MASONIC CANCER CENTER
UNIVERSITY OF MINNESOTA
BLOOD & MARROW TRANSPLANTATION PROGRAM

MT2005-10

TRANSPLANTATION OF UNRELATED UMBILICAL CORD BLOOD FOR PATIENTS WITH HEMATOLOGICAL DISEASES WITH CYCLOPHOSPHAMIDE/FLUDARABINE/TOTAL BODY IRRADIATION MYELOABLATIVE PREPARATIVE REGIMEN

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Revision History

Version Date	Amendment	Details of changes	Consent change?
7/28/05	original		
8/5/05	1A	Clarified criteria for AML eligibility	
9/6/2005	1B	Added to ineligibility section "patients who have received Y-90 ibritumomab (Zevalin) or I-131 tostumomab (Bexxar), as part of their salvage therapy"	
10/28/2005	2	Modified section 6.3.2 MMF to read Patients ≥ **40** kilograms will receive MMF at the dose of 3 grams/day divided into 2 or 3 doses (every 12 or 8 hours). Pediatric patient (<**40** kilograms) will receive MMF at the dose of 15 mg/kg.	
11/22/2005	3	Minor clarifications to clean up typos, change study contact from Meadow Schroeder to Marilee Larkin, removed eligibility checklists and SAE form from appendices Section6.3.2 Clarified that pediatric patients will receive their MMF dosing is three times per day Section 7.2.16, table 7.1 clarified that pediatric pre-transplant evaluation qualification: for children that are not able to cooperate to have a MUGA and/or pulmonary function tests, an echocardiogram should be attempted and pulse oxymetry with exercise tolerance obtained. If not possible at all, it should clearly be documented in the physicians note. Minor edits to table 7.1	
12/21/2005		Minor wording changes to clarify language and minimize chances of protocol deviations.	
4/6/2006		Fixed typo in section 6.2.2. Cyclophosphamide total dose should be measured as mg/kg	
11/1/2006	4	Fred Hutchinson Cancer Research Center will no longer be an affiliate in this study. All references to FHCRC removed.	
4/24/2007		Section 6.2, added noted about the study calendar for subjects co-enrolled on MT2005-01	
8/20/2010	5	Eligibility, section 4.3.1, modified the protocol eligibility to allow LFTs up to 5x ULN	
10/22/2010	6	Revised to remove Appendix IX: cord blood selection algorithm (U of MN Institutional Guidelines will now be a separate document).	
12/16/2011	7	Eligibility modified to allow plasma cell leukemia	No
1/17/2012	8	Eligibility modified to allow subjects up to 55 years, and revised definitions of high risk	No
8/30/2012	9	Added Myeloproliferative Syndromes to the eligibility	No
02/06/2013	10	Schema, section 11: update total enrollment to 300 patients over 10 years (IRB approved Jan 2012);	No

Version Date	Amendment	Details of changes	Consent change?
		Section 7: Remove research samples as patients are co-enrolled on MT2009-22R (immune reconstitution), add standard language regarding flexibility around targeted procedure/treatment dates; Schema: change follow-up to 2 years to match objectives and current institutional procedures; Section 11: delete stopping rules as they are no longer needed as sufficient patients enrollment has occurred; Generalized updating to current institutional template deleting outdated or unnecessary information and referring to institutional guidelines where appropriate	

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PROTOCOL SYNOPSIS

MT 2005-10
TRANSPLANTATION OF UNRELATED UMBILICAL CORD BLOOD FOR PATIENTS WITH HEMATOLOGICAL DISEASES WITH CYCLOPHOSPHAMIDE/FLUDARABINE/TOTAL BODY IRRADIATION MYELOABLATIVE PREPARATIVE REGIMEN

Study design: Single arm non-randomized phase II trial

Primary objective: Determine one year survival

Secondary Objectives: Determine the incidence of 6 month transplant related mortality; chimerism at multiple time points; neutrophil and platelet recovery by day 42 and 6 months, respectively; grades II-IV and III-IV acute GVHD at 100 days; chronic GVHD at 1 year; relapse at 1 year, and probability of progression-free survival for each diagnosis at one and two years.

Eligibility criteria: Subjects must be ≤ 55 years old without an HLA matched sibling donor; absence of recent active mold infection; adequate organ function; performance status >80% (Karnofsky - adults) or >50 (Lansky - children).
UCB graft may be composed of one or two partially HLA matched units. Each unit must be matched at 4-6 HLA loci to the recipient (and to each other if two units are utilized).

Eligible diseases (refer to section 4.2 for detailed disease requirements):
Acute myeloid leukemia (AML)
Very high risk pediatric patients with AML
Acute lymphocytic leukemia (ALL)
Very high risk pediatric patients with ALL Chronic myelogenous leukemia excluding refractory blast crisis
Plasma Cell leukemia
Advanced myelofibrosis. Myelodysplasia (MDS) IPSS Int-2 or High risk (i.e. RAEB, RAEBt) or refractory anemia
Chronic lymphocytic leukemia/small lymphocytic lymphoma (CLL/SLL), marginal zone B-cell lymphoma or follicular lymphoma Lymphoplasmacytic lymphoma, mantle-cell lymphoma, prolymphocytic leukemia Large cell NHL > CR2/> PR2Lymphoblastic lymphoma, Burkitt's lymphoma, and other high-grade NHL Multiple myeloma beyond PR2

Exclusion Criteria: Pregnancy or breastfeeding; evidence of HIV infection or known HIV positive serology; current active serious infection; patients CML in refractory blast crisis; large cell lymphoma and mantle cell lymphoma that is progressive on salvage therapy.

Accrual Objective: 300 patients over 10 years

Duration of Study Participation: The patients will be followed for a minimum of 2 years post-transplant

TREATMENT PLAN

Day	
-8	Fludarabine 25 mg/m^2 IV over 1 hour
-7	Fludarabine 25 mg/m^2 IV over 1 hour Cyclophosphamide 60 mg/kg IV
-6	Fludarabine 25 mg/m^2 IV over 1 hour Cyclophosphamide 60 mg/kg IV
-5	rest
-4	TBI 165 cGy twice daily
-3	TBI 165 cGy twice daily Begin CSA, MMF
-2	TBI 165 cGy twice daily
-1	TBI 165 cGy twice daily
0	Transplant
+1	began G-CSF

1.0 OBJECTIVES

1.1 Primary objective

The primary objective of this study is to determine the one year survival of patients undergoing umbilical cord blood transplantation (UCBT) after a myeloablative preparative regimen consisting of cyclophosphamide (CY), fludarabine (FLU) and fractionated total body irradiation (TBI).

1.2 Secondary objectives

Determine the incidence of transplant-related mortality (TRM) at 6 months after UCBT
Evaluate pattern of chimerism after double UCBT
Determine incidence of neutrophil engraftment at day 42 after UCBT
Determine the incidence of platelet engraftment at 1 year after UCBT
Determine the incidence of acute graft-versus-host disease (GVHD) grade II-IV and grade III-IV at day 100 after UCBT
Determine the incidence of chronic GVHD at 1 year after UCBT
Determine the disease free survival at 1 and 2 years after UCBT
Determine the incidence relapse at 1 and 2 years after UCBT

2.0 BACKGROUND

Bone marrow transplantation (BMT) is a standard treatment option for an increasing number of malignant and non-malignant disorders. To reconstitute hematopoiesis after an intensive myeloablative therapy, the transplantation of pluripotential hematopoietic stem cells (HSCs) is required. Such HSCs are typically recovered from the bone marrow of related or unrelated donors or the bone marrow or apheresed peripheral blood of the patients themselves [1]. Unfortunately, suitable marrow is frequently not available [2]. Either the patient's own marrow is contaminated with tumor cells or potential allogeneic marrow donors are unsuitable most often on the basis of HLA mismatch. Human umbilical cord blood (UCB) is an alternative source of HSCs that is capable of reconstituting hematopoiesis after intensive myeloablative therapy [3-10].

2.1 Umbilical Cord Blood Transplantation (UBCT)

As a result of the early successes with umbilical cord blood transplantation (UCBT) from sibling donors, pilot programs for the banking of unrelated donor UCB were initiated in many countries around the world. Known benefits of banked UCB include: 1) rapid availability, 2) absence of donor risk, 3) absence of donor attrition, and 4) very low risk of transmissible infectious diseases, such as CMV and EBV, and 5) low risk of acute GVHD despite HLA mismatch. UCB is particularly beneficial for patients of ethnic and racial minority descent for whom adult marrow and blood donors often cannot be identified.

2.2 UCBT Experience at the University of Minnesota

2.2.1 Single Unit UCBT [11].

Between 1994 and 2001, 102 consecutive patients (median age 7.4 years) received a single, unrelated UCB unit after a myeloablative conditioning for malignant (n = 65; 68% high-risk) and non-malignant diseases (n = 37). Median infused cell dose of UCB was 3.1 x 10^7 NC/kg (range 0.7-57.9), and 2.8 x 10^5 CD34+ cells/kg (range 0.4-39.1). Fourteen percent had an HLA matched unit and 86% had a 1-3 HLA-match. Neutrophil recovery occurred at a median of 23 days (range 9-54) after UCBT with cumulative incidence of engraftment of 88% (95% CI: 81-95) by day 42. Speed and likelihood of neutrophil recovery were strongly associated with cell dose, with markedly inferior engraftment (72% at a median of 34 days) in patients receiving a CD34+ cell dose <1.7 x 10^5 cells/kg.

The incidences of grade II-IV and III-IV acute GVHD were 39% (95% CI: 29-49) and 11% (95% CI: 5-17), respectively, at day 100, with 10% (95% CI: 4-14) of patients having chronic GVHD at 1 year. One year transplant-related mortality (TRM) was 30% (95% CI: 21-39) which was strongly associated with CD34+ cell dose. The probabilities of 1 and 2-year survival were 58% (95% CI: 49-70) and 47% (95% CI: 36-57), respectively. Importantly, with a graft cell dose ≥1.7 x 10^5 CD34+ cells/kg, survival was 70% (95% CI: 49-90) at 1 year.

The principal conclusions of this study were: 1) an adequate cell dose (≥1.7 x 10^5 CD34+ cells/kg or >2.5 x 10^7 nucleated cells/kg) consistently engraft; 2) GVHD is low despite HLA mismatch; 3) survival and risk of relapse are comparable to that observed after BMT, and; 4) cell dose significantly limits the applicability of UCB, particularly in adult size recipients.

2.2.2 Multi-Unit UCBT [12].

Since cell dose has been clearly identified as a major limitation, often preventing the consideration of UCB for adult recipients, we have explored the possibility of infusing two partially HLA matched units to augment cell dose. Between 2000 and 2005, we have increasingly utilized two UCB units rather than one, particularly in adults and adolescents who cannot find a suitable single UCB unit. The underlying hypothesis of the original study (MT2000-15) was that the addition of the second unit would enhance the engraftment and the speed of hematopoietic recovery.

We have analyzed of the results in the first 31 adult and adolescent patients [median age 24 years (range: 13-53); median weight 73 kg (range: 48-120)] with high-risk hematologic malignancy who were transplanted with two partially HLA-matched UCB units after a myeloablative conditioning. Patient breakdown by diagnosis was: acute myelogenous leukemia (AML, n=15), acute lymphocytic leukemia (ALL, n=12), chronic myelogenous leukemia (CML, n=3) and non-Hodgkin Lymphoma (NHL, n=1). The median total infused dose 3.7 x 10^7 nucleated cells per kilogram (range 1.1-6.3) and 4.9 x 10^5 CD34 per kilogram (range, 0.9-14.5).

In this study, 100% of evaluable patients (i.e., survived for 21 days [n=29]) engrafted at a median of 23 days (range 14-41). This compares favorably with a 65-75% incidence of engraftment reported for adults [9, 10]. In each case, one unit predominated. Thus far, no

factor (nucleated cell dose, CD34+ cell dose, HLA-match, ABO, sex, order infusion) predicts which unit will eventually be responsible for long term marrow recovery. The incidence of platelet recovery (>50,000/uL) was 73% (95% CI, 51-95) at day 180. Incidence of grades II-IV and III-IV acute graft-versus-host disease (GVHD) was 65% (95% CI, 42-88) and 17% (95% CI, 2-32) at day 100. Disease-free survival remains 72% at 1 year for patients transplanted in CR with no relapse in this cohort (median follow up: 1.2 years).

These data indicate that: 1) double unit UCBT is safe in terms of engraftment (bi-directional immune rejection between the units was never observed); 2) one unit always predominates within the first 100 days; 3) >90% of adults will be able to identify two units that are partially HLA matched with the patient and each other; 4) incidence GVHD is similar to that observed after single UCBT; and 5) survival exceeds that of historical data with a single UCB unit.

As a result of these promising early data, we plan to extend this trial in order to 1) confirm the initial observations and 2) establish survival outcomes and relapse risk for individual disease groups.

3.0 STUDY DESIGN

This is a single arm, phase II study to determine the one year survival of patients undergoing umbilical cord blood transplantation (UCBT) after a myeloablative preparative regimen consisting of cyclophosphamide , fludarabine and fractionated total body irradiation (TBI) for the treatment of study specific hematological malignancies.

4.0 PATIENT SELECTION

4.1 Graft Criteria

4.1.1 The unrelated cord blood donor(s) must be 4-6/6 HLA-A, B, DRB1 matched with the recipient (HLA matching using molecular techniques: A and B to antigen level resolution and DR to allele level resolution).

4.1.2 No existing HLA-identical related donor is available.

4.1.3 Suitable UCB units available according to the current University of Minnesota Umbilical Cord Blood Graft selection algorithm. The UCB graft may consist of one or two UCB units.

4.2 Age and Disease Criteria

4.2.1 Patients aged ≤ 55 years must have a hematological malignancy as described below:

4.2.2 Acute myeloid leukemia (AML): high risk CR1 (as evidenced by preceding MDS, high risk cytogenetics, ≥ 2 cycles to obtain CR, erythroblastic or megakaryocytic leukemia;

CR2+. All patients must be in CR as defined by hematological recovery, AND <5% blasts by light microscopy within the bone marrow with a cellularity of ≥15%.

4.2.3 Very high risk pediatric patients with AML. Patients <21 years, however, are eligible with (M2 marrow) with ≤ 25% blasts in marrow after having failed one or more cycles of chemotherapy. This group of patients will be analyzed separately.

4.2.4 Acute lymphocytic leukemia (ALL): high risk CR1 as defined by cytogentics (such as t(9;22), t (1:19), t(4;11), other MLL rearrangements, hypodiploidy, or IKZF1 abnormalities), DNA index < 0.81, > 1 cycle to obtain CR or presence minimal residual disease (MRD). Patients in CR2+ are eligible. All patients must be in CR as defined by hematological recovery, AND <5% blasts by light microscopy within the bone marrow with a cellularity of ≥15%.

4.2.5 Very high risk pediatric patients with ALL. patients <21 years are also considered high risk CR1 if they had M2 or M3 marrow at day 42 from the initiation of induction or M3 marrow at the end of induction. They are eligible once they achieved a complete remission

4.2.6 Chronic myelogenous leukemia excluding refractory blast crisis. To be eligible in first chronic phase (CP1) patient must have failed or be intolerant to imatinib mesylate.

4.2.7 Plasma Cell leukemia after initial therapy, who achieved at least a partial remission

4.2.8 Advanced myelofibrosis

4.2.9 Myelodysplasia (MDS) IPSS Int-2 or High risk (i.e. RAEB, RAEBt) or refractory anemia with severe pancytopenia or high risk cytogenetics. Blasts must be < 10% by a representative bone marrow aspirate morphology.

4.2.10 Chronic lymphocytic leukemia/small lymphocytic lymphoma (CLL/SLL), marginal zone B-cell lymphoma or follicular lymphoma are eligible if there was disease progression/relapse within 12 of achieving a partial or complete remission. Patients who had remissions lasting > 12 months, are eligible after at least two prior therapies. Patients with bulky disease (nodal mass greater than 5 cm) should be considered for debulking chemotherapy before transplant.

4.2.11 Lymphoplasmacytic lymphoma, mantle-cell lymphoma, prolymphocytic leukemia are eligible after initial therapy in CR1+ or PR1+.

4.2.12 Large cell NHL > CR2/> PR2. Patients in CR2/PR2 with initial short remission (<6 months) are eligible.

4.2.13 Lymphoblastic lymphoma, Burkitt's lymphoma, and other high-grade NHL after initial therapy if stage III/IV in CR1/PR1 or after progression if stage I/II < 1 year.

4.2.14 Multiple myeloma beyond PR2. Patients with chromosome 13 abnormalities, first response lasting less than 6 months, or β-2 microglobulin > 3 mg/L, may be considered for this protocol after initial therapy.

4.2.15 Myeloproliferative syndromes

4.3 Organ function and Performance Status Criteria

4.3.1 Recipients must have a Karnofsky score (adults) ≥ 80 % or Lansky play score ≥ 50 (pediatrics) (appendix II) and have acceptable organ function defined as:
Renal: creatinine ≤ 2.0 (adults) or creatinine clearance > 40 ml/min (pediatrics),
Hepatic: bilirubin, AST/ALT, ALP ≤ 5 x upper limit of normal,
Pulmonary function: $DLCO_{corr}$ > 50% normal,
Cardiac: left ventricular ejection fraction ≥ 45%.

4.3.2 Voluntary written consent before performance of any study-related procedure not part of normal medical care.

4.4 Exclusion Criteria

4.4.1 Active infection at time of transplantation (including active infection with Aspergillus or other mold within 30 days.)

4.4.2 History of HIV infection

4.4.3 Pregnant or breast feeding. The agents used in this study may be teratogenic to a fetus and there is no information on the excretion of agents into breast milk. Females of childbearing potential must have a blood test or urine study within 2 weeks prior to registration to rule out pregnancy

4.4.4 Chemotherapy refractory large cell and high grade NHL (ie progressive disease after > 2 salvage regimens)

4.4.5 If ≤ 18 years old, prior myeloablative transplant within the last 6 months. If >18 years old prior myeloablative allotransplant or autologous transplant

4.4.6 Extensive prior therapy including > 12 months alkylator therapy or > 6 months alkylator therapy with extensive radiation.

4.4.7 Patients who have received Y-90 ibritumomab (Zevalin) or I-131 tostumomab (Bexxar), as part of their salvage therapy are not eligible for myeloablative umbilical cord blood transplant.

5.0 PATIENT REGISTRATION

To be eligible for registration to this study, the patient must meet each criteria listed on the eligibility checklist based on the eligibility assessment documented in the patient's medical record.

5.1 Registration with the University of Minnesota Clinical Trials Office (CTO)

Upon completion of the screening evaluation, eligibility confirmation and obtaining written consent, the study coordinator or designee will enroll the patient into OnCore.

5.2 Patients who are registered and do not begin study treatment

If a patient is registered to the study, and is later found not able to begin the planned study treatment, for whatever reason, the patient will be removed from study and treated at the physician's discretion. Study staff will update OnCore of the patient's non-treatment status. Study data will be collected until the time the patient is off study. The reason for removal from study will be clearly indicated on the case report forms.

6.0 TREATMENT PLAN

6.1 Treatment Plan

In order to provide optimal patient care and to account for individual medical conditions, investigator discretion may be used in the prescribing of all supportive care drug therapy (i.e. acetaminophen, diphenhydramine, G-CSF, antimicrobials, etc.).

Day	Preparative Therapy	Supportive Care
-8	Fludarabine 25 mg/m^2 IV over 1 hour	
-7	Fludarabine 25 mg/m^2 IV over 1 hour Cyclophosphamide 60 mg/kg IV	
-6	Fludarabine 25 mg/m^2 IV over 1 hour Cyclophosphamide 60 mg/kg IV	
-5	Rest	
-4	TBI 165 cGy twice daily	
-3	TBI 165 cGy twice daily	Begin CSA, MMF (section 6.3)
-2	TBI 165 cGy twice daily	
-1	TBI 165 cGy twice daily	
0	UCBT	
+1		Begin G-CSF (section 6.5)

6.2 Conditioning Regimen

6.2.1 Fludarabine

Fludarabine 25 mg/m^2/day IV x 3 days, total dose 75 mg/m^2 (days –8 to –6)

6.2.2 Cyclophosphamide

Cyclophosphamide will be administered as a 2 hour intravenous infusion with a high volume fluid flush and mesna per institutional guidelines on days -7 and -6.

Cyclophosphamide 60mg/kg/day IV x 2 days, total dose 120 mg/kg (days –7 and –6)

Dosing is calculated based on Actual Body Weight (ABW) unless ABW > 30 kg above Ideal Body Weight (IBW), in which case the dose should be computed using adjusted body weight. Ideal body weight is calculated using Devine's formula or per institutional guidelines: 50 kg + (2.3 kg x (height in inches - 60)) for men; 45.5 kg + (2.3 kg x (height in inches - 60)) for women. Adjusted body weight = IBW + 0.5(ABW - IBW)

6.2.3 Total Body Irradiation (TBI)

The recommended TBI is 165 cGy given twice daily for a total dose of 1320 cGy (days -4 to –1). Recommended TBI Guidelines are described in Appendix III. TBI may be delivered by local guidelines provided the effective dose is equivalent to what is recommended in the TBI Guidelines.

6.3 Immunosuppressive Therapies

All patients will receive GVHD prophylaxis with 2 drugs as follows:

6.3.1 Cyclosporine (CSA)

Cyclosporine A (CSA) will start day -3 and will be administered PO/IV maintaining a trough level between 200 and 400 ng/mL. For adults the initial dose will be 2.5 mg/kg IV over 1 hour every 12 hours. For children < 40 kg the initial dose will be 2.5 mg/kg IV over 1 hour every 8 hours.

CSA dosing will be monitored and altered as clinically appropriate per institutional pharmacy guidelines. Dose adjustments will be made on the basis of toxicity and/or low CSA levels.

Patients will receive CSA until day +100. If no GVHD, the dose will be tapered 10% per week beginning on day 101, to discontinue at approximately day +180.

6.3.2 Mycophenolate mofetil (MMF)

Mycophenolate mofetil (MMF) 3 gram/day IV/PO for patients who are ≥ 40 kg divided in 2 or 3 doses. Pediatric patient (<40 kilograms) will receive MMF at the dose of 15 mg/kg/dose every 8 hours beginning day -3. MMF dosing will be monitored and altered as clinically appropriate based on institutional guidelines. Patients will be eligible for MMF dosing and pharmacokinetics studies.

Stop MMF at day +30 *or* 7 days after engraftment, whichever day is later, if no acute GVHD. (Definition of engraftment is 1st day of 3 consecutive days of absolute neutrophil count [ANC] $\geq 0.5 \times 10^9$ /L).

If no donor engraftment, do not stop MMF. If no evidence of donor engraftment on the day +21 bone marrow biopsy, notify URD search coordinator to pursue back-up UCB and arrange day +28 bone marrow biopsy per current institutional slow engraftment guidelines.

If the patient has acute GVHD requiring systemic therapy, MMF may be stopped 7 days after initiation of systemic therapy for acute GVHD.

6.4 Umbilical Cord Blood Transplantation (UCBT)

Cord blood products are thawed and filtered (170-micron) in the Molecular and Cellular Therapeutics (MCT) Lab using the method of Rubinstein et al.

Infusion will be per current institutional policies. The infusion of the first UCB unit should begin within 15 minutes, and no later than 30 minutes after arrival on the Unit. If 2 units are used, both cords will be infused within 30-60 minutes of each other as deemed clinically safe by the BMT attending or designee.

6.5 Growth Factor

All patients will receive G-CSF 5 mcg/kg/day IV (dose rounded to vial size) based on the actual body weight IV beginning on day +1 after UCB infusion. G-CSF will be administered daily until the ANC exceeds 2.5×10^9/L for three consecutive days and then discontinued. If the ANC decreases to $<1.0 \times 10^9$/L, G-CSF will be reinstituted.

6.6 Treatment Related Toxicities

Potential toxicities associated with preparative therapies

Cyclophosphamide		
Common	**Less Common**	**Rare**
• low white blood cell count with increased risk of infection • hair loss or thinning, including face and body hair	• low platelet count (mild) with increased risk of bleeding • darkening of nail beds	• heart problems with high doses, with chest pain, shortness of breath, or swollen feet • severe allergic reactions • skin rash

Cyclophosphamide		
Common	**Less Common**	**Rare**
(usually grows back after treatment) • nausea • vomiting • loss of appetite • sores in mouth or on lips • bleeding from bladder, with blood in urine • diarrhea • long-term or short-term infertility in women and men	• acne • tiredness • infection • fetal changes if pregnancy occurs while taking cyclophosphamide	• scarring of bladder • kidney damage (renal tubular necrosis) which can lead to kidney failure • heart damage, with trouble getting your breath, swelling of feet, rapid weight gain • scarring of lung tissue, with cough and shortness of breath • second cancer, which can happen years after taking this drug • death from infection, bleeding, heart failure, allergic reaction, or other causes

Fludarabine		
Common	**Less Common**	**Rare**
• low white blood cell count with increased risk of infection • low platelet count with increased risk of bleeding • low red blood cell count (anemia) with tiredness and weakness • tiredness (fatigue) • nausea • vomiting • fever and chills • infection	• pneumonia • diarrhea • loss of appetite • weakness • pain	• numbness and tingling in hands and/or feet related to irritation of nerves • changes in vision • agitation • confusion • clumsiness • seizures • coma • cough • trouble breathing • intestinal bleeding • weakness • death due to effects on the brain, infection, bleeding, severe anemia, skin blistering, or other causes

Total Body Irradiation (TBI)		
Common	**Less Common**	**Rare**
• nausea and vomiting • diarrhea • cataracts • sterility • endocrinopathies • growth failure • intestinal cramps • mucositis	• parotitis • interstitial pneumonitis • generalized mild erythema • veno-occlusive disease	• dysphagia • vertebral deformities • nephropathy • risk of 2nd malignancy years later (when given along with chemotherapy)

6.6.2 Toxicities potentially associated with the infusion of the UCB graft

- nausea and vomiting
- possible allergic reaction (including itching, hives, flushing [red face], shortness of breath, wheezing, chest tightness, skin rash, fever, chills, stiff muscles, or trouble breathing)
- graft-versus-host-disease (GVHD)
- veno-occlusive disease
- infections (sepsis)
- acute hemolytic reactions
- febrile nonhemolytic reactions
- anaphylactoid or anaphylactic reactions
- transfusion-related acute lung injury (TRALI)
- DMSO toxicity
- transmission of bacterial, viral or protozoal infection
- transfusion-associated circulatory overload (TACO)
- hypothermia
- non-immunologic hemolysis
- granulocyte-related complications
- cardiotoxicity

6.6.3 Potential Toxicities Associated with Immunosuppressive Therapies

Cyclosporine A (CSA) can cause high blood pressure, abnormalities in blood chemicals, seizures, headaches and kidney problems. The effect of CSA on the kidneys ranges from mild (noted only on blood tests) to severe (requiring dialysis). Blood levels of CSA and kidney function tests will be monitored closely to minimize side effects.

Mycophenolate mofetil (MMF) can cause nausea and vomiting, diarrhea or constipation, a lowering of blood counts, leg cramps, skin rash, difficulty sleeping, chemical imbalances including high blood sugar, headaches, dizziness and high blood pressure.

6.6.4 Toxicities associated with Neupogen (G-CSF)

- bone pain
- headaches
- body aches
- fatigue
- nausea/vomiting
- insomnia
- dyspnea
- rash
- edema

6.7 Supportive Care Guidelines

Supportive care will be provided per University Of Minnesota institutional guidelines for transplant patients including any supportive care research protocols.

All patients will receive standard supportive transfusion care according to transfusion committee guidelines or as modified based on clinical parameters.

Antimicrobial prophylaxis directed towards bacteria, fungi and viruses will be per University Of Minnesota current institutional guidelines for transplant patients.

7.0 STUDY PARAMETERS

Scheduled evaluations for days 21, 28 may be performed (+/-3 days) from the targeted date; assessments to be performed on day 60 and 100 may be done on (+/-) 7 days of the targeted date; assessments on day 180, 1 year, 2 years and 3 years may be performed (+/- 30) days of the targeted date. In addition, targeted days may be altered as clinically appropriate.

	Screen	Day 1 to engraftment				Days 31-100			Follow-up
		daily	weekly	Day +21	Day +28	weekly	Day +60	Day +100	6 months, 1 and 2 years
Informed consent	X								
Medical history	X	X							
Physical exam	X	X				X			X
Performance status	X							X	X
Height/Weight	X								
GVHD evaluation		X	X			X			X
CMV Surveillance			X			X			prn
Severe Adverse event notation		X				X			X
Laboratory									
CBC, diff	X	X[3]				X			X
Platelet	X								
PT/PTT	X								
Serum chemistries	X								
Creatinine, Na, K, HCO3	X	X							
Viral tests	X								
Urinalysis	X		prn						
Urine pregnancy test	X[1]								
Bone marrow bx/asp	X			X				X	X
Chimerism – BM	X			X				X	X
Chimerism – PB					X		X		
Procedures									
EKG	X		prn			prn			prn
MUGA or echocardiogram[5]	X		prn			prn			prn
Chest x-ray or CT	X[2]		prn			prn			prn
PFT's[5]			prn			prn			prn
Disease evaluation	X		X					X	X

1- within 14 days of registration in females of childbearing potential

2- CT without contrast to exclude occult infection for patients with a history of the following:

- MDS/CML blast crisis
- 2 or more consecutive leukemia inductions
- prolonged neutropenia, as defined as ≥ 4 weeks of neutropenia within the 2 months prior to BMT

3- Complete blood count with leukocyte differential daily until the absolute neutrophil count (ANC) ≥ 5 X 10^8/L for 3 consecutive measurements

4- Continue longer if clinically indicated

5- Pediatric pre-transplant evaluation qualification: for children that are not able to cooperate to have a MUGA and/or pulmonary function tests, an echocardiogram should be attempted and pulse oxymetry with exercise tolerance obtained. If not possible at all, it should clearly be documented in the physicians note.

7.1 UCB Engraftment Evaluation

Chimerism studies will be performed on the bone marrow on days 21 and 100 and at 6 months 1 year and 2 years. In cases of slow engraftment a BM biopsy may be repeated on day +28. Preparative

therapy for a second graft should not be commenced prior to day +30. Subsequently, if the patient's peripheral blood counts drop after an initial recovery, the peripheral blood and bone marrow should again be evaluated unless a cause has been determined (e.g., use of Ganciclovir for treatment of CMV). Patients diagnosed with graft failure (failure of ANC $\geq$ 5 x 10^8/L of donor origin by day +42) must be reported to the Principal Investigator.

7.2 GVHD Evaluation

GVHD evaluations will be done at least weekly until discharge and at each follow-up visit and continue at each follow-up using the current institutional criteria (acute GVHD appendix II and consensus for late acute and chronic GVHD).

7.3 Residual/Recurrent Disease Evaluation

Patients will be evaluated routinely for evidence of recurrent malignancy. If at any time the attending physician suspects recurrent disease, additional analyses will be performed as clinically indicated.

8.0 STUDY ENDPOINTS

8.1 Primary Objective

Survival at 1 year as measured from date of UCBT

8.2 Secondary objectives

Based on clinical and laboratory evaluations the following will be recorded for each subject:

- 8.2.1 Incidence of transplant-related mortality (TRM) at 6 months after UCBT
- 8.2.2 Pattern of chimerism after double UCBT - Chimerism studies will be performed on the bone marrow on days 21 and 100 and at 6 months 1 year and 2 years.
- 8.2.3 Incidence of neutrophil engraftment at day 42 after UCBT
- 8.2.4 Incidence of platelet engraftment at 1 year after UCBT
- 8.2.5 Acute graft-versus-host disease (GVHD) grade II-IV and grade III-IV at day 100 after UCBT
- 8.2.6 Chronic GVHD at 1 year after UCBT
- 8.2.7 Disease free survival at 1 and 2 years after UCBT
- 8.2.78 Relapse at 1 and 2 years after UCBT

9.0 ADVERSE EVENT MONITORING and REPORTING

Toxicity and adverse events will be classified according to NCI's Common Terminology Criteria for Adverse Events V 3.0 (CTCAE) and reported on the schedule below. A copy of the CTCAE can be downloaded from the CTEP home page http://www.eortc.be/services/doc/ctc/ctcaev3.pdf

9.1 Definitions

The following definitions are based on the Code of Federal Regulations Title 21 Part 312.32 (21CFR312.32(a)).

Adverse Event: Any untoward medical occurrence associated with the use of a drug in humans, whether or not considered drug related.

Suspected Adverse Reaction: Any adverse event for which there is a reasonable possibility that the drug caused the adverse event.

Life-Threatening Adverse Event Or Life-Threatening Suspected Adverse Reaction: An adverse event or suspected adverse reaction is considered "life-threatening" if, in the view of either the investigator or sponsor, its occurrence places the patient or subject at immediate risk of death.

Serious Adverse Event Or Serious Suspected Adverse Reaction: An adverse event or suspected adverse reaction is considered "serious" if, in the view of either the investigator or sponsor, it results in any of the following outcomes:

- Death
- A life-threatening adverse event
- Inpatient hospitalization or prolongation of existing hospitalization
- A persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions
- A congenital anomaly/birth defect.

Unanticipated (unexpected) problems/events as defined by the University Of Minnesota IRB are those that are not already described as potential risks in the consent form, not listed in the Investigator's Brochure or not part of an underlying disease.

Event Attribution Categories:
CTCAE does not define an AE as necessarily 'caused by a therapeutic intervention.' The clinical investigator must assign attribution for an adverse event after naming and grading of the event.

Attribution	Description
Unrelated	The AE is clearly NOT related to the intervention
Unlikely	The AE is doubtfully related to the intervention
Possible	The AE may be related to the intervention
Probable	The AE is likely related to the intervention
Definite	The AE is clearly related to the intervention

UPIRTSO: Federal regulations [45CFR46.103(b)(5) and 21CFR56.108(b)(1)] require the IRB to ensure that researchers promptly report "any unanticipated problems involving risk to subjects or others" (UPIRTSOs). The University of Minnesota IRB defines a UPIRTSO as any problem or event which in the opinion of the local researcher was unanticipated, reflects new or increased risk to the subjects and at least possibly related to the research procedures.

In addition, the IRB has defined the following problems/events as reportable within 10 working days using the UPRITSO form found on the IRB website:

- Any accidental or unintentional change to the IRB-approved protocol that increases risk or has the potential to recur
- Any deviation from the protocol taken without prior IRB review to eliminate apparent immediate hazard to a research subject
- Any publication in the literature, safety monitoring report (including Data and Safety Monitoring Reports), interim result or other finding that indicates an unexpected change to the risk/benefit ratio of the research
- Any breach in confidentiality that may involve risk to the subject or others
- Any complaint of a subject that cannot be resolved by the research staff
- Any other possibly related event which in the opinion of the investigator constitutes an unanticipated risk

Expedited (Rapid) Reporting: Certain events may require rapid notification to entities providing patient safety oversight (e.g. IRB) as detailed in section 9.2. For the IRB this is 10 working days.

9.2 Adverse Event Monitoring

All subjects will be monitored from the initiation of any study treatment through day 100 as it is expected that most serious adverse events will occur during this period.

After day 100, monitoring for adverse events will become less frequent based on the schedule in section 7.0. However, the investigator is obligated, upon knowledge of, to report any event meeting the definition of UPIRTSO and requires expedited reporting per section 9.3.

9.3 Required Reporting

The reporting period for this study is from initiation of any study treatment through day +100; however after day +100, the investigator must report upon knowledge any study treatment related event meeting the expedited reporting criteria below.

Agency	Criteria for reporting	Timeframe	Form to Use	Submission address/ fax numbers	Copy AE to:
U of MN IRB	**UPIRTSO**: any event which is unanticipated, involved new or increased risk to subjects, and was at least possibly related to study procedures	10 Working Days	UMCC SAE	University Of Minnesota IRB MMC 820	Masonic Cancer Center SAE Coordinator mcc-saes@umn.edu
	Other Problems or Events meeting the definition of UPIRTSO in section 9.1		UPIRTSO form		

The SAE Coordinator will provide the Cancer Center's Data and Safety Monitoring Council (DSMC) with the SAE in an appropriate format depending on the individual SAE (as reported or in a summary format).

FOR UNLICENSED UCB UNITS ONLY: Selected expected adverse reactions determined to be caused by or at least possibly caused by the UCB unit based on objective evidence will be reported in an expedited manner to the FDA under University of Minnesota IND BB-14797 (C. Brunstein, MD, PhD – sponsor/investigator).

10.0 DATA AND SAFETY MONITORING PLAN

This study's Data and Safety Monitoring Plan will be in compliance with the University of Minnesota Masonic Cancer Center's Data & Safety Monitoring Plan, which can be accessed at http://www.cancer.umn.edu/exfiles/research/dandsmplan.pdf

For the purposes of data and safety monitoring, this study is classified as moderate risk. Therefore the following requirements will be fulfilled:

- The PI will complete and submit a twice yearly Trial Progress Report to the Masonic Cancer Center Data and Safety Monitoring Council (DSMC) with the understanding more frequent reporting may be required by the Cancer Protocol Review Committee.
- The PI will comply with at least twice yearly monitoring of the project by the Masonic Cancer Center monitoring services.
- The PI will oversee the reporting of all adverse events meeting the definition of reportable in section 9.3 to the University Of Minnesota IRB and the Masonic Cancer Center SAE Coordinator.

At the time of the University of Minnesota IRB continuing review, the Principal Investigator will submit to the CPRC a copy of all documentation submitted to the IRB for continuing review.

11.0 STATISTICAL CONSIDERATIONS

11.1 Objectives

11.1.1 Primary objective

The principal aim of this study is to estimate one year overall survival of UCBT after a myeloablative preparative regimen for the combined experience of single unit UCB transplants and double unit UCB transplants.

11.1.2 Secondary objectives

- the incidence of transplant-related mortality (TRM) at 6 months after UCBT
- the pattern of chimerism at multiple time points after double UCBT
- the incidence of neutrophil engraftment at day 42 after UCBT
- the incidence of platelet engraftment at 1 year after UCBT
- the incidence of acute graft-versus-host disease (GVHD) grade II-IV and grade III-IV at day 100 after UCBT
- the incidence of chronic GVHD at 1 year after UCBT
- the disease free survival at 1 and 2 years after UCBT
- the incidence relapse at 1 and 2 years after UCB T

11.2 Statistical Analysis

Survival and progression-free survival will be estimated by the Kaplan-Meier method. Non-relapse mortality, relapse, neutrophil and platelet engraftment, acute and chronic GvHD will be estimated by cumulative incidence using competing risk methods. 95% confidence intervals will be used to show precision for the estimates.

In order to show that survival after double unit UCB transplants are no worse than single unit UCB transplants, a non-inferiority test will be performed. The null and alternative hypotheses will be:

H_o: 1 year survival for double UCB ≤ 1 year survival for single UCB – 0.2 (20%)
H_a: 1 year survival for double UCB > 1 year survival for single UCB – 0.2 (20%)

Comparison of the single UCB cohort will be made to a historical cohort using a superiority test.

H_o: 1 year survival for new cohort = 1 year survival for old cohort
H_a: 1 year survival for new cohort > 1 year survival for old cohort by at least 0.2(20%)

For inferences of 1 year survival among patients that have been followed for at least 1 year, comparisons will be made by a normalized Z-test for non-inferiority. A Chi-square test will be used for the superiority comparison. Comparisons in which patients have not been followed for the complete follow-up period will be completed by the Log-rank test. Univariate comparisons of factors other than the number of units will be stratified by the number of units. Cox regression will be utilized to correct for any potential confounders.

11.3 Rationale for Sample Size

In order to achieve a precise estimate of the probability of survival at one year post transplant we would like to have an estimate of survival with a 95% confidence interval that has a band width of less than 0.10. We expect to enroll approximately 300 patients over the 10 year study period. We will also be able to use 40 patients from the previous study using this same conditioning regimen. Assuming that the proportion surviving at one year is 65%, the 95% confidence interval should have a band width of approximately 0.08. This is assuming that there are no patients lost to follow-up and that all patients have been followed for at least one year post transplant at the time of analysis.

Based on the accrual goal of 300 patients, the equivalence test will have a power of 97% and a type I error of 5%. The superiority test will have a power of 83% and a type I error of 5%.

We expect 50% to be double unit UCBT.

11.4 Gender and Ethnicities Statement

This study is open to both males and females and to all racial/ethnic groups. The patient enrollment pattern is expected to be similar to that of other hematological malignancy studies. It is not anticipated that the outcome will be affected by either race or gender. The study will not have separate accrual targets for different subgroups.

12.0 RECORDS RETENTION

The investigator will retain study records, including source data, copies of CRF's and all study correspondence in a secure facility for at least 6 years after the study file has been closed with the

IRB. In addition, the Clinical Trials Office will keep a master log of all patients participating in the study with sufficient information to allow retrieval of the medical records.

13.0 REFERENCES

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Appendix I - Diagnosis and Treatment Of Acute Graft Versus Host Disease

Patients will be considered evaluable acute GVHD if they demonstrate donor cell engraftment and survive to day 28. Organ involvement will be staged using the criteria outlined in the table below. Biopsy of each organ site at diagnosis or major change in disease activity will be performed unless clinical circumstances make it impossible.

Consensus Clinical Stage and Grade of Acute GVHD (Przepiorka *et al*, 1995)

Stage	Skin	Liver	Lower Gastrointestinal Tract	Upper Gastrointestinal Tract
1	Maculopapular rash <25% of body surface	Bilirubin 2.0 – 3.0 mg/dl	Diarrhea 500 – 1000 mL/day or 280 – 555 mL/m^2	No protracted nausea and vomiting
2	Maculopapular rash 25-50% body surface	Bilirubin 3.1 – 6.0 mg/dl	Diarrhea 1000 – 1500 mL/day or 556 – 833 mL/m^2	Persistent nausea, vomiting or anorexia
3	Generalized erythroderma	Bilirubin 6.1 – 15.0 mg/dl	Diarrhea >1500 mL/day or >833 mL/m^2	
4	Generalized erythroderma with bullous formation and desquamation	Bilirubin > 15 mg/dl	Severe abdominal pain, with or without ileus, or stool with frank blood or melena	

Grading for Treatment Criteria

Mild GVHD = Skin stage I-II only (Equivalent to Seattle Grade I).

Moderate GVHD = Skin stage I-III and/or liver I-IV and/or Gastrointestinal tract (GI) I-III and/or Upper GI (UGI). (Equivalent to Seattle Grade II, III).

Severe GVHD = Any stage IV along with severe clinical illness.

Patients progressing during initial therapy or not improving sufficiently after 2 courses of therapy are to be treated as severe GVHD.

Acute GVHD treatment may be by institutional guidelines. The recommended treatment for patients demonstrating moderate or severe GVHD (Grade II-IV) is methylprednisolone at 48 mg/m2/day or prednisone 60 mg/m2/day followed by a taper which starts a day 8 of acute GVHD therapy. If there is no response in 7 days or there is progression of the disease, ATG 15 mg/kg every 12 hours for 5 days will be added.

Appendix II - Karnofsky and Lansky Performance Status Scales

Percentage	
100	Normal, no complaints, no evidence of disease
90	Able to carry on normal activity; minor signs or symptoms of disease
80	Normal activity with effort; some signs or symptoms of disease
70	Cares for self; unable to carry on normal activity or do active work
60	Requires occasional assistance, but is able to care for most of his/her needs
50	Requires considerable assistance and frequent medical care
40	Disabled; requires special care and assistance
30	Severely disabled, hospitalization indicated. Death not imminent
20	Very sick, hospitalization necessary, active supportive treatment necessary
10	Moribund, fatal processes, progressing rapidly
0	Dead

REFERENCE

Karnofsky DA: Meaningful clinical classification of therapeutic responses to anti-cancer drugs. Editorial: Clin Pharmacol Ther 2:709-712, 1961.

LANSKY PLAY PERFORMANCE STATUS SCALE

Percentage	
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, play activities
60	Up and around, but minimal active play; keeps busy with quieter activities
50	Gets dressed but lies around much of the day, no active play; able to participate in all quiet play and activities
40	Mostly in bed; participates in quiet activities
30	In bed; needs assistance even for quiet play
20	Often sleeping; play entirely limited to very passive activities
10	Unresponsive
0	Dead

Appendix III - Recommended TBI Guidelines

Fractionated Total Body Irradiation (In Lateral Position):

1320 cGy administered in an 8 fractions of 165 cGy each with 2 fractions being given each day.

Total body irradiation is given at a dose rate of 10-19 cGy/minute prescribed to the midplane of the patient at the level of the umbilicus.

The total body irradiation will be delivered with right and left lateral fields, with the patient supine on a specially designed couch.

Based on measurements of transverse thicknesses, aluminum compensators will be used to ensure that the dose homogeneity across the field is within 10 % of the prescribed dose. Usually head/neck, leg and lung compensators are used (although based on calculated mid-mediastinal doses, lung compensators are often not needed).

Total body irradiation will be delivered with a linear accelerator using 6, 18, 24 MeV X-rays. The energy used will be based on the calculated dose to the midline at points up and down the patient's torso. The lowest energy that gives 90-100% of the prescription point dose will be used.

A beam "spoiler" will be used to ensure a full skin dose.

Half value layer lung and kidney blocks will not be utilized.

Testicular boosts should be used for all males with ALL (and according to institutional practice for other diseases). The testicular boost is given in a single 400 cGy fraction with either electrons prescribed to Dmax or photons prescribed to the midplane of the scrotum. If electrons are used, the energy for the testicular boost depends on the thickness of the testicles and is chosen so that the D90 corresponds > to the posterior surface of the scrotum.