 11 hours. It is primarily excreted in the urine. In children, approximately 55% of the dose is excreted in the urine as etoposide in 24 hours. The mean renal clearance of etoposide is 7 to 10 mL/min/m² or about 35% of the total body clearance over a dose range of 80 to 600 mg/m². Etoposide, therefore, is cleared by both renal and non-renal processes, i.e., metabolism and biliary excretion. The effect of renal disease on plasma etoposide clearance is not known. Biliary excretion appears to be a minor route of etoposide elimination. Only 6% or less of an intravenous dose is recovered in the bile as etoposide. Metabolism accounts for most of the non renal clearance of etoposide.

The maximum plasma concentration and area under the concentration time curve (AUC) exhibit a high degree of patient variability. Etoposide is highly bound to plasma proteins (~94%), primarily serum albumin. Pharmacodynamic studies have shown that etoposide systemic exposure is related to toxicity. Preliminary data suggests that systemic exposure for unbound etoposide correlates better than total (bound and unbound) etoposide. There is poor diffusion into the CSF < 5%.

Etoposide phosphate is a water-soluble ester of etoposide which is rapidly and completely converted to etoposide in plasma. Pharmacokinetic and pharmacodynamic data indicate that etoposide phosphate is bioequivalent to etoposide when it is administered in molar equivalent doses.

Toxicity:

	Common Happens to 21-100 children out of every 100	**Occasional** Happens to 5-20 children out of every 100	**Rare** Happens to < 5 children out of every 100
Immediate: Within 1-2 days of receiving drug	Nausea, vomiting	Anorexia	Transient hypotension during infusion; anaphylaxis (chills, fever, tachycardia, dyspnea, bronchospasm, hypotension)
Prompt: Within 2-3 weeks, prior to next course	Myelosuppression (anemia, leukopenia), alopecia	Thrombocytopenia, diarrhea, abdominal pain, asthenia, malaise, rashes and urticaria	Peripheral neuropathy, mucositis, hepatotoxicity, chest pain, thrombophlebitis, congestive heart failure, Stevens-Johnson Syndrome, exfoliative dermatitis
Delayed: Any time later during therapy			Dystonia, ovarian failure, amenorrhea, anovulatory cycles, hypomenorrhea, onycholysis of nails
Late: Any time after completion of treatment			Secondary malignancy (preleukemic or leukemic syndromes)
Unknown Frequency and Timing:	Fetal toxicities and teratogenic effects of etoposide have been noted in animals at 1/20th of the human dose. It is unknown whether the drug is excreted in breast milk.		

Formulation and Stability:
Etoposide for Injection is available as a 20 mg/mL solution in sterile multiple dose vials (5 mL, 25 mL, or 50 mL each). The pH of the clear, nearly colorless to yellow liquid is 3 to 4. Each mL contains 20 mg etoposide, 2 mg citric acid, 30 mg benzyl alcohol, 80 mg modified polysorbate 80/tween 80, 650 mg polyethylene glycol 300, and 30.5 percent (v/v) alcohol. Vial headspace contains nitrogen. Unopened vials of etoposide are stable until expiration date on package at controlled room temperature (20˚-25˚C or 68˚- 77˚F).

Etoposide phosphate for injection is available for intravenous infusion as a sterile lyophilized powder in single-dose vials containing etoposide phosphate equivalent to 100 mg etoposide, 32.7 mg sodium citrate *USP*, and 300 mg dextran 40. Etoposide phosphate must be stored under refrigeration (2°-8°C or 36°-46°F). Unopened vials of etoposide phosphate are stable until the expiration date on the package.

Guidelines for Administration: See Treatment and Dose Modification sections of the protocol.

<u>Etoposide:</u>
Dilute etoposide to a final concentration ≤ 0.4 mg/mL in D5W or NS. Etoposide infusions are stable at room temperature for 96 hours when diluted to concentrations of 0.2 mg/mL; stability is 24 hours at room temperature with concentrations of 0.4 mg/mL. The time to precipitation is highly unpredictable at concentrations > 0.4 mg/mL. Use in-line filter during infusion secondary to the risk of precipitate formation. However, the use of an in-line filter is not mandatory since etoposide precipitation is unlikely at concentrations of 0.1-0.4 mg/mL. **Do not administer etoposide by rapid intravenous injection.** Slow rate of administration if hypotension occurs.

Leaching of diethylhexyl phthalate (DEHP) from polyvinyl chloride (PVC) bags occurred with etoposide 0.4mg/mL in NS. To avoid leaching, prepare the etoposide solution as close as possible, preferably within 4 hours, to the time of administration or alternatively as per institutional policy; glass or polyethylene-lined (non-PVC) containers and polyethylene-lined tubing may be used to minimize exposure to DEHP.

Etoposide Phosphate:
Reconstitute the 100 mg vial with 5 or 10 mL of Sterile Water for Injection, D5W, NS, Bacteriostatic Water for Injection with Benzyl Alcohol, or Bacteriostatic Sodium Chloride for Injection with Benzyl Alcohol for a concentration equivalent to 20 mg/mL or 10 mg/mL etoposide equivalent (22.7 mg/mL or 11.4 mg/mL etoposide phosphate), respectively. **Use diluents without benzyl alcohol for neonates and infants < 2 years of age or patients with hypersensitivity to benzyl alcohol.**

When reconstituted as directed, etoposide phosphate solutions can be stored in glass or plastic containers under refrigeration for 7 days. When reconstituted with a diluent containing a bacteriostat, store at controlled room temperature for up to 48 hours. Following reconstitution with SWFI, D5W, or NS store at controlled room temperature for up to 24 hours.

Following reconstitution, etoposide phosphate may be further diluted to a concentration as low as 0.1 mg/mL of etoposide with D5W or NS. The diluted solution can be stored under refrigeration or at controlled room temperature for 24 hours.

Supplier:
Commercially available from various manufacturers. See package insert for more detailed information.

9.4 GEMTUZUMAB OZOGAMICIN
(Mylotarg®, CDP-771, CMA-676, hP67.6-calicheamicin) NSC # 720568

Source and Pharmacology: Gemtuzumab ozogamicin is a combination of recombinant humanized anti-CD33 monoclonal antibody, conjugated with the cytotoxic antibiotic calicheamicin, isolated from fermentation of a bacterium, Micromonospora echinospora subsp. calichensis. The antibody portion of Gemtuzumab binds specifically to the CD33 antigen expressed on the surface of leukemic blasts and immature normal cells of myelomonocytic lineage, but not on normal hematopoietic stem cells. This antigen is expressed on the surface of leukemic blasts in more than 80% of patients with acute myeloid leukemia (AML). The binding of anti-CD33 antibody with the CD33 antigen results in the formation of a complex that is internalized, releasing the calicheamicin inside the lysosomes of the myeloid cell. The calicheamicin derivative binds to DNA in the minor growth groove, causing DNA double-strand breaks and cell death.

The first administration of a 9 mg/m^2 dose of gemtuzumab ozogamicin, given as a 2 hour infusion, results in elimination half-lives of total and unconjugated calicheamicin of about 41 and 143 hours, respectively. After the second 9 mg/m^2 dose, the half-life of total calicheamicin was increased to about 64 hours and the area under the concentration-time curve (AUC) was about twice that in the first dose period. The AUC for the unconjugated calicheamicin increased 30% after the second dose.

Toxicity:

	Common Happens to 21-100 children out of every 100	**Occasional** Happens to 5-20 children out of every 100	**Rare** Happens to <5 children out of every 100
Immediate: Within 1-2 days of receiving drug	Post infusion reaction (fever, chills, and rigors) dyspnea, headache, abdominal pain, asthenia, nausea, vomiting, anorexia, constipation, hypokalemia	Hypertension or hypotension, dizziness, tachycardia, back pain, diarrhea, dyspepsia, hypocalcemia, magnesemia, phosphatemia, anxiety, insomnia, depression	Anaphylaxis, hypoxia, hyperglycemia, tumor lysis syndrome
Prompt: Within 2-3 weeks, prior to the next course	Myelosuppression, increased liver enzymes, mucositis or stomatitis	Rash, pruritis, increased cough , infection, hemorrhage, hyperbilirubinemia, VOD (patients who receive gemtuzumab before or after HSCT) (L)	Intracranial bleed, DIC, GI bleed, VOD (L), ARDS (hypoxemia, pulmonary infiltrates, non–cardiogenic pulmonary edema) (L)
Delayed: Any time later during therapy, excluding the above conditions			
Late: Any time after completion of treatment			
Unknown Frequency and Timing: Fetal toxicities and teratogenic effects of gemtuzumab have been noted in animals at doses muc than those used in humans. Toxicities include: decreased skeletal ossification, multiple anomalies, low birth weight and increased f mortality. It is not known if gemtuzumab is excreted into breast milk.			

(L) Toxicity may occur later

Formulation and Stability: Available as single dose 5 mg amber vials of lyophilized powder. Store in the refrigerator at 2º to 8ºC (36º-46º F), protected from light. Bring the vials to room temperature before diluting. Reconstitute vial contents with 5 mL of sterile water, and gently swirl the solution. Inspect the final solution for particulates. The reconstituted solution has a concentration of 1 mg/mL and is stable ≤ 8 hours if refrigerated and protected from light or ≤ 2 hours at room temperature prior to a 2-hour infusion.

Note: The product is very light sensitive. The vials should be diluted, and solution prepared, in a biologic safety cabinet with the fluorescent lights turned off.

Guidelines for Administration: See Treatment and Dose Modifications sections of the protocol.

Add appropriate dose to 100 mL of normal saline in either a polyvinyl chloride (PVC) or ethylene/polypropylene copolymer (non-PVC) IV bag covered by an ultraviolet (UV) light protector.

Gemtuzumab should only be diluted with 0.9% sodium chloride solution.
The diluted solution is stable ≤ 16 hours at room temperature prior to a 2-hour infusion.

Infuse over a 2-hour period in a peripheral or central line. **Do not give IV push or bolus.** Give through a separate IV line, using a 0.22 micron or 1.2-micron low protein-binding in-line filter.

Premedicate with acetaminophen and diphenhydramine and repeat every 4 hours as needed to prevent post-infusion symptom complex. Handle as with other cytotoxic agents. **Protect solution from light at all times**. Monitor vital signs during infusion and for 4 hours following infusion.

Supplier: Commercially available in the U.S. See package insert for further information.

9.5 ASPARAGINASE (ERWINIA CHRYSANTHEMI)

(Erwinia *chrysanthemi*, Erwinase®, ErwinazeTM, Crisantaspase)

Source and Pharmacology:

L-asparagine is a nonessential amino acid synthesized by the transamination of L-aspartic acid by a reaction catalyzed by the enzyme L-asparagine synthetase. Neoplastic cells associated with acute lymphoblastic leukemia, acute myeloid leukemia and lymphoblastic lymphosarcoma are asparagine-dependent but lack asparagine synthetase activity. The administration of L-asparaginase produces an anti-neoplastic effect by catalyzing asparagine into aspartic acid and ammonia. As a result, these cells lack the ability to produce the asparagine necessary for protein metabolism and survival. Deamination of glutamine may also play a role in the antineoplastic activity of asparaginase.

Asparaginase *Erwinia chrysanthemi* (Erwinaze®) is asparaginase derived from cultures of *Erwinia chrysanthemi*. L-asparaginase is a tetrameric enzyme; each of the four identical subunits has a molecular weight of approximately 35 kDa. Asparaginase *Erwinia chrysanthemi* is immunologically distinct from *E. coli* L-asparaginase and may allow continued asparaginase therapy when a hypersensitivity reaction occurs to *Escherichia coli*-derived asparaginase. The package labeling states that there is insufficient information to characterize the incidence of antibodies to asparaginase *Erwinia chrysanthemi*. Several factors are involved in immunogenicity assay results and the assessment of antibodies, including assay methodology, assay sensitivity and specificity, sample handling, timing of sample collection, concomitant medications, and the underlying disease state. The following data have been reported on each of the three preparations of asparaginase:

Clinical Pharmacology of Asparaginase Formulation	Elimination half-life (IM)	% Anti-Asparaginase Antibody positive patients
Native *Escherichia Coli*	26-30 hours	45-75
Pegylated-asparaginase	5.5-7 days	5-18
Erwinia Asparaginase	16 hours (7-13 hrs package insert)	30-50

From: Avramis, V; Panosyan, E; Pharmacokinetic/Pharmacodynamic Relationships of Asparaginase Formulations: The Past, the Present and Recommendations for the Future. Clin Pharmacokinet 2005; 44 (4): 367-393.

Effective asparaginase levels have been defined as activity of ≥ 0.1 International Units per mL. Clinical trials with asparaginase *Erwinia chrysanthemi* demonstrated that 100% of patients achieved effective asparaginase levels at 48 and 72 hours (n=35 and n=13, respectively) following the third total dose when given on a Monday, Wednesday, Friday schedule using the IM route of administration. In a multicenter study characterizing the pharmacokinetic profile of 25,000 International Units/m^2 Erwinaze® given intravenously over one hour on the same dosing schedule of Monday, Wednesday, Friday for 2 consecutive weeks, 83% (20/24) and 43% (9/21) of evaluable patients achieved an asparaginase activity level of ≥ 0.1 International Units/mL at 48 post-dose 5 and 72 hours post-dose 6, respectively. No formal drug interaction studies have been performed with asparaginase *Erwinia chrysanthemi*.

Toxicity:

	Common Happens to 21-100 children out of every 100	Occasional Happens to 5-20 children out of every 100	Rare Happens to < 5 children out of every 100
Immediate: Within 1-2 days of receiving drug		Allergic reactions, anaphylaxis, urticaria	Local injection site reactions, fever
Prompt: Within 2-3 weeks, prior to the next course			Pancreatitis, glucose intolerance, thrombosis, hemorrhage, transient ischemic attack, disseminated intravascular coagulation, hyperbilirubinemia, alanine aminotransferase increased, aspartate aminotransferase increased, hyperglycemia, hyperammonemia, vomiting, nausea, abdominal pain, headache, diarrhea, seizure
Unknown Frequency and Timing:	Fetal toxicities and teratogenic effects of L-asparaginase have been noted in animals. It is unknown whether the drug is excreted in breast milk. Adequate, well-controlled studies of asparaginase *Erwinia chrysanthemi* have NOT been conducted. It is not known whether asparaginase *Erwinia chrysanthemi* will cause fetal harm or affect the ability to reproduce. It is not known if asparaginase *Erwinia chrysanthemi* is excreted into breast milk. The use of asparaginase *Erwinia chrysanthemi* should be avoided in pregnant or lactating patients.		

(L) Toxicity may also occur later.

Formulation and Stability:
Asparaginase *Erwinia chrysanthemi* is supplied as a sterile, white lyophilized powder for reconstitution in a clear glass vial with a 3 mL capacity. Each vial contains 10,000 International Units of asparaginase *Erwinia chrysanthemi* and the following inactive ingredients: glucose monohydrate (5.0 mg), sodium chloride (0.5 mg). Store intact vials between 2°C and 8°C (36° to 46ºF). Protect from light.

Guidelines for Administration: See Treatment and Dose Modification sections of the protocol. *Erwinia* asparaginase can be administered by intramuscular injection or by intravenous infusion. Use appropriate precautions for preparation of a hazardous agent. Visually inspect the powder in vial for foreign particles or discoloration prior to reconstitution.

For intramuscular administration, the contents of each vial should be reconstituted by slowly adding 1 mL or 2 mL of sterile, preservative-free NS to the inner vial wall. The final concentration is 10,000 International Units per mL when using 1 mL for reconstitution or 5,000 International Units per mL when using 2 mL for reconstitution. Gently mix or swirl the contents to dissolve the contents of the vial. Do not shake or invert the vial. The resulting solution should be clear and colorless. Discard if any particulate matter or protein aggregates are visible. **Withdraw the appropriate dosing volume into a polypropylene syringe within 15 minutes of reconstitution.** Polycarbonate luer-lok syringes from B-D (1 mL) are also acceptable (personal communication, EUSA Pharma). Discard any unused drug; do not save or use any unused drug remaining the in the vial. No more than 2mL should be given at any one injection site. Doses larger than 2 mL should be divided and given in separate administration sites.
For intravenous use, slowly inject the appropriate volume of reconstituted solution into a Normal Saline 100 mL infusion bag; do not shake or squeeze the bag. Infuse *Erwinia* asparaginase over 1 hour. Do not infuse other intravenous drugs through the same intravenous line while infusing *Erwinia* asparaginase.

Administer the dose within a 4 hour time period from reconstitution. If the dose is not used within this time period, discard the dose. Do not freeze or refrigerate the reconstituted solution. Have available during and after the infusion: antihistamine, epinephrine, oxygen, and IV corticosteroids. Observe patient for ONE hour after administration for signs of hypersensitivity reactions.

9.6 SORAFENIB TOSYLATE

(BAY 43-9006 tosylate, BAY 54-9085 Nexavar®)

Source and Pharmacology: Sorafenib tosylate has the chemical name: 4-(4-{3-[4-Chloro-3-(trifluoromethyl) phenyl] ureido} phenoxy) *N*2-methylpyridine-2-carboxamide 4-methylbenzenesulfonate. Sorafenib is a kinase inhibitor that decreases tumor cell proliferation *in vitro.* Sorafenib was shown to inhibit multiple intracellular (CRAF, BRAF and mutant BRAF) and cell surface kinases (KIT, FLT-3, RET, VEGFR-1, VEGFR-2, VEGFR-3, and PDGFR-ß). Several of these kinases are thought to be involved in tumor cell signaling, angiogenesis, and apoptosis. Sorafenib inhibited tumor growth and angiogenesis of human hepatocellular carcinoma, human renal cell carcinoma, and several other human tumor xenografts in immunocompromised mice.

The mean relative bioavailability of sorafenib administered as an oral tablet was 38–49% when compared to an oral solution. Pharmacokinetics studies in adults who received oral sorafenib showed that sorafenib reaches peak plasma levels in approximately 3 hours. When given with a moderate-fat meal (30% fat; 700 calories), the bioavailability was similar to that in the fasted state, however, with a high-fat meal (50% fat; 900 calories), sorafenib bioavailability was reduced by 29% compared to administration in the fasted state. Therefore, it is recommended that sorafenib be administered without food.

The mean maximum plasma concentration (Cmax) and area under the concentration curve (AUC) increased less than proportionally when doses greater than 400 mg are given orally twice daily. *In vitro* binding of sorafenib to human plasma proteins is 99.5%. Sorafenib is metabolized mainly in the liver through oxidation by cytochrome P-450 (CYP) isoenzyme 3A4, and glucuronidation by uridine diphosphate-glucuronosyltransferase (UGT) 1A9. Sorafenib accounted for approximately 70–85% of the circulating analytes in plasma at steady state. Eight metabolites of sorafenib have been identified. The primary metabolite, a pyridine *N*-oxide derivative, is pharmacologically active, shows *in vitro* potency similar to that of sorafenib, and accounts for approximately 9-16% of total plasma concentrations of the drug. Approximately 77% of an oral dose of sorafenib is excreted in feces; 19% is eliminated in urine as glucuronidated metabolites. Unchanged sorafenib is recovered in feces and accounts for 51% of a single dose. No unchanged sorafenib is recovered in the urine.

The mean elimination half-life of sorafenib is approximately 25 to 48 hours. Multiple dosing of sorafenib for 7 days resulted in a 2.5 to 7-fold accumulation compared to single dose administration. Steady-state plasma sorafenib concentrations are achieved within 7 days, with a peak-to-trough ratio of mean concentrations of less than 2. A study of the pharmacokinetics of sorafenib indicated that the mean AUC of sorafenib in Asians (N=78) was 30% lower than the mean AUC in Caucasians (N=40). In adult patients, gender and age do not appear to have a clinically meaningful effect on the pharmacokinetics of sorafenib. In children with refractory solid tumors or leukemias, single agent sorafenib pharmacokinetics were similar to adults. Significant interpatient variability was observed both on Day 1 and at steady state. Drug exposure increased only marginally with dose escalation from 150 to 200 mg/m2 for day 1 pharmacokinetics. The terminal half-life in children appeared to be prolonged (≥24 hours) but could not be estimated. Sorafenib accumulated after multiple doses and steady state appeared to be achieved after 5 to

7 days. Steady-state sorafenib concentrations (AUC0–12h/12) were 4.1 ± 2.1 µg/mL at the 150 mg/m2 and 5.4 ± 1.8 µg/mL at 200 mg/m2 dose levels, respectively. The median apparent sorafenib clearance at the MTD was 56.0 mL/min/m2. The apparent sorafenib clearance increased with patient age. The observed single agent sorafenib MTD in pediatric patients was 200 mg/m2/dose twice daily for solid tumors and 150 mg/m2/dose twice daily for leukemias. The pharmacokinetics of sorafenib in patients with varying degrees of renal and hepatic impairment have been studied. No dose adjustment is necessary in patients with renal impairment or with mild to moderate hepatic impairment.

Studies in human liver microsomes demonstrated that sorafenib competitively inhibited CYP2B6, CYP2C8, CYP2C9, CYP2C19, CYP2D6, and CYP3A4. However, sorafenib 400 mg twice daily for 28 days with substrates of CYP3A4, CYP2D6 and CYP2C19 did not increase the systemic exposure of these substrates. Studies with cultured human hepatocytes demonstrated that sorafenib did not increase CYP1A2 and CYP3A4 activities, suggesting that sorafenib is unlikely to induce CYP1A2 or CYP3A4 in humans. Drug interaction studies with the CYP3A4 inhibitor ketoconazole did not demonstrate a clinically important decrease in sorafenib exposure, however, the concentration of the active *N*-oxide metabolite decreased. Therefore, the use of strong or clinically relevant moderate CYP3A4 inhibitors should be avoided or used with caution. Continuous concomitant administration of sorafenib and rifampicin resulted in an average 37% reduction of sorafenib AUC. Other inducers of CYP3A4 activity (e.g., St. John's wort, phenytoin, carbamazepine, phenobarbital, and dexamethasone) may also increase the metabolism of sorafenib and thus decrease sorafenib concentrations. Please see Appendix II.

In vitro studies indicate that sorafenib inhibits glucuronidation by the uridine diphosphate-glucuronosyltransferase (UGT) 1A1 and 1A9 pathways. Systemic exposure to UGT 1A1 or 1A9 substrates may increase when co-administered with sorafenib. Caution is recommended when sorafenib is used concomitantly with drugs predominantly metabolized by the UGT 1A1 pathway such as irinotecan, whose active metabolite SN-38 is metabolized by UGT 1A1.
Sorafenib also inhibited P-glycoprotein *in vitro* and could increase the concentrations of concomitantly administered drugs that are P-glycoprotein substrates.
Concomitant use of docetaxel or doxorubicin with continuous sorafenib administration resulted in an increase in the AUC of both docetaxel and doxorubicin. With fluorouracil, both increases and decreases in AUC were seen. Therefore, caution is recommended when administering sorafenib with these chemotherapeutic drugs. Sorafenib does not appear to affect the metabolism of warfarin (a CYP2C9 substrate) *in vivo*; however, infrequent bleeding events or elevations of the International Normalized Ratio (INR) have been reported in some patients receiving concomitant therapy with warfarin. Patients taking concomitant warfarin should be monitored regularly for changes in prothrombin time, INR or clinical bleeding episodes.
Repeat dosing of sorafenib to young and growing dogs resulted in irregular thickening of the femoral growth plate at daily doses ≥ 600 mg/m2 (approximately 0.3 times the AUC at the recommended human dose), hypocellularity of the bone marrow adjoining the growth plate at 200 mg/m2/day (approximately 0.1 times the AUC at the recommended human dose), and alterations of the dentin composition at 600 mg/m2/day. Similar effects were not observed in adult dogs when dosed for 4 weeks or less.

Toxicity

Adverse Events with Possible Relationship to Sorafenib (BAY 43-9006; Nexavar) (CTCAE 4.0 Term) [n= 2571]		
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**
BLOOD AND LYMPHATIC SYSTEM DISORDERS		
Anemia		
CARDIAC DISORDERS		
		Acute coronary syndrome
	Chest pain - cardiac	
		Heart failure
		Left ventricular systolic dysfunction
		Myocardial infarction
GASTROINTESTINAL DISORDERS		
Abdominal pain		
	Ascites	
	Constipation	
Diarrhea		
	Gastrointestinal hemorrhage[2]	
		Gastrointestinal perforation[3]
	Mucositis oral	
Nausea		
	Vomiting	
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS		
	Edema limbs	
Fatigue		
	Fever	
HEPATOBILIARY DISORDERS		
		Hepatic failure
IMMUNE SYSTEM DISORDERS		
		Anaphylaxis
INFECTIONS AND INFESTATIONS		
	Infection[4]	
INVESTIGATIONS		
	Activated partial thromboplastin time prolonged	
Alanine aminotransferase increased		
Alkaline phosphatase increased		
Aspartate aminotransferase increased		
Blood bilirubin increased		
Creatinine increased		
		Electrocardiogram QT corrected interval prolonged
	GGT increased	
INR increased		
	Investigations - Other (bicarbonate-serum low)	
Lipase increased		

Adwrse Ewnts nith Possil:ie Relationship to Sorafenih (BAY 43-9006; exa\:lr) (CTCAE 4.0 Term) [n= 2 571]		
Likely (=:20%)	Less Likely «20%)	Rare but Serious (<3%)
Lymphocytecotmt decreased		
	Neutrophi1coun:t decreased	
Platelet cotmt decreased		
Serum amylase increased		
Weight loss		
\Vhite blood cell decreased		
MErABOilSM AND NUTRITION DISORDERS		
Anorexia		
	Hypercalcemia	
Hyperglycemia		
	Hyperkalemia	
	Hypernatremia	
Hypoalbllllinemia		
Hypocalcemia		
	Hypoglycemia	
	Hypokalemia	
Hyponatremia		
Hypophosphatemia		
MUSCULOSKELErAL AND CONNECTIVE TISSUE DISORDERS		
	Arthralgia	
	Back pain	
	Bonepain	
	Musculoskeletal and cowiective tissuedisorder - Other(muscle spasm)	
	Myalgia	
	Pain in extremity	
NEOPIASMS BENICN, MAUCNANT AND UNSPECIFIED (INCL CYSTS AND POLYPS)		
	Treatmentrelated secondary malicy	
NERVOUS SYSTEM DISORDERS		
	Dizziness	
	Headache	
		Intracranial henlorrhage
		Reversible posterior leukoencephalopathy syndrome
PSYCHIATRIC DISORDERS		
	Insomnia	
RENAL AND URINARY DISORDERS		
	Acutekidney injury	
RESPIRATORY, THORAOC AND MEDIASTINAL DISORDERS		
	Coug)l	
	Dyspnea	
	Respiratory henuorrhage>	
	Voicealteration	
SKIN AND SUBCUTANEOUS TISSUE DISORDERS		

Adverse Events with Possible Relationship to Sorafenib (BAY 43-9006; Nexavar) (CTCAE 4.0 Term) [n= 2571]		
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**
Alopecia		
	Dry skin	
		Erythema multiforme
Palmar-plantar erythrodysesthesia syndrome		
	Pruritus	
Rash maculo-papular		
		Stevens-Johnson syndrome
		Toxic epidermal necrolysis
VASCULAR DISORDERS		
	Hypertension	
		Thromboembolic event

Formulation and Stability:
Sorafenib is supplied as a yellow-orange 50 mg and a red-colored 200 mg, round, immediate-release filmcoated tablet containing the excipients croscarmellose sodium, microcrystalline cellulose, hydroxypropylmethylcellulose, sodium lauryl sulfate, and magnesium stearate. The 200 mg film-coat consists of hydroxypropylmethyl cellulose, polyethylene glycol, titanium dioxide and red iron oxide. The 50 mg film- coat consists of hypromellose, PEG 3350, titanium dioxide, and yellow ferric oxide. The film coating has no effect on active BAY 43-9006 tosylate release rate.

Sorafenib 200 mg tablets are supplied in high-density polyethylene (HDPE) bottles of 140 tablets and the 50 mg tablets are supplied in HDPE bottles of 100 tablets. The bottles should be stored at controlled room temperature (15-25ºC; 59-77°F) and storage temperature should not exceed 25ºC (77°F). Stability testing is ongoing.

10.1 CRITERIA FOR REMOVAL FROM PROTOCOL THERAPY AND OFF STUDY CRITERIA

10.2 Criteria for Removal from Protocol Therapy

1. Failure to attain or maintain remission following Induction II (≥ 5% malignant blasts or extramedullary disease).
2. Relapse after Induction II of any site following remission.
3. Refractory CNS leukemia following 6 doses of IT cytarabine therapy in Induction I or Induction II.
4. CNS leukemia at start of Induction II after treatment for CNS leukemia in Induction I.
5. Grade 3 or greater neurotoxicity from HD cytarabine or,

6. Other intolerable or unacceptable toxicity secondary to the standard chemotherapy backbone.
7. Refusal of further protocol therapy by patient/parent/guardian.
8. Completion of planned therapy.
9. Physician determines it is in patient's best interest.
10. Development of a second malignancy.
11. Graft failure

Patients who are off protocol therapy are to be followed until they meet the criteria for Off Study (see below). Follow-up data will be required unless consent was withdrawn.

10.3 Off Study Criteria

1. Death.
2. Lost to follow-up.
3. Patient enrollment onto another study with tumor therapeutic intent (e.g., at recurrence).*
4. Withdrawal of consent for any further data submission.
5. Tenth anniversary of study entry.
6. Excluded constitutional condition or oncologic diagnosis is indicated following enrollment
7. FLT3/ITD status is unknown at End of Induction I.

11.1 STATISTICAL CONSIDERATIONS

Sample size: This is a pilot study aimed at utilizing a risk-stratified approach along with pharmacogenomics to treat newly diagnosed patients with AML. Annually we expect to have 10-20 de novo AML patients to be diagnosed and treated at our institution. This two-year study would be expected to enroll approximately 30 patients (both low and high risk) during that time period. As a pilot study, this trial is not hypothesis-driven but rather estimation-driven, and the sample size is based on a feasible two-year target for our institution.

Descriptive statistics will be conducted for demographic and patient characteristic variables, including toxicity related variables. Mean and standard deviation will be used for continuous variables and frequency and percent will be used for categorical variables.

Primary Endpoints:

1. Event-free survival: Event-free survival (EFS) is defined as the time from on study to death, failure to achieve remission or relapse, where those patients who do not experience these events will be censored at last follow-up date. EFS will be estimated using the Kaplan-Meier method for newly diagnosed patients with pediatric acute myeloid leukemia using a risk stratified approach.

Secondary Endpoints:

1. Disease-free survival: Disease-free survival (DFS) from end of first course of therapy is defined as time from end of first course of therapy to death or relapse, where those patients who do not experience these events will be censored at last follow-up date. DFS will be estimated using the Kaplan-Meier method for newly diagnosed patients with pediatric acute myeloid leukemia using a risk stratified approach.

2. Overall survival: Overall survival (OS) is defined as (1) time from study entry and (2) from end of first course of therapy to death, where those who do not die will be censored at

last follow-up date. OS will be estimated using the Kaplan-Meier method for newly diagnosed patients with pediatric acute myeloid leukemia using a risk stratified approach.

3. Minimal residual disease: Minimal residual disease (MRD) is the proportion of patients that are in remission (MRD negative) after one cycle of induction chemotherapy in both the GO and non-GO groups. Descriptive statistics (frequency and percent) will be used to estimate the frequency of MRD.

4. Disease-free-survival MRD negative: Disease-free-survival, defined as time from end of first course of therapy to death or relapse, will be estimated for MRD negative patients who lack high or low risk molecular and cytogenetic features using the Kaplan-Meier method.

5. Cardiac ejection fraction: Descriptive statistics (frequency and percent) will be used to estimate the frequency of patients that develop cardiac ejection fraction <50% (CTCAE V5.0 grade 2 or greater dysfunction) either during therapy (early cardiotoxicity) or after completion of therapy (late cardiotoxicity).

Exploratory Endpoints:

1. Molecular abnormalities: Descriptive statistics will be used to describe the molecular abnormalities seen in de-novo AML samples using comprehensive genomic testing.

Analysis Datasets:

All eligible patients who receive at least one dose of protocol therapy will be included in the primary efficacy analysis. Safety analysis will be conducted using the Safety Population, defined as any participant receiving at least one dose of study treatment.

Interim Analysis:

The rates of infectious and hematologic toxicities with each chemotherapy course are well established from prior COG study AAML0531 (refer to table below). These toxicity rates will be monitored after enrollment of the first 10 patients and subsequently after the next 10 patients are enrolled. While this is highly unlikely to happen, if toxicity rates are exceeding those expected for each course, the study will be stopped.

AAML0531	ADE	ADE +GO	MA	AE	Capizzi
Infection incidence					
Documented infection	35 %	36 %	69%	49 %	67%
Neutropenic fever	31 %	32 %	23%	23 %	18%
Hematologic Recovery (days)					
Median time to platelets >50K	26	28	38	25	42
Median time to ANC >500	30	30	37	27	40

APPENDIX I – SORAFENIB DOSING NOMOGRAM

AAML1031 Sorafenib Dosing Nomogram			
BSA Range (m^2)	**Dose Level 0** 100 mg/m^2/day once daily (DOSE/WEEK)	**Dose Level 1** 200 mg/m^2/day once daily (DOSE/WEEK)	**Dose Level 2** 300 mg/m^2/day divided BID (DOSE/WEEK)
≤ 0.35	50 mg x 4 days/week, 0 x 3 day/week (200 mg)	50 mg x 5 days/week, 100 mg x 2 days/week (450 mg)	50 mg twice daily x 7 days/week (700 mg)
0.36 - 0.4	50 mg x 5 days/week, 0 x 2 day/week (250 mg)	50 mg x 4 days/week, 100 mg x 3 days/week (500 mg)	50 mg twice daily x 6 days/week + 100 mg twice daily x 1 day/week (800 mg)
0.41 - 0.45	50 mg x 6 days/week, 0 x 1 day/week (300 mg)	50 mg x 2 days/week, 100 mg x 5 days/week (600 mg)	50 mg twice daily x 5 days/week + 100 mg twice daily x 2 days/week (900 mg)
0.46 - 0.55	50 mg x 7 days/week (350 mg)	100 mg x 7 days/week (700 mg)	50 mg q AM +100 mg q PM x 7 days/week (1050 mg)
0.56 - 0.6	50 mg x 6 days/week, 100 mg x 1 day/week (400 mg)	100 mg x 6 days/week, 200 mg x 1 day/week (800 mg)	50 mg twice daily x 2 days/week + 100 mg twice daily x 5 days/week (1200 mg)
0.61 - 0.7	50 mg x 5 days/week, 100 mg x 2 days/week (450 mg)	100 mg x 5 days/week, 200 x 2 days/week (900 mg)	100 mg twice daily x 7 days/week (1400 mg)
0.71 - 0.8	50 mg x4 days/week, 100 mg x3 days/week (500 mg)	150 mg x 7 days/week (1050mg)	100 mg twice daily x 5 days/week + 150 mg twice daily x 2 days/week (1600 mg)
0.81 - 0.9	50 mg x 2 days/week, 100 mg x 5 days/week (600 mg)	150 mg x 4 days/week, 200 mg x 3 days/week (1200 mg)	100 mg q AM + 150 mg q PM x 7 days/week (1750 mg)
0.91 - 1.05	100 mg x 7 days/week (700 mg)	200 mg x 7 days/week (1400 mg)	150 mg twice daily x 7 days/week (2100 mg)
1.06 - 1.2	100 mg x 6 days/week, 200 mg x1 day/week (800 mg)	200 mg x 3 days/week, 250 mg x 4 days/week (1600 mg)	150 mg q AM + 200 mg q PM x 7 days/week (2450 mg)
1.21 - 1.35	100 mg x 5 days/week, 200 mg x 2 days/week (900 mg)	250 mg x 7 days/week (1750 mg)	200 mg twice daily x 7 days/week (2800 mg)
1.36 - 1.6	100 mg x 4 days/week, 200 mg x 3 days/week (1000 mg)	300 mg x 7 days/week (2100 mg)	200 mg q AM + 250 mg q PM x 7 days/week (3150 mg)
1.61 - 1.7	100 mg x 2 days/week, 200 mg x 5 days/week (1200 mg)	350 mg x 7 days/week (2450 mg)	250 mg twice daily x 7 days/week (3500 mg)
1.71 - 1.85	100 mg x 1 day/week, 200 mg x 6 days/week (1300 mg)	350 mg x 3 days/week 400 mg x 4 days/week (2650 mg)	250 mg q AM + 300 mg q PM x 7 days/week (3850 mg)
≥ 1.86	200 mg x 7 days/week (1400 mg)	400 mg x 7 days/week (2800 mg)	300 mg twice daily x 7 days /week (4200 mg)

APPENDIX II – CYP3A4 INDUCERS AND INHIBITORS

Strong CYP3A4 Inducers:	
Generic Name	*Common Trade Name*
Carbamazepine	Tegretol
Phenobarbital	Luminal
Phenytoin	Dilantin
Rifampin	Rifadin
St. John's wart	N/A
Systemic dexamethasone	Decadron
Strong* and clinically relevant moderate CYP3A4 Inhibitors:	
Generic Name	*Common Trade Name*
Aprepitant	Emend
Clarithromycin*	Biaxin
Erythromycin	Eryc, EryPed
Fluconazole	Diflucan
Grapefruit and its juice	N/A
Itraconazole*	Sporanox
Ketoconazole*	Nizoral
Voriconazole	VFend
Posaconazole*	Noxafil

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