

MEDICAL RESEARCH COUNCIL  
WORKING PARTIES ON LEUKAEMIA IN ADULTS AND CHILDREN  
ACUTE MYELOID LEUKAEMIA TRIAL 15

**PROTOCOL FOR PATIENTS AGED UNDER 60  
(Trial Reference ISRCTN 17161961)**

Through the use of an efficient factorial design, AML15 will evaluate several relevant therapeutic questions in acute myeloid leukaemia (AML) as defined by WHO. The trial is open to all patients aged less than 60 years, whether adults or children, and also to patients aged 60 years or over for whom intensive therapy is considered appropriate. At least 2500 patients will be recruited. For patients who do not have the Acute Promyelocytic (APL) subtype, an induction randomisation will compare the standard ADE and DA regimens. Patients who have a FLT3 mutation at diagnosis will be randomised to combine, or not, a FLT3 inhibitor after each course of the allocated induction and consolidation chemotherapy. A consolidation randomisation will compare MRC chemotherapy (MACE + MidAC) with high-dose Ara-C, at doses of either 1.5 g/m<sup>2</sup> or 3.0 g/m<sup>2</sup>. The 4 versus 5 courses randomisation from AML12 will continue in patients under 45 years, but the fifth course will be Ara-C at a dose of 1.5 g/m<sup>2</sup>. The role of the immunoconjugate Mylotarg will be evaluated in consolidation (course 3) in patients who do not enter the FLT3 inhibitor randomisation. The role of allogeneic transplant, either standard or “mini”, will be assessed in standard and poor risk patients. Poor risk patients, with resistant disease after Course 1 or adverse genetics, may either continue in AML15 or may be entered into the current NCRI high risk trial when available.

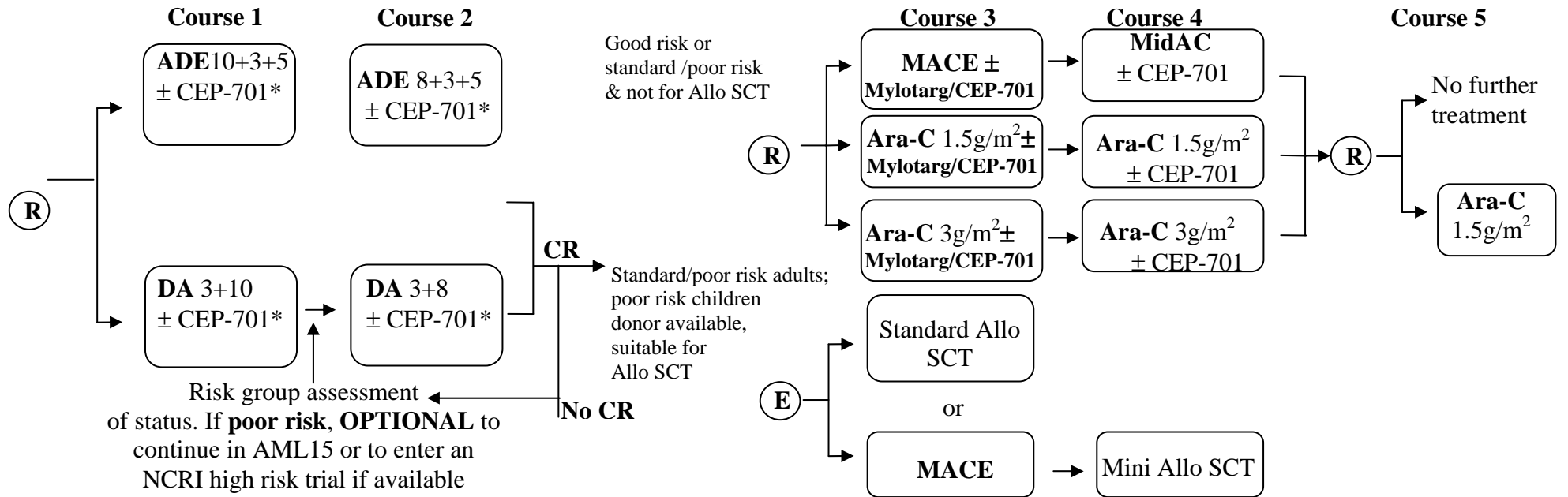
Adult patients with APL will be treated with the “Spanish” approach. APL patients will be eligible for the consolidation Mylotarg randomisation. Children will only enter selected randomisations within the trial which are described in the Paediatric Variations section of the protocol. There are about 700 cases of AML each year at ages 0-59 in the British Isles alone. About 400 adult patients and 100 children were entered annually into AML12, so with a continuation of accrual at this, or a higher level, clear evidence on the relative benefits of the therapeutic options being tested in AML15 will be obtained in just a few years. This information will contribute to the continuing improvement of the treatment available to many future patients with AML.

This protocol is intended to describe a Medical Research Council collaborative trial in acute myeloid leukaemia in adults and children, which is being undertaken by the NCRI Haematological Oncology Study Group under the sponsorship of Cardiff University, and to provide information about procedures for the entry, treatment and follow-up of patients. It is not intended that this protocol should be used as an aide-memoir or guide for the treatment of other patients. Every care has been taken in its drafting, but corrections or amendments may be necessary. Before entering patients into the trial, clinicians must ensure that the trial protocol has received clearance from their Local Research Ethics Committee. During the course of this 5-year trial, not all randomisation options will be open at all times and some additional options may be included by protocol amendment.

**Clinicians are asked to read the whole protocol before  
commencing treatment**

**AML15 Protocol Flow Chart 1 - Trial Overview** (Please refer to the back of the protocol for more detailed flow diagrams)

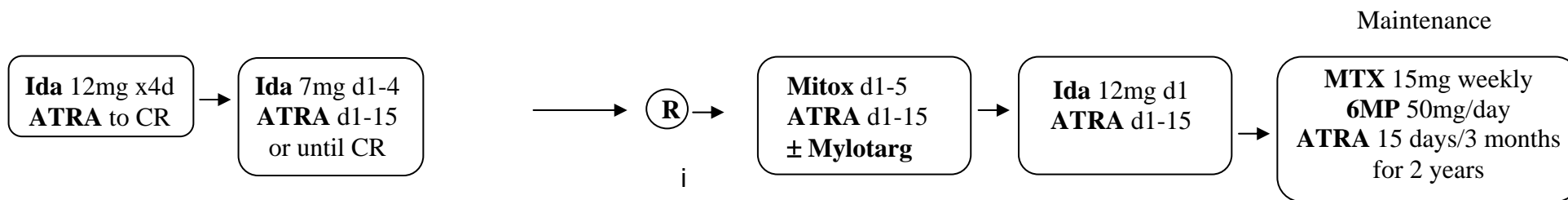
**AML patients, other than APL**



\* Patients with a FLT3 mutation are eligible for the CEP-701 randomisation after course 1; patients who do not enter the CEP-701 randomisation after course 1 are eligible for the mylotarg consolidation randomisation.

**(R)** Randomise  
**(E)** Elect

**Spanish Treatment**



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09.00-17.00 hours, Monday to Friday (except bank holidays)

### 24 hour internet randomisation and data entry:

<https://www.trials.bham.ac.uk/aml15>

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## 1 ETHICAL CONSIDERATIONS

The AML15 Trial Protocol has been approved by the Welsh Multicentre Research Ethics Committee (MREC) and must also be approved by the Local Research Ethics Committee (LREC) at each centre before patients are entered. A copy of a centre's LREC approval must be lodged with the Trial Office at BCTU before entry of patients can commence at that centre. Centres are required to go through a registration process with the Trial Office before recruitment is started.

The right of a patient to refuse to participate in the trial without giving reasons must be respected. After the patient has entered the trial, the clinician is free to give alternative treatment to that specified in the protocol at any stage if he/she feels it to be in the patient's best interest, and the reason for doing so should be recorded. Similarly, the patient must remain free to withdraw at any time from protocol treatment without giving reasons and without prejudicing any further treatment. All patients who come off protocol therapy for whatever reason will still need to remain within the study for the purposes of follow-up and data analysis.

The AML15 trial will be conducted in accordance with the Medical Research Council's Guidelines for Good Clinical Practice in Clinical Trials (a copy of these may be obtained from the MRC or from the Trial Office).

## 2 OBJECTIVES

The AML15 trial has two separate parts:

- i. For patients with acute myeloid leukaemia (AML), other than acute promyelocytic leukaemia (APL), as defined by the WHO Classification (2001) (Appendix A).
- ii. For adults with acute promyelocytic leukaemia (APL).

**Children with APL and children with Down Syndrome and AML are not eligible for AML15.**

The objectives for each of these components are summarised below.

### 2.1 Therapeutic questions for patients with non-APL AML

For patients with acute myeloid leukaemia (AML) the aims of the AML15 trial are:

- To compare two induction schedules (namely **ADE** and **DA**).
- To assess the value of the FLT3 inhibitor CEP-701 for patients with a FLT3 mutation
- To compare the standard MRC consolidation chemotherapy (i.e. **MACE + MidAC**) versus **high-dose Ara-C**.
- For those allocated to high-dose Ara-C to compare high dose Ara-C during consolidation (see above) at two different doses (**1.5 g/m<sup>2</sup>** versus **3.0 g/m<sup>2</sup>**).
- To assess the value of **Mylotarg** during **consolidation**.
- To compare **four** versus **five** courses of treatment in total in patients under 45 years (where the final course is Ara-C at a dose of 1.5 g/m<sup>2</sup>).

- In standard and poor risk adults and poor risk children, to evaluate, by means of a genetic randomisation (i.e. the presence or absence of a matched donor), the value of **allogeneic stem cell transplantation** (SCT), whether standard allogeneic (allo-SCT) or non-myeloablative “mini” allogeneic (mini-SCT).

## 2.2 Therapeutic questions for patients with APL

For patients with APL the aims of the AML15 trial are:

- Adults with APL will be treated with the **Spanish approach** (based on anthracyclines with maintenance therapy).
- In adults to assess the value of **Mylotarg** during **consolidation** (i.e. with Course 3).

## 2.3 Endpoints

The main endpoints for each comparison will be:

- Complete remission (CR) achievement and reasons for failure (for induction questions).
- Duration of remission, relapse rates and deaths in first CR.
- Overall survival.
- Toxicity, both haematological and non-haematological, and quality of life (QoL).
- Supportive care requirements (and other aspects of health economics).

## 2.4 Subsidiary objectives

Blood and bone marrow will be required at diagnosis, during remission and at relapse to evaluate the therapeutic relevance of morphological, cytogenetic, molecular-genetic and immunophenotypic assessments, with particular respect to:

- The relevance of the presence of a cytogenetic abnormality in the bone marrow of patients in morphological remission.
- The relevance of the molecular detection of residual disease in patients in morphological remission.
- The prospective validation of the incidence and prognostic value of FLT3 mutations.
- The relevance of CD33 expression and resistance protein expression to the effectiveness of Mylotarg therapy.
- To correlate the serum level of anti-FLT3 activity and the extent of dephosphorylation of the FLT3 receptor with response.
- Future research questions.

# 3 TRIAL DESIGN

AML15 is a randomised controlled Phase III trial for patients with AML that uses a factorial design for maximum efficiency. The design may, at first sight, appear complicated. However, if

the trial is broken down into separate sections, each phase is straightforward and should be readily understandable to both clinicians and patients:

AML (other than APL):

- A. Induction phase: one randomisation (three arms in total)
- B FLT3 inhibition for patients with FLT3 mutations: one randomisation (two arms).
- C Initial consolidation phase: two randomisations (3x2 factorial design)
- D. Late consolidation phase: one randomisation (two arms)

APL:

- A Consolidation phase: One randomisation (two arms) (for adults only)

Looked at this way, AML15 is no more complicated than AML12, to which over 400 consultants from about 200 hospitals contributed over 3400 patients.

### 3.1 AML (other than APL)

There are five randomised comparisons within the trial:

- At diagnosis: (i) ADE versus DA
- End of Course 1 (ii) FLT3 inhibitor (CEP-701) versus not
- Before Course 3: (iii) MRC consolidation versus high-dose Ara-C (at two doses levels in adults; at one dose level in children)  
(iv) Mylotarg versus not in consolidation
- After Course 4: (v) 4 versus 5 courses of therapy in total for patients under 45 years of age

In standard and poor risk adults and poor risk children, the role of standard allogeneic SCT will be assessed by means of a genetic randomisation (i.e. donor versus no donor comparison). In adults the role of 'mini' allogeneic SCT will be assessed.

Full details of the rationale for these comparisons, progress through the trial and treatment can be found in the relevant sections of the protocol, but are summarised below (and in the flow diagrams at the front and back of the protocol):

1. At diagnosis: randomise between ADE and DA as induction therapy,.  
The three induction treatment arms will therefore be:
  - Arm A Two courses of ADE
  - Arm B Two courses of DA
2. By the end of the first course of induction chemotherapy, the FLT3 mutation status of each non-APL patient should be available. Patients with a FLT3 mutation can then be randomised to start FLT3 inhibition or not (see Section 10).

3. After haematological recovery from Course 1 in all patients, assess remission status and assign patient to the appropriate risk group (see Section 11.2):
  - If good or standard risk, continue with AML15 protocol.
  - If poor risk, patient should continue with AML15 protocol or may be entered into a current NCRI high risk AML trial (when available).
  - Carry out tissue typing to ascertain donor availability (in patients classified as poor risk this should include unrelated donor).
  - Blood and bone marrow should be sent for minimal residual disease monitoring (see Section 18).

Administer the second course of the allocated AML15 induction therapy.

4. After Course 2, assess remission status:
  - If in CR, continue with AML15 protocol.
  - If not in CR, patient can continue in the AML15 trial or can enter a current NCRI high risk trial (when available).
  - Patients who are suitable for minimal residual disease monitoring should have a bone marrow assessment (see Section 18).
5. If the patient is standard or poor risk (for adults) or poor risk (for children) and has a donor available – the search should be initiated as soon as possible after diagnosis – and the patient is considered suitable for an allogeneic Stem Cell Transplant (allo-SCT) they should receive either:
  - (i) Standard allo-SCT as Course 3, if considered young and fit enough for the procedure. Standard allo-SCT beyond age 45 years is not recommended.
  - (ii) A course of MACE followed by non-ablative “mini” allo-SCT as Course 4, if not considered suitable for standard allo-SCT.
6. If patient is intended not to receive SCT, randomise between:
  - (i) MRC consolidation chemotherapy (MACE plus MidAC) versus high-dose Ara-C (with a sub-randomisation between Ara-C at 1.5 g/m<sup>2</sup> versus 3.0 g/m<sup>2</sup> dose levels)
  - (ii) Mylotarg versus not with Course 3.

The six main consolidation treatment arms will therefore be:

Arm D	MACE followed by MidAC (with no Mylotarg)
Arm E	MACE followed by MidAC (with Mylotarg in Course 3)
Arm F	Ara-C at 1.5 g/m <sup>2</sup> for two courses (with no Mylotarg)
Arm G	Ara-C at 1.5 g/m <sup>2</sup> for two courses (with Mylotarg in Course 3)
Arm H	Ara-C at 3.0 g/m <sup>2</sup> for two courses (with no Mylotarg)
Arm I	Ara-C at 3.0 g/m <sup>2</sup> for two courses (with Mylotarg in Course 3)

This is a factorial 3x2 design, so clinicians may elect to undertake only one or other of the two randomisations, though it is intended that most will undertake both.

Patients who have entered the FLT3 inhibition randomisation are **not eligible to enter the Mylotarg consolidation randomisation**. They should be randomised between arms D, F and H and, if allocated, will receive CEP-701 after each course of consolidation therapy as described in section 13.

**Children are eligible for the Mylotarg consolidation randomisation but will only be randomised between MRC consolidation and Ara-C at 3.0 g/m<sup>2</sup> (i.e. the options will be Arms D, E, H, and I).**

7. After Course 4, for patients under 45 years, randomise between:

4 versus 5 courses of therapy in total - the fifth course will be Ara-C at 1.5 g/m<sup>2</sup>.

**No patient will receive CEP-701 treatment after course 5.**

### 3.2 Acute Promyelocytic Leukaemia (APL)

There is one randomisation within the trial (adults only):

Before Course 3: Mylotarg versus not with Course 3.

Full details of the rationale for this comparison, progress through the trial and treatment can be found in the relevant sections of the protocol, but are summarised below (and in the flow diagrams on the front and back covers):

1. At diagnosis: Adults only will be treated by the Spanish approach (4 courses of less intensive anthracycline based therapy with maintenance treatment). All APL patients receive all-trans retinoic acid (ATRA) until remission is achieved.
2. After Course 1, assess remission status (see Section 11.1).
3. After Course 2, reassess remission status:
  - If CR, continue with AML15 protocol.
  - If not in morphological CR, the patient should be treated as high risk APL as set out in Section 17.11.
  - Bone marrow should be sent for MRD monitoring.
4. Before Course 3, randomise to receive Mylotarg or not with Course 3.
5. After Course 4, regular assessment of minimal residual disease should take place (see Section 18).

**Children with APL are not eligible for randomisation in AML15.**

## 4 JUSTIFICATION OF TRIAL DESIGN AND TREATMENT SCHEDULES

### 4.1 AML (excluding APL)

#### 4.1.1 Experience from AML12

The rationale behind the design of AML15 and the treatment choices is based on the experience gained from AML12 and earlier MRC AML trials.

It is clear that AML12 was a highly successful trial with recruitment at an unprecedented level, a high overall CR rate of 84%, and survival which is significantly improved compared with the previous MRC AML10 trial and which compares very favourably with any international protocol. Thus, the therapy used in AML12 forms the backbone of the AML15 trial.

#### 4.1.2 Induction therapy

The MRC AML10 trial randomised more than 1800 patients between the standard DAT regimen and the etoposide containing regimen ADE for the first two courses<sup>(1)</sup>. CR rate, disease-free survival or overall survival were very similar with the two schedules. There was slightly greater non-haemopoietic toxicity with ADE (alopecia, mucositis, nausea, diarrhoea). Despite the greater toxicity of etoposide, it is considered appropriate to use ADE as the standard induction arm of AML15, because of concerns over the liver toxicity of thioguanine (the “T” component of DAT) and doubts over its long-term availability. The DA schedule is widely used around the world as standard of care, and has been found to be more feasible when combined with Mylotarg.

The randomisation between daunorubicin and mitoxantrone in the first two courses of AML12 (i.e. ADE versus MAE) accrued over 1900 patients. There were no significant differences in CR rate, disease-free survival or overall survival between the two schedules, but mitoxantrone caused significantly longer neutropenia and thrombocytopenia after course 2 (an effect which also carries over into later courses), thereby leading to substantially increased resource usage and fewer patients being able to undergo the second randomisation<sup>(2)</sup>. Thus, daunorubicin will be used in the standard arm of AML15 and the standard induction regimen will be ADE.

A number of trials have compared different doses of Ara-C during induction<sup>(3-7)</sup>. The CALGB 7921 and 8321 trials showed that relatively modest increases in Ara-C dose (100 mg/m<sup>2</sup>/d for 10 versus 7 days and 200 mg/m<sup>2</sup>/d versus 100 mg/m<sup>2</sup>/d respectively) produced substantial reductions in the number of patients failing to achieve CR due to resistant disease (19% versus 29%, p=0.003). The small UCLA trial showed a similar 10% reduction in resistant disease with 1 g/m<sup>2</sup>/d<sup>(5)</sup>. Neither the ALSG study of 6 g/m<sup>2</sup>/d for 4 days versus 100 mg/m<sup>2</sup>/d for 7 days<sup>(6)</sup> nor the SWOG study<sup>(7)</sup> of 4 g/m<sup>2</sup>/d for 6 days versus 200 mg/m<sup>2</sup>/d for 7 days demonstrated higher CR rates with the higher dose of Ara-C, but in both trials the relapse risk was reduced with the higher dose (p=0.002 and p=0.04 respectively). Thus, there is some evidence that increasing the dose of Ara-C both reduces the number of failures due to resistant disease and the subsequent risk of relapse. There is no clear evidence that higher doses of Ara-C (greater than 1 g/m<sup>2</sup>/d) are more effective than intermediate doses. These data provided the rationale for testing an intermediate dose of Ara-C (200 mg bd) versus a standard dose (100 mg bd) in the H-DAT v. S-DAT randomisation in the second half of the MRC AML12 trial. 1400 patients were

randomised and a preliminary analysis suggests that there are no important differences in efficacy, but there may be some more toxicity in the H-DAT arm. For these reasons the standard 100 mg/m<sup>2</sup> bd dose of Ara-C for courses 1 and 2 of AML15 will be used in the ADE and DA regimens.

#### 4.1.3 FLT3 Inhibition

A number of prognostic factors have been identified for CR and relapse. Among the adverse prognostic factors is an fms-like tyrosine kinase 3 (FLT3) activating mutation. These mutations spontaneously initiate ligand-independent autophosphorylation of the receptor, stimulating proliferation of AML cells<sup>(8)</sup>. Two types of FLT3 activating mutations have been identified in patients: an internal tandem duplication (ITD) and a point mutation, usually at aspartate 835<sup>(9, 10)</sup>. The presence of FLT3 ITD mutations has been shown to be associated with a decreased remission induction rate and poorer outcome in paediatric AML<sup>(11)</sup> and a higher rate of relapse and poorer overall survival in adult AML<sup>(12)</sup>.

Studies in animals suggest that inhibition of mutated FLT3 improves response to chemotherapy and/or overall survival. CEP-701 is a potent FLT3 inhibitor and induces a cytotoxic-like effect on both FLT3/ITD transfected cells and primary leukaemic myeloblasts in patients with AML with the FLT3/ITD mutation. In a mouse model of FLT3/ITD leukaemia, treatment with CEP-701 significantly prolonged survival<sup>(13)</sup>. In a recently completed Phase II clinical trial in patients with relapsed disease, CEP-701 at doses of 60 and 80 mg bd was associated with transient decreases in the number of peripheral AML myeloblasts<sup>(14)</sup>. A similar response was seen in a UK Phase II study in untreated older patients<sup>(15)</sup>. A pharmacokinetic/ pharmacodynamic analysis indicated that this anti-leukaemic activity required a high degree of inhibition of the target kinase, FLT3<sup>(16)</sup>. *In vitro* studies have shown that AML cells that survive chemotherapy treatments remain responsive to CEP-701 (Levis, *personal communication*, 2003). There is *in vitro* data to suggest that combining CEP-701 with Ara-C has a synergistic effect<sup>(17)</sup>. This has led to an ongoing randomised study in relapsed AML which compares high dose Ara-C with and without sequential CEP-701.<sup>(18)</sup> An analysis performed after 24 patients had combination arm and 6 responses in the Ara-C alone arm (P Brown, *personal communication*). The objective of the current study is to determine whether CEP-701, given in sequence with standard chemotherapy, can reduce the risk of relapse and improve survival in patients who have a FLT3 mutation.

Background information on the nonclinical pharmacology and pharmacokinetics, toxicology, and clinical experience both in healthy subjects and patients with cancer is given in Appendix C. Of potential clinical relevance is the theoretical interaction with azole antifungal agents which use CYP3A4 in metabolism, with the potential effect of increasing blood levels of CEP-701. The extent to which this happens and whether it is clinically relevant is not known. As part of the assessment of CEP-701 in this trial, blood levels of free CEP-701 and azole blood levels will be measured on day 14 of each course of CEP-701 treatment

## 4.2 Mylotarg (Gemtuzumab Ozogamicin)

The MRC AML10 trial suggested that “more treatment is better”<sup>(19)</sup>. However, adding still more conventional chemotherapy is difficult because of toxicity. The possibility of targeting chemotherapy using the immunoconjugate Mylotarg has become a realistic option<sup>(20,21)</sup>. This agent has been developed by Wyeth-Ayerst Research and comprises a humanised anti-CD33 monoclonal antibody which is linked to Calicheamicin, a potent anti-tumour agent. As a single agent it achieved a remission rate of 34% in relapsed patients<sup>(22)</sup>. However, of greater interest is that it has, by comparison with chemotherapy, a favourable toxicity profile. Conceptually it offers more anti-leukaemic treatment with less ‘cost’ in terms of toxicity.

As a single agent Mylotarg results in cytopenias to a degree equivalent to chemotherapy. In the post-transplant setting, veno-occlusive disease (VOD) has been reported in approximately 4% of cases and a transient transaminitis in 20% of cases. In the AML15 trial to date, over 1000 patients have been randomised to receive mylotarg or not in induction. The possibility of benefit will continue to be assessed in consolidation (course 3) in patients who have not entered the FLT3 inhibition randomisation.

## 4.3 Risk Group Designation

Univariate and multivariate analyses of AML10 indicated that two parameters were of very highly significant prognostic importance in relation to relapse — namely, cytogenetic group and percentage of blasts in the bone marrow after Course 1<sup>(23)</sup>. This finding has been confirmed in AML12 and the risk group definitions are now well-validated.

Favourable genetic abnormalities are t(8;21) or inv(16), while adverse abnormalities are -5, -7, del(5q), abn(3q), t(9;22) or complex karyotype (5 or more abnormalities). All other abnormalities and normal karyotypes are classified as intermediate risk<sup>(24)</sup>. Occasionally, a favourable abnormality may occur in conjunction with other genetic changes and, in such cases, the patient should still be regarded as having favourable genetics. In AML12, the 5 year survival of patients in each of these three groups is 73% in favourable, 44% in intermediate and 17% in adverse genetic groups. Patients may be placed in the favourable or adverse groups if relevant genetic abnormalities are detected by any method (e.g. cytogenetics, PCR, FISH).

Patients also fall into three status groups based on the bone marrow assessment after Course 1 (see Section 11.1 for full definitions).

1. Complete remission (CR) — i.e. <5% leukaemic cells by morphology.
2. Partial remission (PR) — i.e. 5-15% leukaemic cells by morphology.
3. Resistant disease (RD) — i.e. >15% leukaemic cells by morphology.

In AML12, patients with resistant disease have a very poor outlook (5 year survival of 26%), while partial remitters do only slightly worse than complete remitters (5 year survivals of 44% and 56% respectively).

There is a correlation between genetics and bone marrow status, i.e. patients with favourable genetics tend to be in CR or PR after one course, while patients with adverse genetics tend to have resistant disease. Patients with favourable genetics who have resistant disease after Course 1 appear to have a good prognosis, while patients with adverse genetics who are in CR or PR after Course 1 tend to do badly (though the numbers in both of these groups are small).

Therefore, genetics at diagnosis and bone marrow status after the first course can be combined to give three risk groups defined as follows:

- Good risk:** Any patient with favourable genetic abnormalities — i.e. t(8;21), inv(16)/t(16;16), including those molecularly detected — irrespective of marrow status after Course 1 or the presence of other genetic abnormalities.
- Standard risk:** Any patient not in either good risk or poor risk groups — i.e. neither favourable nor adverse genetic abnormalities and not more than 15% blasts in the bone marrow after Course 1.
- Poor risk:** Any patient with more than 15% blasts in the bone marrow performed after Course 1 or with adverse genetic abnormalities — -5, -7, del(5q), abn(3q), t(9;22), complex (≥5 abnormalities) — and without favourable genetic abnormalities.

Data from AML12 suggest that about half of patients will fall into the standard risk group, with about 30% in the good risk group and 20% in the poor risk group. The survival and relapse rates of these three groups at 5 years are shown below:

Risk group	Survival	Relapse rate
Good	76%	25%
Standard	48%	52%
Poor	21%	73%

#### 4.4 Consolidation Chemotherapy

Since most younger patients with AML enter complete remission, a major therapeutic issue is to prevent relapse. We have established again the importance of cytogenetics in predicting relapse<sup>(24)</sup> which has been incorporated into a simple risk score<sup>(23)</sup>. This has been used and prospectively validated in the recent AML12 Trial<sup>(2)</sup>. Using this risk directed approach we have shown that good risk patients do not benefit from transplant in terms of overall survival or quality of life<sup>(19,25-27)</sup>. We will continue this risk directed approach in AML15. Poor risk patients identified after Course 1 of treatment should however continue in AML15 and should receive the second course as allocated but should also be considered as eligible for any current NCRI AML trial of relapsed or high risk disease. The AML15 trial will also be used to identify new risk factors and to enable several add-on studies to take place.

The major issues in the consolidation phase are: (a) what chemotherapy schedule is best? (b) how many courses are needed? (c) is transplantation needed? The established MRC

consolidation (MACE and MidAC) is effective. High dose Ara-C is most commonly used in the USA based on the major dose comparison by the CALGB group<sup>(28)</sup>. It has been suggested that Ara-C is the treatment of choice for good risk patients<sup>(29)</sup> but similar good results have been obtained using the MRC schedule<sup>(30)</sup>, so it is particularly relevant to compare these approaches in good risk patients. For standard risk patients, who are the majority group, the question of which and how much therapy is of crucial importance. This group may benefit from transplantation as shown in the MRC AML10 Trial<sup>(19,25)</sup>. With respect to the chemotherapy question we will compare a high dose Ara-C approach with the MRC approach. It is not clear whether 3 g/m<sup>2</sup> dose levels of Ara-C are needed. The CALGB trial<sup>(28)</sup> demonstrated that a 3 g/m<sup>2</sup> dose schedule was better than 0.4 or 0.1 g/m<sup>2</sup>, but no studies have compared intermediate doses. We will recruit sufficient patients to compare a dose level of 1.5 g/m<sup>2</sup> with 3 g/m<sup>2</sup>, as this lower dose may be as effective but less toxic, and have a higher compliance. The availability of the immunoconjugate, Mylotarg offers an additional way of improving consolidation treatment. The pilot study has established the feasibility of combining Mylotarg with MACE or high-dose Ara-C, whether or not Mylotarg was used in Course 1<sup>(29)</sup>. Mylotarg will be tested in the first consolidation course of each chemotherapy regimen (Course 3) including the consolidation phase for APL (see below), given on day 1 of each course at a dose of 3 mg protein/m<sup>2</sup>.

The MRC AML12 trial has addressed the issue of how many courses of treatment in total are required. Even with 1200 cases randomised there remains uncertainty. No other collaborative group is currently addressing this question where the level of intensity of each course is high. In this trial, standard risk and good risk patients who complete four courses will be randomised to a fifth course or not. Either longer follow-up of the 1200 randomised patients in AML12 or, more likely, a combined analysis with AML15 of a projected 2000 patients will be available to address this issue. A recent follow-up analysis on this issue in the AML12 trial has suggested that it is unlikely that patients over 45 years will benefit from a fifth treatment course. For this reason and the continuing uncertainty in younger patients the randomisation to a fifth course will only be open to patients under 45 years.

#### 4.5 Stem Cell Transplantation

There was a modest overall survival advantage of allogeneic or autologous SCT in the MRC AML10 Trial<sup>(19,25)</sup>, but there was sufficient uncertainty to justify continuing to address the question in standard risk patients in the MRC AML12 trial. The AML10 trial suggested that transplant, while able to reduce relapse risk in good risk patients, did not improve survival. The policy of not using transplant in good risk patients in AML12 has been validated. With an additional 330 randomised patients the preliminary conclusion is that there is no advantage in using an autograft as Course 4 or 5 (i.e. instead of chemotherapy). Careful analysis of other trials<sup>(31-33)</sup> demonstrates that autograft is not superior with respect to survival, either overall or within any risk group. It does however reduce the risk of relapse in all subgroups. This is confirmed in a meta-analysis<sup>(34)</sup>. We therefore do not propose to evaluate autograft further. When the role of allograft is evaluated on a donor versus no donor basis and within risk groups, the survival benefit is modest<sup>(24,35)</sup>. However, a very important feature of these recent transplant trials has been that only a proportion (around 50-60%) of patients with donors receive the transplant. An intent to treat analysis may therefore underestimate the value of transplant – which clearly has the greatest antileukaemic effect. In AML15, patients who are standard risk with a sibling donor will have a conventional allograft as Course 3. Poor risk patients will receive an allogeneic transplant from a sibling or volunteer donor. The antileukaemic effect of a

conventional allograft is well known and a recent International Bone Marrow Transplant Registry analysis suggested that the result of the transplant was not improved by prior consolidation chemotherapy<sup>(36)</sup>, so bringing the allograft to Course 3 is justifiable. This should enable a higher proportion of those with donors to receive the transplant. It will also address the question of whether a transplant given early will involve less overall care when compared with 4 or 5 courses of intensive chemotherapy. The comparison will be on a donor versus no donor basis. In AML10 we demonstrated that there was no significant overall survival benefit with SCT for patients over the age of 35 years or in children. Transplant will only be offered to patients under 15 years who are designated poor risk. The use of peripheral blood rather than bone marrow has been suggested to be more effective<sup>(37,38)</sup>. At this stage, the stem cell source will be left to investigator choice until more data emerge.

The concept of “minigrafting” has expanded rapidly in the last two years<sup>(39)</sup>. By using a non-ablative schedule it is possible to achieve a high level of donor engraftment and thereby exert a graft versus leukaemia effect. The long-term efficacy of this approach is not yet known. Studies to evaluate its efficacy within disease groups are only now being planned. In the proposed trial we will evaluate the application of mini-transplants for standard risk older patients. This is of particular relevance to older patients who tolerate a conventional transplant less well or are too old to be offered a transplant. Ideally we would prefer to randomise patients in the middle age (35-44 years) range to a conventional versus mini-transplant, but the numbers available will be insufficient. Data from AML10 suggest that there is no survival benefit for patients over 35 years. We therefore will permit investigators to choose conventional or mini-allograft in patients 35-44 and recommend all older patients  $\geq 45$  receive a minigraft. In AML10, patients  $\geq 35$  years showed a significant reduction in relapse risk (45% v. 58%) but this was counterbalanced by an excess of deaths in CR. It is recognised that mini-allograft is an emerging technique with differing protocols available. In the context of AML in older patients it may deliver the anti-leukaemic benefit with a reduced risk. The data of efficacy in AML are preliminary. Data from Seattle, Houston and London<sup>(40)</sup> and from the IBMTR (R Storb, S Giralt, S McKinnon, *personal communications*) in a total of 200 patients suggest a similar efficacy to conventional allograft. During the course of the AML15 trial this technique will mature, so within the trial design a ‘mini-graft’ will be an available option as a phase II study where its efficacy can be compared on a donor versus no donor analysis in age stratified analyses. Similarly it can be expected that as the technique evolves the preparative schedule will alter, so this will be allowed for within the protocol. Since the anti-leukaemic effect of a minigraft is not known in AML, and the possibility exists that a minigraft as Course 3 could represent under treatment, the minigraft will take place as Course 4 with a preceding course of MACE chemotherapy, until more data emerge.

#### **4.6 Acute Promyelocytic Leukaemia (APL)**

The combination of retinoic acid with chemotherapy in induction and/or consolidation phases with or without maintenance treatment in recent randomised trials has produced a cure rate of 70-75% in APL<sup>(41-44)</sup>. Included in these trials was the MRC ATRA trial. Having established in that trial that all-trans retinoic acid (ATRA) given continuously until CR combined with chemotherapy was the best approach, a subsequent 179 patients have been treated and have achieved an 83% three year survival. This approach involves the standard 4 or 5 courses of chemotherapy with associated severe myelosuppression. The Spanish trial group<sup>(44)</sup> have recently shown, based on a protocol development from the

Italian Study Group<sup>(45)</sup>, a 92% two year survival using retinoic acid combined with an anthracycline based approach and oral maintenance, i.e. eliminating the other myelosuppressive drugs used in the MRC schedule. This raises the issue as to whether the MRC approach involves unnecessary myelosuppression necessitating more supportive care and reduced QoL. A limited comparison between the two protocols suggests a substantially greater number of days cytopenic and in hospital in the MRC schedule. In APL, in spite of good survival, we propose to evaluate the low intensity (Spanish) approach versus the established MRC approach. The primary endpoints will be resource use (measured by hospital days; day unit and OP attendance; antibiotics and transfusion support; neutropenic days) and QoL at baseline, 3, 6, 12 and 24 months.

Since APL cells have high CD33 expression, they may be a good target for an anti-CD33 antibody. Early experience using an unconjugated humanised antibody supported this concept with a patient entering CR and becoming RT-PCR negative for the PML-RAR $\alpha$  fusion transcript<sup>(49)</sup>. Recent data from the MD Anderson Hospital (Houston) in a series of 18 patients demonstrated rapid RT-PCR negativity in patients treated with ATRA – which typically does not achieve molecular negativity as a single agent – with Mylotarg<sup>(47)</sup>. This is supported by evidence that Mylotarg alone was able to achieve molecular negativity in relapsed APL patients. Thus Course 3 in each arm will have a randomised addition of Mylotarg.

In spite of this success there are still issues to be resolved in how best to manage high risk situations in APL. The MRC experience suggests that these are mainly patients who remain or who re-develop molecularly positive disease or who relapse. It is now clear that the most efficient way of restoring molecular negativity to these patients is the use of Arsenic Trioxide followed by stem cell transplantation<sup>(48)</sup>. The protocol provides guidance on how to manage these high risk situations.

**Although a FLT3 mutation is frequently found in patients with APL, these patients are not eligible for the FLT3 inhibition randomisation.**

## **4.7 Molecular Screening and Minimal Residual Disease Monitoring**

### **4.7.1 Risk Definition**

At diagnosis all cases will have molecular screening. The particular target lesions concern the definition of favourable genetic abnormalities, i.e. AML1-ETO, CBF $\beta$ -MYH11 and PML-RAR $\alpha$  corresponding to t(8;21)(q22;q22), inv(16)(p13q22) and t(15;17)(q22;q12-21). Previous analyses suggest that approximately 15% of cases with these lesions that were not detected by conventional cytogenetics can be detected molecularly<sup>(49, 50)</sup>. In several cases this was due to technical failure, but may also be explained by more complex rearrangements. Although the number of cases is small they seem to respond in a similar way to cases defined by cytogenetics, and therefore can be used to define the favourable risk group.

Recent studies have revealed that 20-27% of AML cases are associated with a mutation of the FLT3 gene, which is an independent prognostic factor<sup>(8-12)</sup>. All samples received by the University College Hospital London tissue bank will be analysed for FLT3 mutations as a quality control for banked nucleic acid and to

prospectively validate the prognostic implications of FLT3 mutations within cytogenetic risk groups. At a later stage in the trial this information may be incorporated into the risk stratification.

#### 4.7.2 Minimal Residual Disease Monitoring

Endpoint PCR analysis has been able to predict patients with APL who are at highest risk of relapse, when applied after Course 3<sup>(43)</sup>. Conventional RT-PCR assays are relatively insensitive and hence a single MRD assessment performed at the end of consolidation therapy fails to identify all patients destined to relapse. However, the Italian GIMEMA group has shown that subsequent molecular surveillance successfully predicts the majority of relapses, with recent data suggesting that pre-emptive treatment at the point of molecular relapse achieves a superior outcome to historical controls in frank haematological relapse<sup>(51)</sup>. Experience with AML1-ETO and CBF $\beta$ -MYH11 is much less, and the results are inconclusive with respect to predictive testing<sup>(52,53)</sup>. Patients known to have these three lesions will be serially followed using 'real-time' quantitative RT-PCR. These three lesions will only be useful in 25-30% of patients, with the lowest risk of relapse. A more universal target gene is WT1 (Wilm's Tumour gene) that is over expressed in 90% of AML cases<sup>(54)</sup>. While the suitability of WT1 as a marker for MRD is controversial, this may be the result of the use of non-quantitative assays of differing sensitivity. The trial will assess the value of quantitative monitoring by 'real-time' RT-PCR in appropriate cases for these lesions.

## 5 RANDOMISATION AVAILABILITY

Investigators are invited to regard this protocol as an evolving investigation into AML treatment. The statistical power calculations differ with each randomisation, so recruitment to some randomisations may be completed before others. This will mean that a randomised component of the trial may close or be changed before completion of the trial. Similarly, because individual components might require alteration in the light of trial monitoring or other experience this will be a feature of the trial.

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## 7 INCLUSION AND EXCLUSION CRITERIA

### 7.1 Inclusion Criteria

Patients are eligible for the AML15 trial if:

- They have one of the forms of acute myeloid leukaemia as defined by the WHO Classification (Appendix A) — this can be any type of *de novo* or secondary AML. Adult patients with acute promyelocytic leukaemia (APL) are eligible and should be entered into the randomisations specifically for APL (see Section 17).
- They are considered suitable for intensive chemotherapy.
- They should normally be under the age of 60, but patients over this age are eligible if intensive therapy is considered a suitable option.
- They have given written informed consent.

### 7.2 Exclusion criteria

Patients are not eligible for the AML15 trial if:

- They have previously received cytotoxic chemotherapy for AML. [Hydroxyurea, or similar low-dose therapy, to control the white count prior to initiation of intensive therapy is not an exclusion.]
- They are in blast transformation of chronic myeloid leukaemia (CML).
- They have a concurrent active malignancy.
- They are pregnant or lactating.
- The physician and patient consider that intensive therapy is not an appropriate treatment option. **(Such patients should be entered into the current NCRI trial for older or less fit patients).**

**CHILDREN WITH APL AND CHILDREN WITH DOWN SYNDROME AND AML ARE NOT ELIGIBLE FOR AML 15 – SEE SECTION 23.**

## 8 PROCEDURES FOR ENTRY INTO THE TRIAL AND DATA RECORDING

### 8.1 Centre Registration

Centres will be sent trial information by way of an invitation to participate in the trial. New regulations on the conduct of clinical trials place obligations on the investigators. In order to be registered as a trial centre, investigators (as an institution) will be asked to confirm: (1) that they have received and have read the MRC guidelines for good clinical practice in clinical trials, (2) that the study has LREC approval, (3) that the institution has accepted the responsibilities under the Research Governance Framework, (4) that written consent will be obtained for each patient and a copy retained in the notes, (5) that they agree to report serious unexpected adverse events as set out in Section 25 of this protocol, or in any subsequent guidance, (6) that they agree to participate in random audit if requested,

(7) that they will report data in a timely fashion, (8) that material to be stored for research is obtained using the trial consent documentation, (9) that copies of completed consent forms will be sent to BCTU.

For administrative reasons, investigators will also be asked to supply details of the location of their immunophenotyping, cytogenetic, molecular, genetic, pharmacy, tissue typing and transplant services, whether they wish to transmit data using the web based data collection system, and investigator contact e-mail addresses. In addition a limited amount of biochemical data will be collected and, as part of the centre registration process, relevant institutional normal ranges (bilirubin, AST, ALT and LDH) will be registered.

### 8.1.1 Patient Recruitment

Patients may be recruited only once a centre is fully registered. Patients should be consented for overall entry into the trial using **Patient Information Sheet 1 and Consent Form 1**. Further consent documents will be used at each randomisation point.

## 8.2 Randomisation

There are four randomisation points in the trial for which contact must be made with the Birmingham Clinical Trials Unit (BCTU). Patients fulfilling the criteria for entry into the trial (see Section 7) should be entered into the first randomisation by telephoning the BCTU in Birmingham (tel: 0800 953 0274). Telephone randomisation is available Monday to Friday, 09.00–17.00; internet randomisation is available seven days a week at: <https://www.trials.bham.ac.uk/aml15>.

### 8.2.1 First randomisation

Note: For this randomisation **Patient Information Sheet 2 and Consent Form 2** should be used. During the course of the trial certain randomisation options may not be available permanently or on a temporary basis. Investigators will be informed in advance so that only relevant information is given to the patient during the consent procedure.

Induction chemotherapy allocation will be given once the required patient details have been supplied. Patients will be allocated to one of three induction treatment arms, namely:

- Arm A Two courses of ADE
- Arm B Two courses of DA

Patients have a 33% chance of receiving each of the treatments.

### 8.2.2 Information required at first randomisation

- Centre and name of consultant in charge of management
- Patient's name (family name and given name)
- Sex

- Date of birth
- WHO performance status: 0=normal activity, 1=restricted activity, 2=in bed <50% waking hours, 3=in bed >50% waking hours, 4=completely disabled. For children under 10 use the Play Performance Scale (see Appendix D)
- Type of disease: *de novo* AML / secondary AML
- Whether APL (FAB type M3) or not
- Which laboratory will be undertaking FLT3 mutation analysis

For APL patients, you will be asked to confirm that you wish to randomise between the MRC and Spanish approaches.

For non-APL patients, you will be asked whether you wish to randomise ADE versus DA.

Clinicians will also be asked to state whether, based on a preliminary assessment, the patient is a potential candidate for standard allograft, mini-allograft or not for transplant if a matched donor were to be identified. It is obviously difficult to make such judgements at diagnosis, and clinicians will not be expected to stick with their initial evaluation, but this information is necessary to give an idea of the patient's possible course.

### 8.3 Diagnostic material

One objective of the trial is to investigate the therapeutic relevance of new techniques for detecting minimal residual disease and the quality of remission. Diagnostic material is essential for these studies. It is of particular importance to define the cytogenetic abnormalities, and where possible the molecular characteristics, of each patient as this may be relevant to the treatment strategy.

#### 8.3.1 Morphology

Central morphological review of adults will be provided by Dr David Swirsky as in previous MRC AML trials. Six unstained unfixed marrow slides should be sent to him at diagnosis (see page ii for address). For children, slides should be sent to Mrs J. Britton (see Section 23.2)

#### 8.3.2 Cytogenetics and Molecular Genetics

Cytogenetics should be carried out locally and reports sent directly to Dr Christine Harrison in Southampton for coding and storage on the central cytogenetic database. Cell pellets should be stored locally. If there are difficulties locally, central facilities will be provided by Mrs Yashma Patel at UCH — please indicate clearly on the samples that cytogenetic analysis is required. Dr Christine Harrison will coordinate cytogenetics in children (see Section 23.2).

#### 8.3.3 FLT3 Mutation Status

Molecular definition is intended for all patients, initially for characterisation of FLT3 mutation, for identification of cases with cryptic gene rearrangements that reassign patients to the favourable risk group, and for the identification of cases suitable for minimal residual disease monitoring. To enable this to be achieved in the timescale

required samples should be sent to either Dr Paul White in Cardiff or Prof Rosemary Gale at University College Hospital using the dispatch collection described below and on the trial website. Investigators will be informed of the FLT3 mutation status of patients to determine eligibility for the FLT3 inhibition randomisation. Additionally, they will be told of patients in whom molecular screening alters the risk group assignment and/or reveals a molecular marker suitable for MRD detection before the first bone marrow assessment of remission status after Course 1.

FLT3 mutation analysis will be analysed in real time at two reference laboratories (see below). Diagnostic material will also be stored for studies of resistance proteins, WT-1 gene expression, DNA microarray and future research studies, for which patient informed consent must be obtained. Reference laboratories for molecular monitoring have been established (listed below), to which more may be added; however for cases of suspected APL, **all samples** should be directed to Yashma Patel at UCH, London.

**It is essential that a sample is sent to a designated laboratory for the identification of patients with a FLT3 mutation.** These laboratories will pass samples on to the laboratories designated for MRD monitoring. It is intended that investigators will have the results of FLT3 assays by the end of the first course of chemotherapy to enable eligible patients to be randomised between FLT3 inhibition and not.

#### **Laboratories for FLT3 Mutation and other Molecular Analysis:**

Prof Rosemary Gale	Dr Paul White
Department of Haematology	Department of Haematology
University College Hospital	School of Medicine
2 <sup>nd</sup> Floor	Cardiff University
60 Whitefield Street	Heath Park
London W1T 4EU	Cardiff CF14 4XN
Tel: 0207 387 9300 (Ext: 8519)	Tel: 02920 742375
Fax: 0207 380 9911	Fax: 02920 744655
Email : yashma.patel@uclh.nhs.uk	Email: millski@cardiff.ac.uk

#### **Samples at diagnosis for molecular analysis:**

4 ml of bone marrow and 30ml of blood in EDTA.

(To be sent to UCH or Cardiff Laboratories)

#### **Samples at diagnosis for cytogenetic analysis:**

4 ml of bone marrow in tissue culture medium with preservative-free heparin

30 ml of heparinised blood

Ideally, both marrow and blood should be sent, but if only one is available please send that.

### 8.3.3 Immunophenotyping

Immunological definition is essential and should be carried out locally at the regional service — a copy of the report should be sent to BCTU with the "Notification of Entry" form.

### 8.3.4 Follow-up Material

Investigators will be informed of patients who are of particular interest for minimal residual disease monitoring. Arrangements for monitoring these patients are set out in Section 18. The labs undertaking initial characterisation and MRD are listed. **Note: these labs will automatically receive some of the initial sample which has been sent to either the UCH or Cardiff laboratories.**

#### Laboratories for Molecular Monitoring:

: Department of Haematology, University College Hospital, London.  
(Contact: Mrs Yashma Patel)

Department of Haematology, University Hospital of Wales, Cardiff  
(Contact: Dr Paul White)

Department of Haematology, Manchester Royal Infirmary, Manchester  
(Contact: Dr Abida Awan)

Haematological Malignancy Diagnostic Services, Algernon Firth Building, Leeds General Infirmary, Leeds, LS1 3EX (Contact: Paul Evans)

Haematology Department, City Hospital, Hucknell Road, Nottingham, NG5 1PB  
(Contact: Ian Carter)

Department of Haematology, Royal Hallamshire Hospital, Glossop Road, Sheffield, S10 2JF (Contact: Gill Wilson)

Regional Genetics Laboratory, Birmingham Woman's Hospital, Edgbaston, Birmingham, B15 2TG (Contact Mike Griffiths)

### 8.4 Data recording

It is intended to develop data recording for this trial as a web-based system. This is a secure encrypted system accessed by an institutional password, and complies with Data Protection Act standards. The system can be accessed on:

<https://www.trials.bham.ac.uk/aml15>

A user password will be supplied to investigators on receipt of the letters of LREC approval and site specific assessment, and centre registration information (see Section 8.1).

For investigators who do not wish to use the internet system, a patient record book will be sent to the consultant in charge of management following entry.

Forms should be completed and either entered via the web-based system or returned to BCTU as follows:

**Notification of Entry** (Section A) — return when all the diagnostic data requested are available (but not later than 1 month after entry).

**Induction Chemotherapy** (Section B) — return when blood counts have recovered after the second induction course, or at prior death (but not later than 2 months after completion of Course 2).

**Consolidation Chemotherapy** (Section C) — return when blood counts have recovered after the final course of consolidation chemotherapy, or at prior death (but not later than 2 months after the final course).

**Transplant** (Section D - only for patients receiving a transplant) — return when blood counts have recovered post transplant, or at prior death (but not later than 3 months after transplant).

**One Year Follow-up** (Section E) — return at one year after the end of treatment in 1st CR (i.e. last consolidation chemotherapy or transplant), or at death if the patient dies within 1 year of finishing therapy.

**Relapse** (Section F) — return at the completion of reinduction (and consolidation) therapy or at death (but not later than 4 months after relapse).

### **8.5 Quality of Life Assessments**

Subgroups of patients will be invited to participate in Quality of Life assessment at times specified in the protocol. All non-APL patients will be asked to complete a quality of life assessment, if in remission, at 12 and 36 months. The instrument will be the EORTC QLQC-30, plus the leukaemia specific module, which has been used in previous MRC AML trials and the Hospital Anxiety and Depression Scale (HADS). In addition at 36 months a Late Effects Questionnaire, as used in previous MRC trials, will be included. The patients should complete this at the specified time point and return it (Freepost) to the Trial Office at BCTU.

### **8.6 Health Economics**

Basic information on resource usage will be collected in the data forms B to F on all patients. Selected patients will be invited to provide additional information in the form of a patient diary that will be issued to the patient by the investigator.

Once a patient has been randomised, it is very important to have full details of the subsequent course of events, even if allocated therapy has been abandoned. Although clinical decisions remain with the physician (see Section 1, Ethical Considerations), follow-up data must continue to be collected on such patients and trial forms must be filled in, as far as possible, giving details of the therapy actually received and its outcome.

## **9 INDUCTION CHEMOTHERAPY: Courses 1 and 2**

Each induction schedule comprises two courses of allocated chemotherapy. Remission status will be determined after each course. If after Course 1, the patient is defined as poor risk (see Section 11.2), they should continue in AML15 or enter any currently available NCRI high risk

trial. If remission is not confirmed after the second course, the patient is off protocol for the AML15 study and can be entered into an NCRI high risk AML trial if one is available. Follow-up data must continue to be collected on all patients who go off protocol.

## 9.1 ADE schedule

### Course 1 **ADE 10+3+5**

Cytosine Arabinoside 100 mg/m<sup>2</sup> 12-hourly by i.v. push on days 1-10 inclusive (20 doses).

Daunorubicin 50 mg/m<sup>2</sup> daily by slow (1 hour) i.v. infusion on days 1, 3 and 5 (3 doses).

Etoposide 100 mg/m<sup>2</sup> daily by 1 hour i.v. infusion on days 1-5 inclusive (5 doses). (In children Etoposide is to be given as a 4 hour infusion.)

### Course 2 **ADE 8+3+5**

Cytosine Arabinoside 100 mg/m<sup>2</sup> 12-hourly by i.v. push on days 1-8 inclusive (16 doses).

Daunorubicin 50 mg/m<sup>2</sup> daily by slow (1 hour) i.v. infusion on days 1, 3 and 5 (3 doses).

Etoposide 100 mg/m<sup>2</sup> daily by 1 hour i.v. infusion on days 1-5 inclusive (5 doses). (In children Etoposide is to be given as a 4 hour infusion.)

## 9.2 DA schedule

### Course 1 **DA 3+10**

Daunorubicin 50 mg/m<sup>2</sup> daily by slow (1 hour) i.v. infusion on days 1, 3 and 5 (3 doses).

Cytosine Arabinoside 100 mg/m<sup>2</sup> 12-hourly by i.v. push on days 1-10 inclusive (20 doses).

### Course 2 **DA 3+8**

Daunorubicin 50 mg/m<sup>2</sup> daily by slow (1 hour) i.v. infusion on days 1, 3 and 5 (3 doses).

Cytosine Arabinoside 100 mg/m<sup>2</sup> 12-hourly by i.v. push on days 1-8 inclusive (16 doses).

NB: Prednisolone eye drops (Predsol 0.5%) should be given during each course of Ara-C.

Once blood counts have recovered after the second course of induction therapy, either the completed "Induction Chemotherapy" form (Section B) from the patient record book should be returned to BCTU, or the data should be entered into the web-based system.

## 10 FLT3 INHIBITION

Patients who have a confirmed FLT3 mutation are eligible to enter the FLT3 inhibition randomisation. **Patient Information Form 2B and Consent Form 2B** should be used. Patients

should be randomised immediately following the end of the first course of chemotherapy. For patients randomised to receive CEP-701, treatment should commence 2 days after the completion of each course of chemotherapy and should continue until two days before the commencement of the next chemotherapy course, or for a maximum of 28 days after the end of the chemotherapy course.. CEP-701 should not be administered concurrently with other chemotherapy. Patients allocated to inhibitor are intended to receive CEP-701 after each of the first four courses of chemotherapy. It is expected that CEP-701 will be administered at home, and investigators must ensure that the patient has received adequate instruction in reconstituting the drug. An instruction sheet is given in Appendix D and is available on the trial website (<http://www.aml15.bham.ac.uk>). The CEP-701 randomisation is only open to patients over 15 years old.

### 10.1 CEP-701 RANDOMISATION (Randomisation 1B)

The CEP-701 randomisation should take place as soon as the FLT3 status is known, so that treatment can start 2 days after completion of the allocated chemotherapy. Investigators are advised to discuss the possibility of randomisation to CEP-701 with the patient before initial entry to the trial, and before mutation status is known. Randomisation should take place irrespective of disease response.

To randomise a patient, (i) telephone the BCTU (tel: 0800 953 0274) during office hours (09:00 to 17:00 hrs, Monday to Friday); or (ii) use the 24 hour internet randomisation available at: <https://www.trials.bham.ac.uk/aml15>.

Before treatment allocation is given, investigators will need to provide:

- The name and hospital of the randomising clinician
- The AML15 trial number (or full name and date of birth)
- The approved laboratory where mutation analysis was done
- Confirmation of the patient's FLT3 ITD/TKD mutation status
- Date of completion of the first course of chemotherapy
- Whether or not the patient is receiving prophylactic Azole anti-fungal treatment.

### 10.2 CEP-701 TREATMENT

Patients randomised to chemotherapy plus sequential CEP-701 will receive a dose of **80 mg bd** starting 2 days after the last administration of each course of induction chemotherapy. **Note: because of a likely interaction with azole antifungal agents, where the patient is to be treated with azoles, the initial CEP-701 dose should be 40mg bd, for the first 7 days of each course of treatment to avoid gastrointestinal symptoms, if tolerated the dose can be continued at 60mg bd. If it emerges that the patient cannot tolerate the 60mg dose they should continue on 40mg.** Investigators may have to continue to adjust the dose based on the patient's symptoms

**Under no circumstances should CEP-701 be co administered with any course of chemotherapy treatment.**

Treatment will be given daily for **up to 28 days** or until 2 days before the next course of chemotherapy, whichever is sooner. When subsequent courses of chemotherapy are given, treatment with CEP-701 must stop 2 days before the first administration of the next course of chemotherapy, because elevated CEP-701 plasma concentrations may interfere with the effect of chemotherapy. Treatment with CEP-701 will recommence 2 days after the last administration of chemotherapy in the subsequent courses. In patients who are allocated to receive one course of consolidation treatment (see section 12 for details) the fourth course of CEP-701 should be commenced 7 days after the completion of the 28 days of CEP-701 given after consolidation course 1.

Patients randomised to receive chemotherapy plus sequential CEP-701 who have not achieved complete remission after the second course of chemotherapy will discontinue the study. After patients have completed two courses of induction treatment those in CR will be expected to enter the consolidation randomisation as described in sections 12 and 13.

### 10.2.1 Dose Adjustment

In patients who are **not receiving azole anti-fungals** the dose of CEP-701 may be reduced to 60 mg bd at any time for any patient receiving CEP-701 if the 80 mg bd dosage is not well tolerated; a return to 80 mg bd is permitted if tolerance improves. The dose may be increased to 100 mg bd under the following circumstances:

- Where patients have tolerated CEP-701 treatment at 80 mg bd well and have met the criteria for a second course of chemotherapy. (CEP-701 may be administered at 100 mg bd following completion of the second course of chemotherapy.)
- Where patients have tolerated CEP-701 treatment at 80 mg bd well and have achieved at least a PR, CEP-701 may be administered at 100 mg bd after the marrow assessment.)

In both cases, the dosage of CEP-701 treatment must be reduced to 80 mg bd (and if necessary 60 mg bd) in the event of poor tolerability.

In patients who are receiving azole anti-fungals the dose of CEP-701 should commence at 40 mg bd for the first 7 days of each treatment course. If well tolerated the dose can be increased thereafter to 60mg bd. If the 60mg dose is well tolerated the 80mg dose can be attempted, but if the 60mg dose is not well tolerated the patient should continue at the 40mg dose. Investigators are requested to record the sequence of dosing administered on the appropriate data collection form (Form D).

The dose can be increased to 100 mg bd under the following circumstances:

- Where patients have tolerated CEP-701 treatment at 80 mg bd well and have met the criteria for a second course of chemotherapy. (CEP-701 may be administered at 100 mg bd following completion of the second course of chemotherapy.)
- Where patients have tolerated CEP-701 treatment at 80 mg bd well and have achieved at least a PR, CEP-701 may be administered at 100 mg bd after the marrow assessment.)

The most likely side effect associated with CEP-701 treatment is nausea, so patients should be given precautionary anti-emetic treatment.

CEP-701 is supplied in 100 ml glass bottles as a clear yellow oral solution at a concentration of 25 mg/ml in polysorbate 80 NF (10 ml) and propylene glycol USP (10 ml). Prior to administration, CEP-701 should be diluted in fruit juice. The following juices are approved for use to administer CEP-701:

- grape
- pineapple
- apple
- V8<sup>®</sup> 100% vegetable juice
- orange juice (pulp-free)

During the study, patients will receive multidose bottles, each containing 100 ml of CEP-701 solution, from the investigator. Patients must be formally assessed on or near day 14 of CEP-701 treatment, and issued with further supplies as required. At each visit, patients should return all used and unused bottles to monitor dosage and compliance. Appendix D gives the dosing regimen.

Patients who are being treated with CEP-701 should be reviewed on or as near as possible to day 14 from the start of CEP-701 treatment. They should be evaluated for drug tolerance and a dose adjustment made if required. At the day 14 assessment, 20ml of blood should be collected and sent to the Cardiff laboratory in a standard lithium heparin container, using the Royal Mail collection service provided, for the assessment of free CEP-701 and FLT3 inhibitory activity and azole blood levels

Patients randomised to chemotherapy plus sequential CEP-701 will receive a dose of **80 mg bd** starting 2 days after the last administration of each course of induction chemotherapy. **Under no circumstances should CEP-701 be co administered with any course of chemotherapy treatment.** Treatment will be given daily for **up to 28 days** or until 2 days before the next course of chemotherapy, whichever is sooner. When subsequent courses of chemotherapy are given, treatment with CEP-701 must stop 2 days before the first administration of the next course of chemotherapy, because elevated CEP-701 plasma concentrations may interfere with the effect of chemotherapy. Treatment with CEP-701 will recommence 2 days after the last administration of chemotherapy in the subsequent courses.

Patients randomised to receive chemotherapy plus sequential CEP-701 who have not achieved complete remission after the second course of chemotherapy will discontinue the study. Patients for whom a stem cell transplant is planned can continue on CEP-701 until 28 days after their last pre-transplant course of chemotherapy.

### 10.2.2 Dose Adjustment

The dose of CEP-701 may be reduced to 60 mg bd at any time for any patient receiving CEP-701 if the 80 mg bd dosage is not well tolerated; a return to 80 mg bd is permitted if tolerance improves. The dose may be increased to 100 mg bd under the following circumstances:

- where patients have tolerated CEP-701 treatment at 80 mg bd well and have met the criteria for a second course of chemotherapy. (CEP-701 may be administered at 100 mg bd following completion of the second course of chemotherapy.)
- where patients have tolerated CEP-701 treatment at 80 mg bd well and have achieved at least a PR (CEP-701 may be administered at 100 mg bd after the marrow assessment.)

In both cases, the dosage of CEP-701 treatment must be reduced to 80 mg bd (and if necessary 60 mg bd) in the event of poor tolerability.

The more likely side effect associated with CEP-701 treatment is of nausea, so patients should be given precautionary anti-emetic treatment.

CEP-701 is supplied in 20 ml amber glass vials as a clear yellow oral solution at a concentration of 25 mg/ml in polysorbate 80 NF (10 ml) and propylene glycol USP (10 ml). Prior to administration, CEP-701 should be diluted in fruit juice. The following juices are approved for use to administer CEP-701: grape, pineapple, apple, V8<sup>®</sup> 100% vegetable juice, and orange juice (pulp-free).

During the study, patients will receive multidose vials, each containing 20 ml of CEP-701 solution, from the investigator. No more than two weeks of treatment should be dispensed at any one time. Patients must be formally assessed on day 14 of CEP-701 treatment, and issued with the second two week supply. At each visit, patients should return all used and unused vials to monitor dosage and compliance. Appendix D gives the dosing regimen, and also the minimum and maximum numbers of doses (based on a 16 to 20 ml withdrawal volume) obtainable per vial and these figures determine the number of vials supplied to the patient at each visit.

Patients who are being treated with CEP-701 should be reviewed on or as near as possible to day 14 from the start of CEP-701 treatment. They should be evaluated for drug tolerance and a dose adjustment made if required. On the morning of the day 14 assessment, the patient should delay taking CEP701 until blood has been collected. 20ml of blood should be collected as close as possible to 12 hours after the previous evening's dose, and sent to the Cardiff laboratory in a standard lithium heparin container, using the Royal Mail collection service, for the assessment of free CEP-701 and FLT3 inhibitory activity. Following sampling, the patient should take the scheduled day 14 morning dose of CEP701.

## **11 ASSESSMENT OF RESPONSE AND RISK GROUP ASSIGNMENT.**

A bone marrow aspirate to assess remission status should be carried out at 18-21 days after the end of Course 1. If the bone marrow is of adequate cellularity for the assessment of haematopoiesis, the patient's remission status should be ascertained. If the marrow is hypoplastic and assessment of status is not possible, a repeat marrow should be performed after a further 7-10 days and remission status be assessed. In order to achieve a subsidiary aim of the trial (i.e. assessing the relevance of residual cytogenetic or molecular existence of disease in morphological CR) investigators should also request cytogenetic analysis on this

sample and, if known to be relevant, molecular analysis. The patient should also be assigned to the appropriate risk group (see Section 11.2).

### 11.1 Definitions of Complete Remission, Partial Remission and Resistant Disease

**Complete Remission (CR):** The bone is regenerating normal haemopoietic cells and contains <5% blast cells by morphology.

**Partial Remission (PR):** The bone marrow is regenerating normal haemopoietic cells and contains between 5 and 15% leukaemic cells.

**Resistant Disease (RD):** The bone marrow contains >15% leukaemic cells.

### 11.2 Risk Group Assignment

The definitions of risk group for non-APL patients are detailed in Section 4.3 and are summarised below:

**Good risk:** Any patient with favourable genetic abnormalities — i.e. t(8;21), inv(16)/t(16;16) — irrespective of marrow status after Course 1 or the presence of other genetic abnormalities.

**Standard risk:** Any patient not in either good risk or poor risk groups — i.e. neither favourable nor adverse genetic abnormalities and not more than 15% blasts in the bone marrow after Course 1.

**Poor risk:** Any patient with more than 15% blasts in the bone marrow performed after Course 1 or with adverse genetic abnormalities — -5, -7, del(5q), abn(3q), t(9;22), complex ( $\geq 5$  abnormalities) — and without favourable genetic abnormalities.

**NB:** The presence of favourable or adverse genetic abnormalities may be established by any appropriate technique (e.g. cytogenetics, PCR, FISH). Patients without a genetic result can not be assigned to the good risk group, even if their morphology, e.g. M4<sub>EO</sub>, is suggestive of good prognosis. Such patients should be discussed with one of the clinical coordinators.

Patients should be assigned to the appropriate risk group based on the above criteria by their physician. If there is any doubt about the assignment, please contact one of the clinical coordinators.

### 11.3 Progression Through Induction Therapy

FLT3 mutation status should be available by the end of the first course of chemotherapy, and patients over the age of 15 with a FLT3 mutation can be entered in the CEP-701 randomisation (see Section 10). Irrespective of mutation status and result of CEP-701 randomisation, chemotherapy treatment following course 1 should continue according to the patient's risk group as follows:

- (i) If defined as poor risk, the patient may either continue with the AML15 protocol – see point (v) below – or may be entered into an NCRI high risk AML trial if one is available. (Note variation for children (Section 23).)
- (ii) If defined as good or standard risk, the patient should progress to Course 2 when neutrophils recover to  $1.0 \times 10^9/l$  and platelets to  $100 \times 10^9/l$ . (Note that dates of blood count recovery should be recorded in the data return.)
- (iii) The marrow should be re-assessed at 18-21 days after the end of Course 2 (patients who were confirmed to be in complete remission after Course 1 do not necessarily require a marrow assessment after Course 2 unless they are candidates for minimal residual disease monitoring).
- (iv) After Course 2, when patients in complete remission have regenerated to  $1.0 \times 10^9/l$  neutrophils and  $100 \times 10^9/l$  platelets, they are ready for the consolidation randomisation (see Section 12) and commencement of consolidation treatment, i.e. Course 3 (see Section 13). Standard and poor risk adults, and poor risk children, with a matched sibling donor should be considered for transplant (see Section 16).
- (v) For patients who are not in complete remission after Course 2 treatment will be deemed to have failed. They may be entered into any current NCRI high risk AML trial or, in the case of children, can be entered into the European Poor Risk AML Protocol 2001/01. All patients off protocol will still continue to be followed up within AML15.

#### 11.4 Donor Search

As early as possible during the induction phase of treatment (and before Course 3), tissue typing of siblings should be initiated in order to ascertain the availability of a HLA-matched sibling donor. Matching should be carried out on all patients, irrespective of age, who have siblings available. Allogeneic transplantation from volunteer matched donors for patients who are in the poor risk group is permitted.

## 12 CONSOLIDATION RANDOMISATION (Randomisation 2)

For this randomisation **Patient Information Sheet 3** and **Consent Form 3** should be used.

It is anticipated that during the life of the trial certain randomisations will be unavailable on a permanent or temporary basis. If this happens investigators will be informed in advance so that only relevant information is given to the patient during the consent procedure (see Section 5).

### 12.1 Randomisation Options

The consolidation randomisation is only available to patients who have achieved complete remission within 2 courses and who are not scheduled for SCT. Patients who have entered the FLT3 inhibition randomisation are eligible for the chemotherapy randomisation, **but not for the associated Mylotarg randomisation**. Randomisation will take place **before Course 3**. The randomisation has two components: i) patients may be randomised to receive either standard MRC consolidation chemotherapy (i.e. **MACE then MidAC**) or two courses of **high-dose Ara-C** (with a sub-randomisation between **1.5 g/m<sup>2</sup>** and **3 g/m<sup>2</sup>** for adults, but with all children to receive a dose of **3g/m<sup>2</sup>**); and ii) at the same time, adults

and children may be randomised to receive either **Mylotarg** or **not** as part of consolidation Course 3. Only patients not entering the FLT3 inhibitor randomisation are eligible for this part of the randomisation. Additionally, patients who received Mylotarg in induction, and who experienced side-effects which are clearly related to the Mylotarg treatment, should **not be randomised for Mylotarg in consolidation**.

Therefore, the six possible treatment arms are:

Arm D	MACE followed by MidAC (with no Mylotarg)
Arm E	MACE followed by MidAC (with Mylotarg in Course 3)
Arm F	Ara-C at 1.5 g/m <sup>2</sup> for two courses (with no Mylotarg)
Arm G	Ara-C at 1.5 g/m <sup>2</sup> for two courses (with Mylotarg in Course 3)
Arm H	Ara-C at 3.0 g/m <sup>2</sup> for two courses (with no Mylotarg)
Arm I	Ara-C at 3.0 g/m <sup>2</sup> for two courses (with Mylotarg in Course 3)

**Note:** It is expected that most patients who did not enter the FLT3 inhibitor randomisation will be entered into both randomisations. Exceptionally, a clinician may elect to undertake just one or other randomisation. If a patient is not randomised to MRC consolidation versus high-dose Ara-C, they must be given MRC consolidation; if a patient is not randomised to Mylotarg versus not, they must not receive Mylotarg. Despite the availability of this choice to opt out of one or other of the randomisations, participating clinicians are strongly encouraged to randomise eligible patients between all six arms when available. However, it is very important that patients should be randomised only between treatment arms that are likely to be feasible and acceptable in the individual case, since non-compliance with the allocated treatment can substantially reduce the power of the trial.

**Children: Children are also eligible for the Mylotarg randomisation but will only be randomised to MRC consolidation versus Ara-C at 3.0 g/m<sup>2</sup> – they will not be randomised to Ara-C at 1.5 g/m<sup>2</sup> (i.e. only Arms D,E, H and I will apply).**

## 12.2 Stem Cell Transplantation for Standard and Poor Risk Adults and Poor Risk Children

Standard risk adults who have a matched sibling donor, and poor risk adults and children who have a sibling or volunteer donor should receive an allogeneic SCT if considered fit enough for the procedure. This may be either standard allo-SCT or “mini”-SCT for adults (for full details, see Section 16 for adults and Section 23 for children). If it is decided that a transplant is to be carried out, the patient should not be randomised for consolidation chemotherapy options.

[Autologous bone marrow harvest and/or peripheral blood stem cell collection are not a mandatory part of the AML15 protocol. Harvest/collection may be undertaken (e.g. for potential use in 2nd CR autograft) at the discretion of the physician.]

## 12.3 Timing of Consolidation Randomisation

Statistically, it is preferable for the randomisation to take place as close as possible to the start of Course 3, and ideally on the day that Course 3 is scheduled to start. This will reduce non-compliance, which would have an adverse impact on the power of the trial.

Although randomisation should be carried out as close to Course 3 as possible, it is recommended that the options available are discussed with the patient at an earlier stage, e.g. during induction therapy, in order to ensure that the patient has plenty of time to

consider the options and arrive at an informed decision. This should reduce the risk of non-compliance with allocated treatment.

#### 12.4 Information Required at Consolidation Randomisation

Before carrying out the consolidation randomisation please make sure that:

- (a) The patient's risk group is known (see Section 11.2).
- (b) It has been decided whether the patient is willing to be randomised between MRC consolidation chemotherapy and high-dose Ara-C (at one of two doses).
- (c) It has been decided whether the patient (if not already in the FLT3 inhibitor randomisation) is willing to be randomised between Mylotarg versus not during consolidation, and that the liver function tests do not exceed twice the upper limit of normal.

For randomisation: (i) telephone the BCTU (tel: 0800 953 0274) during office hours (09:00 to 17:00 hrs, Monday to Friday); or (ii) use the 24 hour internet randomisation available at: <https://www.trials.bham.ac.uk/aml15>.

Treatment allocation will be given once the following patient details have been supplied:

- AML15 trial number (or full name and date of birth).
- Confirmation that the patient has received two courses of induction therapy, is in complete remission and is not scheduled for SCT.
- Patient's risk group (good, standard, poor — see Section 11.2 for definitions).
- Whether the patient is to be randomised between MRC consolidation chemotherapy versus Ara-C chemotherapy (if not, MRC therapy must be elected).
- If not already enrolled in the FLT3 inhibitor randomisation, whether the patient is to be randomised between Mylotarg versus not (if not, Mylotarg must not be given).
- In order to be entered into Mylotarg randomisation liver function tests must not exceed twice normal.

### 13 CONSOLIDATION CHEMOTHERAPY: Courses 3 and 4

Each consolidation schedule comprises two courses of chemotherapy, with or without CEP-701. The consolidation chemotherapy to be given depends on the allocation at second randomisation (or the elected therapy, if not randomised).

**Note: All drug doses should be reduced by 25% for children aged less than one year.**

#### 13.1 MRC Consolidation

Course 3

##### **MACE**

Amsacrine 100 mg/m<sup>2</sup> daily by 1 hour i.v. infusion (in 5% dextrose) on days 1-5 inclusive (5 doses).

Cytosine Arabinoside 200 mg/m<sup>2</sup> daily by continuous i.v. infusion on days 1-5 inclusive.

Etoposide 100 mg/m<sup>2</sup> daily by 1 hour i.v. infusion on days 1-5 inclusive (5 doses). (In children Etoposide is to be given as a 4 hour infusion.)

**Note:** Amsacrine must not be infused in 0.9% sodium chloride: precipitation or flocculation occurs. This will necessitate a 2-hour interruption of the Cytosine Arabinoside infusion. Where venous access is limited to a single lumen line a 2 hour (adults) or 5 hour (children) interruption to the Ara-C infusion may be required to enable the Amsacrine and Etoposide administration.

Course 4                    **MidAC**  
Mitoxantrone 10 mg/m<sup>2</sup> daily by slow (1 hour) i.v. infusion on days 1-5 inclusive (5 doses).  
  
Cytosine Arabinoside 1.0 g/m<sup>2</sup> 12-hourly by 2-hour i.v. infusion on days 1-3 inclusive (6 doses).

### 13.2 High-dose Ara-C (1.5 g/m<sup>2</sup>)

Course 3                    Cytosine Arabinoside 1.5 g/m<sup>2</sup> 12-hourly by 4 hour i.v. infusion on days 1, 3 and 5 (6 doses).  
  
Course 4                    Cytosine Arabinoside 1.5 g/m<sup>2</sup> 12-hourly by 4 hour i.v. infusion on days 1, 3 and 5 (6 doses).

### 13.3 High-dose Ara-C (3.0 g/m<sup>2</sup>)

Course 3                    Cytosine Arabinoside 3.0 g/m<sup>2</sup> 12-hourly by 4 hour i.v. infusion on days 1, 3 and 5 (6 doses).  
  
Course 4                    Cytosine Arabinoside 3.0 g/m<sup>2</sup> 12-hourly by 4 hour i.v. infusion on days 1, 3 and 5 (6 doses).

**Note:** Prednisolone (0.5% Predsol) eye drops should be used during each course of high-dose Ara-C, and during the course of MidAC which also contains high-dose Ara-C, and be continued for 5 days after the course finishes.

**In children under 1 year the Ara-C dose should be calculated on body weight; i.e, the 3.0g/m<sup>2</sup> dose will be calculated as 100mg/kg.**

In all arms, Course 4 should be given once counts have recovered to 1.0 x 10<sup>9</sup>/l neutrophils and 100 x 10<sup>9</sup>/l platelets following Course 3. Delay in count recovery regularly occurs, and problem cases should be discussed with the clinical coordinators.

Once blood counts have recovered after the fourth course of chemotherapy, the "Consolidation" form (Section C) from the patient record book should be filled in and returned to BCTU, or completed online, unless the patient is randomised to receive a fifth course.

**Patients who have entered the FLT3 inhibitor randomisation, and who have been allocated CEP-701 should continue to receive the drug after courses 3 and 4.**

## 14 MYLOTARG THERAPY DURING CONSOLIDATION

### 14.1 Administration of Mylotarg

Patients are only eligible to enter the Mylotarg randomisation if they have not already entered the FLT3 inhibition randomisation. They should only receive Mylotarg if their liver function tests do not exceed twice normal. Mylotarg will be given at a dose of 3 mg protein/m<sup>2</sup> on day 1 of Course 3. Details of the premedication, and other procedures for Mylotarg administration, are set out in Appendix B.

## 15 FOUR vs. FIVE COURSE RANDOMISATION (Randomisation 3)

For this randomisation **Patient Information Sheet 4** and **Consent Form 4** should be used.

**Only patients under 45 years are eligible for this randomisation.** Patients who have completed four courses of treatment are eligible to be randomised to receive a fifth course or not. This should be initiated once the neutrophil count has exceeded  $1.0 \times 10^9/l$  and the platelet count has exceeded  $80 \times 10^9/l$ .

For randomisation: (i) telephone the BCTU (tel: 0800 953 0274) during office hours (09:00 to 17:00 hrs, Monday to Friday), or (ii) use the 24 hour internet randomisation available at: <https://www.trials.bham.ac.uk/aml15>.

Treatment allocation will be given once the following patient details have been supplied:

- AML15 trial number (or full name and date of birth).
- Confirmation that the patient has received four courses of chemotherapy and remains in first complete remission.

### 15.1 Late Consolidation Therapy: Course 5

**Course 5** Cytosine Arabinoside 1.5 g/m<sup>2</sup> 12-hourly by 4 hour i.v. infusion on days 1, 3 and 5 (6 doses).

**Note:** Prednisolone (0.5% Predsol) eye drops should be used during the course and be continued for 5 days after the course finishes.

**In children under 1 year the Ara-C dose should be calculated on body weight; i.e. the 1.5g/m<sup>2</sup> dose will be calculated as 50mg/kg.**

Once blood counts have recovered after the fifth course of chemotherapy, the "Consolidation" form (Section C) from the patient record book should either be filled in and returned to BCTU, or completed using the web based data entry system.

**FLT3 inhibitor treatment with CEP-701 will *not* be given to patients allocated to receive a fifth course of chemotherapy.**

## 16 STEM CELL TRANSPLANTATION

The protocol provides for allogeneic transplantation for all adult patients with an HLA-matched sibling donor who are **not** designated as good risk, and for children designated as poor risk. As soon as a potential donor is identified the transplant centre should be informed. The transplant should be carried out 6-8 weeks after the final course of chemotherapy. The type of transplant and the transplant protocol will be determined by the transplant centre's usual policy. As a guide based on prior evidence:

1. Patients <35 years should receive a conventional allogeneic transplant with Cyclophosphamide and Total Body Irradiation (8 x 180cGy fractions). [For children aged less than 2 years, conditioning should be with Busulphan and Cyclophosphamide] (See Appendix E).
2. Patients 35-44 years can receive a conventional allogeneic transplant or a "mini" allograft depending on investigator or patient choice.
3. Patients ≥45 years should receive a "mini" allograft.

### 16.1 Conventional Allogeneic Transplantation

If the patient meets the criteria of the transplant centre, he/she will receive the transplant **as Course 3**. The most widely used myeloablative schedule is Cyclophosphamide and Total Body Irradiation (8 x 180 cGy). The source of stem cells can be bone marrow or peripheral blood. If peripheral blood is used, a dose of at least  $4 \times 10^6$  CD34 cells/kg should be given. Graft versus host prophylaxis will be determined by the transplant centre, but the most widely used is Methotrexate and Cyclosporin. It is required that patients who receive a transplant will provide written consent in line with the transplant centre policy. Children intended for SCT will have a conventional allograft following UKCCSG Protocols (Section 23).

### 16.2 Mini-allografting

Patients who will receive a mini-allograft must first receive **MACE as Course 3** (see Section 13.1) and the **mini-allograft as Course 4**. The mini-allograft should only be carried out at centres with experience of this approach and should **not be carried out in centres who do not perform conventional allografts**. The precise protocol to be used in the AML15 trial will be prescribed and, as the field develops over the next five years, will be subject to changes in light of experience.

Transplant centres initially may choose one of two mini-allograft protocols:

#### **FBC Protocol:**

Fludarabine	30 mg/m <sup>2</sup> /day	days -9 to -5 inclusive
Busulphan	4 mg/kg/day	days -3 and -2
Campath 1H	20 mg/day i.v.	days -5 to -1 inclusive

(use of phenytoin and low molecular weight heparin for VOD prophylaxis is optional)

#### **Fludara, Melphalan, Campath (UCL) Protocol:**

Fludarabine	30 mg/m <sup>2</sup> /day	days -7 to -3 inclusive
Melphalan	140 mg/m <sup>2</sup>	on day -2
Campath 1H	20 mg/day	days -8 to -4 inclusive

Since patient and donor will require time to be counselled about the transplant option which may be delivered as early as course 3, investigators are encouraged to identify donor availability as soon as possible after diagnosis.

On completion of the transplant the “Transplant” form (Section D) should be completed and returned to BCTU or entered via the web-based system.

### 16.3 Collection of Autologous Stem Cells

Autologous stem cell transplantation is not part of the AML15 trial. However investigators may wish to collect stem cells during treatment for use in second remission. If peripheral blood cells are collected, mobilisation should take place after Course 2 if the patient was confirmed to be in CR after Course 1, and after Course 3 if remission was not confirmed until after Course 2. A minimum collection of  $2 \times 10^6$  CD34 cells/kg should be collected. Mobilisation should be attempted using G-CSF [Lenograstim, 263µg (1 vial)] daily for a maximum 10 days starting day +8 from the end of the chemotherapy course.

If bone marrow cells are to be collected, the harvest should take place immediately before Course 4.

## 17 ACUTE PROMYELOCYTIC LEUKAEMIA

Patients may enter this part of the protocol at diagnosis with *de novo* or secondary acute promyelocytic leukaemia (APL) recognised morphologically as FAB-M3. Molecular genetic confirmation is required by either: i) cytogenetics, ii) demonstration of PML-RAR $\alpha$  rearrangement by PCR, or iii) the characteristic staining with the PML antibody. Diagnostic bone marrow and peripheral blood from all patients with suspected APL should be sent to Yashma Patel at UCH, London Tissue Bank (for contact details see page ii). Investigators will be contacted by the laboratory of Dr David Grimwade within 2 weeks to co-ordinate MRD assessment (contact details, page ii). Confirmation of the molecular lesion is important because a non-APL case allocated to the ‘Spanish’ arm may be under treated. Patients who enter the APL part of this trial will be eligible for minimal residual disease (MRD) monitoring (Section 18)

### 17.1 ATRA Therapy

All patients with APL should receive ATRA therapy as follows:

45 mg/m<sup>2</sup>/day starting on day 1 on induction therapy until first CR is achieved or until completion of 2 courses of chemotherapy.

If the patient is not in morphological CR after Course 2, ATRA will be discontinued. Such patients may benefit from Arsenic Trioxide therapy (see section 17.11).

Patients who present with a WBC  $>10 \times 10^9/l$  should receive steroid (Dexamethasone 10 mg i.v. 12-hourly) for the first 5 days of chemotherapy as prophylaxis against retinoic acid syndrome (see Section 17.10.1). Leucopheresis is not indicated. Heparin and Tranexamic Acid should not be routinely used.

The comparison between the MRC and Spanish treatments is now closed and is being analysed. The preliminary advice is that the Spanish arm is superior, so it is recommended the patients with APL are allocated to the Spanish treatment. Patients should still be registered at diagnosis, because these patients are still eligible for the Mylotarg randomisation in course 3. Monitoring of disease will continue to be available. Patients in the Spanish arm will receive ATRA on days 1-15 of Courses 3 and 4.

## 17.2 Spanish Treatment

### Course 1

Idarubicin 12 mg/m<sup>2</sup> by short (10 minute) i.v. infusion on days 2, 4, 6 and 8 (4 doses).

ATRA will be given until CR (see Section 17.1).

Following recovery of neutrophils to 1.0 x 10<sup>9</sup>/l and platelets to 100 x 10<sup>9</sup>/l patients will receive Course 2.

Bone marrow assessment should be carried out 18-23 days from the end of Course 1 and should include material for minimal residual disease (see Section 18). The marrow response should be documented.

### Course 2

Idarubicin 7 mg/m<sup>2</sup> by short (10 minute) i.v. infusion on days 1, 2, 3, 4 (4 doses).

ATRA 45 mg/m<sup>2</sup>/day on days 1-15 (15 doses) if patient is in CR after Course 1, otherwise ATRA until morphological CR.

Following recovery of neutrophils to 1.0 x 10<sup>9</sup>/l and platelets to 100 x 10<sup>9</sup>/l patients will receive Course 3.

### Course 3

Mitoxantrone 10 mg/m<sup>2</sup> as a 30 minute infusion on days 1-5 inclusive (5 doses).

Mylotarg 3 mg/m<sup>2</sup> on day 1 if allocated (see second randomisation – Section 17.5 and Appendix B).

ATRA 45 mg/m<sup>2</sup>/day on days 1-15 (15 doses).

Following recovery of neutrophils to 1.0 x 10<sup>9</sup>/l and platelets to 100 x 10<sup>9</sup>/l patients will receive course 4.

### Course 4

Idarubicin 12 mg/m<sup>2</sup> as a short (10 minute) i.v. infusion on day 1 only (1 dose).

ATRA 45 mg/m<sup>2</sup>/day on days 1-15 (15 doses).

Note: After each course of treatment blood and bone marrow should be sent for minimal residual disease monitoring (see Section 18).

### 17.3 Second Randomisation

Note: For this randomisation, **Patient Information Sheet 6** and **Consent Form 6** should be used.

Following completion of Courses 1 and 2 patients are eligible for the second randomisation.

Patients (adults only) will be randomised to receive Mylotarg or not during Course 3.

For randomisation: (i) telephone the BCTU (tel: 0800 953 0274) during office hours (09:00 to 17:00 hrs, Monday to Friday); or (ii) use the 24 hour internet randomisation available at: <https://www.trials.bham.ac.uk/aml15>.

Treatment allocation will be given once the following patient details have been supplied:

- AML15 trial number (or full name and date of birth).
- Confirmation that the patient has received two courses of induction therapy and is in complete morphological remission.
- Confirmation that liver function tests are normal.

### 17.4 Mylotarg Treatment

Patients are only eligible to receive Mylotarg if the liver function tests do not exceed twice the normal range. Mylotarg will be given at a dose of 3 mg protein/m<sup>2</sup> on day 1 of Course 3. Details of the premedication, and other procedures for Mylotarg administration, are set out in Appendix B.

### 17.5 Maintenance Therapy

Maintenance therapy is **given with the Spanish treatment** and should commence following full haemopoietic recovery from Course 4 (neutrophils > 1.5 x 10<sup>9</sup>/l and platelets > 100 x 10<sup>9</sup>/l), but not less than 1 month after Course 4. Marrow reassessment for molecular assessment is essential. If RT-PCR is positive, the patient is eligible for treatment as a high-risk patient (see Section 17.11).

- **6-Mercaptopurine (6-MP)**, 50 mg/m<sup>2</sup>/day orally. The dose will be adjusted according to toxicity during the follow-up period. The treatment must be continued for two years.
- **Methotrexate (MTX)**, 15 mg/m<sup>2</sup> weekly orally, starting one month after recovery from the last consolidation. The dose will be adjusted according to toxicity during the follow-up period. This weekly treatment must be continued for two years.
- **ATRA**, 45 mg/m<sup>2</sup>/day orally, for 15 days every three months for 2 years. The first ATRA maintenance course will begin four months after recovery from the last consolidation course. During the days of ATRA administration, the treatment with MTX and 6-MP will be discontinued.

### Modification of the Maintenance Treatment

The doses of Methotrexate and 6-Mercaptopurine will be modified as a function of the peripheral blood counts:

- WBC between  $2.5$  and  $3.5 \times 10^9/l$ : dose reduction by 50%.
- WBC  $<2.5 \times 10^9/l$ : temporary discontinuation of maintenance.

In addition selected patients will be asked to complete a patient diary concerning medical events. For patients who are selected the diary will be sent to the investigator.

## **17.6 Treatment Modification**

During induction treatment, ATRA may be temporarily discontinued in the presence of one of the following complications: ATRA syndrome, pseudotumour cerebri, hepatotoxicity.

### **17.6.1 ATRA Syndrome**

This is accurately defined by the presence of: unexplained fever, weight gain, respiratory distress, interstitial pulmonary infiltrates, and pleural or pericardial effusion, with or without hyperleukocytosis. No single sign or symptom itself may be considered diagnostic of the syndrome. However, at the earliest manifestations of suspected ATRA Syndrome (e.g. unexplained respiratory distress), and prior to development of a full blown syndrome, the following measures should be immediately undertaken:

- temporary discontinuation of ATRA treatment.
- prompt initiation of dexamethasone 10 mg i.v. 12-hourly until disappearance of symptoms and signs, and for a minimum of 3 days.
- frusemide when clinically required.

### **17.6.2 Pseudotumour Cerebri**

This is defined as presence of: severe headaches with nausea, vomiting, and visual disorders. In this case, generally developing in patients under 20 years of age, it is often necessary to temporarily discontinue ATRA treatment and to administer opiates.

### **17.6.3 Hepatotoxicity**

This is defined as: an increase in serum bilirubin, AST/ALT, or alkaline phosphatase  $>5$  times the normal upper level. This requires a temporary suspension of the ATRA. The Idarubicin doses should not be changed if on the Spanish schedule.

As soon as the symptoms and the patient's clinical condition improves, treatment with ATRA will be resumed at 50% of the previous dose during the first 4 days after the disappearance of retinoic acid syndrome, amelioration of pseudotumour cerebri or when serum bilirubin, AST/ALT or alkaline phosphates are reduced to  $<4$  times the normal upper level. Thereafter, in absence of worsening of the previous toxicity, ATRA should be resumed at full dosage.

In case of reappearance of signs and symptoms of ATRA toxicity, the drug must be discontinued indefinitely during induction therapy. However, patients who enter the maintenance phase of the Spanish schedule should receive ATRA where possible.

### **17.7 Treatment of High Risk APL (relapse, molecular relapse, persistent MRD positivity)**

Initial treatment of APL may fail, in which case patients will either relapse or be at high risk of relapse. In this study adult patients who relapse, or who are deemed to be at high risk of relapse based on molecular data, should be treated with Arsenic Trioxide. It is anticipated that during the course of the trial molecular criteria will become more precise as a result of the monitoring data (Section 18). As this evidence emerges investigators will be informed of patients who are considered high risk and who should be offered further treatment.

**Note: At relapse CNS should be checked for occult disease.**

#### **17.7.1 Treatment of High Risk APL**

It is recommended that patients who are designated as high risk either because of molecular persistence or recurrence or haematological relapse are treated with Arsenic Trioxide (ATO). This should initially be given on an inpatient basis, but can be continued as an outpatient.

Patients should have an ECG assessment before and up to twice weekly during treatment to ensure that the QT interval does not exceed 460 msec. Drugs which can prolong the QT interval should be avoided (a list of such drugs is given on the website [www.torsades.org](http://www.torsades.org)). During therapy the serum potassium must be kept above 4mmol/l and the serum magnesium above 1.8mg/dl.

During treatment patient may develop signs of the “differentiation syndrome” and should be treated as recommended for the “ATRA” syndrome (see section 17.10.1). If hepatic or renal function is abnormal then consider interrupting the treatment and/or consult with a trial coordinator.

Patients should have molecular assessments (organised by Dr David Grimwade, see page ii).

#### **Induction Treatment**

One of two treatment schedules can be followed for induction:

- A) 0.15mg/kg daily by a slow (1-2 hour) infusion for a maximum of 50 days.
- B) 0.30mg/kg daily for 5 days followed by 0.25mg/kg twice weekly for up to 7 weeks

Patients should have a marrow re-assessment at 28 days which should include molecular assessment. If the patient is in morphological and molecular remission they should enter consolidation treatment. If the patient is not in haematological remission or is in haematological but not molecular remission two doses of Idarubicin (10mg/m<sup>2</sup>) should be added on 2 consecutive days and the morphological and molecular status re-checked 2 weeks later. The ATO treatment should be continued at the induction dose. When the patient is confirmed to be in haematological remission they should enter the consolidation phase.

### **Consolidation Treatment**

When the patient is confirmed to be in haematological remission, but not necessarily molecular remission, they should receive an additional 4 weeks of consolidation treatment with the same schedule as chosen for induction. At the end of this period the marrow should be re-assessed for molecular response.

If the patient has achieved a molecular remission arrangements should be made to undertake autologous stem cell transplant. Patients who remain in remission but are molecularly positive should be assessed for allogeneic stem cell transplant. If no transplant option is available the patient should commence maintenance chemotherapy as described in protocol section 17.7.

## **18 ARRANGEMENTS FOR MOLECULAR SCREENING AND MINIMAL RESIDUAL DISEASE MONITORING**

All diagnostic material will be collected into the MRC AML tissue bank at the UCH (Mrs Yashma Patel) or Cardiff (Dr P White) laboratories, where it will be analysed for FLT3 status and passed on to the other molecular laboratories for the studies described and also for future research. Investigators should note that patients must consent to this donation, and documentation concerning this is included in the main trial consent documentation (**Patient Information and Consent Form 6**).

The Tissue Bank is under the direction of Professor David Linch but material can be deposited in five sites under the aegis of this bank:

Mrs Yashma Patel  
Department of Haematology - Cytogenetics  
University College Hospital  
Gower Street  
LONDON WC1E 6AU  
Tel: 0207 387 9300 (Ext: 8519)  
Fax: 0207 380 9911  
Email: [Yashma.Patel@uclh.nhs.uk](mailto:Yashma.Patel@uclh.nhs.uk)

Dr Paul White  
Department of Haematology, 7<sup>th</sup> Floor  
University Hospital of Wales  
Heath Park  
Cardiff CF14 4XN  
Tel: 02920 744524  
Fax: 02920744655  
Email: [WhitePC@Cardiff.ac.uk](mailto:WhitePC@Cardiff.ac.uk)

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Dr Paul Evans  
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Algernon Firth Building  
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Patients will be characterised by the screening process (see Section 4.7) as suitable for MRD monitoring, and this will be led by different molecular laboratories. Investigators will be informed if their patient is suitable for monitoring before the marrow assessment after Course 1.

**PML-RAR $\alpha$  Rearrangements:**

Dr David Grimwade  
Division of Medical and Molecular Genetics  
8th Floor, Guy's Tower  
Guy's Hospital  
London SE1 9RT  
Tel: 0207 188 3699  
Fax: 0207 188 2585  
Email: [david.grimwade@kcl.ac.uk](mailto:david.grimwade@kcl.ac.uk)

**Note:** Please address all APL samples for diagnosis and MRD assessment to Mrs Yashma Patel (see above for address).

**AML1-ETO, CBF $\beta$ -MYH11 rearrangements and WT1 expression:**

Dr Sarah Daley  
Molecular Diagnostics Centre  
Top Floor, Multi-purpose Building  
Manchester Royal Infirmary  
Oxford Road  
Manchester  
M13 9WL  
Tel: 0161 276 4137  
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**Frequency of Monitoring**

On entering the trial, it should be explained to patients that, if it emerges that the leukaemia cells have an appropriate molecular lesion, they will be invited to participate in the molecular disease monitoring. Once patients have been identified, investigators will be alerted by the molecular monitoring group (Dr. Grimwade/Professor Yin). The intention is routinely to monitor patients (blood and bone marrow) after each course of chemotherapy, 3-monthly in year 1, 4-monthly in year 2 and 6-monthly in year 3. The frequency of monitoring may change during the trial as new information becomes available. Since it has become clear that persistent or recurrent RT-PCR positivity powerfully predicts relapse, it is important to ensure that the test is completely reliable for that patient. This may result in advice to repeat the test within the interval planned.

Samples from children for MRD monitoring should be sent to their local MRD laboratory for appropriate forwarding to other laboratories. These are listed below:

Bristol Children's Hospital  
Haematology and Bone Marrow

Department of Haematology  
Royal Hospital for Sick Children

North Trent Molecular Genetics  
Sheffield Children's Hospital

Transplant Research Group  
Dept of Pathology and  
Microbiology,  
School of Medical Sciences,  
University of Bristol,  
Bristol BS8 1TD  
Tel: 0117 928 9158/7888  
Fax: 0117 928 7896

Dalnair Street  
Glasgow  
G3 8SJ  
Tel: 0141 201 0689  
Fax: 0141 201 0857

Western Bank  
Sheffield  
S10 2TH  
Tel: 0114 271 7003  
Fax : 0114 273 7467

## 19 MANAGEMENT OF HIGH RISK, REFRACTORY OR RELAPSED PATIENTS

Patients who are defined as high risk (see definition in Sections 4.3 and 11.2) after Course 1, who do not achieve morphological CR after Course 2, or who relapse, are eligible for any current NCRI high risk AML trial. It is permissible for patients defined as high risk, but who are in morphological CR after Course 1 to continue with the AML15 protocol. APL patients who relapse morphologically or are designated as having a high risk molecular response should be managed as described in the APL section of this protocol (see Section 17.11).

Children who are defined as high risk or who fail to achieve CR after 2 courses or who relapse may enter any current NCRI high risk AML trial or leave the trial and enter the European Paediatric Poor Risk Trial 2001/01.

## 20 SUPPORTIVE CARE

The remission induction and consolidation phases of therapy are intensive and will be associated with a risk of infection and haemorrhage. The care of patients will make stringent demands on supportive care. Some information regarding aspects of supportive care will be collected in the patient record books, since this will be one factor to be taken into account in assessing the schedules.

Participants should have local supportive care protocols. It is considered that policies related to the following aspects should be decided in advance to ensure that treatment-related complications are minimised.

1. Venous access via Hickman-type catheter
2. Control of nausea and vomiting
3. Mouth care
4. Prophylactic gut decontamination (if considered appropriate)
5. Antifungal prophylaxis
6. Response to a significant pyrexia — i.e. two readings of  $\geq 38^{\circ}\text{C}$  two hours apart, or a single reading  $\geq 39^{\circ}\text{C}$
7. Antibiotic treatment of febrile episodes — including antibiotic choice(s) and monitoring, duration of therapy, and the treatment of non-response
8. G-CSF therapy [Lenograstim 263  $\mu\text{g}$  (1 vial) s.c. daily in adults or 5 $\mu\text{g}/\text{kg}$  i.v. in children] may be given in case of prolonged neutropenia but it is **not** intended that it should be part of routine supportive care
9. Irradiated blood products should be given to patients who receive Fludarabine or Stem Cell Transplant.

## 21 CNS TREATMENT

### 21.1 Adults

The routine administration of treatment to the central nervous system is not recommended for patients with no evidence of CNS disease at diagnosis.

Patients who present with CNS disease may be entered into the trial and be randomised at the same points as patients without obvious CNS involvement. If a patient presents with physical signs suggesting CNS disease, an intrathecal injection of Cytosine Arabinoside (50 mg) should be given when the diagnostic lumbar puncture is performed. If blast cells are identified in the CSF sample, a series of intrathecal injections with Cytosine Arabinoside should be given on 3 days each week until CSF samples are clear. This may need to be modified if the platelet count is very low or coagulation is abnormal. Thereafter treatment should be repeated at intervals of approximately 2 weeks until consolidation treatment has been completed.

### 21.2 Children

A lumbar puncture should be performed at the time of diagnosis in all children. CNS disease is defined by the presence of  $>5 \times 10^6/l$  leukaemic blasts in a CSF cytopsin preparation.

All children who enter the Paediatric AML Poor Risk Protocol 2001/01 after Course 2 or who relapse on treatment should complete their CNS therapy already started on AML15 as outlined below under '**No CNS disease**' if they have not already done so. Patients who relapse after completing CNS prophylaxis need not have further CNS prophylaxis but should have a diagnostic lumbar puncture performed to exclude active CNS disease. Patients found on diagnostic LP to have active CNS disease for the first time should follow the protocol (see below) for '**CNS disease at diagnosis**'. Patients relapsing in the CNS with active CNS disease for a second time should be discussed with the trial co-ordinators.

#### 21.2.1 No CNS disease

If there is no evidence of CNS disease at diagnosis patients should receive a total of two courses of "triple" intrathecal chemotherapy, one after each of the first two courses of chemotherapy.

### Triple Therapy

AGE (years)	METHOTREXATE	CYTARABINE	HYDROCORTISONE
<1	5mg	15mg	5mg
1	7.5mg	20mg	7.5mg
2	10mg	25mg	10mg
3+	12.5mg	30mg	12.5mg

#### 21.2.2 CNS disease at diagnosis

If CNS disease is present at diagnosis, patients should receive two courses of “triple” intrathecal therapy (as in Section 21.2.1 above) each week until the CNS is clear, plus two further courses. A minimum of six courses should be given in a period of three weeks following diagnosis.

This intensive phase is followed by monthly courses of the same “triple” therapy until after the final course of systemic chemotherapy has been completed.

Children aged 2 years or over with CNS disease not receiving allo-SCT should receive cranial irradiation (2400 cGy) after the final course of chemotherapy. The need for cranial irradiation in children presenting with CNS disease and still less than 2 years old on completion of systemic chemotherapy should be discussed with the clinical coordinators.

## 22 RELAPSE

Relapse will be diagnosed either on morphological or cytogenetic grounds. Studies will be conducted within AML15 to assess the significance of detecting recurring disease by molecular genetics. The management of disease detected molecularly, in the absence of morphological or cytogenetic relapse is unclear and should be discussed with the coordinators.

Adults who relapse from first CR **should be entered into the current NCRI high risk AML trial** (if available) if an attempt to induce second remission is considered appropriate.

The "Relapse" form (Section F) from the patient's record book should be completed giving details of the relapse, subsequent therapy and its outcome. This form should be returned to BCTU when all the necessary data are available, or entered using the online data entry system.

## 23 MODIFICATIONS FOR CHILDREN

There are a number of differences between the treatment of adults and children within the AML15 protocol. These are indicated in the relevant sections and are summarised below.

## 23.1 Treatment Variations

- Children with APL and children with Down syndrome and AML are not eligible for AML 15. Guidelines for the treatment of these patients are available from the paediatric co-ordinators or the Trial Office.
- Children will only be randomised between MRC consolidation and high-dose Ara-C at 3.0 g/m<sup>2</sup>.
- Only poor risk children are eligible for allogeneic SCT, which may be sibling or unrelated.
- SCT will follow the recommendation of the UKCCSG BMT Subcommittee (copies available from the paediatric co-ordinators or the Trial Office).
- Cardiotoxicity should be assessed using the UKCCSG guidelines (copies available from the paediatric co-ordinators or the Trial Office).
- If a patient is not in CR after Course 2, he/she is off protocol and can enter the European Paediatric AML Poor Risk Protocol 2001/01.
- All children will receive CNS prophylaxis (Section 21.2).
- Etoposide should be administered as a 4 hour infusion.
- Idarubicin should be administered as a 1 hour infusion.
- G-CSF should be given intravenously.
- Add on studies will include an investigation of the Fanconi Anaemia Pathway in childhood AML (Dr S Meyer, Manchester) and the DCLSG studies will be carried on from AML12.

## 23.2 Management Group

### Clinical Coordinators

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### Cytogenetics Coordinator

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## **Morphology**

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## **24 STATISTICAL CONSIDERATIONS**

### **24.1 Patient numbers**

Over the last 40 years, 5-year survival of younger patients in MRC AML trials has gone from 0% in AML4 to about 45% in AML12. This dramatic improvement, which has changed AML from an invariably fatal disease into a potentially curable one, has been achieved not by any single major advance but through a series of small, but nonetheless important, increases in survival over a number of trials. It is therefore unrealistic to expect any of the treatment options in AML15 to lead to improvements in survival of more than 10%, while even a 5% increase would be worth knowing about. In order to be able to detect improvements of this order, large trials are needed. For example, to demonstrate, at a 2-tailed  $p=0.05$ , a proportional improvement of about 20% in five-year survival from 45% on one treatment to 55% on the other (a 10% absolute difference) requires approximately 1000 patients to have a 90% chance of detecting this difference. If, however, a smaller — but still worthwhile — 15% proportional improvement in survival from 45% to 52% is to be detected, this would require approximately 3000 patients to have a 90% chance of detecting this difference.

There are approximately 700 cases of AML under the age of 60 diagnosed each year in the British Isles, of whom about 15% have the APL sub-type. It is hoped that the majority of suitable patients will be entered into the trial. Indeed, towards the end of AML12, over 500 patients were entered annually. Thus, if AML15 can recruit successfully for about 5 years, over 2500 patients will be entered.

Over 2000 of these patients will not have APL and could be randomised to the induction options. Two primary comparisons are specified: ADE versus DA. With at least 1000 patients in each comparison, this will give good power to detect differences between these treatments: 90% to detect at  $2p=0.05$  a 10% difference in survival (from 45% to 55% at 5 years). If during the course of the trial it becomes clear that sufficient patients have been randomised to answer one of the questions (see Section 24.2, DMEC), the trial could be modified to introduce one or more new induction questions.

It is likely that only about 25-30% of patients will have a FLT3 mutation, and therefore be eligible for the CEP-701 randomisation. This means that over the course of the trial, it is unlikely that more than 250-300 patients will enter this randomisation. Survival is worse in patients with a FLT3 ITD mutation, with an analysis of patients from AML10 and AML12 giving an overall 5-year survival rate of only around 30%. At 80% power, the sample size collected would be sufficient to detect an absolute improvement in survival of around 15-17% (to 45-47% at five years). In the light of the history of progress in AML, with small but consistent improvements in survival, to expect a proportional reduction in mortality of one third may be overoptimistic, but, as with the 4 vs. 5 course randomisation in AML12, the results of AML15 can be combined in a meta-analysis with future trials to provide a definitive answer.

In AML12, there was considerable drop-out of patients before the consolidation randomisations, so accrual to these randomisations was about 200 patients per annum. With  
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earlier randomisation in AML15 (after Course 2, not Course 3), there will be less patient drop out. However, there will be patients with a sibling donor who will be going down the allograft route and will not be available for the chemotherapy randomisations. Taking this into account it is still envisaged that at least 1000 patients will be entered into the AML15 consolidation randomisations (MACE+MidAC versus HD-Ara-C 1.5 g/m<sup>2</sup> versus HD-Ara-C 3.0 g/m<sup>2</sup>; Mylotarg versus not). This will give 90% power to detect (at 2p=0.05) a 10% difference in survival (60% to 70%) in the MACE + MidAC versus HD-Ara-C (at either dose) and Mylotarg versus control comparisons, and about 70% power to detect a similar difference in the comparison of HD-Ara-C at 1.5 g/m<sup>2</sup> versus HD-Ara-C 3.0 g/m<sup>2</sup>. To reduce the potential for multiple testing, two primary comparisons have been specified: MACE + MIDAC v. HD-Ara-C (at either dose), with a subsidiary comparison of the two Ara-C doses. Patients will be allocated to MACE + MidAC, HD-Ara-C 1.5 g/m<sup>2</sup> and HD-Ara-C 3.0 g/m<sup>2</sup> in a 2:1:1 ratio (children will be allocated to MACE + MidAC and HD-Ara-C 3.0 g/m<sup>2</sup> in a 1:1 ratio).

By the time of the 4 v. 5 courses randomisation, there will have been further patient drop out, so it is unlikely that this comparison will accrue more than 750 patients in total (150 per year), giving about 80% power to detect a 10% survival difference. This randomisation is a continuation of the similar one in AML12, so meta-analysis of the two studies will have over 2000 patients and thus very good power to detect any worthwhile difference in survival.

It is estimated that at least 400 patients will have a sibling donor and will be considered suitable for allogeneic transplant, with a much larger number having no suitable donor. These numbers will give a power of at least 80% to detect (at 2p=0.05) a 10% survival difference in the donor versus no donor biological randomisation.

For the APL question, about 375 patients (75 each year) will be available. With this number, there will be reasonable power to investigate the quality of life and economic aspects of the comparison. The power to detect a 10% difference in survival between MRC and Spanish approaches will only be about 60%, although the power to detect a 15% difference will be about 90%. APL patients are eligible for the Mylotarg consolidation randomisation, so will help increase the power of this comparison.

In the absence of interactions between treatments, the sample size for each comparison in a factorial design is not different from that in a simple two arm trial addressing (inefficiently) a single question. Empirical evidence from previous MRC AML trials suggests that such interactions are rare, but if they were to be observed the sample size would need to be adjusted upwards somewhat. Given the good power for most comparisons, this will not be a problem.

It is not realistic to expect any of the treatments in AML15 to improve outcome by more than 10%, and a moderate benefit of this magnitude would nevertheless be clinically important. The trial will be monitored regularly by the DMEC (see Section 24.2) so, if clear differences were to be observed in one or more of the comparisons, they would bring this to the attention of the Trial Steering Committee.

Most previous trials in leukaemia, and indeed in most disease areas, have been too small to give reliable answers, so the large size of AML15 is essential to provide the high quality of evidence needed to guide future practice.

Even if AML15 is not large enough on its own to provide completely reliable answers to all the questions addressed, the possibility exists to undertake analyses in conjunction with other trials that have addressed similar questions (i.e. meta-analyses).

## 24.2 Data analysis

Interim analyses of the main endpoints will be supplied periodically, in strict confidence, to the MRC Leukaemia Data Monitoring and Ethics Committee (DMEC). In the light of these interim analyses, the DMEC will advise the chairman of the Trial Steering Committee if, in their view, one or more of randomised comparisons in the trial have provided proof beyond reasonable doubt\* that for all, or for some, types of patient one treatment is clearly indicated or clearly contraindicated.

The main analyses will be performed using standard contingency table and log-rank methods based on the intention to treat — i.e. **all** patients believed to be eligible at the time of randomisation will be included in the analysis, irrespective of protocol compliance, early death, etc. The randomisations — and subsidiary data analyses — will be stratified by age (0-14, 15-29, 30-39, 40-49, 50-59, 60+), performance status, and type of disease (*de novo*/secondary AML). Consolidation randomisations will also be stratified by initial allocation and by risk group. All analyses will assume that there may be some **quantitative** differences in the size of any treatment effects in these different strata, but that there is unlikely to be any **qualitative** difference (i.e. harm in one group, benefit in another).

## 25 TRIAL GOVERNANCE AND ADVERSE EVENT REPORTING

Investigators have obligations described in the MRC handbook “MRC Guidelines for Good Clinical Practice in Clinical Trials”. In the use of unlicensed drugs the trial is conducted under a CTA issued by the MHRA which requires the investigators to report Serious Adverse Events (SAEs). The trial will be monitored by an independent Data Monitoring and Ethics Committee.

### 25.1 ADVERSE EVENT REPORTING

Principal Investigators at each participating institution have an obligation to report relevant Serious Adverse Events (SAEs) which occur in this trial to the trial office in a timely manner. It is recognised that adverse events which may be life-threatening are a normal consequence of acute myeloid leukaemia or its effective treatment, and many clinical changes in the patient’s condition are expected.

#### Definitions:

For the purpose of this trial a **Serious Adverse Event** is defined as:

- Development of a non-haematological toxicity of grade 3 as defined in the NCI Common Toxicity Criteria\*\*, which does not resolve to grade 2 or less within 7 days
- Development of any grade 4 non-haematological toxicity (excluding alopecia)

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\* Appropriate criteria of proof beyond reasonable doubt cannot be specified precisely, but a difference of at least three standard deviations in an interim analysis of a major endpoint may be needed to justify halting, or modifying, a randomisation prematurely. If this criterion were to be adopted, it would have the practical advantage that the exact number of interim analyses would be of little importance, and so no precise schedule is proposed.

\*\* A copy of the NCI Common Toxicity Criteria is available from the Trial Office and the trial website.

- Development of neutropenia ( $<1.0 \times 10^9/l$ ) or thrombocytopenia ( $<50 \times 10^9/l$ ) for longer than 42 days after the end of chemotherapy in the absence of significant disease in the bone marrow ( $>5\%$  blasts)
- Any event which results in persistent or significant disability or incapacity
- Any event which results in a congenital abnormality or birth defect
- Death in the absence of persistent or progressive disease

The following **do not** require to be reported as **SAEs**:

- Grade 4 haematological toxicity is an expected consequence of effective treatment, and is only required to be reported if it fulfils the criteria as defined above.
- Patients may present with some pre-existing toxicities which meet the criteria set out above, but it is only the *development* of these toxicities after entering the trial which should be reported.
- Neutropenic fever is an expected severe adverse event which may occur as a result of the disease or the treatment. This or its consequences do not have to be reported unless fulfilling the criteria set out above.

### **Causality**

Investigators will be asked to record their opinion as to whether the SAE as defined above was related to the study medication. This will be further reviewed by the Trial Management Group.

### **Collection of Data**

Preliminary discussion of the event may take place with a clinical co-ordinator. SAEs should be recorded on the Adverse Event Form which is available on the trial website, and sent to the Trial Office at BCTU.

### **Time of Report**

Any death that is clearly **not** due to, or associated with, persistent or progressive disease should be reported to the trial office within 24 hours.

### **Reporting to the Regulatory Authorities**

The Chief Investigator or his nominee will review and record all SAEs. He will be responsible for reporting the events to the MHRA, COREC, and the Trial Steering Committee in the appropriate timelines. He will also report, where relevant, to the provider of the IMP (Investigational Medicinal Product) and produce periodic reports for all investigators to forward to the LREC.

## APPENDIX A: WHO Histological Classification of Acute Myeloid Leukaemias

	ICD Code
<b>Acute myeloid leukaemia with recurrent genetic abnormalities</b>	
Acute myeloid leukaemia with t(8;21)(q22;q22); (AML1(CBF $\alpha$ )/ETO)	9896/3
Acute myeloid leukaemia with abnormal bone marrow eosinophils Inv(16)(p13q22) or t(16;16)(p13;q22); (CBF $\beta$ /MYH11)	9871/3
Acute Promyelocytic leukaemia (AML with t(15;17)(q22;q12-21), (PML/RAR $\alpha$ ) and variants.	9866/3
Acute myeloid leukaemia with 11q23 (MLL) abnormalities	9897/3
<b>Acute myeloid leukaemia with multilineage dysplasia</b>	9895/3
<b>Acute myeloid leukaemia and myelodysplastic syndromes, therapy-related</b>	9920/3
<b>Acute myeloid leukaemia not otherwise categorised</b>	
Acute myeloid leukaemia minimally differentiated	9872/3
Acute myeloid leukaemia without maturation	9873/3
Acute myeloid leukaemia with maturation	9874/3
Acute myelomonocytic leukaemia	9867/3
Acute monoblastic and monocytic leukaemia	9891/3
Acute erythroid leukaemias	9840/3
Acute megakaryoblastic leukaemia	9910/3
Acute basophilic leukaemia	9870/3
Acute panmyelosis with myelofibrosis	9931/3
Myeloid sarcoma	9930/3
<b>Acute leukaemia of ambiguous lineage</b>	9805/3
Undifferentiated acute leukaemia	9801/3
Bilineal acute leukaemia	9805/3
Biphenotypic acute leukaemia	9805/3

## APPENDIX B: Preparation, Administration and Toxicity of Drugs used in AML15

### **Daunorubicin** (Cerubidin<sup>TM</sup> - May & Baker Ltd)

Daunorubicin is presented as a red powder in glass vials containing 20 mg with mannitol as a stabilising agent. The drug is reconstituted in sodium chloride 0.9% or water for injection. Following reconstitution, further dilution with sodium chloride 0.9% to a concentration of 1mg/ml is recommended. The resultant solution is given by a one hour infusion into a swiftly flowing drip. In children Daunorubicin should be administered as a 6 hour infusion. For hepatic dysfunction with a bilirubin 20-50  $\mu\text{mol/l}$  reduce by 25%; for bilirubin >50  $\mu\text{mol/l}$  reduce by 50%. In patients with renal impairment dose reduction should take place: Serum Creatinine 105-265, reduce dose by 25%; Serum Creatinine >265 reduce dose by 50%.

Side effects include nausea, alopecia, chronic and acute cardiac failure and dysrhythmias. Subcutaneous extravasation may cause severe tissue necrosis.

### **Cytosine Arabinoside** — Ara-C, Cytarabine (Cytosar<sup>TM</sup> – Pharmacia & Upjohn)

Cytosar is available as a freeze dried powder containing 100 mg or 500 mg of Cytosine Arabinoside in a rubber capped vial. The diluent provided in the drug pack is water for injection containing 0.9% w/v benzyl-alcohol. Following reconstitution with the manufacturer's diluent the solution contains 20 mg/ml of Cytosine Arabinoside. At this concentration it is suitable for direct intravenous bolus injection into a central or peripheral line.

Cytarabine solution is also available in a non-proprietary form from Pharmacia & Upjohn and Faulding DBL. These are presented as 20mg/ml and 100mg/ml solutions of cytarabine in a variety of vial sizes. It is recommended that before administration by intravenous bolus injection the hypertonic 100mg/ml solution is further diluted in water for injection, sodium chloride, 0.9%, or glucose, 5% solution, to produce a solution of 20mg/ml concentration. In patients with impaired hepatic function (bilirubin >34 $\mu\text{mol/l}$ ) the dose should be reduced by 50%. No reductions are necessary for renal impairment.

Side effects at the doses prescribed for remission induction include nausea, diarrhoea, oral ulceration and hepatic dysfunction. A Cytosar syndrome has also been described. It is characterised by fever, myalgia, bone pain, occasional chest pains, maculopapular rash, conjunctivitis and malaise. It usually occurs 6-12 hours following administration, and is more common with higher doses.

### **Etoposide** (VP16-213) (Vepesid<sup>TM</sup> - Bristol-Myers Pharmaceuticals)

Etoposide is available as a solution in ampoules containing 100 mg in 5 ml. The appropriate dose should be diluted in sodium chloride 0.9% for infusion. The company recommend that the infusion solution concentration does not exceed 0.25 mg/ml of etoposide.

**Give over at least 30 minutes as hypotension may be produced by excessively rapid infusion.**

For hepatic dysfunction with bilirubin of 26-51 $\mu\text{mol/l}$  reduce dose by 50%. At higher bilirubin levels the decision to administer is a clinical one. For renal dysfunction dose reduction should be: Creatinine Clearance (Cr Cl) of 60ml/min the dose should be 85%; Cr Cl of 45ml/min reduce dose by 20%; for a Cr Cl of 30ml/min reduce dose by 25%.

Side effects include tissue necrosis if extravasation should occur, nausea, mucositis and alopecia. Anaphylactic-like reactions have been reported rarely and have responded to stopping the infusion and the administration of an antihistamine and hydrocortisone.

**Gemtuzumab Ozogamicin** — Mylotarg™ (CMA-676), Wyeth Genetic Institute  
MYLOTARG (gemtuzumab ozogamicin for Injection) is supplied as an amber glass vial containing 5mg of MYLOTARG lyophilised powder. This vial should be refrigerated (2-8°C).

### **Preparation**

The drug product is light sensitive and must be protected from direct and indirect sunlight and unshielded fluorescent light during the preparation and administration of the infusion. **All preparation should take place in a biologic safety hood with the fluorescent light off.** Reconstitute the contents of each vial with 5ml Water for Injection. Gently swirl each vial. Each vial should be inspected to ensure dissolution and for particulates. (The final concentration of drug in the vial is 1mg/ml). This solution may be stored refrigerated (2-8° C) and protected from light for up to 8 hours. (Reconstituted vials of drug should not be frozen.) Before administration, withdraw the desired volume from each vial and inject into a 100ml IV bag of 0.9% Sodium Chloride Injection. Place the 100ml IV bag into an UV protectant bag. The following time intervals for reconstitution, dilution, and administration should be followed for storage of the reconstituted solution: reconstitution ≤ 2 hours; dilution ≤ 16 hours at room temperature; administration; 2 hour infusion; i.e. **a total of a maximum of 20 hours.**

### **Administration**

#### **DO NOT ADMINISTER AS AN INTRAVENOUS PUSH OR BOLUS**

Once the reconstituted Mylotarg™ is diluted in 100ml sodium chloride 0.9% for infusion, the resulting solution should be infused over 2 hours. Prior to infusion inspect visually for particulate matter and discoloration.

**A separate IV line equipped with a low protein-binding 1.2-micron terminal filter must be used for administration of the drug** (see note). MYLOTARG may be given peripherally or through a central line.

Premedication, consisting of an antihistamine (such as chlorphenamine), should be given before each infusion to reduce the incidence of a post-infusion symptom complex. Vital signs should be monitored during infusion and for four hours following infusion.

### **Instructions for Use, Handling and for Disposal**

Procedures for handling and disposal of cytotoxic drugs should be applied.

### **Cautions**

**Hepatic Insufficiency:** Patients with hepatic impairment will not be included in the clinical studies.

**Renal Insufficiency:** Patients with renal impairment will not be included in the clinical studies.

## Note

The recommended in-line filter for Mylotarg administration is a 1.2-micron polyether sulfone (PES) filter, e.g. “intrapur lipid” (Braun product number 4099702). If that filter is not available, the following filters may be used: 0.22 micron PES, 0.20 micron cellulose acetate, 0.8 to 1.2 micron cellulose acetate/cellulose nitrate (mixed ester), or 1.2 micron acrylic copolymer.

## Adverse Events

The most important serious adverse event may be hepatotoxicity or myelosuppression. These should be reported to the Chief Investigator as described in Section 25.1. Other events which have been reported in at least 10% of recipients of single agent Mylotarg include fever, nausea, chills, vomiting, headache, dyspnoea, hypotension, and hyperglycaemia. It is not necessary to report these events.

## Amsacrine (Amsidine™ - Parke Davis Co.)

Amsacrine is presented as two sterile liquids which are combined immediately prior to use. The drug ampoule contains 1.5 ml of amsacrine, a bright orange-red liquid at a concentration of 50 mg/ml. The diluent vial contains 13.5 ml of 0.0353 M L-lactic acid. When 1.5 ml (75mg) of concentrated amsacrine is added to 13.5 ml of lactic acid diluent the resulting solution **contains 5 mg/ml of amsacrine** (i.e. 75mg in 15ml).

It is recommended that preparation of the drug should be carried out using a **GLASS SYRINGE** due to possible extraction of components of rubber or certain plastic material. The solution should be added to 500 ml of **5% dextrose** (in adults) and infused over a period of 60 – 90 minutes. **Amsacrine is incompatible with sodium chloride 0.9%**. In patients with impaired hepatic function (bilirubin > 34µmol/l) the dose should be reduced to 60%. If renal function is reduced (Cr Cl < 60ml/min) the dose should be reduced by 25%.

**Side-effects:** Some nausea and mucositis occur fairly frequently. Cardiac toxicity has been described as with anthracyclines; the risk of arrhythmias is increased by hypokalaemia. Hepatotoxicity is uncommon but is associated with a rise in serum bilirubin and alkaline phosphatase. Phlebitis may be a problem with peripheral venous access points and local necrosis is described. The risk of phlebitis can be decreased by a slow rate of infusion. The degree of alopecia is variable but sometimes severe.

## Mitoxantrone (Novantrone™ - Lederle laboratories)

Mitoxantrone is presented as a dark blue aqueous solution in vials of 20 mg, 25 mg and 30 mg (2 mg/ml) with saline and a buffer of sodium acetate and acetic acid.

The required dose should be diluted to at least 50ml in 0.9% saline or 5% dextrose. It should be injected slowly (over >5 minutes) into a fast flowing infusion of 5% dextrose or 0.9% saline. (Alternatively the solution can be diluted in at least 50ml and given by short intravenous infusion). In children Mitoxantrone should be given as a 6 hour infusion. In hepatic dysfunction with a bilirubin > 60µmol/l maximum dose should be 8mg/m<sup>2</sup>.

Side effects include tissue necrosis following extravasation outside a vein. It is probably slightly less cardiotoxic than daunorubicin but care should be taken to avoid low serum

potassium levels. Anorexia, diarrhoea, stomatitis, fatigue and mild alopecia have also been described.

### **Idarubicin** (Zavedos™ - Pharmacia)

Idarubicin is available as a sterile pyrogen-free, orange-red freeze-dried powder, in vials containing 5 or 10 mg of idarubicin hydrochloride with 50 or 100 mg of lactose respectively.

For administration the vial contents should be dissolved in water for injection to give a solution of 1mg/ml. The resultant solution should be administered intravenously into the side arm of a freely running intravenous infusion of 0.9% sodium chloride over 5 to 10 minutes. In children Idarubicin should be given as a 1 hour infusion.

In cases with hepatic dysfunction dose reduction is required: bilirubin 21 – 34µmol/l reduce the dose by 50%. Greater rises contraindicate administration. For renal impairment with a serum creatinine 100 – 175µmol/l reduce the dose to 50%. Administration at higher creatinine levels is a clinical decision.

Side-effects: The major side effect is myelosuppression. Cardiac toxicity may occur, manifested by cardiac failure, arrhythmias or cardiomyopathies, either during therapy or several weeks later. The cumulative dose associated with cardiotoxicity is not known, but it is believed that a total dose of 60-80 mg/m<sup>2</sup>, which is considerably higher than that used in AML15, is not problematic. Idarubicin may cause a red discoloration of the urine for 1-2 days after administration. Reversible alopecia will occur, and some nausea or vomiting and oral mucositis should be expected. Elevation of liver enzymes and bilirubin may occur in a minority of patients.

Idarubicin should not be given to patients with severe renal or liver impairment.

### **Cyclophosphamide** (Endoxana™ – ASTA Medica)

Endoxana is available as a powder in vials containing 100 mg, 200 mg, 500 mg or 1000 mg of anhydrous cyclophosphamide and sufficient sodium chloride to render the reconstituted solution isotonic. The vial should be reconstituted with a suitable volume of Water for Injection to produce a 20mg/ml solution. This solution can then be administered by slow intravenous bolus injection or further diluted for infusion. The dose should be reduced in renal impairment: for GFR 10-50ml/min reduce by 25%; for GFR <10 the dose should be reduced by 50%.

Side-effects: Cystitis, mucositis, nausea and vomiting, and hypoglycaemia and hyperglycaemia may occur.

### **Fludarabine** (Fludara™ - Schering-Plough)

Fludara contains 50mg fludarabine phosphate per vial. It should be given by slow intravenous infusion after dilution in 2ml Water for Injection.

For hepatic dysfunction no dose change is required. For renal impairment a Cr Cl of 30 – 70 ml/min requires a dose reduction of 50%; greater impairment excludes administration.

The most frequent adverse events are myelosuppression. Patients less commonly suffer nausea, vomiting or alopecia. Fludarabine is a prolonged inhibitor of T-cells and has been

associated with the development of transfusional GVHD and pneumocystis pneumonia. Rarely, fludarabine has caused CNS side-effects with agitation, confusion and visual disturbance.

#### **ATRA** (Vesanoid™ - Roche Products)

The most common adverse effect of ATRA has been headaches of mild to moderate severity. Younger (paediatric) patients appear to be more sensitive to this particular effect. Bone pain, occasionally requiring analgesic treatment, has also been observed. Biochemical abnormality of liver function has occasionally been reported, specifically raised transaminases, alkaline phosphatase and bilirubin, but these are reversible on stopping the drug.

The most serious adverse event has been a syndrome characterized by fever, respiratory distress and episodic hypotension, usually in association with leucocytosis (now known as "ATRA Syndrome"). The onset of this syndrome has usually been in the first 1-2 weeks of drug treatment. Should this occur the ATRA should be stopped. Some cases are reported to respond well to high-dose corticosteroid therapy (dexamethasone 10 mg i.v. 12 hourly for 3 or more days).

Prolonged ATRA treatment may cause dryness of the skin. ATRA is also believed to be highly teratogenic.

#### **G-CSF**- Human Granulocyte Colony-Stimulating Factor: (Granocyte™ - rHuG-CSF, lenograstim - Chugai Pharma UK Limited)

Granocyte is available as lyophilised powder, each single use vial containing either 105µg of lenograstim (13.4 MIU rHuG-CSF) or 263µg of lenograstim (33.6 MIU rHuG-CSF). A pre-filled syringe of Water for Injection (1 ml) for each vial of Granocyte is provided for reconstitution before administration. Granocyte can now be stored at room temperature, up to 30°C.

Dose in adults:

In autologous PBPC mobilisation:	1 vial/day sc or as per local protocol
In allogeneic PBPC mobilisation:	10µg/kg/day for 4-6 days
Post BMT:	1 vial/day sc or as per local protocol
Chemotherapy induced neutropenia:	1 vial/day sc days 1-7

Dose in children: 5µg/kg up to a maximum of 1 vial (263µg) should be administered intravenously over 1 hour.

Bone pain and injection site reaction have been associated with Granocyte treatment in some patients.

Granocyte is distributed by Aventis UK, telephone 08705 133347, Fax: 08705 133329.

#### **Arsenic Trioxide** (Trisenox™ - Cephalon Inc.)

Trisenox is 1mg/ml concentrate for solution for infusion (arsenic trioxide). It is presented as a sterile, clear, aqueous solution in a single-use 10ml ampoule. ATO is a trivalent inorganic arsenical. The active substance is a white crystalline powder that is very poorly soluble in water.

Trisenox must be diluted with 100-250 ml of glucose (5%) injection or sodium chloride 9mg/ml (0.9%) injection immediately after withdrawal from the ampoule and must not be mixed with or concomitantly administered in the same intravenous line with other medicinal products.

Aseptic technique must be strictly observed throughout the handling of Trisenox since no preservation is present.

After dilution in intravenous solutions, Trisenox is chemically and physically stable for 24 hours at 15-30° C and 48 hours at refrigerated temperatures (2-8°C). From a microbiological point of view, the product must be used immediately. If not used immediately in-use storage times and conditions prior to use are the responsibility the user and would normally not be longer than 24 hours at 2-8°C, unless dilution has taken place in controlled and validated aseptic conditions.

Trisenox is given as a slow infusion over 1-2 hours daily until bone marrow remission is achieved. The daily infusions should be given on an inpatient basis at the beginning of induction therapy, followed, when the acute symptoms of APL have resolved and the patient's condition is stable, by outpatient administration for the remaining induction and consolidation treatment period.

**CEP-701 (Cephalon Inc.)**

The study drug should be stored, under adequate security, in the pharmacy at the study centre at a controlled room temperature of 20°C to 25°C (68°F to 77°F), protected from light, until taken by the study patients or until returned to Cephalon or its designated agent. The study drug is stable for at least 30 months from the day of manufacture if stored in amber vials, protected from light, at a controlled room temperature below 25°C

## APPENDIX C: Background Information on CEP-701

### Name and Description of Investigational Product

CEP-701 is an orally bioavailable receptor-tyrosine kinase inhibitor. The compound is a chemically synthesized derivative of K-252a, a fermentation product of *Nonomurea longicatena*, and belongs to a class identified as indolocarbazole alkaloids. CEP-701 drug product is supplied in a carrier of polysorbate 80 NF and propylene glycol USP, 25 mg CEP-701/ml, in 100-ml amber glass bottles. A more detailed description of the product is provided in Section 10.2.

### Findings From Nonclinical and Clinical Studies

#### Nonclinical Pharmacology

CEP-701 exhibits most potency in the inhibition of the neurotrophin receptor Trk A (tropomyosin receptor kinase) (concentration at which 50% of the enzyme is inhibited [IC<sub>50</sub>] 3 nM) and the fms-like tyrosine kinase, FLT3 (IC<sub>50</sub> 3 nM). In addition, CEP-701 can be classed as a moderately potent inhibitor of vascular endothelial growth factor receptors (VEGFR) 1 to 3 (IC<sub>50</sub> 37 to 76 nM), mixed lineage kinases 1 to 3 (IC<sub>50</sub> 11 to 71 nM), and c-Jun N-terminal kinases 1 $\beta$ 1 (IC<sub>50</sub> 81 nM), 2 $\alpha$ 2 (IC<sub>50</sub> 113 nM), and 3 $\alpha$ 1 (IC<sub>50</sub> 31 nM). It displays weaker inhibitory activity against platelet-derived growth factor receptor  $\beta$  (IC<sub>50</sub> 216 nM), rat brain protein kinase C (IC<sub>50</sub> 226 nM), and fibroblast growth factor receptor (IC<sub>50</sub> 420 nM). CEP-701 does not show appreciable inhibition of epithelial growth factor receptor or the  $\beta$ -insulin receptor kinase. When added to intact cells under plasma-free conditions, CEP-701 inhibits TrkA and FLT3 with IC<sub>50</sub> of 25 and 1 to 2 nM, respectively.

The inhibition of Trks by CEP-701 may be of value as therapy for both prostate cancer and pediatric neuroblastoma, where this family of tyrosine-receptor kinases is believed to play a role in tumour cell survival and/or invasive growth. Treatment of rats bearing the Dunning H rat prostate cancer with CEP-701 caused a significant regression of established tumours, (Cephalon data on file). CEP-701 was also shown to inhibit the growth of DU145, PC3, and CWR22Rv1 human androgen-independent prostate cancer xenografts in nude mice.

Studies with human neuroblastoma cell lines have been performed with CEP-751, a close analog of CEP-701 that is metabolized to CEP-701 in vivo. Treatment of nude mice bearing such xenografts with CEP-751 resulted in a significant inhibition of tumour growth.

#### Nonclinical Pharmacokinetics

Pharmacokinetic parameters for intravenous (iv) CEP-701 are similar in rats and dogs, with a plasma clearance of 0.8 l/hr-kg, a volume of distribution of approximately 1.4 l/kg, and an elimination half-life (t<sub>1/2</sub>) of 1.2 hours. Orally administered CEP-701 is rapidly absorbed in both rats and dogs, with peak plasma concentrations occurring within 2 to 4 hours of dosing. In dogs, the absolute oral bioavailability of CEP-701 from a vehicle composed of polysorbate 80 and propylene glycol is 39%. The oral bioavailability of CEP-701 from solution formulations in dogs is more than 10-fold higher than in rats. In vitro, CEP-701 binds with high affinity to human alpha-1-acid glycoprotein (hAGP), resulting in an estimated unbound, biologically active fraction of less than 1% in human plasma. The binding appears saturable. In an intact cell assay, hAGP (1 mg/ml) shifted

the IC<sub>50</sub> value of CEP-701 for TrkA 750-fold, from 8 nM to 6000 nM. In the same assay, 2 samples of human plasma caused 250- and 500-fold shifts in this IC<sub>50</sub> value. The significance of these findings with respect to the clinical activity of CEP-701 is not known; however; they most probably explain the need to attain micromolar plasma concentrations of CEP-701 in order to observe biological effects in patients.

CEP-701 is primarily metabolized via hydroxylation and glucuronidation. Results from an in vitro study with cDNA-expressed human cytochrome P450 enzymes (CYP) indicate that CYP3A4, CYP1A2, and CYP2B6 may be involved in the metabolism of CEP-701. CEP-701 inhibits several CYP isoforms in human liver microsomal preparations, with Ki values below 5 µM (approximately 2 µg/mL) for CYP1A2, CYP2C9, and CYP3A4; and 10 µM (approximately 4 µg/mL) for CYP2C19. The potential for CEP-701 to inhibit these CYP isoforms in vivo has not been assessed. Caution regarding concomitant administration of drugs that are substrates for these CYP isoforms, and especially those with narrow therapeutic indices, would be prudent.

### Nonclinical Toxicology

The toxicity of CEP-701 has been evaluated in single-dose oral administration studies in mice, rats, and dogs (Table C1). In addition, repeated-dose oral administration studies have been completed in rats (6 months) and dogs (9 months). In the single-dose studies, the lethal dose at which 10% of mice died after experiencing adverse events (LD<sub>10</sub>) was 73 mg/kg, and an LD<sub>50</sub> of 103 mg/kg was determined in rats. A maximum tolerated dose (MTD) of 100 mg/kg was determined in dogs.

**Table C1: Summary of Key Toxicology Parameters**

Species	Parameter	Dosage (mg/kg/day)	Dosage (mg/m <sup>2</sup> /day)	Human dosage <sup>a</sup> (mg/day)
Mouse	LD <sub>10</sub>	73	219	379
Rat	LD <sub>50</sub>	103	618	1069
Dog	Acute MTD	100	2000	3460
Rat	6-month MTD	20	120	208
Dog	9-month MTD	5	100	173

<sup>a</sup> Extrapolated data, assuming a body surface area of 1.73 m<sup>2</sup>.

LD<sub>10</sub> = the dose at which 10% of the species experienced adverse events that resulted in death.

LD<sub>50</sub> = the dose at which 50% of the species experienced adverse events that resulted in death.

MTD = maximum tolerated dose.

In a 6-month study, rats received CEP-701 by oral gavage at 0.8, 4, or 20 mg/kg/day. There was no compound-related mortality. A reduction in body weight gain was observed in all dose groups relative to the deionised water control group, including the CEP-701 vehicle group. However, the most prominent reduction in body weight gain (9% to 11% relative to vehicle) was noted in the 20-mg/kg/day CEP-701 dose group. No CEP-701-related changes were noted in ophthalmology, haematology, serum chemistry, or urinalysis data. Changes interpreted to be related to administration of CEP-701 were detected microscopically in the epithelium of the anorectal junction, caecum, extrahepatic

bile duct, and the main pancreatic duct. Microscopic changes were described as “epithelial cell alteration” (bile duct, pancreatic duct, and caecum) and inflammation (anorectal junction). Treatment-related changes in these epithelia were subtle, did not occur in all treated animals/sections of affected groups, and, in some sites, were morphologically similar to microscopic changes that occurred in some control animals. No treatment-related changes were found histologically in rats following a 6-week recovery period. On the basis of the results of this 6-month oral toxicity study, 20 mg/kg/day of CEP-701 was identified at the MTD in rats.

In a 9-month study, dogs received CEP-701 by oral gavage at dosages of 0.2, 1, and 5 mg/kg/day. There was no compound-related mortality or morbidity. An increased incidence of emesis was noted in both male and female dogs in the 5.0 mg/kg/day dose group relative to dogs in both control groups. In general, increased salivation was noted in male and female dogs only in the 5.0 mg/kg dose group. There were no other compound-related clinical observations. There were no compound-related changes noted in body weights, food consumption, ophthalmology, haematology, serum chemistry, urinalysis, electrocardiograms, or organ weight data. Changes interpreted to be related to CEP-701 were detected microscopically in the mucosa of the anorectal junction of both male and female dogs in the 5.0 mg/kg/day dose group. Many dogs from the other groups, including the deionised water and vehicle control groups, also had evidence of inflammation within the anorectal mucosa. However, in the high-dose group, the inflammation was more severe and there was evidence of hyperplastic changes within deeper layers of the anorectal epithelium. In addition, the high-dose group had a shift in cell populations that included large numbers of neutrophils, especially in the squamous mucosa of the anal area. There were no new treatment-related histologic changes following the 9-week recovery period. Basal cell hyperplasia also resolved, indicating that the chronic irritation with CEP-701 administration had subsided. On the basis of the results of this 9-month oral toxicity study, 5 mg/kg/day of CEP-701 was identified as the MTD in dogs.

In addition to these findings, it is noted that a dosage of 15 mg/kg/day in dogs was found to be associated with significant toxicity when administered for 28 days. Clinical signs included hunched posture, decreased activity, weight loss, vomiting, and increased salivation. Faecal occult blood was increased in these animals, and gastrointestinal haemorrhage was noted in some males. Four of the 10 dogs dosed at 15 mg/kg/day were euthanized *in extremis* prior to the completion of the 28-day study. Terminal serum chemistry findings in these animals were consistent with cholestasis, and hepatic inflammation and necrosis. Treatment-associated lesions were observed throughout the gastrointestinal tract as well as in the liver, gallbladder, bile duct, prostate, and thymus. The day-28 toxicokinetics revealed a maximum plasma concentration ( $C_{max}$ ) of 385 ng/ml in males and 606 ng/ml in females, and an area under the plasma concentration-time curve from time zero to 24 hours ( $AUC_{0-24}$ ) of 1365 and 733 ng-hr/ml, respectively.

A battery of *in vitro* and *in vivo* tests for genotoxicity has been conducted with CEP-701. CEP-701 was negative in the bacterial point (Ames) assay and the *in vivo* aneuploidy/micronucleus assay. CEP-701 was positive in the mouse lymphoma mutagenesis assay (–S9 5.0 µg/ml, +S9 2.5 µg/ml), and also caused increased percentages of polyploidy (–S9  $\geq 0.4$  µg/ml) and endo-reduplicated cells (+S9  $\geq 5.0$  µg/ml) in the Chinese hamster ovary cell chromosome aberration assay. In a chromosome aberration assay with human peripheral blood lymphocytes (HPBL), CEP-701 caused an increase in the number of

polyploidy cells ( $-S9 \geq 0.38 \mu\text{g/ml}$ ,  $+S9 \geq 2.0 \mu\text{g/ml}$ ). In a separate study with HPBLs, CEP-701 was able to induce micronuclei ( $-S9 0.74 \mu\text{g/ml}$ ,  $+S9 1.4 \mu\text{g/ml}$ ). Subsequent investigation into the mechanism of action via the application of a pain-centromeric DNA probe indicated that micronuclei were generated via an aneugenic mechanism.

Ancillary pharmacology studies of CEP-701 suggest little potential for interaction with the cardiovascular and gastrointestinal smooth muscle systems. Effects on renal excretion and interactions with the central nervous system were apparent only at oral doses of 100 mg/kg or above.

## Clinical Experience

### Healthy Subjects

The safety and pharmacokinetics of single doses of oral CEP-701 from 3 to 20 mg were evaluated in healthy subjects (study M97-792). Nine subjects were treated at each dose; 6 received active drug and 3 received a placebo.  $C_{\text{max}}$  of CEP-701 ranged from 114 to 1041 ng/ml, mean AUC from 929 to 7889 ng·h/ml, and mean apparent  $t_{1/2}$  from 6.8 to 9.2 hours. There was no significant departure from dose proportionality. In the second stage of the study, in which 12 subjects received 10 mg CEP-701 under fasting and non-fasting conditions, the time to maximum plasma concentration ( $t_{\text{max}}$ ) was lengthened from 1.8 hours to 3.7 hours ( $p < 0.001$ ) and  $C_{\text{max}}$  reduced from 403 to 294 ng/mL ( $p = 0.002$ ) when CEP-701 was taken with food. CEP-701 was well tolerated and treatment-emergent adverse events were mild in nature (headache, sweating, constipation). No clinically significant abnormal haematology, chemistry, urinalysis, or stool laboratory values were observed. In a separate study (study M99-067), the tolerability of oral CEP-701 vehicle (polysorbate 80 and propylene glycol, USP) was assessed in 24 healthy subjects who were administered twice-daily volumes of 3.2 to 9.6 ml of the vehicle diluted to 30 ml in apple juice for 14 days. Results indicated that the vehicle alone was well tolerated at volumes used in clinical studies.

### Patients With Cancer

The safety and/or efficacy of CEP-701 in cancer patients has been explored in 5 Phase 1 or Phase 2 studies. In a pilot study, CEP-701 was administered preoperatively for 5 days at 40 mg bd to 9 men with localized prostate cancer who had elected to have radical prostatectomy (study M99-108). In 5 patients who completed the study, the mean plasma concentration of CEP-701 was 427.9 ng/ml immediately prior to surgery, and the mean prostate tissue concentration was 97.1 ng/g, indicating reasonable tissue penetration.

The MTD and dose-limiting toxicities (DLTs) of repeated doses of CEP-701 were defined in a study of patients with advanced solid tumours (study M98-909). A total of 30 patients were enrolled at twice-daily doses of 5 mg ( $n=3$ ), 10 mg ( $n=3$ ), 20 mg ( $n=3$ ), 40 mg ( $n=13$ ), 80 mg ( $n=7$ ), and 120 mg ( $n=1$ ). The majority of patients (67%) received a single 28-day cycle of CEP-701. However, 7 patients received study drug for at least 3 months, including 3 patients who were treated for more than 6 months. One patient received 13 cycles of treatment. The most frequently reported adverse events were nausea (63%), diarrhoea (47%), anorexia (37%), asthenia (30%), constipation (27%), and vomiting (27%). The incidence of adverse events tended to

be greater in patients who received at least 40 mg bd. Most of these events were intermittent, but lasted several days.

Dose-limiting toxicities were reported for 1 patient at 80 mg bd (grade 3 nausea) and 1 patient at 120 mg bd (grade 3 hypotension). The formal definition of MTD was not met for either of these dosages, but treatment-related adverse events, especially of the gastrointestinal system, made CEP-701 poorly tolerated at 80 mg and above. After expanding the MTD group (40 mg bd) to 13 patients, DLTs were reported in 2 patients at this dosage (grade 3 anorexia, nausea, dyspepsia and grade 3 asthenia). Thus, the MTD of CEP-701 without antiemetic prophylaxis was considered to be 40 mg bd. However, in view of the intermittent nature of many of the gastrointestinal events, the tolerability of higher dosages (60 and 80 mg bd) is considered worthy of further study when CEP-701 is given with appropriate antiemetic support.

The pharmacokinetics of CEP-701 in patients with cancer were found to be similar to those in healthy subjects. CEP-701 was rapidly absorbed, with mean  $t_{max}$  values ranging from 0.8 to 2.7 hours on days 1 and 28 across all dosages. At 40 mg bd, the day-28 mean  $C_{max}$  was 3973 ng/mL, and the mean  $AUC_{0-12}$  was 26630 ng·hr/mL. At 80 mg bd, the day-28 mean  $C_{max}$  was 12117 ng/mL, and the mean  $AUC_{0-12}$  was 114857 ng·hr/ml. Plots of dose-normalized CEP-701  $AUC_{0-12}$  on days 1 and 28 indicated greater than dose-proportional increases in  $AUC_{0-12}$  after multiple doses. As a result there was greater accumulation of CEP-701 with bd administration at higher doses, with mean accumulation factors of 0.9 at 5 mg, 1.7 at 10 and 20 mg, 3.0 at 40 mg, and 4.1 at 80 mg.

CEP-701 did not produce an objective tumour response in any patient. The median duration of treatment was 5 weeks. Three patients had stable disease for more than 6 months and 1 of the patients with small cell lung cancer was stable for almost a year.

In study M99-051, a 40-mg bd dosage of CEP-701 was shown to be ineffective in maintaining or lowering prostate specific antigen concentrations in patients with hormone-refractory prostate cancer. Eight-week trough plasma concentrations of 1538 ng/ml (n=37) were similar to plasma concentrations observed in earlier studies. Nausea and diarrhea were the most common adverse events, observed in 42% and 30% of patients, respectively. Severe gastrointestinal events (nausea, vomiting, abdominal pain, diarrhoea) considered possibly or probably related to CEP-701 were recorded for 7 of the 159 patients enrolled in the study. In addition, there were 4 reports of moderate to severe gastrointestinal hemorrhage, of which 1 event was considered possibly related and the other 3 unrelated to study drug.

Preliminary findings are available from 2 recently completed studies. In the first study (study C0701a/102/ON/US), 19 patients with advanced pancreatic cancer received CEP-701 at 20 and 40 mg bd in combination with a standard regimen of gemcitabine. The study was closed early because of lack of efficacy. Preliminary assessment of safety data indicated that the combination of CEP-701 with gemcitabine did not result in significant additive toxicity. A single serious adverse event of acute renal failure was considered possibly related to CEP-701. The second study (study C0701a/202/

ON/US) explored the activity of CEP-701 in 18 patients with refractory or relapsed AML with activating mutations of the receptor tyrosine kinase FLT3. The starting dosage of CEP-701 was raised in this study from 40 mg bd to 60 mg bd after the first 4 patients showed no response and a cell-based ex vivo assay of plasma indicated that the inhibition of the FLT3 target may have been suboptimal. Fourteen patients were subsequently enrolled at 60 mg bd, and the dosage for 3 patients was increased to 80 mg bd after approximately 1 month of treatment. The higher dosages of 60 and 80 mg bd appeared to be relatively well tolerated in this group of patients. At a dosage of 60 mg bd, ex vivo assay indicated that a high degree of inhibition (>90%) of the FLT3 target was maintained over the 12-hour interval between doses. Transient decreases in the number of peripheral blast counts and the recovery of elements of the normal peripheral cell population were documented in several patients. Serious adverse events of gastrointestinal haemorrhage, fatigue and congestive heart failure in 1 patient each were considered possibly related to CEP-701. These events were also considered possibly related to the patients' disease and/or prior treatment.

### **Known and Potential Risks and Benefits to Human Subjects**

The principal treatment-related side effects of CEP-701 are gastrointestinal in nature (nausea, diarrhoea, anorexia, constipation, and vomiting). Asthenia has also been reported. Severe events are relatively uncommon, and clinical experience suggests that the nausea and vomiting should respond to 5-HT<sub>3</sub> receptor antagonist antiemetics. These effects are consistent with the gastrointestinal signs and symptoms observed in repeat-dose toxicology studies in animals. More severe findings of cholestasis and hepatic inflammation that were observed in the 28-day dog toxicology study have not been manifested in the clinical program to date. Gastrointestinal haemorrhage has been reported in a small number of patients. It is not known to what extent these hemorrhagic events are related to CEP-701 treatment.

The benefits of CEP-701 to patients remain to be determined. Transient decreases in peripheral blast count have been observed in some patients with refractory AML treated with CEP-701. A scientific rationale, based on in vitro studies and animal cancer model data, exists to support the potential benefit of inhibiting FLT3 in patients with AML and of inhibiting Trks in patients with prostate cancer and neuroblastoma.

Additional information regarding risks to human subjects may be found in the Investigator's Brochure.

## APPENDIX D: Instructions to the Patient/Caregiver for Administering CEP-701

### General Instructions

1. Store at room temperature of 20 to 25°C (68–77°F). Protect from light. Keep used and unused vials in original vial box. (Note: For centres outside the US, solutions of study drug in juice should be stored below 25°C.)
2. Keep Out of Reach of Children. For Oral Administration Only.
3. Return all used and unused bottles in original box at each study visit.
4. CEP-701 may be taken with food. If you vomit after a dose, do not repeat this dose. You should take your next dose at the regular scheduled time.
5. Administer this study drug in the morning and in the evening, approximately every 12 hours (there must be at least 8 hours between doses).
6. See your physician or pharmacist if you have any questions.

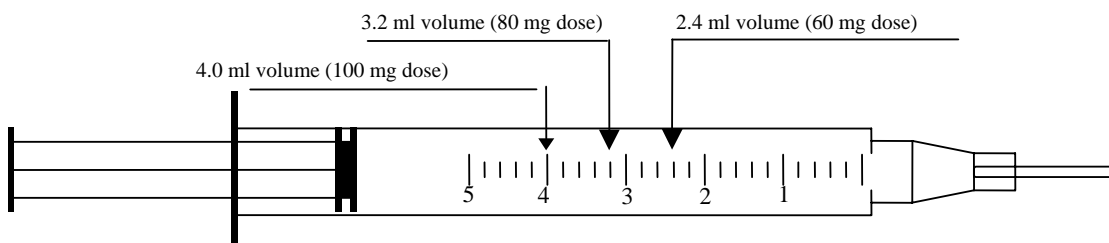
### Withdrawal of CEP-701 solution from 100ml bottle

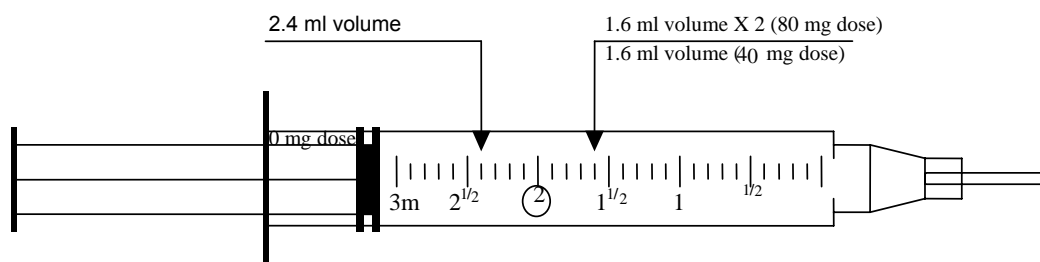
1. Remove the child resistant cap from the bottle and attach the push-in bottle adaptor to expose the rubber stopper.
2. Insert the 5ml oral syringe through the adaptor.
- 3.
4. Turn the bottle upside down and withdraw the prescribed amount of study drug solution (1.6 ml, 2.4 ml, 3.2 ml, 4.0 ml, or per label on bottle box) from the bottle into the syringe by pulling down on the syringe plunger.

Note: There may be a vacuum created in the bottle which will make it difficult to pull down plunger. If this occurs, the vacuum can be removed by first inserting the syringe with the plunger already pulled out or completely removed from the syringe barrel.

If air bubbles or air gaps are visible in the syringe, remove air by pushing the plunger back into syringe until all of the air is forced out of the syringe and into the bottle. Then pull down on the plunger to withdraw the prescribed amount of study drug solution. See diagram.

5. Remove the syringe from the bottle. Leave the adaptor on the bottle.





## Preparation of Diluted CEP-701/Juice Solution and Administration

1. Dispense ENTIRE contents of syringe into plastic dosing cup containing 30 ml (1 ounce) of approved juice. The following juices or cocktails are approved for use for the administration of the study drug: grape, pineapple, apple, or V8® 100% vegetable juice, orange juice (pulp-free).

CEP-701 should NOT be taken with grapefruit juice or drinks containing grapefruit juice.

2. Mix or stir the solution for approximately 1 minute or until well mixed. Do not shake the solution.
3. Drink the diluted solution immediately after mixing.
4. Rinse dosing cup 3 times with 30 ml (1 ounce) of juice and drink juice after each rinse. Discard syringe and dosing cup.

### Number of Doses Available From Each 20-ml Vial of CEP-701 Solution

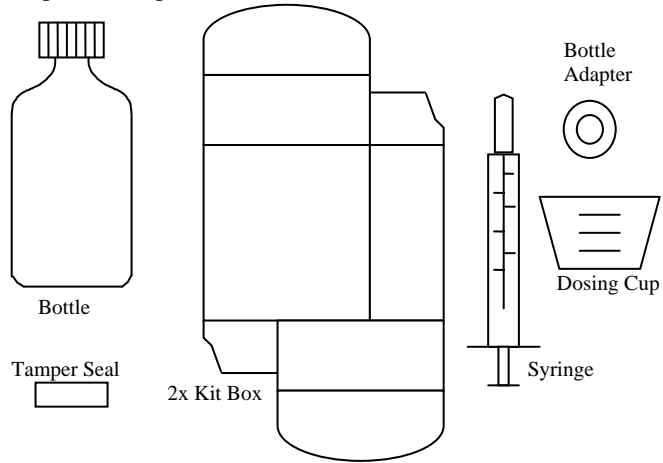
Dose of CEP-701 (mg bd)	Volume of CEP-701 stock solution dispensed per dosing syringe (ml)	Minimum number of doses (days) per vial	Maximum number of doses (days) per vial
60	2.4	6 (3)	8 (4)
80	3.2	5 (2.5)	6 (3)
100	4.0	4 (2)	4 (2)

The patient/caregiver will prepare each dose as instructed by the study investigator/research nurse, using syringes to withdraw the appropriate volume of CEP-701 as instructed. Patients/caregivers should be instructed to add the entire contents of the syringe to 30 ml of the approved juice in a dosing cup. Patients should then drink an additional 90 ml of juice (3 x 30 ml in the dosing cup). (See above for patient instructions.) The investigator/research nurse will ensure that the patient/caregiver is able to demonstrate competence in preparing CEP-701 prior to receiving the drug supply. CEP-701 may be taken with food. If the patient vomits after receiving a dose, the dose should not be re-administered and the patient should take the next dose at the scheduled time. The minimum time between doses is 8 hours.

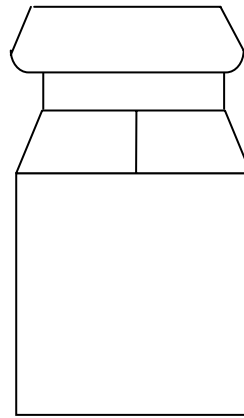
Solutions of study drug in juice may be stored (protected from light) for up to 1 hour at room temperature (approximately 20°C to 25°C). (Note: for centres outside the US,

solutions of study drug in juice should be stored below 25°C.) Solutions may be stored up to 8 hours refrigerated at approximately 2°C to 8°C.

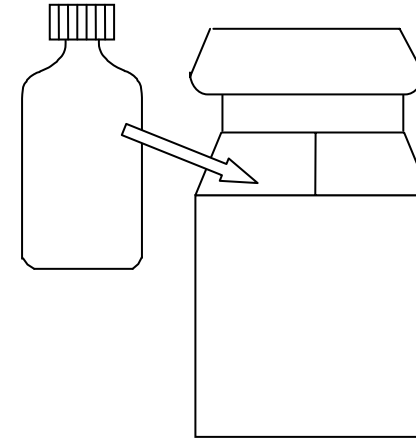
**Step 1:** Obtain (1) CEP-701 25mg/mL Bottle (1) 2x Kit Box, (1) 5ml Syringe, (1) 4oz. Bottle Adapter, (1) 30ml Dosing Cup, (2) Tamper Seals



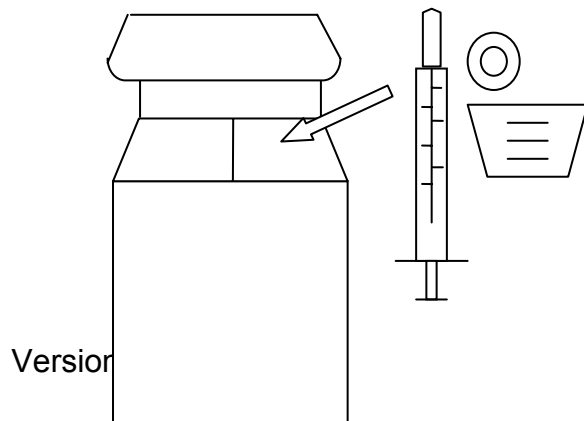
**Step 2:** Assemble 2x Kit Box leaving one end open



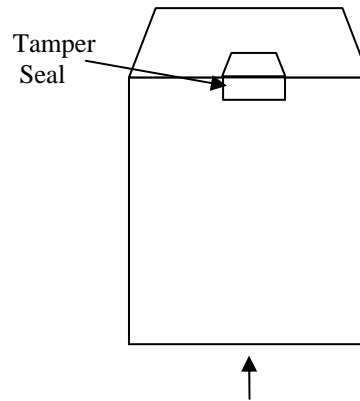
**Step 3:** Place Bottle of CEP-701 into left side of 2x Kit Box



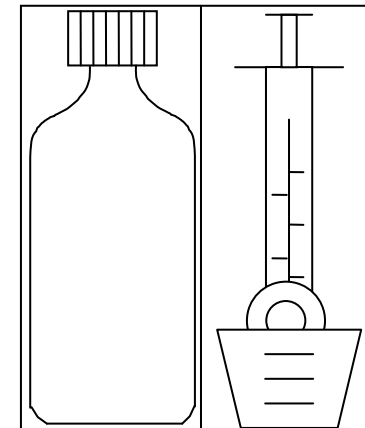
**Step 4:** Place 5ml Syringe, 4 oz. Bottle Adapter, and 30mL Dosing Cup into right side of 2x Kit Box.



**Step 6:** Close top of 2x Kit Box and secure both ends with Tamper Seals



**CROSS SECTION EXAMPLE OF INSIDE COMPLETED KIT**



Version

Tamper Seal (bottom/centre/rear)

(bottom/centre/rear)

## **APPENDIX E: Procedures for Bone Marrow Transplantation**

### **Pre-transplant investigations**

Centres will wish to perform their own pre-transplant investigations but the following are strongly recommended because they may reveal possible contraindications for proceeding with marrow-ablative therapy.

1. Bone marrow aspiration to confirm remission (ABSOLUTELY ESSENTIAL)
2. Chest x-ray
3. ECG
4. MUGA scan or Echocardiogram
5. Lung function studies

### **Pre-graft ablative therapy with TBI and cyclophosphamide**

The patient should receive allopurinol 300 mg/day for at least two days before the cyclophosphamide. One of the most distressing and dose-limiting side-effects of cyclophosphamide is haemorrhagic cystitis. This may be prevented by MESNA, a compound that inactivates toxic metabolites of cyclophosphamide in the bladder. Patients should also receive intensive hydration during the giving of cyclophosphamide and TBI.

### **Cyclophosphamide**

#### **Dosage**

Cyclophosphamide is administered at a dose of 60 mg/kg for each of 2 successive days (use lean body weight for obese patients). It is dissolved in 250 ml of 5% glucose and administered over 60 min. Following the cyclophosphamide a clear 24 hours should elapse before TBI commences. The marrow is thawed and reinfused within 24 hours of completing TBI whether the TBI was given by single or multiple fractions.

#### **MESNA**

During cyclophosphamide administration MESNA is given in 4 divided doses by i.v. push at time 0 (time of commencement of cyclophosphamide), time +3 hours, and +6 and +9 hours. Each dose of MESNA is 40% of the total dose of cyclophosphamide, i.e. the total MESNA dose is 160% of the total cyclophosphamide dose. Each individual dose of MESNA must be prescribed separately and the time of administration clearly noted. The hydration regimen (up to 3l/m<sup>2</sup>/day), unless used with MESNA, is itself insufficient to prevent cystitis.

#### **Diuresis**

Adequate urine flow must be maintained before and following cyclophosphamide administration to prevent urate nephropathy and haemorrhagic cystitis. All patients should receive i.v. fluids at twice the maintenance rate beginning at 6-12 hours before the cyclophosphamide dose. This will ensure adequate hydration.

#### **Total body irradiation**

TBI procedures cannot be completely standardized throughout the UK because of constraints of machine characteristics and availability. It is recognised that many schedules in use at present are effective and safe, but the adoption of a limited number for this study is recommended to make it possible to evaluate the significance of fractionation



and lung shielding for control of leukaemia and normal; tissue toxicity. This study should not obscure in any way the primary aims of the trial

### **Single fraction TBI**

- No lung shielding
- 1050 cGy if the dose rate is less than 5 cGy per minute.
- 950 cGy if the dose rate is 5-10 cGy per minute.
- 750 cGy if the dose rate is more than 10 cGy per minute.

### **Fractionated TBI**

- 1440 cGy in 8 fractions over 4 days, 180 cGy per fraction.

Treatment will be given using a linear accelerator or cobalt unit operating at the SSD/FSD which gives an adequate, or the largest available, field size. The whole body dose should be defined as the maximum dose to the lung measured by thermoluminescent dosimetry or diodes over 20 minutes for single fraction treatments and for one whole fraction for fractionated treatments. Patient separations will be taken at, and calculation of dose made for, the following sites:

Lung  
Abdomen (at umbilicus)  
Pelvis

Additional measurements can be made at the discretion of the participating clinician. No lung shielding will be used and the prescribed dose will be that to the lung. Compensators may be used to give homogenous whole body dose if required: doses will then be measured under compensators. Depth dose data, built up depth and beam flatness must be determined by phantom measurement at the extended treatment distance. A central review of machine operating data and calculated doses will be undertaken.

**Note:** For patients with initial CNS involvement, additional cranial irradiation (3 x 200 cGy over 3 to 5 days) will be given before TBI using lateral fields encompassing the whole brain down to C2 and including the orbit with shielding of the lens. Additional radiotherapy will not be given to sites of initial bulk disease unless there is persistent extra-medullary disease in one site only which is not thought to be a contra-indication to transplantation. A dose of 1000 cGy in 5 fractions will then be given before TBI.

If you are unable to use TBI ablation please contact one of the transplant coordinators about possible alternatives.

### **Sedation and anti-nausea**

Combinations of metoclopramide (20 mg i.v.), lorazepam (1-3 mg i.v.), ondansetron (8 mg i.v.) or other 5HT antagonists and dexamethasone (10 mg i.v.) may be used.

### **Prevention of infection**

Specific prophylactic measures are not laid down and procedures may vary slightly from centre to centre. Infection prophylaxis is of great importance because of the difficulties in diagnosing and treating infection in immunocompromised patients.

### **Infusion of marrow**

The marrow should be infused intravenously through a normal giving. This may be at any time up to 24 hours following the TBI. Toxicity of the marrow infusion includes volume overload, pulmonary emboli and allergic reactions.

### **Other supportive care**

Red cell or platelet transfusions will be necessary in the period following the graft. It is recommended that platelets be given if the peripheral platelet count is less than  $20 \times 10^9 /l$ . All blood products, including platelets, must be irradiated to at least 2500 cGy post transplant. CMV negative recipients should receive CMV negative blood products whenever possible.

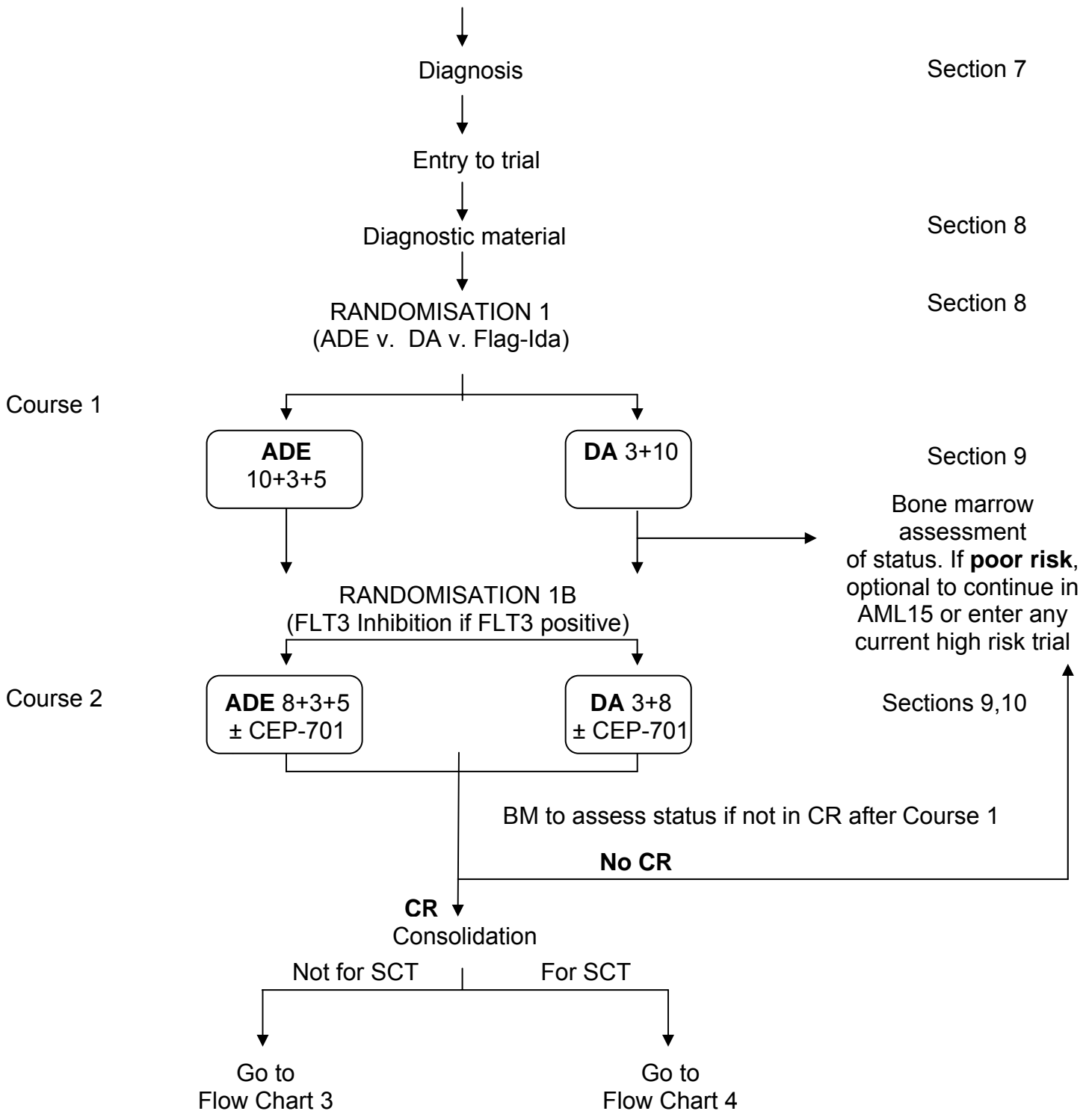
### **GVHD**

Prophylaxis and treatment of graft versus host disease following allo-SCT should follow the practice of the individual transplant centre.

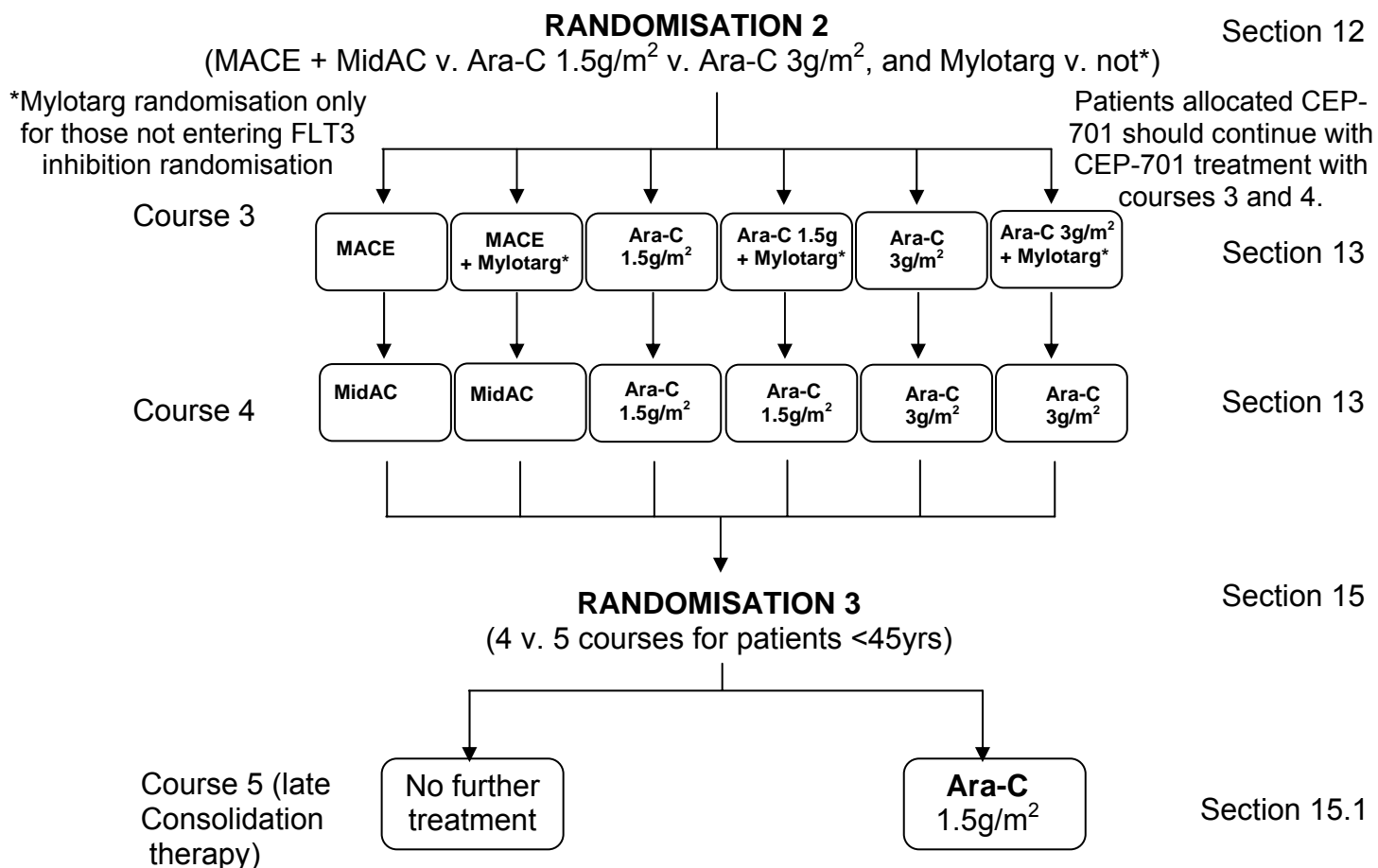
## **APPENDIX F: WHO Play Performance Scale For Children Aged 0-9 Years**

- 100 Fully active, normal.
- 90 Minor restrictions in physically strenuous activity.
- 80 Active but tires more quickly.
- 70 Both greater restriction of, and less time spent, in active play.
- 60 Up and about but minimal active play; keeps busy with quieter activities.
- 50 Gets dressed but lies around most of the day; no active play; able to participate in quiet play and activities.
- 40 Mostly in bed; participates in quiet play and activities.
- 30 In bed; needs assistance even for quiet play.
- 20 Often sleeping; play entirely limited to very passive activities.
- 10 No play; does not get out of bed; unresponsive.

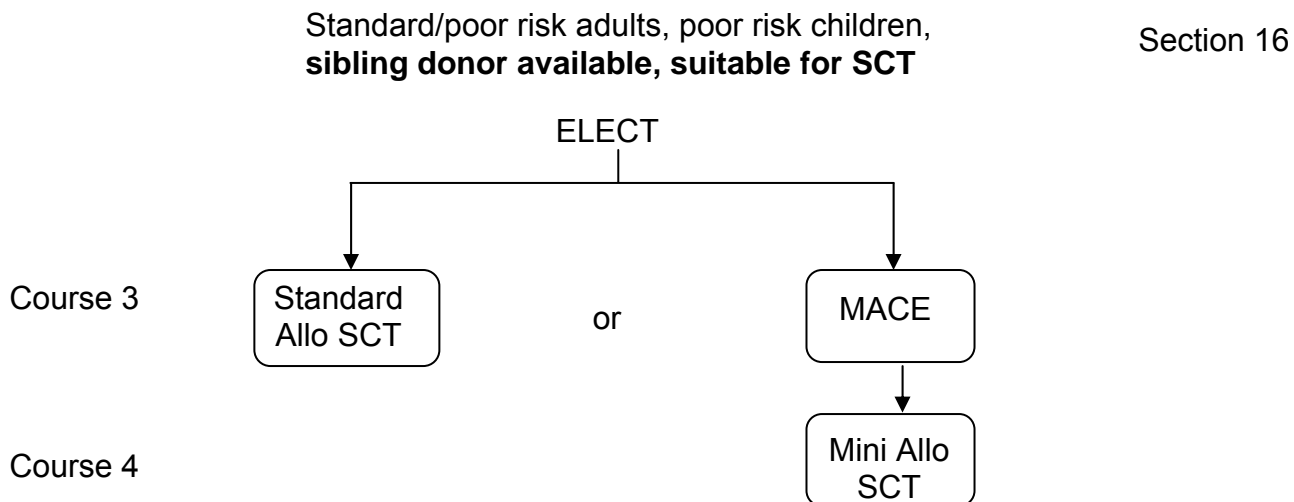
**AML15 Protocol Flow Chart 2**  
 Induction (courses 1 and 2 of treatment) for **NON APL** Patients



**AML15 Protocol Flow Chart 3**  
 Consolidation (courses 3 and 4) for **NON APL** Patients  
 Good/standard/poor risk patients **not for Allograft SCT**

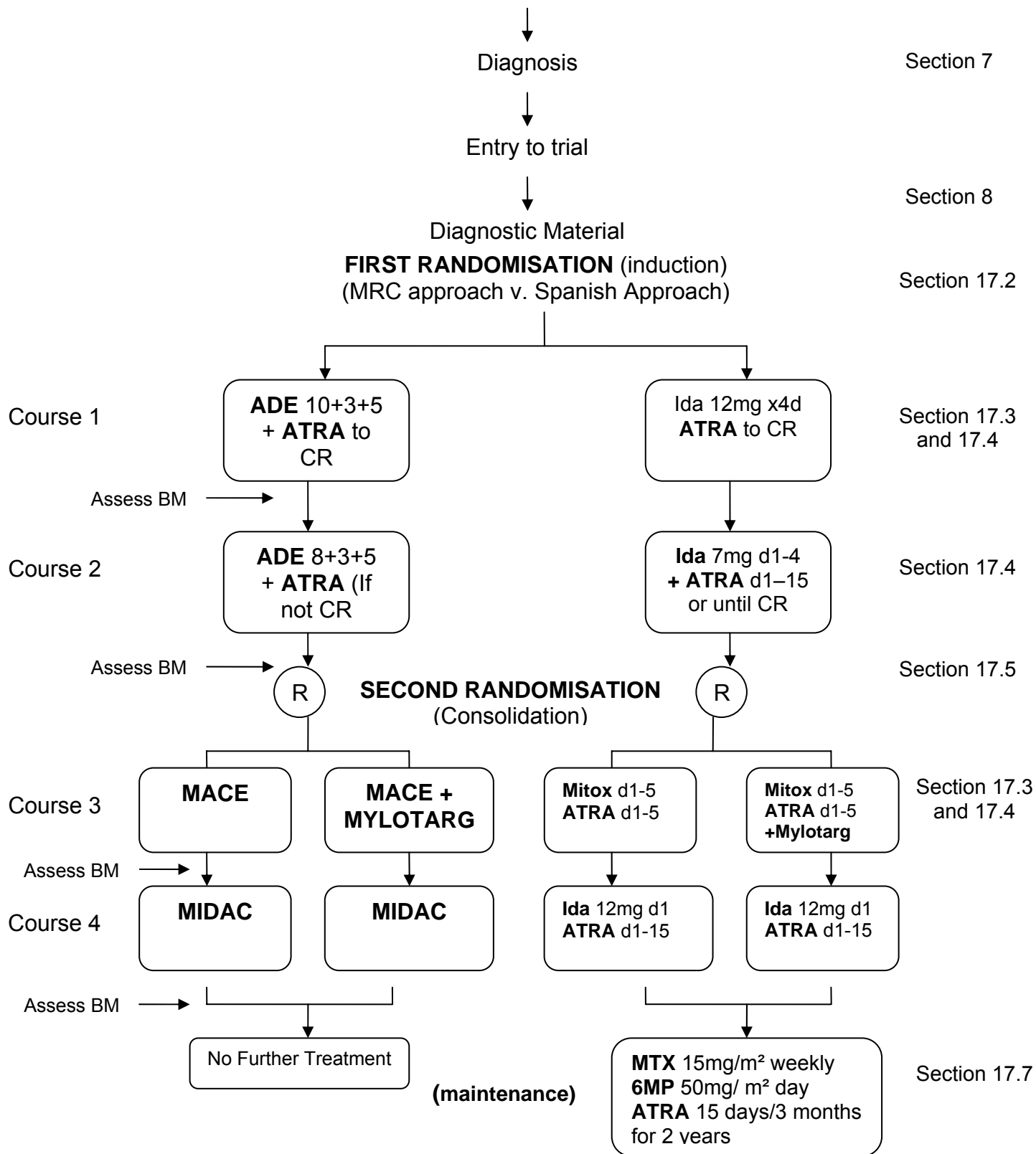


**AML15 Protocol Flow Chart 4**  
 Consolidation for **NON APL** Patients **Elected for Allo SCT**



# Aml15 Protocol Flow Chart 5

Treatment for **APL** Patients



For further Bone marrow assessments see Section 18