

Myechild 01

Statistical Analysis Plan

International Randomised Phase III Clinical Trial in Children
with Acute Myeloid Leukaemia -
Incorporating an Embedded Dose Finding Study for
Gemtuzumab Ozogamicin in Combination with Induction
Chemotherapy

Version:1.0
28 July 2016

Sponsor: University of Birmingham

EudraCT number: 2014-005066-30

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Document Control Sheet

Statistical Analysis Plan version:	Reason for update:

1. INTRODUCTION

1.1 Purpose of the Statistical Analysis Plan

This Statistical Analysis Plan (SAP) provides guidelines for the analysis and presentation of results for the interim independent data monitoring committee (DMC) and the final analysis report for the Myechild 01 trial. This plan, along with all other documents relating to the analysis of this trial, will be stored in the 'Statistical Documentation' section of the Trial Master File. The statistical analysis will be carried out by the Trial Statistician.

1.2 Summary of the Trial

Trial Design

An international randomised phase III clinical trial incorporating an embedded dose finding study.

Primary Objectives

In newly diagnosed AML and high risk myelodysplastic syndrome (MDS) (>10% blasts in the bone marrow):

Non-randomised

To establish the optimum tolerated number of 3 mg/m² doses of gemtuzumab ozogamicin (up to a maximum of 3 doses) that can be delivered safely in combination with cytarabine plus mitoxantrone or liposomal daunorubicin in induction

Randomised

1. To compare mitoxantrone (anthracenedione) & cytarabine with liposomal daunorubicin (anthracycline) & cytarabine as induction therapy.
2. To compare a single dose of gemtuzumab ozogamicin 3 mg/m² with the optimum tolerated number of doses of gemtuzumab ozogamicin (identified by the dose-finding study) when combined with induction chemotherapy.
3. To compare two consolidation regimens: high dose cytarabine (HD Ara-C) and fludarabine & cytarabine (FLA) in standard risk patients.
4. To compare the toxicity and efficacy of two haemopoietic stem cell transplant (HSCT) conditioning regimens of different intensity: conventional myeloablative conditioning (MAC) with busulfan/cyclophosphamide and reduced intensity conditioning (RIC) with fludarabine/busulfan.

Secondary Objectives

1. To compare the predictive value of flow and molecular MRD monitoring for relapse risk.
2. To evaluate a number of prognostic factors with a view to defining a Risk Score for children and adolescents with AML

Exploratory Objectives

Exploratory objectives for each sub study are stated in the respective Appendix.

Outcome Measures

Primary Outcome Measures

Gemtuzumab Ozogamicin Dose Finding Study

- The incidence of dose limiting toxicities (DLTs)

Randomisation 1: Induction Randomisation (R1)

- Event-free survival (EFS) from date of randomisation 1 (R1)

Randomisation 2: Gemtuzumab Ozogamicin Randomisation (R2)

- EFS from date of randomisation 2 (R2)

Randomisation 3: Consolidation Randomisation (R3)

- Relapse-free survival (RFS) from date of randomisation 3 (R3)

Randomisation 4: HSCT Conditioning Randomisation (R4)

- Early treatment related adverse events (AEs)
- RFS from date of randomisation 4 (R4)

Secondary outcome measures

Gemtuzumab Ozogamicin Dose Finding Study

- The nature, incidence and severity of AEs.
- Response measured by bone marrow morphology and MRD assessment
- Serum Pharmacokinetic (PK) parameters of gemtuzumab ozogamicin including clearance (CL) and volume of distribution (Vd)

All randomisations

- Complete Remission (CR) (R1 and R2 only)
- Reasons for failure to achieve CR (R1 and R2 only)
- Cumulative incidence of relapse (CIR)
- Death in CR (DCR)
- EFS
- Overall Survival (OS)
- Incidence of toxicities
- Incidence of cardiotoxicity (R1, R2 and R4 only)
- Incidence of bilirubin of grade 3 or higher (R2 and R4 only)
- Incidence of veno-occlusive disease (R2 and R4 only)
- MRD clearance after course 1 and course 2 and MRD negativity post-therapy (R1 and R2 only)
- Time to haematological recovery
- Days in hospital after each course of treatment
- Incidence of mixed chimerism at day 100 post-transplant (R4 only)
- Treatment Related Mortality (TRM) (R4 only)
- Gonadal function at 1 year post-transplant and end of study follow up (R4 only)

Exploratory outcome measures

Sub-study outcomes are stated for each sub-study in the respective Appendix.

Patient Population

Children and young adults up to their 18th birthday with newly diagnosed AML, high risk MDS or isolated myeloid sarcoma (MS).

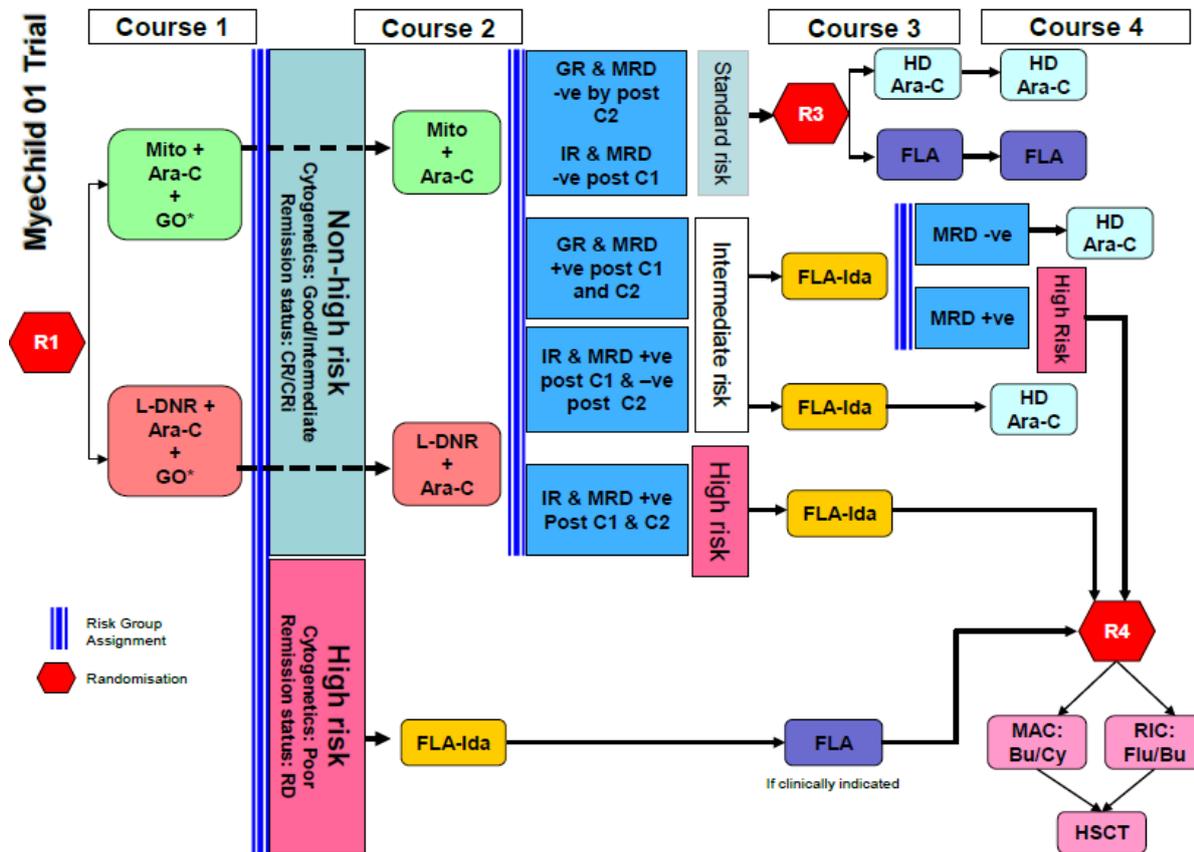
Sample Size

The target recruitment for the trial is up to 700 patients.

Trial Duration

The trial will recruit for approximately 5-6 years, and all patients will be followed up for a minimum of 1 year.

Figure 1: Trial schema prior to the opening of R2.



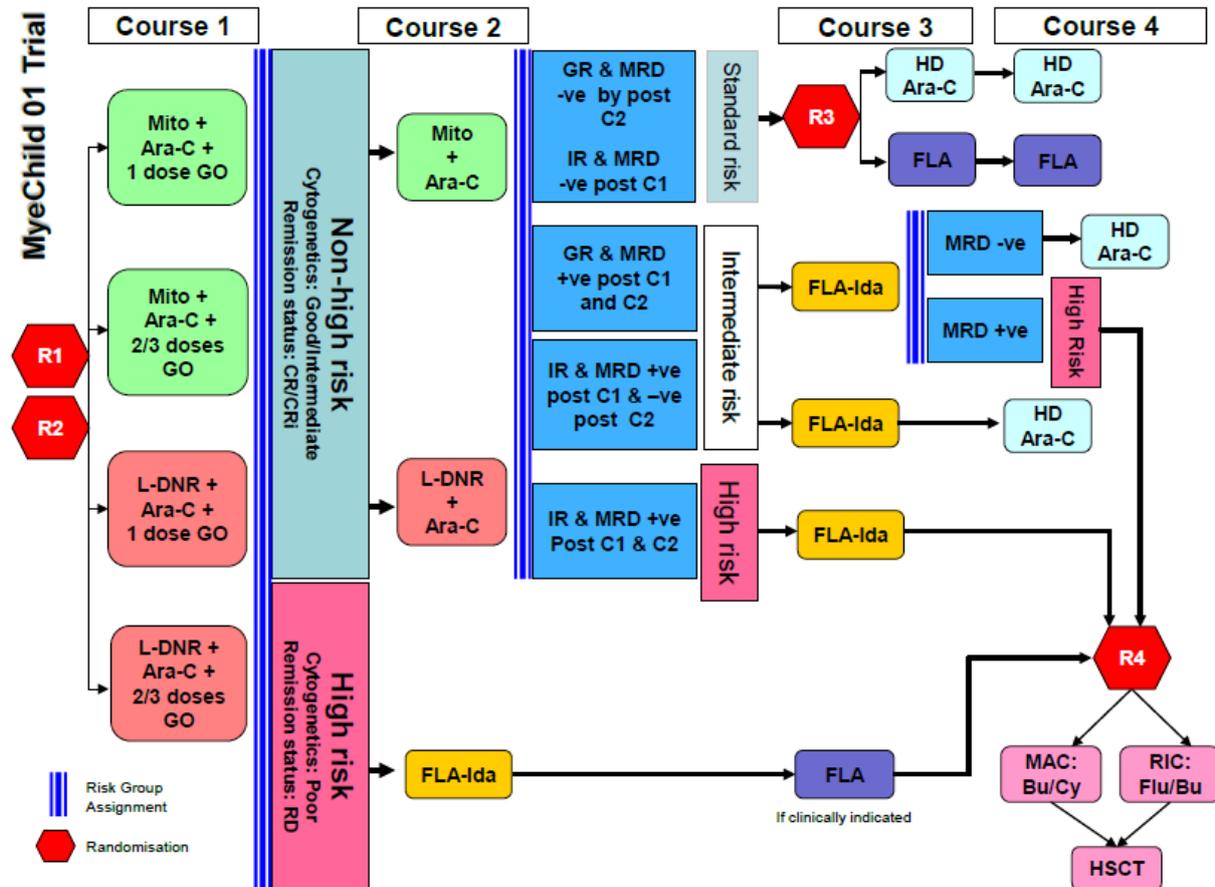
*Patients will only receive GO with induction chemotherapy as part of the embedded gemtuzumab ozogamicin dose finding study (restricted centres), or after the first dose finding cohort has been completed and it has been shown that one dose of GO is safe when given in combination with induction chemotherapy.

Note: where liposomal daunorubicin is unavailable due to national or local restrictions, patients may enter the embedded gemtuzumab ozogamicin dose finding study without participating in R1. Such patients will receive mitoxantrone & cytarabine induction chemotherapy. Such patients will follow the same patient pathway as patients entered at R1 (including risk group assignment) and will be eligible for subsequent randomisations.

Ara-C: Cytarabine
 Bu/Cy: Busulfan & cyclophosphamide
 CR: Complete remission
 CRi: Complete remission with incomplete blood count recovery
 FLA: Fludarabine & cytarabine
 FLA-Ida: Fludarabine, cytarabine & idarubicin
 Flu/Bu: Fludarabine & busulfan
 GO: Gemtuzumab ozogamicin
 GR: Good risk cytogenetics/molecular genetics
 HD-Ara-C: High dose cytarabine
 HSCT: Haemopoietic stem cell transplant
 IR: Intermediate risk cytogenetics
 L-DNR: Liposomal daunorubicin
 MAC: Myeloablative conditioning
 Mito: Mitoxantrone
 MRD: Minimal residual disease
 R1: Randomisation 1: Induction randomisation
 R3: Randomisation 3: Consolidation randomisation

R4: Randomisation 4: Haemopoietic stem cell transplant conditioning randomisation
 RIC: Reduced intensity conditioning
 RD: Resistant disease

Figure 2: Trial schema following the opening of R2



- Ara-C: Cytarabine
- Bu/Cy: Busulfan & cyclophosphamide
- CR: Complete remission
- CRi: Complete remission with incomplete blood count recovery
- FLA: Fludarabine & cytarabine
- FLA-Ida: Fludarabine, cytarabine & idarubicin
- Flu/Bu: Fludarabine & busulfan
- GO: Gemtuzumab ozogamicin
- GR: Good risk cytogenetics/molecular genetics
- HD-Ara-C: High dose cytarabine
- HSCT: Haemopoietic stem cell transplant
- IR: Intermediate risk cytogenetics
- L-DNR: Liposomal daunorubicin
- MAC: Myeloablative conditioning
- Mito: Mitoxantrone
- MRD: Minimal residual disease
- R1: Randomisation 1: Induction randomisation
- R2: Randomisation 2: Gemtuzumab ozogamicin randomisation
- R3: Randomisation 3: Consolidation randomisation
- R4: Randomisation 4: Haemopoietic stem cell transplant conditioning randomisation
- RIC: Reduced intensity conditioning
- RD: Resistant disease

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2. TIMING AND REPORTING OF INTERIM AND FINAL ANALYSES

The main analysis of all the outcome measures will be performed when all patients have been followed up for a minimum of 1 year. This is expected to be roughly 8 years following the opening of the trial.

Accumulating data and analysis from the whole trial will be reported by treatment arm to the DMC on a regular basis. Throughout the duration of the dose finding study interim reports will be produced and presented to the DMC when 50 and 100 of each cohort have been evaluated for DLTs. Following the opening of randomisation 2 interim reports containing data and analysis on all randomisations will be produced and presented to the DMC on a 6 monthly basis.

3. RECRUITMENT AND RANDOMISATION

3.1 Recruitment

The trial began recruiting in June 2016. At each point of analysis the following will be presented:

- Date the snapshot was taken
- Dates when the trial opened and closed (if appropriate) for recruitment
- Recruitment over time (monthly or quarterly) and an average monthly recruitment rate
- Overall recruitment by site and clinician
- Cross-tabulation of recruitment by site – time when sites are not open will be shaded out
- A plot comparing the actual cumulative recruitment to the target recruitment.
- A plot comparing the actual monthly recruitment to the monthly target recruitment.
- The number of patients recruited to each cohort of both the major and minor dose finding studies.

3.2 Randomisation

The number of patients randomised by treatment arm will be presented for each randomisation.

Patients will be randomised to treatments based on minimisation algorithms. All randomisations will be minimised by their age at randomisation, diagnosis and type of disease.

In R1, patients will be randomised 1:1 to receive either mitoxantrone with cytarabine or liposomal daunorubicin with cytarabine. Patients in this randomisation will also be minimised by their WCC and the number of doses of gemtuzumab ozogamicin assigned.

In R2, patients will be randomised 1:1 to receive either 1 dose of gemtuzumab ozogamicin or the optimum tolerated dose of gemtuzumab ozogamicin. Patients in this randomisation will also be minimised by WCC and the allocated treatment in R1.

In R3, patients will be randomised 1:1 to receive either high dose cytarabine or fludarabine & cytarabine.

In R4, patients will be randomised 1:1 to receive either busulfan/cyclophosphamide MAC or fludarabine/busulfan RIC.

Patients in both R3 and R4 will also be minimised by their treatment allocation in R1 and the number of doses of gemtuzumab ozogamicin assigned. Patients in R4 will also be minimised by donor type.

Stratification variables are defined as follows:

R1 treatment assignment:

- Randomised to mitoxantrone and cytarabine
- Randomised to liposomal daunorubicin and cytarabine
- Registered to receive mitoxantrone and cytarabine

Dose allocation of gemtuzumab ozogamicin

- Dose finding 1 dose
- Dose finding 2 doses
- Dose finding 3 doses
- Randomised to 1 dose via R2
- Randomised to 2 doses via R2
- Randomised to 3 doses via R2
- No gemtuzumab ozogamicin allocated

Age at randomisation:

- <12 months
- ≥1 year and <2 years
- ≥2 years and <10 years
- ≥10 years

Diagnosis:

- AML
- High risk MDS
- Isolated MS

Type of disease

- De novo
- Secondary

WCC

- <100 x 10⁹ /l
- ≥100 x 10⁹ /l

Donor type

- Related
- Unrelated
- Cord

3.3 Ineligible Patients

Despite treatment withdrawal, patients will continue to be followed in the trial. All statistical analyses will be carried out on an intention-to-treat (ITT) basis, retaining patients in their randomised treatment groups and including patients who were protocol deviations and ineligible patients. Ineligible patients are defined as those registered or randomised patients who are subsequently found to not meet the eligibility criteria of the trial. The number of ineligible patients and reasons for their ineligibility will be reported; a sensitivity analysis may be conducted and reported if the number of ineligible patients is substantial. Protocol deviations relating to treatment will be reported as part of treatment compliance (section 6).

4. DATA QUALITY

All patient data will be checked in accordance with the trial specific data validation plan version 1.

4.1 Return rate of CRFs

The number of CRFs returned along with the number expected will be presented for the whole trial.

4.2 Length of patient follow up

All patients will be followed up until the final patient has a minimum of 1 year of follow up post treatment.

At each point of analysis reverse Kaplan-Meier plots will be presented to show the length of follow up by treatment arm in each randomised question.

5. TRIAL POPULATION

5.1 Baseline patient characteristics

A descriptive comparison of patient demographics and clinical baseline characteristics at trial entry will be presented for the trial overall as well as for each individual randomised question. These characteristics will include, but will not be limited to the stratification factors.

5.2 Definition of patients population for analysis

All statistical analysis will be carried out on an intention to treat (ITT) basis, retaining patients in their randomised treatment groups and including patients who were protocol deviations or ineligible patients. Where specified analysis will be carried out on a per protocol basis, restricting analysis to patients who have received one dose of the allocated treatment and fulfil the protocol in terms of eligibility.

6. TREATMENT RECEIVED

In order to assess treatment compliance across the trial the following will be presented for patients in the dose finding study:

- The number and proportion of patients who received the correct number of doses
- The number and proportion of doses that were given in accordance with the protocol
- The number and proportion of doses that were modified.
- A cross tabulation of the reasons for modification by the type of modification.

For all randomised questions the following will be presented by allocated treatment and treatment arm:

- The number and proportion of patients who received treatment according to the protocol by course
- The number and proportion of patients whose treatment was modified and the type of modification by course
- A cross tabulation of the reasons for modification by the type of modification by course

7. TOXICITY AND SAFETY ANALYSIS

Toxicity will be assessed by treatment arm within each randomised question as well as within each dose cohort for both the major and minor dose finding study on the safety population (defined in section 5.2).

At each interim analysis (during the course of the dose finding studies) the DMC reports will focus on data from the relevant dose cohorts and will restrict analysis of previous (fully assessed) cohorts that have been deemed safe by the DMC to summary data. For the primary analysis of the dose finding study full analysis will be presented on all dose cohorts.

For all dose cohorts a summary table showing the number of AEs, SAEs and DLTs reported alongside the number of patients registered and details on the treatment they received will be presented.

For relevant dose cohorts:

- The number and proportion of patients experiencing one or more adverse events
- Where applicable, line listings of adverse events.
- Given large enough numbers cross tabulations of the adverse events and number of patients experiencing the type of event by event category, type of event and grade.
- The number and proportion of patients experiencing one or more serious adverse events (SAEs).
- Where applicable, line listings of adverse events.
- Given large enough numbers cross tabulations of SAEs and the number of patients experiencing each SAE by event category, type of event and grade.
- Cross tabulations of SAEs by grade and relatedness.
- Cross tabulations of SAEs by grade and outcome.

Prior to the opening of R2 toxicities will be reported by randomised question. However, once R2 has been activated toxicities will be reported together for R1 and R2 and separately for R3 and R4.

The following will be reported by treatment arm for each randomised question:

- The number and proportion of patients experiencing one or more grade 3 or greater adverse events
- Cross tabulations of grade 3 or greater adverse events and number of patients experiencing the type of event by event category, type of event and treatment arm.
- The number and proportion of patients experiencing one or more serious adverse events (SAEs).
- Tabulations of SAEs and the number of patients experiencing each SAE by event category and type of event.
- Cross tabulations of SAEs by grade and relatedness.
- Cross tabulations of SAEs by grade and outcome.

8. ANALYSIS

8.1 Definition and Calculation of Outcome Measures

Primary outcome measures

Gemtuzumab ozogamicin dose finding study

- Incidence of DLTs from date of randomisation to R1 or registration to the dose finding study to count recovery after course 2 of treatment (up to day 45).

DLTs is defined as any of the following events that are assessed as being possibly, probably or definitely related to any of the induction chemotherapy Investigational Medicinal Products (IMPs):

- Haematological DLT:

- Failure to recover neutrophil count to $1.0 \times 10^9/L$ by day 45 post course 1 or 2 of treatment
- Failure to recover non-transfusion dependent platelet count to $80 \times 10^9/L$ due to documented bone marrow aplasia/hypoplasia.
If failure to recover peripheral count by day 45 after the start of course 1 is due to leukaemic infiltration, this will render the patient non-evaluable for haematological DLT. These patients will however be evaluable for non-haematological DLT.
- Non-haematological DLTs:
 - Death from any cause other than AML
 - VOD
 - Cardiac Disorders: Any grade ≥ 3 reduction of left ventricular systolic function, confirmed by local cardiology review
 - Any grade 3 or higher non-haematological toxicity persisting for >48 hours without resolution to grade ≤ 2 , with the exception of:
 - Alopecia
 - Anorexia
 - Nausea
 - Grade 3 or 4 mucositis that resolves to grade ≤ 2 within 14 days
 - Grade 3 or 4 vomiting that resolves to grade ≤ 2 within 7 days
 - Grade 3 or 4 diarrhoea that resolves to grade ≤ 2 within 7 days
 - Grade 3 or 4 elevation in amylase, lipase, or direct or total bilirubin that is asymptomatic and that returns to grade ≤ 2 elevation within 14 days
 - Grade 4 elevation in hepatic transaminases (aspartate transaminase (AST), alanine transaminase (ALT) or gamma-glutamyl transferase (GGT)) and alkaline phosphatase that returns to grade ≤ 3 elevation within 14 days
 - Grade 3 or 4 fever with neutropenia, with or without infection
 - Grade 3 or 4 infection
 - Grade 3 or 4 electrolyte abnormalities that are not associated with clinical sequelae
 - Grade 3 or 4 hypotension that can be explained by sepsis

Randomisation 1: Induction Randomisation (R1)

- EFS defined as the time from randomisation 1 (R1) to the first event.
An event is defined as failure to achieve Complete remission (CR) (which will be recorded as an event on day 1), relapse, secondary malignancy, or death from any cause. Patient who do not experience an event during the course of the trial will be censored at date last seen.
CR is defined by Creutzig et al as CR or CRi.

Randomisation 2: Gemtuzumab Ozogamicin Randomisation (R2)

- EFS defined as time from randomisation 2 (R2) to the first of failure to achieve CR (recorded as an event on day 1), relapse, secondary malignancy or death from any cause. For patients who do not experience an event during the course of the trial, EFS time will be censored at the date last seen.

Randomisation 3: Consolidation Randomisation (R3)

- RFS defined as time from randomisation 3 (R3) to the first of relapse or death from any cause. For patients who have not experienced an event during the course of the trial, RFS time will be censored at date last seen.

Randomisation 4: HSCT Conditioning Randomisation (R4)

- Early treatment related adverse reactions defined as the incidence by day 100 post-transplant of grade 3-5 toxicity for the following systems using the National Cancer Institute (NCI) Common Terminology Criteria v4:
 - Cardiac (pericardial effusion/Left ventricular systolic dysfunction)
 - Respiratory, thoracic and mediastinal (hypoxia/ [pneumonitis)
 - Gastrointestinal (GI) (diarrhoea/typhilitis/upper and lower GI haemorrhage)
 - Investigations (bilirubin)
 - Renal and Urinary (acute kidney injury/haematuria)
 - Nervous system (seizure)
- RFS defined as time from randomisation 4 (R4) to the first of relapse or death from any cause. For patients who have not experienced an event during the course of the trial, RFS time will be censored at date last seen.

Secondary outcome measures

Gemtuzumab Ozogamicin Dose Finding Study

- The nature, incidence and severity of AEs.
- Responses measured by bone marrow assessment using morphology and MRD assessment between day 21-45 post day 1 of study drugs.

All randomisations

- CR after course 1 and 2 of treatment defined by Creutzig et al as CR or CRi. (R1 and R2 only)
- Reasons for failure to achieve CR after course 1 and 2 of treatment (R1 and R2 only)
- Cumulative incidence of relapse (CIR) defined as time from randomisation to the relevant question to relapse. Death in remission will be treated as a competing risk in this analysis and patients who do not relapse or die within the duration of the trial will be censored at their date last seen.
- Death in CR (DCR) defined as time from randomisation to relevant question to date of death from any cause in patients who have achieved CR. Relapse will be treated as a competing risk in this analysis and patients who do not relapse or die prior to relapse during the trial being censored at date last seen.
- EFS defined as time from randomisation to the relevant question to the first of failure to achieve CR (recorded as an event on day 1), relapse, secondary malignancy or death from any cause. Patients who do not experience an event during the course of the trial will be censored at date last seen.
- OS defined as time from randomisation to the relevant question to death from any cause or date last seen for patients who are alive at the end of the trial.
- Incidence of toxicity within 30 days following the end of treatment. Toxicity defined as a grade 3 or 4 toxicity using the NCI Common Toxicity Criteria v4.
- Incidence of Cardiotoxicity within 30 days following the end of treatment. Cardiotoxicity defined as a fall in fractional shortening to <28% or ejection fraction <55% (R1, R2 and R4 only)
- Incidence of Bilirubin of grade 3 or higher within 30 days following the end of treatment. Bilirubin defined by CTCAE 4 (R2 and R4 only)
- Incidence of VOD within 30 days following the end of treatment. (R2 and R4 only)
- MRD clearance after course 1 and course 2 and MRD negativity post-therapy (R1 and R2 only)
- Time to haematological recovery defined as time from date of randomisation to the relevant question to date of neutrophil recovery to $>1 \times 10^9/l$ and platelet recovery to $>80 \times 10^9/l$.
- Days in hospital per course of treatment
- Incidence of mixed chimerism at day 100 post-transplant. (R4 only)

- Treatment related mortality (TRM) defined as the time between randomisation to R4 and death which is unrelated to the underlying disease and considered related to the transplant procedure. Non transplant related deaths will be treated as a competing risk and patients who are still alive at the end of the trial will be censored at date last seen. (R4 only)
- Gonadal function at 1 year post-transplant and end of study follow up assessed by Tanner Stage, gonadotrophins and serum AMH (females)/inhibin B (males). (R4 only)

8.2 Analyses of Primary outcome measures

Gemtuzumab ozogamicin dose finding study

- Incidence of confirmed DLTs will be tabulated and presented by dose cohort.
- Line listings of all confirmed events will be presented and will include details on grade, relatedness, type of event, clinical significance and relatedness.
- Further line listings of suspected but not confirmed line listings will be presented.

Randomisation 1: Induction Randomisation (R1)

- EFS will be calculated using the method of Kaplan and Meier. A Kaplan Meier plot will be presented with estimates at 1 and 6 year being provided along with 95% confidence intervals (CI). A log rank test will be used to compare the treatment effect of liposomal daunorubicin and cytarabine v. mitoxantrone and cytarabine.
- As a Bayesian approach has been applied to this randomisation posterior probability plots of the treatment effect together with the probability that the true effect is less than 1 will be produced.

Randomisation 2: Gemtuzumab Ozogamicin Randomisation (R2)

- EFS will be calculated using the method of Kaplan and Meier and a log rank test will be used to compare the treatment effect between 1 dose of gemtuzumab ozogamicin and the optimum tolerated number of doses. A Kaplan Meier plot will be presented with estimates at 1 and 6 year being provided along with 95% CIs. A HR along with 95% CI will be produced.

Randomisation 3: Consolidation Randomisation (R3)

- RFS will be calculated using the method of Kaplan and Meier and a log rank test will be used to compare the treatment effect of FLA with HD Ara-C. A Kaplan Meier plot will be presented with estimates at 1 and 6 year being provided along with 95% CIs.
- As a Bayesian approach has been applied to this randomisation posterior probability plots of the treatment effect together with the probability that the true effect is less than 1 will be produced.

Randomisation 4: HSCT Conditioning Randomisation (R4)

- The number of patients who experience one or more episode of toxicity (adverse event grade 3 and above or SAE) will be assessed using a chi-squared test to compare Bu/Cy with Bu/Flu.
- RFS will be calculated using the method of Kaplan and Meier and a log rank test will be used to compare the treatment effect of Bu/Cy vs. Bu/Flu. A Kaplan Meier plot will be presented with estimates at 1 and 6 year being provided along with 95% CIs.
- As a Bayesian approach has been applied to this randomisation posterior probability plots of the treatment effect together with the probability that the true effect is less than 1 will be produced.

8.2.1 Sample Size Calculations

Randomisation 1 is between liposomal daunorubicin and mitoxantrone during induction. With 700 patients, 280 events are anticipated (based on EFS of 54% in AML12). If the observed HR was 0.89 or better in favour of a particular treatment, we could be >80% sure that this was indeed the more effective treatment. If the true HR was 0.8 in favour of a particular treatment, then we would have an 81% chance of observing this HR or better.

R2 is a comparison between 1 dose of gemtuzumab ozogamicin and the optimum tolerated number of doses of gemtuzumab ozogamicin and will be analysed using a classical hypothesis testing approach. As this randomisation will not open initially (i.e., once the optimum tolerated number of doses has been identified), only 550 patients will be eligible for this randomisation. If the higher dose of gemtuzumab ozogamicin leads to an increase in EFS from 60% to 70%, then 550 patients will provide >84% power to detect this difference on a 2-sided alpha of 0.15. Based on this it is expected that around 188 events will occur. The size of the study is based on a large treatment effect (HR 0.7) and a relaxed alpha, and we will use results from paediatric patients in other ongoing studies (e.g. AML17) to provide extra power to reliably detect smaller effects via a Meta-analysis.

R3 is a comparison between two consolidation therapies with known activity. An assumption is that only 60% of patients enrolled to the study qualify for this randomisation. Given this assumption and a historical event rate of 33%, 140 events are expected. This would mean that an observed HR of 0.86 or less would be required to be 81% sure that a particular treatment was better. If the true HR is 0.8 in favour of a particular treatment, then we have a 66% chance of achieving this level of certainty, but a 90% chance that the better treatment will be observed to perform best (HR<1).

R4 is a comparison between the current standard MAC and RIC regimens for SCT. The primary outcomes will be early regimen-related toxicity and DFS. We expect 150 patients to enter R4. We expect the severe toxicity rate (proportion of grade 3/4/5 toxicity) in the MAC arm to be 40%. In order for RIC to be considered worthwhile in this patient population, it would have to have a reduced toxicity and therefore a standard hypothesis testing approach will be used. If the true toxicity rate in the RIC arm is 20%, then this study will have 85% power to demonstrate the difference with a 2-sided alpha of 0.15. A relaxed alpha is necessary in this study as a more conventional alpha (0.05) would require an unfeasibly large number of patients. For RFS, a posterior probability plot of the underlying treatment effect will be produced as before. This analysis will not have great reliability. If uncertainty remains, this randomisation will be continued in the next trial.

8.3 Analysis of Secondary Outcome Measures

Each outcome will be analysed for each randomisation in its own right. All outcomes, unless otherwise stated, apply to all randomisations. No secondary outcomes have been powered.

- **CR rates (R1 and R2 only):** A table showing the number and proportion of patients with a CR in each treatment arm will be presented and a chi squared test will be used to compare treatment arms. Odds ratios (OR) and 95% CIs will be presented.
- **Reasons for failure to achieve CR (R1 and R2 only):** A table categorising the reasons for failure to achieve CR will be presented by treatment arm and a chi squared test will be used to compare between treatment arms. ORs and 95% CIs will be presented.
- **Cumulative incidence of relapse:** A cumulative incidence curve will be presented with estimates and 95% CIs at 1 and 6 years. Grey's test will be used to compare between the treatment arms.
- **Death in CR:** A cumulative incidence curve will be presented with estimates and 95% CIs at 1 and 6 years. Grey's test will be used to compare between the treatment arms.
- **EFS:** Kaplan Meier curves will be presented with estimates and 95% CIs at 1 and 6 years. Log rank tests will be used to compare between treatment arms.
- **OS:** Kaplan Meier curves will be presented with estimates and 95% CIs at 1 and 6 years. Log rank tests will be used to compare between treatment arms.
- **Incidence of toxicity:** The number and proportion of patients who experience one or more episode of toxicity (adverse event grade 3 and above or SAE) will be tabulated

by treatment arm and assessed using a chi-squared test. Odds ratios and 95% CIs will also be presented.

- **Incidence of Cardiotoxicity (R1, R2 and R4 only):** The number and proportion of patients who have experienced one or more episode of cardiotoxicity will be presented by treatment arm and compared using a chi squared test. Odds ratios and 95% CIs will be presented.
- **Incidence of Bilirubin (R2 and R4 only):** The number and proportion of patients who have experienced one or more episode of bilirubin will be tabulated by treatment arm and compared using a chi squared test. Odds ratios and 95% CIs will be presented.
- **Incidence of VOD (R2 and R4 only):** The number of patients who have experienced one or more episode of VOD will be tabulated by treatment arm and compared using a chi squared test. Odds ratios and 95% CIs will be presented.
- **MRD clearance after course 1 and 2 and MRD negativity post-therapy (R1 and R2 only):** The number and proportion of patients with MRD clearance/negativity will be tabulated by treatment arm and time point. Chi squared tests will be used to compare treatment arms at each time point and odds ratios with 95% CIs will be presented.
- **Time to haematological recovery:** Kaplan Meier curve will be presented with estimates and 95% CIs at 2, 4 and 6 weeks. Log rank tests will be used to compare between treatment arms.
- **Days in hospital per course of treatment:** The median number of days in hospital alongside the interquartile range will be presented by treatment arm for each course of treatment. The Mann-Whitney U test will be used to compare the two arms for each course of treatment.
- **Incidence of mixed chimerism (R4 only):** The number and proportion of patients who have incidence of mixed chimerism will be tabulated by treatment arm and will be compared using a chi squared test. An odds ratio and 95% CI will be presented.
- **Treatment related mortality (R4 only):** A cumulative incidence curve will be presented with estimates and 95% CIs at 1 and 2 years. Grey's test will be used to compare between the treatment arms.
- **Gonadal function assessed by Tanner Stage, gonadotrophins and serum AMH (females)/inhibin B (males). (R4 only):** Categorical variables will be presented as the number and proportion in each category at 1 year post transplant and end of study follow up. Chi squared tests will be used to compare between treatment arms at both time points with odds ratios and 95% confidence intervals will be presented. Continuous variables will be presented using means and sds at 1 year post transplant and end of study follow up given the relevant assumptions hold. T-tests will be used to compare between treatment arms at both time points. Given the relevant assumptions are shown to be invalid medians and interquartile ranges will be presented and the Mann-Whitney U test will be used to compare between treatment arms.

8.4 Additional Analyses

Additional multivariable analysis of each primary outcome will be conducted to account for the other randomisations in the trial.

R1 EFS:

Exploratory multivariable cox regressions will be used to compare EFS between the two arms adjusting for the dose of gemtuzumab ozogamicin allocated in either the dose finding study or throughout randomisation 2 as specified in section 3.2.

R2 EFS:

Exploratory multivariable cox regressions will be used to compare EFS between the two arms adjusting for the assigned treatment in R1 as specified in section 3.2.

R3 RFS:

Exploratory multivariable cox regressions will be used to compare RFS between the two arms adjusting for both the assigned treatment in R1 and the dose of gemtuzumab ozogamicin received as specified in section 3.2.

R4 toxicity:

- Exploratory multivariable logistic regression will be used to compare the incidence of toxicities between the treatment arms while adjusting for both the assigned treatment in R1 and the dose of gemtuzumab ozogamicin received as specified in section 3.2.

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R4 DFS:

- Exploratory multivariable cox regressions will be used to compare RFS between the two arms adjusting for both the assigned treatment in R1 and the dose of gemtuzumab ozogamicin received as specified in section 3.2.

8.5 Subgroup Analysis

Subgroup analysis will be carried out on the primary outcome for WCC, age, type of disease and cytogenetics/molecular genetic risk group for all randomisations. Further sub group analysis of the primary outcome will be carried out for donor type in R4 patients. Heterogeneity tests will also be performed.

9. STATISTICAL SOFTWARE

All analysis will be carried out in appropriate statistical software, SAS 9.4, Stata 14 or R 3.2.2. Later versions of this software may also be used.

10. STORAGE AND ARCHIVING

Raw data will be stored in the following location

T:\Trials Work\CCTT\Leukaemia\MyeChild\Analysis\Data

Data snapshot names will include the date the snapshot was taken and some indication of what it is intended to be used for e.g. DMC