Reduced Intensity Conditioning for Haploidentical Bone Marrow Transplantation in Patients with Symptomatic Sickle Cell Disease

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Version 6.0

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Reduced Intensity Conditioning for Haploidentical Bone Marrow Transplantation in Patients with Symptomatic Sickle Cell Disease

Co-Chairs:	Robert Brodsky MD, Michael DeBaun, MD, Adetola Kassim, MD, and Mark Walters, MD
Study Design:	This is a Phase II, single arm, multi-center trial, designed to estimate the efficacy and toxicity of haploidentical bone marrow transplantation (BMT) in patients with sickle cell disease (SCD). Patients are stratified into two groups: (1) children with SCD with strokes; and (2) adults with severe SCD. Based on their age and entry criteria, participants are enrolled in 2 strata as defined below. The primary endpoint of event-free survival (EFS) will be estimated in each age-specific stratum.
Primary Objective:	The primary objective is to estimate EFS at 2 years after haploidentical BMT in patients with SCD enrolled in 2 strata – children 5.00 – 14.99 years of age at enrollment with SCD and adults 15.00 - 45.99 years of age at enrollment with severe SCD. Primary or secondary graft rejection, second transplant, or death will count as events for this endpoint. Stratum-specific estimates will be provided. If the protocol implements the planned treatment change in response to safety monitoring for graft failure, then the analysis will be based on the stratum participants under the active treatment plan at the close of the study and results for the other participants will be described.
Secondary Objectives:	Secondary objectives include determining the effect of haploidentical BMT on clinical and laboratory manifestations of SCD by 2 years after transplantation and determining the incidence of other transplant-related outcomes. Secondary outcomes include: overall survival post-haplo BMT at 1 and 2 years post-enrollment; EFS at 1 year; graft rejection; disease progression; donor chimerism; grades II-IV and III-IV acute graft-versus-host disease (GVHD); chronic GVHD, severe chronic GVHD; neutrophil, and platelet recovery; hepatic veno-occlusive disease (VOD); idiopathic pneumonia syndrome (IPS); central nervous system (CNS) toxicity (reversible posterior leukoencephalopathy syndrome [RPLS], hemorrhage, and seizures); cytomegalovirus (CMV) infection; adenovirus infection; Epstein Barr virus post-transplant lymphoproliferative disease (EBV

PTLD); invasive fungal infection; CNS outcomes; 28 day e-pain diary assessments; health-related quality of life (HRQoL) in the adult stratum; lung function; TRJV; 6 minute walk distance; hematological outcomes; viral mold infections/bacterial or fungal sepsis; and proportion on immunosuppression. All objectives are within each stratum.

Eligibility: Eligibility criteria differ by age.

1. Children with SCD (Hb SS or Sß° Thalassemia) aged 5.00 – 14.99 years at Segment A enrollment who have one or more of the following:

    a. A neurological event resulting in focal neurologic deficits that lasted ≥ 24 hours (classical clinical definition of stroke, not requiring imaging studies of the brain) **OR** a focal neurological event resulting in abnormalities on T2-weighted or FLAIR images using a MRI scan, indicative of an acute infarct, with no other reasonable medical explanation (definition of a stroke supported with MRI imaging scans of the brain), **OR** both.

    b. Abnormal transcranial Doppler (TCD) measurement with a timed average maximum mean velocity of ≧ 200 cm/sec in the terminal portion of the internal carotid or proximal portion of middle cerebral artery[1] or if the imaging TCD method is used > 185 cm/sec[2] plus evidence of intracranial vasculopathy.

    c. Silent Cerebral Infarct defined as an infarct-like lesion based on an MRI signal abnormality at least 3 mm in one dimension and visible in two planes on FLAIR or T2-weighted images (or similar image with 3D imaging) and documented neurological examination performed by a neurologist demonstrating the participant has a normal neurologic examination or an abnormality on examination that could not be explained by the location of the brain lesion(s).

    d. Acute severe vaso-occlusive pain episodes requiring hospitalization and recalcitrant to maximum medical therapy. Episodes of pain to be adjudicated by selected committee.

    e. One acute chest syndrome episode resulting in intensive care admission requiring non-mechanical ventilatory support: simple nasal cannula, face mask that requires oxygen content

(venti mask, non-rebreather), simple nasal cannula, face mask O2(e.g. ventimask, rebreather), CPAP, SiPAP, BiPAP, high flow nasal cannula (HFNC) or invasive mechanical ventilatory support (delivered by ETT or trach).

f. Right heart catheterization confirmed pulmonary artery pressure >25 mmHG or mean pulmonary vascular resistance 206(57-421) $dyn \cdot s \cdot cm^{-5}$

g. Essential hypertension on antihypertensive medications >95% upper limit of normal age (as defined according to the American Academy of Pediatrics)

h. Recurrent priapism (episodes lasting at least 4 hours at least twice in the last 12 months or 3 times in the last 24 months) recalcitrant to medical treatment or unable to use hydroxyurea due to SCD phenotype with the approval of the adjudication committee

2. Participants with SCD (any clinically significant sickle genotype, for example, Hb SS, SC, SD Sβ° Thalassemia, $S\beta^+$ Thalassemia, S-OArab) aged 15.00 – 45.99 years at Segment A enrollment who have one or more of the following:

a. A neurological event resulting in focal neurologic deficits that lasted ≥ 24 hours (classical clinical definition of stroke, not requiring imaging studies of the brain) **OR** a focal neurological event resulting in abnormalities on T2-weighted or FLAIR images using a MRI scan, indicative of an acute infarct, with no other reasonable medical explanation (definition of a stroke supported with MRI imaging scans of the brain), **OR** both.

b. History of two or more episodes of acute chest syndrome (ACS) in the 2-year period preceding enrollment despite the institution of supportive care measures (i.e. asthma therapy and/or hydroxyurea);

c. History of three or more severe vaso-occlusive pain crises per year in the 2-year period preceding enrollment despite the institution of supportive care measures (i.e. a pain management plan and/or treatment with hydroxyurea); painful episodes related to priapism, osteonecrosis or any sickle-related complication are acceptable;

d. Administration of regular red blood cell (RBC) transfusion therapy, defined as 8 or more transfusion events per year (in the 12 months before enrollment) to prevent vaso-occlusive

clinical complications (i.e. pain, stroke, or acute chest syndrome);

e. An echocardiographic finding of tricuspid valve regurgitant jet velocity (TRJV) ≥2.7 m/sec.

3. Participants in either age group must have adequate physical function as measured by all of the following:

a. Karnofsky or Lansky performance score ≥60.

b. Cardiac function: Left ventricular ejection fraction (LVEF)

i. 40%; **or** LV shortening fraction > 26% by cardiac echocardiogram or by MUGA scan.

c. Pulmonary function: Pulse oximetry with a baseline O_2 saturation of ≥85% **and** DLCO > 40% (corrected for hemoglobin).

i. Patients unable to perform DLCO due to young age or other inability are not required to have a specified threshold but must have no evidence of dyspnea at rest and O_2 saturation level must be greater than 88% in room air at baseline.

d. Renal function: Serum creatinine ≤ 1.5 x upper limit of normal for age **and** estimated or measured creatinine clearance ≥ 70 mL/min/1.73 m^2.

e. Hepatic function:

i. Serum conjugated (direct) bilirubin ≤ 2x upper limit of normal for age as per local laboratory. Participants are not excluded if the serum conjugated (direct) bilirubin is >2x the upper limit of normal for age as per local laboratory **and:**

1. There is evidence of a hyperhemolytic reaction after a recent RBC transfusion, **OR**

2. There is evidence of moderate direct hyperbilirubinemia defined as direct serum bilirubin < 5 times ULN and not caused by underlying hepatic disease.

ii. ALT and AST < 5x upper limit of normal as per local laboratory.

f. Liver MRI using a validated methodology per institutional preference (T2* or R2* or by ferriscan [R2 MRI]) for estimation of hepatic iron content is required for participants who are currently receiving ≥8 packed red blood cell transfusions per year for ≥1 year or have received ≥20 packed red blood cell transfusions (lifetime cumulative). Participants who have hepatic iron content ≥ 10 mg Fe/g liver dry weight by liver MRI must have a Gastroenterology/hepatology consultation with liver biopsy and histological examination including documentation of the absence of cirrhosis, bridging fibrosis[1], and active hepatitis.

g. Participants must be HLA typed at high resolution using DNA based typing at HLA-A, -B, -C, DRB1, and have available:

i. An HLA haploidentical first degree relative donor (parents, siblings or half siblings, or children) with 2, 3, or 4 (out of 8) HLA-mismatches who is willing and able to donate bone marrow. A unidirectional mismatch in either the graft versus host or host versus graft direction is considered a mismatch. The donor and recipient must be HLA identical for at least one antigen (using high resolution DNA based typing) at the following genetic loci: HLA-A, HLA-B, HLA-C, and HLA- DRB1. Fulfillment of this criterion shall be considered sufficient evidence that the donor and recipient share one HLA haplotype and typing of additional family members is not required. Confirmatory donor HLA typing must be completed≤ 100 days prior to Segment A enrollment

h. Umbilical cord blood or peripheral blood stem cell donors will not be accepted.

Exclusion criteria:

1. Participants who have an HLA-matched sibling who is able and willing to donate bone marrow.

2. Participants with uncontrolled bacterial, viral, or fungal infection in the 6 weeks before enrollment (currently taking medication with evidence of progression of clinical symptoms or radiologic findings).

3. Participants with evidence of HIV infection or seropositivity for HIV.

4. Participants who have received a previous Hematopoietic Cell

Transplant (HCT)

5. Participants who have had an EDAS procedure within the 6 months prior to enrollment (Section 2.4.6).
6. Participants who have received a prior solid organ transplant.
7. Participants who have participated in another clinical trial in which the participant received an investigational or off-label use of a drug or device within 3 months prior to enrollment.
8. Females who are pregnant or breast feeding.
9. Participants with clinically significant, uncontrolled autoimmune disease, requiring active medical management (immunosuppressive therapy or chemotherapy), which, in the judgment of the local Principal Investigator, indicates that the patient could not tolerate transplantation.

   Females of child bearing potential (to include all female participants > 10 years of age, unless postmenopausal for a minimum of 1 year before the time of consent or surgically sterilized) who do not agree to practice two (2) effective methods of contraception at the same time, or who do not agree to practice true abstinence when this is in line with the preferred and usual lifestyle of the subject, from the time of signing of informed consent through 12 months post-transplant.
10. Males (even if surgically sterilized) who do not agree to practice effective barrier contraception, or who do not agree to practice true abstinence from the time of signing informed consent through 12 months post-transplant.
11. Presence of anti-donor specific HLA antibodies. HLA antibody presence and specificity will be determined by solid phase immunoassays. An anti-donor specific HLA antibody will be considered positive when the mean fluorescence intensity (MFI) is higher than the cut-off defined by each center. Recommended cut-off values are MFI >1000 for donor specific antibody to HLA-A, -B, and DRB1 and MFI >2000 for HLA-C, DQB1 and DPB1. This must be measured before the final donor selection, and ≤ 100 days before enrollment in Segment A (preferably ≤ 30 days before Segment A enrollment). If MFI >1000 for donor specific antibody to HLA-A, -B, DRB1 and/or MFI >2000 for HLA-C, DQB1 and DPB1, documentation must be submitted to the DCC coordinator for review and approval by a Protocol Chair and/or Protocol Officer prior to enrollment.

Treatment Description: The BMT preparative regimen will start on Day -70 with hydroxyurea 30mg/kg daily through Day -10. The conditioning regimen will also include Thymoglobulin (rATG) (0.5mg/kg on Day -9, 2mg/kg on Day -8, Day -7), Thiotepa 10mg/kg on Day -7, Fludarabine (30mg/m2 on Day -6 to Day -2), Cyclophosphamide 14.5mg/kg on Day -6 and Day -5, and TBI 200cGy on Day -1. Day 0 is the day of infusion with non T-cell depleted bone marrow. Cyclophosphamide 50 mg/kg and Mesna 40mg/kg is given on Days +3 and +4, and GVHD prophylaxis (sirolimus and mycophenolate mofetil) begin on Day +5.

If it is determined that the graft failure rate in a stratum has met the stopping rule within the first 12 evaluable participants in the stratum, and approval from the DSMB and Sponsor is provided, then the conditioning regimen with TBI 400cGy instead of TBI 200cGy will be used for future participants in the stratum, and an additional 40 participants will be accrued to that stratum.

Accrual Objective: The target sample size is 40 transplanted patients in each stratum; if the stopping rule for graft failure is triggered within the first 12 evaluable participants in either stratum using the 200cGy dose of TBI and the DSMB and sponsor approve the pre-defined switch in conditioning regimen, then accrual for that strata will restart under the modified conditioning regimen.

Accrual Period: The estimated accrual period is 4 years. If the safety monitoring for graft failure passes a stopping boundary within the first 12 evaluable participants, it is estimated that the accrual period will be extended up to an additional 15 months.

Study Duration: Participants will be followed for the 28-day pain diary (prior to 70-day conditioning period) and for 2 years from date of infusion (post-transplant).

Safety Monitoring: The rate of overall mortality by Day 180 post start of hydroxyurea therapy pre-transplant, acute grade III-IV GVHD at 100 days, and severe chronic GVHD at 18 months post-transplant will be monitored by using a sequential probability ratio test (SPRT) for censored exponential data for each of these. Graft failure at 100 days post-transplant will be monitored by using a sequential probability ratio test (SPRT) for binary data.

TREATMENT SCHEMA

Days –70 → –10	Hydroxyurea 30 mg/kg po daily
	Hgb S fraction < 35% prior to administration of Thymoglobulin. If Hbg S > 35%, automated exchange transfusion (RBC apheresis) is the preferred strategy to lower Hgb S with a goal of 20% and the maximum Hgb S fraction not to exceed 35%.
Day –9***	Thymoglobulin 0.5 mg/kg IV with pre-meds
Day –8***	Thymoglobulin 2 mg/kg IV qd with pre-meds
Day –7***	Thymoglobulin 2 mg/kg IV qd with pre-meds Thiotepa 5 mg/kg IV q 12 h (10 mg/kg total)
Days –6, –5	Fludarabine 30 mg/m2 IV over 30-60 minutes, then Cyclophosphamide (CY) 14.5 mg/kg IV over 1-2 hours*
Days –4 → –2	Fludarabine 30 mg/m2 IV over 30-60 minutes
Day –1	TBI 200 cGy with gonadal shielding**
Day 0	Non-T-cell depleted bone marrow
Days +3, +4	Cyclophosphamide 50 mg/kg IV Mesna 40 mg/kg IV*
Day +5	Begin sirolimus (section 2.4.5.1)**, mycophenolate mofetil (MMF) 15 mg/kg pot id with maximum daily dose 3 gm/d
Day +35	Discontinue MMF

*Uroprophylaxis may be altered per institutional preference (see below)
**See Section 2.4.2.10 and section 4.4.6 regarding planned protocol change in TBI dose if safety monitoring guideline for 100-day graft failure is passed in the first 12 evaluable participants per stratum.
***If using corticosteroids as premedication, discontinue before Day -1.

ABBREVIATIONS

ACS	Acute Chest Syndrome
AE	Adverse Event
AjBW	Adjusted Body Weight
ANC	Absolute Neutrophil Count
rATG	rabbit Anti-thymocyte Globulin (Thymoglobulin)
BMT	Bone Marrow Transplant
CBC	Complete Blood Count
CIBMTR	Center for International Blood and Marrow Transplant Research
CMV	Cytomegalovirus
CNS	Central Nervous System
CNT	Clinical Trials Network
CTCAE	Common Terminology for Adverse Events
CY	Cyclophosphamide
DCC	Data and Coordinating Center
EFS	Event-Free Survival
DSMB	Data Safety Monitoring Committee
EBV	Epstein-Barr Virus
EDC	Electronic Data Capture
GVHD	Graft-versus-Host Disease
HCT	Hematopoietic Cell Transplantation
HLA	Human Leukocyte Antigen
HRQoL	Health-Related Quality of Life
HU	Hydroxyurea
IBW	Ideal Body Weight
IPS	Idiopathic Pneumonia Syndrome
IV	Intravenous
6MWD	6 Minute Walk Distance
MMF	Mycophenolate Mofetil
NHLBI	National Heart Lung & Blood Institute
NIH	National Institutes of Health
NMDP	National Marrow Donor Program
OS	Overall Survival
PCR	Polymerase Chain Reaction
PFT	Pulmonary Function Test
PI	Principal Investigator
PRES	Posterior Reversible Encephalopathy Syndrome
PROMIS	Patient Reported Outcomes Measurement Information System
PTLD	Post-transplant lymphoproliferative disease
SAE	Serious Adverse Event
SCD	Sickle Cell Disease
SCD-EOSI	Sickle Cell Disease-related Event of Special Interest
SPRT	Sequential Probability Ratio Test
TBI	Total Body Irradiation
TLC	Total Lung Capacity
TRJV	Tricuspid Regurgitant Jet Velocity
VOD	Veno-occlusive Disease

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CHAPTER 1

BACKGROUND AND RATIONALE

1.1 Introduction

Sickle cell disease (SCD) is caused by a single point mutation in codon 6 of the β-globin chain. This mutation directs an amino acid substitution of valine for glutamic acid, which promotes the formation of long hemoglobin polymers under hypoxic conditions.[3,4,5] This propensity for polymer formation deforms the red blood cell (RBC) and causes significant alterations in red cell integrity, rheologic properties, and life span.[6] The presence of SCD with chronic hemolysis is directly or indirectly responsible for the vasculopathy that forms in virtually all body organs. SCD is a heritable disorder of hemoglobin. One in every 1941 newborns regardless of race[7] and 1 in every 400 African-American newborns[8] in the United States have SCD. Approximately 80,000 to 100,000 persons in the United States are affected by SCD.[9] This disease burden has a considerable impact on individuals affected and health-care systems. In the United States alone, the medical cost of caring for approximately 80,000 affected individuals exceeds $1 billion.[10]

The course of SCD has dramatically changed over the last 40 years. Currently children born with SCD are expected to live through adolescence and young adulthood with > 95% expected to reach their 18th birthday.[11] A recent prospective cohort study in children with the most severe disease (based on recurrent pain or acute chest syndrome episodes or other severe manifestations) have a 15 year estimated survival of 99% when placed on hydroxyurea therapy. In comparison, children from the same cohort with less severe disease that were not placed on hydroxyurea therapy had an overall survival of 95%. These results are similar to a Brazilian cohort demonstrating that children with more severe SCD that were placed hydroxyurea therapy had a higher survival rate than those with less severe disease that were not, (99·5% vs. 94·5%, P = 0·01), for a median of 2 years (range 0·1-6·5) .[12] Taken together, these two studies suggest that with appropriate anticipatory guidance for splenic sequestration, prompt management of fever, routine vaccinations, and penicillin prophylaxis, SCD in children is no longer life threatening.11

1.1.1 Stroke and silent cerebral infarction in children with SCD

The biggest challenge in managing children with SCD is the prevention of strokes and silent cerebral infarcts. Once a stroke occurs, standard care for secondary prevention of overt strokes in children and adults with SCD includes regular blood transfusion therapy to suppress synthesis of hemoglobin S (HbS).[13] Without transfusion therapy, approximately 67% of these children will have second overt strokes.[14]

The SWiTCH trial was the definitive randomized trial to test the hypothesis that hydroxyurea therapy with phlebotomy (alternative therapy) was non-inferior to regular blood transfusion therapy and chelation (standard therapy) for secondary prevention of stroke recurrence. As part of the interim analysis, the trial was stopped prematurely based on futility assessment fordecreasing iron stores in the alternative therapy group. However, the rate of strokes in the alternative and standard therapy group was 10.5% (7 of 67) and 0% (0 of 66), In one of the few prospective trials conducted to determine the efficacy of blood transfusion therapy to prevent both overt and silent cerebral infarcts (infarcts that are not coincident with a focal neurological deficit) 40 children with

SCD and overt strokes were enrolled at 7 academic centers and followed for a mean of 5.5 years. Despite the mean pre-transfusion hemoglobin S concentration of 29%, an optimal transfusion therapy goal, progressive cerebral infarcts occurred in 45% (18 of 40 children).[15]

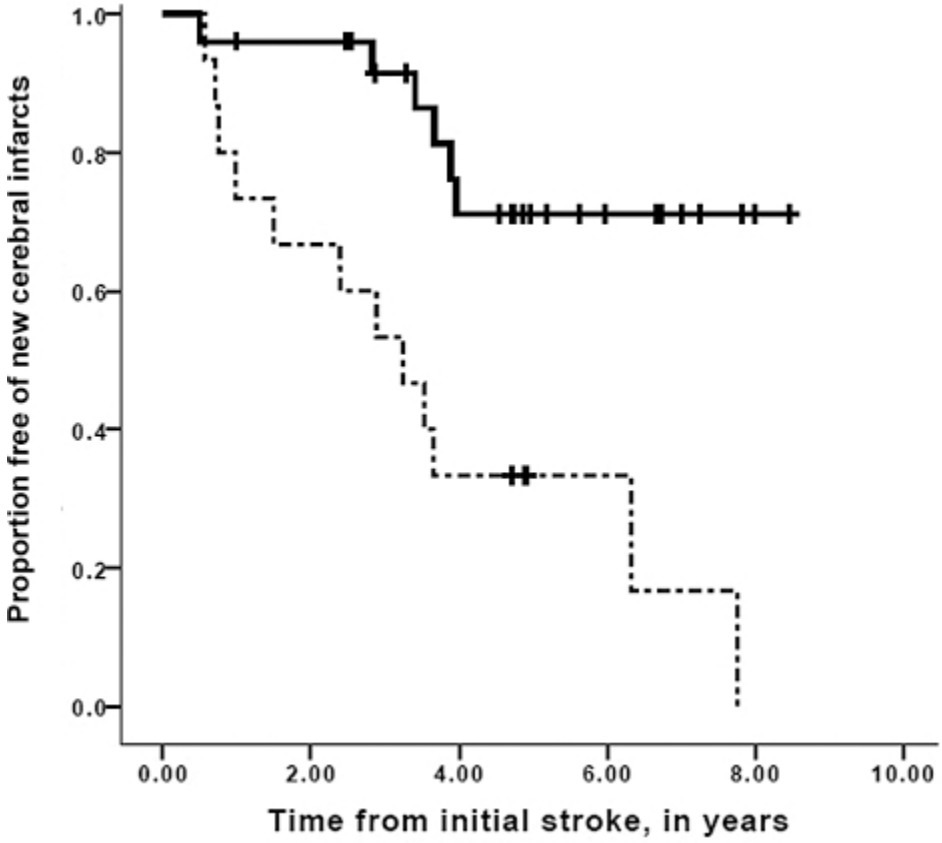


Figure 1. Survival free of new overt or silent cerebral infarcts in children with SCD while on transfusion therapy for secondary stroke prophylaxis. Participants with progressive overt and silent cerebral infarction, stratified for the absence (solid line) or the presence (dashed line) of progressive cerebral vasculopathy during chronic blood transfusion therapy. Vertical lines represent censored cases. Median event-free interval for new silent or overt infarction was 3.2 years for the group with progressive vasculopathy, compared with median event-free interval not reached in the group without progressive vasculopathy (Mantel-Cox log-rank, P = .001).

A much larger, but retrospective cohort consisting of 14 centers and including 137 children with sickle cell disease (SCD) and strokes were followed for a median of a decade. Over the course of a decade, approximately 22% had a second stroke despite regular blood transfusion therapy, and among those with a second stroke, approximately 30% had a third stroke.[16] In both the retrospective and prospective studies, participants had strokes when hemoglobin S levels were 1%, 9%, 22%, 25%, and 26%. The hemoglobin S levels at the time of the stroke recurrence were all < 30%, the maximum target percent for hemoglobin S concentration. Based on the compelling results from these studies and other large retrospective observational studies,[17,18] blood transfusion therapy can only be considered palliative for secondary prevention of strokes.

Silent cerebral infarcts are the most common cause of neurological injury in children with SCD occurring in almost 40% before their 14th birthday.[19] Recently our group completed a controlled randomized trial demonstrating this phenomenon in children with pre-existing silent cerebral infarcts. Those that received regular blood transfusion therapy for 36 months to keep their maximum hemoglobin S level less than 30% had a 58% relative risk reduction of infarct recurrence when compared to observation.[20] Unfortunately, even among the children with silent cerebral infarcts that received regular blood transfusion therapy, recurrent infarcts (both strokes and silent cerebral infarcts) still occurred.[21]

Thus, for children with overt strokes and silent cerebral infarcts, blood transfusion therapy to prevent cerebral infarct recurrence has to be continued indefinitely and is palliative. Alternative therapies must be pursued for this high-risk group. Haplo-identical transplant may provide an alternative to blood transfusion therapy for secondary prevention of cerebral infarct recurrence.

1.1.2 Progressive Disease with a high mortality rate for adults with SCD

In contrast to children where advances in medical care, public health and anticipatory guidelines have decreased the mortality, in adults with SCD the mortality rate has remained unchanged in large population based studies. In the most comprehensive study of SCD conducted, the Cooperative Study for Sickle Cell Disease (CSSCD), from 1977 to 1998, the average life span for men and women with SCD was 42 and 48 years, respectively. Data from 1979 to 2005 from National Center for Health Statistic that included over 16,000 adults with SCD revealed that the median age of death was 38 and 42 years for men and women with SCD, respectively. With improvement in care for the adults, we know the biggest risk factors for earlier death in adults with SCD, however, we lack any definitive strategy to abate the progression of cardiopulmonary disease that leads to earlier mortality. An elevated triscuspid regurgitant jet velocity (TRJV) remains the biggest risk factor associated with earlier death in adults with SCD.[21] In multi-institutional prospective cohort study, the rate of mortality for those with and without TRJV > 3 m/sec and < 3.0 m/sec respectively is shown in Figure 2.[22] Currently there is no efficacious treatment for adults with TRJV > 3.0 m/sec. Hydroxyurea therapy is recommended by the American Thoracic Society; however, no randomized trial has been conducted to demonstrate its efficacy. Sidenifil versus placebo was introduced as a therapy for elevated TRJV in a double blind randomized controlled clinical trial; however, the trial stopped prematurely because of the increased rate of vaso-occlusive pain events in the active treatment arm. In prospective cohort study of 430 adults with SCD followed for approximately 5 years, Kassim et al. demonstrated that low forced expiratory volume in one second % predicted (FEV_1) is associated with a higher mortality rate. Again, understanding the risk factor for earlier death is important, however, there is no therapy for low FEV1% predicted in adults with SCD. Likewise, renal disease, which is common in adults with SCD, is also associated with higher mortality. In a prospective population study conducted by the CDC between 2005 and 2009, in adults with SCD and end stage renal disease that start dialysis, the rate of death in the first year after commencing dialysis was 26%.[23]

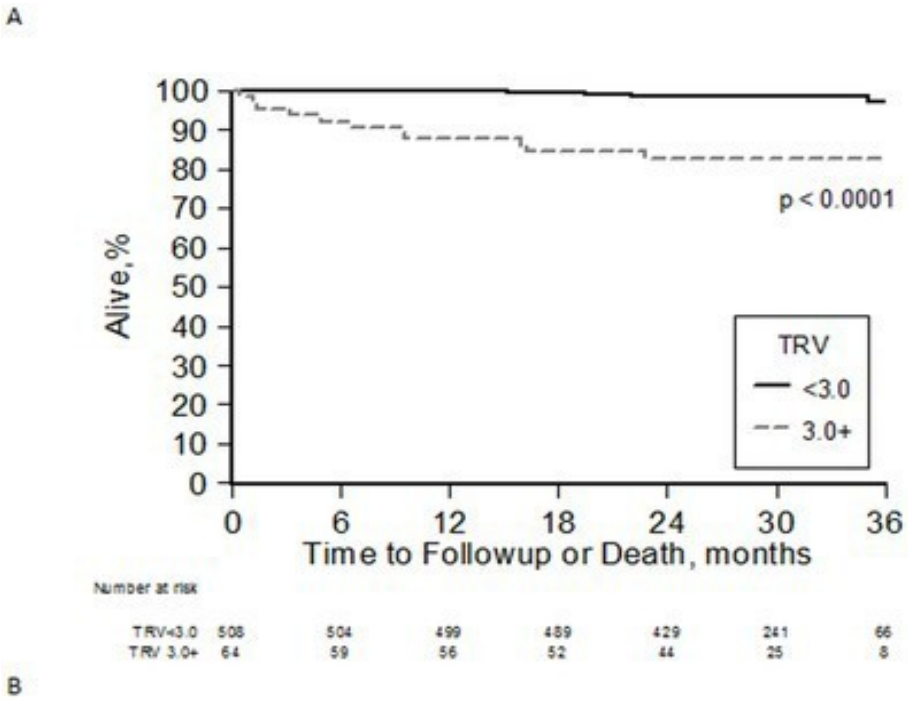


Figure 2. Kaplan Meier plot from the Walk Phast Cohort demonstrating the proportion alive in a cohort of adults with SCD that had TRJV $\geq$ 3 m/sec (n=64) and < 2 m/sec. (n=508).

In summary, adults with SCD have a significant risk of earlier death with no therapeutic option to prevent progression of end organ disease.

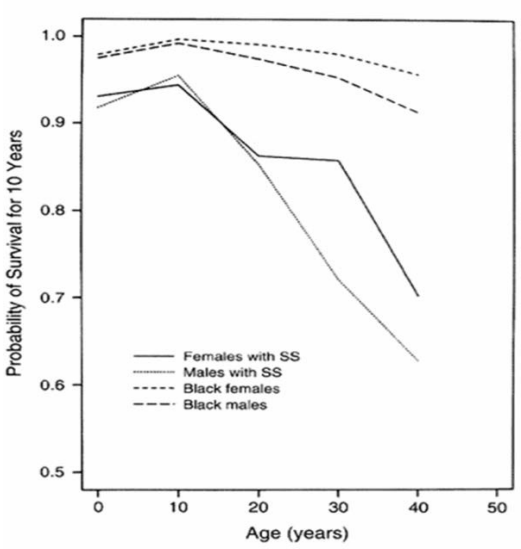
The most debilitating complication is vaso-occlusive pain that occurs in a large proportion of adults with SCD. The only FDA approved treatment for prevention of vaso-occlusive pain events in SCD is hydroxyurea therapy; however, its effectiveness in decreasing mortality, particular in the subset of adults with severe pre-existing end organ disease is unclear. In a prospective cohort study that documented daily pain in a diary Smith et al.,[24] demonstrated that majority 29.3% of adults with SCD reported pain almost every day in their diary days; whereas, only 14.2% reported pain in 5% or fewer diary days (adjusted). Most importantly when a patient reported that they had vaso-occlusive pain episode, referred to as crises in the manuscript, only 22% of these episodes resulted in health care utilization; the remaining 78% of the acute pain episodes resulted in the adult electing to manage the crises at home. Given the increased rate of mortality associated with SCD in adults coupled with the crippling effects of vaso-occlusive pain episodes, individuals with evidence of end-organ-disease or debilitating vaso-occlusive pain events despite medical therapy, should be offered an option for cure.

1.2 Rapid disease progression in adults with SCD

Natural history studies show that in contrast to the improvements in outcomes in childhood, there is a rapid progression in organ damage, morbidity and premature mortality in adulthood. Progression of organ damage in adulthood is marked by pulmonary hypertension, which occurs in

20-40% of adult patients with a 10 fold increase in risk of premature mortality,[37] renal insufficiency with proteinuria in 70% and progressing to renal failure in 11%, abnormal pulmonary function in 90% and progression to irreversible organ damage in 50% of patients by 50 years of age.[42] One third develop chronic pain syndrome and only 20% are employed.[43, 25] Death in adulthood is frequently related to organ damage which is not preventable or easily managed with current medical measures.[42] SCD related complications such as leg ulcers, stroke, priapism, vascular necrosis, anxiety, and depression further worsen the health related quality of life. The mortality rate of patients with SCD is 5.8-20% in the first 10 years after transition to adult care.[28,43] Premature death occurs at a median age of 38 years, a statistic that has not changed in 20 years[42,26] (Figure 3). In a long-term follow-up study of patients with symptomatic SCD who were eligible to participate in the multicenter study of hydroxyurea (MSH), the annual mortality rate was 4.4 per 100 person-years among adults with SCD who satisfied eligibility criteria, which is 4.4%.

Figure 3. Steep decline in probability of survival in adulthood in patients with sickle cell disease[1]



Thus, the inexorable progression of disease and premature mortality in adulthood provides the strong rationale for intensifying the investigation and development of curative therapies in this group of patients.

1.3 HCT is a therapeutic option for SCD

Supportive health care measures instituted during childhood, which include newborn screening and pneumococcal prophylaxis, the administration of hydroxyurea and regular red blood cell (RBC) transfusions, have decreased the risk of serious infections and other life-threatening complications, resulting in improved survival to adulthood. This has, in part, shifted the demographics of SCD to include a growing proportion of young adults with chronic health impairments. Sickle cell disease is considered a life threatening disease in adults. Multiple studies have demonstrated that adults with SCD, particularly those with co-morbidities have a shortened life span.[27] Even with the most recent use of hydroxyurea therapy, the only FDA disease modifying therapy for SCD, the life expectancy for SCD is still in the mid-40s.[28]

As an alternative to chronic supportive care, hematopoietic cell transplantation (HCT) from a human leukocyte antigen (HLA)-identical sibling donor has been used sparingly in children, but is curative in the majority of children treated.[4,10] Event-free probabilities of 90 – 95% have been reported after HLA-ID sibling HCT using cord blood or marrow as the source of hematopoietic cells, for the most part after myeloablative preparation.[9,29,30,31,32,33,34] However, the application of BMT in children with SCD is pursued very infrequently today, despite these compelling results. The most important reason why so few are treated is that most patients lack a suitable donor. Based upon current estimates, which project that 14% will have a HLA-identical donor[35] and recent projections published by the NMDP in which the likelihood of identifying a well-matched unrelated marrow donor is 18% among African-American recipients,[36] it is estimated that two-thirds of patients will not be able to pursue transplantation. We hypothesize that HLA haplo-identical transplant is an alternative to blood transfusion therapy for secondary prevention of cerebral infarct recurrence and in abating the morbidity from the age-dependent chronic organ dysfunction in adult patients who lack a well-matched donor.

1.4 HCT for SCD – conditioning regimens

Even when it is clear that allogeneic BMT can decrease or eliminate sickle cell disease-related complications[30], myeloablative transplantation can cause toxicities such as GVHD, infertility and importantly, a fixed risk of transplantation-related mortality that has caused families and their providers to seek alternative safer therapies, even when the latter lack curative potential. Recently, progress in the development of reduced intensity conditioning regimens that facilitate the sustained engraftment of donor marrow with reduced toxicity has occurred. Most of these regimens incorporate highly immunosuppressive purine analogues, such as fludarabine, which allow the reduction or elimination of myeloablative agents such as busulfan or total body irradiation and still sustain engraftment of HLA-identical allogeneic stem cells.

Early attempts to use a non-myeloablative conditioning regimen for HLA-identical sibling BMT for sickle cell disease were unsuccessful. In these trials, an initial wave of donor engraftment typically was observed, but the disease invariably recurred when post-grafting immunosuppression was withdrawn[6]. To investigate the cause of this failure, a group at the NIH led by John Tisdale studied sirolimus in a preclinical model and determined that this agent was more effective than standard calcineurin inhibitors at establishing bidirectional donor-host tolerance. His team tested this approach in humans, and Hsieh et al demonstrated stable donor engraftment in adult SCD patients following non-myeloablative HCT from HLA-matched sibling donors.[29,30] Between 2004 and 2013, thirty patients with severe disease who were 16 to 65 years of age were treated by a nonmyeloablative combination of alemtuzumab (1mg/kg in divided doses), total-body irradiation (300 cGy), and sirolimus followed by HLA-ID sibling filgrastim mobilized peripheral blood stem cell transplantation. Sirolimus alone was used for GVHD prophylaxis and 87% of recipients had long-term engraftment without acute or chronic GVHD. While sirolimus was discontinued in 15 of 30 recipients, in the remaining recipients it was extended due to lymphohematopoietic chimerism that was judged too low to prevent a late graft rejection. The selection of sirolimus appeared pivotal to the very good outcome because of its inhibitory effect on effector T-cell proliferation and viability while preserving T-regulatory cells that are required for tolerance, and because sirolimus does not appear to cause posterior reversible encephalopathy syndrome (PRES) unlike other calcineurin inhibitors. PRES has been observed in approximately 10 - 20% of sickle

cell recipients who receive cyclosporine post-HCT for GVHD prevention, and is thus a leading contributor to adverse events after HCT for SCD.[31,37] Thus, it appears that reduced intensity transplantation regimens that include immunoablation after treatment might be especially important to ensure donor engraftment after HCT for SCD. This aspect will carry even greater importance in the setting of HLA-mismatched donor transplantation that will be needed to tackle the problem of limited matched donor availability as a barrier to transplantation. Therefore, developing novel strategies that address the issue of expanding the donor pool while ensuring a low rate of graft rejection are potentially transformative to curative therapy for SCD.

1.5 Haploidentical HCT with post-transplant cyclophosphamide

In the past five years, investigators at Johns Hopkins have developed a non-myeloablative conditioning regimen for transplantation of marrow from partially HLA-mismatched, or haploidentical, bone marrow from first-degree relatives. The regimen's main goal, J9966 (RPN 99-11-05-01), was to titrate the dose of pre- and post-transplantation cyclophosphamide (CY), a potent immunosuppressive drug, given in conjunction with pre-transplantation fludarabine and total body irradiation (TBI), to achieve a regimen that had an acceptably low risk of graft rejection and GVHD, the two major complications of haploidentical bone marrow transplantation (BMT). All patients received mycophenolate mofetil (MMF) and tacrolimus (FK), beginning on day 4 or 5 and terminating on days 35 and 50-180, respectively, to reduce the incidence and severity of GVHD. The first cohort of three patients received no pre-transplantation CY and 50 mg/kg CY IV on day 3, and two of the patients rejected their grafts. A second cohort of 20 patients received 14.5 mg/kg CY IV on days –6 and –5 in addition to 50 mg/kg IV on day 3. Of 18 evaluable patients, 13 patients had donor engraftment on day 60, but accrual of patients to this dose level was stopped because 8/13 patients developed severe GVHD, an incidence convincingly in excess of the stopping criterion of 20%. To reduce the incidence of GVHD, a third cohort of patients received an additional dose of CY 50 mg/kg IV on day 4, and MMF dosing was increased from bid to tid, based upon pharmacokinetic data suggesting the need for more frequent dosing. Of seventeen evaluable patients so far, two patients have had non-fatal graft rejection, and only one patient treated according to the protocol has had severe GVHD (an additional patient developed severe GVHD after withdrawal of immunosuppression to treat relapse). Two patients died of causes other than relapse: one from GVHD, and the other from disseminated fungal infection. Of the 16 patients who have been followed up to 100 days for relapse, 8 have relapsed at a median of 64 days (range 24-~100) after transplantation, and 6 patients are alive and disease free at a median of 206 days (range, 100-429 days [as of Feb 8, 2014]) following BMT.[38]

In order to better judge the safety and efficacy of the non-myeloablative BMT protocol, the tables below compare the results of J9966, dose level 3, to the results of the four largest published trials of HLA-identical sibling peripheral blood versus bone marrow transplantation for early stage leukemia.

TABLE 1.5A: ENGRAFTMENT DATA AFTER HAPLOBMT

Author	N (PB/BM)	Median age	ANC 500/mm^{3†}	Plt 20K†	Plt 50K†
Blaise[5]	48/52*	37/36	15/21	13/21	15/26
Bensinger[6]	81/91	42/42	16/21	13/19	NA
Schmitz[7]	163/166	39/37	12/15	15/20	20/26
Couban[8]	109/118	45/44	19/23	16/22	NA
J9966	17	31	16	24	31

*Numbers represent: recipients of peripheral blood/recipients of bonemarrow
†Time fromtransplantation to designated count, sustained without transfusion

TABLE 1.5B: OUTCOMES DATA AFTER HAPLOBMT

Author	aGVHD II-IV (%)	aGVHD III-IV (%)	cGVHD (%)	TRM (%)	Relapse (%)
Blaise	45/42*	17/28	55/30	23/21†	6/11
Bensinger	64/57	15/12	46/35	21/30†	14/25
Schmitz	52/39	28/16	66/50	24/24*	12/7
Couban	44/44	26/18	40/30	7/16†	15/20
J9966	47	13	NA	13	50

*Numbers represent: recipients of peripheral blood/recipients of bone marrow
†Transplant-related mortality (TRM) or relapse over entire study (median f/u ~ 2 years)
†100 day mortality

Compared to the patients receiving HLA-identical sibling bone marrow following myeloablative conditioning, patients on J9966 were younger, took longer to engraft platelets, and had a substantially higher rate of relapse, but were similar in the time to neutrophil recovery, the incidence of GVHD, and transplant-related mortality (TRM). The higher rate of relapse for patients on J9966 may be attributable to a benefit of myeloablative conditioning in reducing the risk of relapse, or that patients on J9966 had advanced, poor prognosis hematologic malignancies, which relapse more frequently than early leukemia after allogeneic BMT.

Since the toxicities of non-myeloablative haploidentical BMT (haploBMT) were not known when the trial was written, eligibility for J9966 was restricted to patients with advanced, poor-risk hematologic malignancies, such as chronic myeloid leukemia in 2nd chronic phase, advanced myelodysplasia, acute leukemia in 2nd remission, and lymphoma in relapse after autologous BMT. Eligibility on the protocol has been expanded to 'standard risk' hematologic malignancies in trial J0457, which is ongoing.

Between J9966 and J0457, 56 patients with hematologic malignancies received cyclophosphamide 50 mg/kg IV, either once (on day 3) or twice (on days 3 and 4) after non-myeloablative

conditioning and haploidentical bone marrow transplant. Most of these patients had advanced disease or failed a previous autologous transplant. All were conditioned as outpatients with fludarabine, cyclophosphamide, and total body irradiation, transplanted with non-T cell-depleted marrow, and treated with tacrolimus and mycophenolate mofetil beginning the day after the last dose of cyclophosphamide. The most interesting finding was that compared to patients receiving a single dose of post-transplant cyclophosphamide, those receiving two doses had significantly less grade II-IV aGVHD (43% vs. 78%; p=.01) and grade III-IV aGVHD (20% vs. 53%; p=.006) by day 200 after transplant. Death from GVHD occurred in 5/13 assessable patients receiving one dose versus 2/28 assessable patients receiving 2 doses of cyclophosphamide.

Since the data to date suggest that this treatment regimen may be as safe as HLA-identical sibling BMT after myeloablative conditioning, non-myeloablative haploidentical BMT may be considered a reasonable treatment option for patients who have severe SCD. Also, the novel use of post-transplant cyclophosphamide with a reduced intensity conditioning emerges as an interesting option of immunotherapy to prevent graft rejection. Moreover, as cancer relapse is not a concern in the setting of SCD, engraftment with non-myeloablative hematopoietic stem cell transplantation should be curative. In clinical transplantation, antithymocyte globulin (ATG) has been used extensively in conventional myeloablative and non-myeloablative conditioning regimens to facilitate engraftment in patients with sickle cell disease.9 This effect is largely mediated by *in vivo* T-cell depletion produced by the ATG, similar to the effect of monoclonal T-cell antibodies in the murine model.

These preliminary data have relied upon the administration of donor marrow in lieu of umbilical cord blood or mobilized peripheral blood progenitor cells, which is an important consideration in this non-malignant condition. The use of mobilized peripheral blood progenitor cells for malignant and non-malignant diseases is associated with higher risk of chronic GVHD compared to bone marrow. It is well recognized that chronic GVHD adds to the burden of morbidity and mortality. In particular, for non-malignant diseases where there is not a need for graft versus tumor effect, there are no advantages to transplantation of peripheral blood. In patients with severe aplastic anemia, the most common non-malignant indication for transplantation, chronic GVHD risks are higher after transplantation of peripheral blood compared to bone marrow and mortality risks are higher after transplantation of peripheral blood from HLA-matched sibling and HLA-matched unrelated donors.[39] In addition, graft failure rates are high after unrelated UCB transplantation for SCD. Data from CIBMTR/Eurocord reported an event-free survival was 50% for children with sickle cell disease (the predominant cause of failure was graft rejection).[40] The cord blood arm of the recently completely BMT CTN 0601 (unrelated donor transplantation for children and adolescents with sickle cell disease) was closed early for excess graft failure.[41] Together, the preliminary data presented below with the published evidence about the use of cord blood and mobilized peripheral blood progenitor cells in sickle cell disease strongly support the use of marrow in this study.

1.6 Haploidentical HCT for SCD – preliminary results

The single center experience from Johns Hopkins includes 38 patients with SCD who were screened and 36 (95%) who proceeded to transplantation. The overall median age of transplanted patients was 23 years. With a median follow-up of 35 (range, 1-93) months, 97% of patients are

alive; one patient died from complications of graft-versus-host disease 7 months after transplant and one patient developed AML requiring a second BMT (now alive and in remission). Of the 36 patients transplanted, 5 had matched sibling donors and 31 had HLA haploidentical donors. All five patients who received matched sibling donor transplants are alive and engrafted (4 mixed chimerism and 1 with full donor chimerism). In patients who received transplants from an HLA haploidentical donor 22/31 had stable engraftment of donor cells (71%). All patients with secondary graft failure recovered host hematopoiesis and survive with autologous reconstitution of SCD. The overall incidence of cGVHD is 5.8%. The last 5 consecutive patients who were transplanted (patients 32-36) received TBI increased from 200cGy to 400cGy. All 5 of these patients survive beyond day 60 with 100% donor chimerism.

While these results are very good with regard to transplant-related toxicity, there is a clear need for improvement with regard to mitigating disease recurrence. Experimental data using high dose cyclophosphamide has clearly shown that in order to increase engraftment efficiency there are strategies that might be followed: increase the intensity of the conditioning (such as increasing the dose of TBI or adding additional agents to the pre-transplant regimen) or increase the cell dose of the graft.[42,43] In this study, the former was adopted in the form of adding thiotepa to the conditioning regimen in order to abrogate the host-versus-graft rejection and to generate partial marrow ablation to promote the engraftment of donor cells. We did not find that G-CSF mobilization as a method to increase the cell dose improved donor engraftment; thus, it was discontinued. We recently increased the TBI dose in our conditioning regimen from 200 to 400 cGy with early results as noted above.

There is evidence that thiotepa is an active agent in the conditioning regimen for hemoglobin disorders that addresses the difficulty of donor engraftment after allogeneic transplantation. In a report by La Nasa et al, an investigation demonstrated that a combination of Bu/TT/CY conditioning in the initial series of 32 patients that received URD transplantation had a Transplant Related Mortality (TRM) of 19% and a disease-free survival probability of 69%. The Bu/TT/CY regimen later was replaced with Bu/Flu/TT in the subsequent series, in 17 patients who received a combination of Bu/Flu with thiotepa, the TRM was 0% and thalassemia free survival was 77% after HCT. Bernardo et al substituted busulfan with its dihyroxy derivative treosulfan and demonstrated updated data in 60 patients (40 URD and 20 matched related donor (MRD) recipients indicating an overall survival of 93% and thalassemia free survival of 84%.[44]) Finally, a recent report of 12 consecutive parental haploidentical transplants (11 with SCD and one with β thalassemia major) from St. Mary's Hospital, London was reported at the 2015 EBMT meeting. Eleven of the 12 patients survive with partial or full donor chimerism after receiving a preparative regimen of fludarabine 150 mg/m^2, thiotepa 10 mg/kg, cyclophosphamide 29 mg/kg, TBI 2 Gy and ATG (Thymoglobulin) 4.5 mg/kg with HU, hyper-transfusion and azathioprine administered 2 months before commencing the conditioning regimen. Patients received cyclophosphamide 50 mg/kg on days +3 and +4, MMF and sirolimus for post-grafting immunosuppression (Delafuente et al EBMT 2015). Taken together, these data demonstrate steady improvement in donor engraftment was associated with thiotepa, and that the safety profile has been acceptable. Thus, we propose to add this drug to the conditioning regimen to address the problem of graft rejection/disease recurrence after haploBMT.

To date, the aforementioned single center experience from St. Mary's London now includes a total of 23 pediatric patients (18 SCD, 5 thalassemia, ages 3 to 18 years). With a median follow up of 10.7 (range, .7 to 23) months, 20 patients are alive and disease free. Of the 3 (13.0%) patients who died, one patient died from secondary graft failure and complications of IPS at Day +90, a second patient died at D +36 of septic shock and meningitis and the third died at Day + 148 due to IPS. cGVHD was observed in 5 patients (29.4%) beyond Day+ 100, however no cGVHD was reported beyond 12 months. Additional data regarding count recovery and GVHD incidence is presented in Table 1.6a below; while details regarding this group's percent donor chimerism is broken down at specific intervals as seen in Table 1.6b.

TABLE 1.6A: ST. MARY'S LONDON EXPERIENCE: OUTCOMES

Stem Cell Source	BM (G-CSF primed)
TNC x 10^8/kg	Median: 9.66 Range: 2.35 - 20.53
CD34 x 10^6/kg	Median: 3.46 Range: 1.12 - 9.21
Survival Months	Median: 10.7 Range: 0.7 - 23.0
Neutrophil engraftment	Median: 17 Range: 15 – 29
Plt >20	Median: 35.5 Range: 20 – 64
Plt>50	Median: 35.5 Range: 20 - 64
Chimerism	See Table 2 for chimerism summary
Graft Failure/Relapse	2 (8.7%) • One secondary graft failure day +60. • One primary graft failure day +28.
aGvHD grade I	7 (30.4%)
aGvHD ≥ grade 2	2 (8.7%): • Day +35 skin stage 3, treated with MSC • Day +16 gut stage 3, treated with MSC
cGvHD >day +100 Limited	2 (8.7%) • Day +320 skin • Day +259 liver, treated with steroids
cGvHD >day +100 Extensive	3 (13.0%) • Day +187 lung, day +308 skin, day +320 muscoskeletal. • Day +257 skin and liver • Day +180 gut, treated with MSC.
cGvHD >day +100 Total	5 (21.7%)
cGvHD >18 months	None

Stem Cell Source	BM (G-CSF primed)
Deaths	3 (13.0%) • Day +90. CoD: Secondary Graft Failure/Infection/IPS • Day +148. CoD: Idiopathic Pneumonia Syndrome • Day +36. CoD: Septic shock and meningitis.
VOD	1 (4.3%) Day +78 post-haplo but day +4 post-auto rescue.
Days to Cessation of immunosuppression:	Median: 145.5 Range: 108 - 432 One patient prolonged course of immunosuppression due to low donor T cell fraction: 432days.
Disease Free Survival	87.0%
Overall Survival	87.0%

TABLE 1.6B: ST. MARY'S LONDON EXPERIENCE: CHIMERISM ANALYSIS

Haplo	**of the total evaluable patients at each time point**							
Time point (days post-BMT)	**Day +28**		**Day +90**		**Day +180**		**≥ day +365**	
Evaluable patients (n)	**22**	**22**	**13**	**13**	**14**	**14**	**9**	**9**
% Donor Chimerism	**WB**	**T cell**	**WB**	**T cell**	**WB**	**T cell**	**WB**	**T cell**
≥95	91.0	72.7	76.9	69.2	85.8	78.6	77.8	88.9
90-94	4.5	9.1	0	23.1	7.1	14.3	11.1	0
50-89	4.5	9.1	23.1	0	7.1	0	11.1	0
<50	0	9.1	0	7.7	0	7.1	0	11.1

The administration of HU in advance of the conditioning regimen and the donor hematopoietic cell administration has been studied in the setting of HCT for hemoglobin disorders as a strategy to mitigate the risk of graft rejection. It was incorporated into protocol 26 in treating children with thalassemia major who had class III risk-group features and it was associated with a lower incidence of graft rejection compared to historical observations.[45] In another contemporary series of 50 children with SCD treated in Belgium between 1988 and 2013, the DFS after HLA-ID sibling HCT was 85.6% and 94.1% of recipients survived.[46] Of interest, all children treated since 1995 had hydroxyurea administered well before HCT, which comprised a group of 38 recipients, as first

reported in 2004.[47] Thirty-seven of these individuals survive free of SCD with an 8-year estimate of DFS that is 97.1%, and which is significantly higher compared to the DFS in those who did not receive HU before HCT (P<0.001). Together, these data strongly suggest that HU might be safely administered in advance of transplantation to exert its immunosuppressive and myelosuppressive effects to promote engraftment of donor cells.

1.7 Disease Severity in SCD and transplant eligibility

The application of experimental HCT therapy in SCD must strike a balance between the underlying disease severity and the possibility of a direct benefit of the treatment, particularly in pediatric populations. Thus, the SCD subjects targeted for enrollment are patients at risk for early mortality and progressive global organ damage, as occurs in adults, and also children at risk for progressive brain injury following an initial cerebral infarction, for whom the possibility of a curative outcome will have the greatest impact. In these two groups, there are compelling reasons to consider an investigational transplant option.

1.7.1 Lack of curative treatments for adults with SCD

The prevailing framework of clinical research in SCD shifts in the transition from childhood to adulthood. Among pediatric hematologists, the dominant view is that survival to adulthood is excellent, and that children, on average, have a very good quality of life as a result of supportive care measures such as antibiotics, family education, and the judicious use of transfusions. In addition, a great deal of effort has focused on identifying children who have high-risk features, so that the risk of any specific intervention might be balanced by the severity of disease in that individual. Thus, clinical research studies in children with SCD have focused primarily on safety and efficacy, often in the setting of a high-risk population, such as children at risk for a stroke. While HCT has curative potential, its routine application remains quite limited in children with SCD, due in part to its toxicities which include a risk of dying from the procedure itself. As a result, clinical studies of transplantation in children with SCD have suffered from poor accrual and despite excellent disease-free survival, HCT is not routinely considered in children with SCD.

In contrast, the prevailing view among clinicians who care for adults is that SCD is, on average, a severe disease with a significant risk of sudden death and the development of chronic medical problems and that supportive care options for young adult patients are not adequate to address the overwhelming nature of this disease. Thus, clinical studies in adults with SCD have focused on interventions that prolong survival and improve the quality of life. Unlike children, adults with SCD are much more likely to have a debilitating complication. As a result, the risk/benefit ratio of HCT is very favorable in adults, particularly if an approach to HCT that defines an acceptable level of toxicity can be established. If successful, such an approach could significantly improve the outlook of many adults with SCD and broaden the therapeutic choices.

CHAPTER 2

STUDY DESIGN

2.1 Study Overview

This study is a phase II, single arm, multi-center study designed to estimate the efficacy and toxicity of haploidentical BMT in patients with SCD who are in two groups: (1) children between 5.00-14.99 years with SCD with strokes; and (2) adults between 15.00-45.99 years with severe SCD.

2.1.1 Primary Objective

The primary objective is to estimate event-free survival (EFS) at 2 years after a reduced intensity conditioning regimen and human leukocyte antigen (HLA)-haploidentical bone marrow transplantation (haploBMT) in children with SCD and adults with severe SCD.

2.1.2 Secondary Objectives

Within each stratum:

1. Estimate overall survival at one year and two years after enrollment and after haploBMT.
2. Estimate EFS at one year after haploBMT.
3. Estimate incidence of primary and secondary graft rejection at one and two years after haploBMT.
4. Estimate incidence and severity of acute GVHD until Day 100 then chronic GVHD at six months, one year, 18 months, and two years post-transplant.
5. Characterize donor hematopoietic chimerism in peripheral blood at days 28, 42, 100, and 180 and at 1 and 2 years after haploBMT.
6. Characterize hematologic and non-hematologic toxicities of haploBMT, including the incidence and severity of acute and chronic graft-versus-host disease; time to and probability of red blood cell, neutrophil, and platelet recovery; hepatic veno-occlusive disease (VOD); idiopathic pneumonia syndrome (IPS); central nervous system (CNS) toxicity (reversible posterior leukoencephalopathy syndrome [RPLS], hemorrhage, and seizures); cytomegalovirus (CMV) infection; adenovirus infection; Epstein Barr virus post-transplant lymphoproliferative disease (EBV PTLD); invasive fungal infection.
7. Evaluate if sickle vasculopathy is halted by successful transplantation as determined by comparing brain MRI pre- and 2 years post-haploBMT. Cerebral MRI/MRA is required for only pediatric patients enrolled under a neurologic indication to assess status of CNS disease. For adults where the indication for the transplant is a stroke, cerebral MRI/MRA is required after transplant to assess progression of CNS disease.
8. Estimate disease recurrence at one and two years after haplo BMT.
9. Evaluate sickle-related events and end organ function in all recipients after haploBMT to

determine if severe and debilitating vaso-occlusive pain and cerebral infarction are stabilized after transplantation.

10. Evaluate patient-reported quality of life (pain and fatigue domains) pre- and 1 and 2 years post-haploBMT in the adult stratum for English and Spanish-speaking patients
11. Lung function pre- and 2 years post-haploBMT
12. TRJV pre- and 1 and 2 years post-haploBMT
13. 6 min walk distance pre- and 1 and 2 years post-haploBMT
14. Pain intensity assessed by an electronic pain diary at baseline, 1 and 2 years post-haploBMT for English-speaking patients ≥ 15.00 years of age at time of enrollment
15. Hematological outcomes at 2 years (hgb, retic, %HbS, LDH, bili, last date of red blood cell transfusion)
16. Viral mold infections/bacterial or fungal sepsis at any time up to 2 years post-transplant
17. Proportion on immunosuppression at 2 years post-haploBMT

2.2 Patient Eligibility

2.2.1 Inclusion Criteria
Some inclusion criteria differ by age.

2.2.1.1 Inclusion Criteria for Both Strata (Stratum 1 and Stratum 2)

Participants in both strata must have adequate physical function as measured by all of the following:

1. A Karnofsky/Lansky performance score of ≥ 60.
2. Cardiac function: Left ventricular ejection fraction (LVEF) > 40%; **or** LV shortening fraction > 26% by cardiac echocardiogram or by MUGA scan.
3. Pulmonary function: Pulse oximetry with a baseline O_2 saturation of ≥ 85% **and** DLCO > 40% (corrected for hemoglobin).
    a. Patients unable to perform DLCO due to young age or other inability are not required to have a specified threshold but must have no evidence of dyspnea at rest and O_2 saturation level must be greater than 88% in room air at baseline.
4. Renal function: Serum creatinine ≤ 1.5 x upper limit of normal for age **and** estimated or measured creatinine clearance ≥ 70 mL/min/1.73 m^2.
5. Hepatic function:
    a. Serum conjugated (direct) bilirubin ≤ 2x upper limit of normal for age as per local laboratory. Participants are not excluded if the serum conjugated (direct) bilirubin is >2x the upper limit of normal for age as per local laboratory **and:**
        i. There is evidence of a hyperhemolytic reaction after a recent RBC transfusion, OR

ii. There is evidence of moderate direct hyperbilirubinemia defined as direct serum bilirubin < 5 times ULN and not caused by underlying hepatic disease.

b. ALT and AST < 5x upper limit of normal as per local laboratory.

6. Liver MRI using a validated methodology per institutional preference (T2* or R2* or by ferriscan [R2 MRI]) for estimation of hepatic iron content is required for participants who are currently receiving ≥8 packed red blood cell transfusions per year for ≥1 year or have received ≥20 packed red blood cell transfusions (lifetime cumulative). Participants who have hepatic iron content ≥ 10 mg Fe/g liver dry weight by liver MRI must have a gastroenterology/hepatology consultation with liver biopsy and histological examination including documentation of the absence of cirrhosis, bridging fibrosis[1], and active hepatitis.

7. Lack of clinical evidence of overt stroke or transient ischemic attack (TIA) within 180 days prior to enrollment in Segment A. Participants with clinical evidence of an overt stroke or TIA within 180 days prior to enrollment in Segment A will require a cerebral MRI/MRA confirming stabilization of the neurologic event prior to proceeding to enrollment.

8. Participants must be HLA typed at high resolution using DNA based typing at HLA-A, - B, -C, DRB1, and have available:

   a. An HLA haploidentical first degree relative donor (parents, siblings or half siblings, or children) with 2, 3, or 4 (out of 8) HLA-mismatches who is willing and able to donate bone marrow. A unidirectional mismatch in either the graft versus host or host versus graft direction is considered a mismatch. The donor and recipient must be HLA identical for at least one antigen (using high resolution DNA based typing) at the following genetic loci: HLA-A, HLA-B, HLA-C, and HLA-DRB1. Fulfillment of this criterion shall be considered sufficient evidence that the donor and recipient share one HLA haplotype and typing of additional family members is not required. Confirmatory donor HLA typing must be completed ≤ 100 days prior to Segment A enrollment

9. Umbilical cord blood or peripheral blood stem cell donors will not be accepted.

2.2.1.2 Inclusion Criteria for Stratum 1: Children Ages 5.00 - 14.99 at enrollment

1. Age 5.00 – 14.99 years at Segment A enrollment

2. Participants with sickle cell anemia (Hb SS or Sß° Thalassemia) who have **one or more** of the following:

   a. A neurological event resulting in focal neurologic deficits that lasted ≥ 24 hours (classical clinical definition of stroke, not requiring imaging studies of the brain) **OR** a focal neurological event resulting in abnormalities on T2-weighted or FLAIR images using a MRI scan, indicative of an acute infarct, with no other reasonable medical explanation (definition of a stroke supported with MRI imaging scans of the brain), **OR** both.

b. Abnormal transcranial Doppler (TCD) measurement with a timed average maximum mean velocity of at least 200 cm/sec in the terminal portion of the internal carotid or proximal portion of middle cerebral artery or if the imaging TCD method is used > 185 cm/sec plus evidence of intracranial vasculopathy.

c. Silent Cerebral Infarct defined as an infarct-like lesion based on an MRI signal abnormality at least 3 mm in one dimension and visible in two planes on FLAIR or T2-weighted images (or similar image with 3D imaging) and documented neurological examination performed by a neurologist demonstrating the participant has a normal neurologic examination or an abnormality on examination that could not be explained by the location of the brain lesion(s).

d. Acute severe vaso-occlusive pain episodes requiring hospitalization and recalcitrant to maximum medical therapy. Episodes of pain to be adjudicated by selected committee.

e. At least one acute chest syndrome episode resulting in intensive care admission requiring non-mechanical ventilatory support: simple nasal cannula, face mask that requires oxygen content (venti mask, non-rebreather), simple nasal cannula, face mask

f. O2 (e.g. ventimask, non-rebreather), CPAP, SiPAP, BiPAP, high flow nasal cannula (HFNC) or invasive mechanical ventilatory support (delivered by ETT or trach).

g. Right heart catherization confirmed pulmonary artery pressure >25 mmHg or mean pulmonary vascular resistance 206(57-421) dyn·s·cm^{-5}.

h. Essential hypertension on antihypertensive medications >95% upper limit of normal for age (as defined according to the American Academy of Pediatrics).

i. Recurrent priapism (episodes lasting at least 4 hours at least twice in the last 12 months or 3 times in the last 24 months) recalcitrant to medical treatment or unable to use hydroxyurea due to SCD phenotype, with the approval of the adjudication committee.

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[1]The absence of bridging fibrosis will be determined using the histological grading and staging scale as described by Ishak and colleagues (1995)[1,2,3].

2.2.1.3 Inclusion Criteria for Stratum 2: Adults Ages 15.00 - 45.99 at enrollment

1. Age 15.00-45.99 years at Segment A enrollment
2. Participants with sickle cell anemia (any clinically significant sickle genotype, for example, Hb SS, SC, SD Sβ° Thalassemia, Sβ^{+} Thalassemia, S-OArab) who have **one or more** of the following:

   a. A neurological event resulting in focal neurologic deficits that

lasted ≥ 24 hours (classical clinical definition of stroke, not requiring imaging studies of the brain) **OR** a focal neurological event resulting in abnormalities on T2-weighted or FLAIR images using a MRI scan, indicative of an acute infarct, with no other reasonable medical explanation (definition of a stroke supported with MRI imaging scans of the brain), **OR** both.

b. History of two or more episodes of acute chest syndrome (ACS) in the 2-year period preceding enrollment despite the institution of supportive care measures (i.e. asthma therapy and/or hydroxyurea);

c. History of three or more severe vaso-occlusive pain crises per year in the 2-year period preceding enrollment despite the institution of supportive care measures (i.e. a pain management plan and/or treatment with hydroxyurea); painful episodes related to priapism, osteonecrosis or any sickle-related complication are acceptable;

d. Administration of regular red blood cell (RBC) transfusion therapy, defined as 8 or more transfusion events per year (in the 12 months before enrollment) to prevent vaso- occlusive clinical complications (i.e. pain, stroke, or acute chest syndrome);

e. An echocardiographic finding of tricuspid valve regurgitant jet velocity (TRJV) ≥ 2.7 m/sec.

2.2.2 Exclusion Criteria

Participants fulfilling any of the following criteria are ineligible for participation on this protocol:

1. Participants who have an HLA-matched sibling who is able and willing to donate bone marrow. Patients with an HLA-matched unrelated donor are not excluded.
2. Uncontrolled bacterial, viral or fungal infection in the 6 weeks before enrollment (currently taking medication with evidence of progression of clinical symptoms or radiologic findings).
3. Evidence of HIV infection or known HIV positive serology.
4. Participants who have received a previous HCT.
5. Participants who have had an Encephaloduroarteriosynangiosis (EDAS) procedure in the 6 months prior to enrollment.
6. Participants who have received a prior solid organ transplant.
7. Participants who have participated in another clinical trial in which the patient received an investigational or off-label use of a drug or device within 3 months of enrollment.
8. Females who are pregnant or breastfeeding.
9. Participants with clinically significant, uncontrolled autoimmune disease, requiring active medical management (immunosuppressive therapy or

chemotherapy), which, in the judgment of the local Principal Investigator, indicates that the patient could not tolerate transplantation.

10. Females of child bearing potential (to include all female participants > 10 years of age, unless postmenopausal for a minimum of 1 year before the time of consent or surgically sterilized), who do not agree to practice two (2) effective methods of contraception at the same time, or do not agree to practice true abstinence when this is in line with the preferred and usual lifestyle of the subject, from the time of signing of informed consent through 12 months post-transplant.
11. Males (even if surgical sterilized) who do not agree to practice effective barrier contraception, or who do not agree to practice true abstinence from the time of signing informed consent through 12 months post-transplant.
12. Presence of anti-donor specific HLA antibodies. HLA antibody presence and specificity will be determined by solid phase immunoassays. An anti-donor specific HLA antibody will be considered positive when the mean fluorescence intensity (MFI) is higher than the cut-off defined by each institution. Recommended cut-off values are MFI >1000 for donor specific antibody to HLA-A, -B, and DRB1 and MFI >2000 for HLA-C, DQB1 and DPB1. This must be measured before the final donor selection, and $\leq$ 100 days before enrollment in Segment A (preferably $\leq$ 30 days before Segment A enrollment). If MFI >1000 for donor specific antibody to HLA-A, -B, DRB1 and/or MFI >2000 for HLA-C, DQB1 and DPB1, documentation must be submitted to the DCC coordinator for review and approval by a Protocol Chair and/or Protocol Officer prior to enrollment.

2.3 Donor Eligibility for Haploidentical-bone marrow

2.3.1 Donor Selection Criteria

Donors must be:

1. HLA-haploidentical first-degree relatives of the patient(Section 2.2.1.1 describes acceptable HLA typing and relationship to the patient)
2. The donors must be willing to donate bone marrow
3. Donor must meet institutional criteria for donation.

2.3.2 Donor Prioritization Schema

In the event that two or more haploidentical donors are identified (section 2.3.1), the following order of priority is recommended:

1. Donor age <40 years
2. Avoid female donors for male recipients
3. Avoid major/minor ABO mismatched transplants
4. Avoid CMV mismatched donor-recipient transplants

2.3.3 Donor Exclusion Criteria

Donors fulfilling the following criteria are ineligible for registration onto this study:

1. All donors will be screened by hemoglobin electrophoresis; donors with a clinically significant hemoglobinopathy are ineligible. Sickle trait is acceptable.

2.4 Treatment Plan

2.4.1 Indwelling central venous catheter

Placement of a double lumen central venous catheter will be required for administration of IV medications and transfusion of blood products.

2.4.2 Preparative regimen

The preparative regimen described below and in Table 2.4.2 will be used for all patients enrolled and transplanted on the study. Any modifications to the preparative regimen must be reported to the BMT CTN DCC protocol coordinator for review and receive written approval by the Protocol Chairs/Officer *prior* to initiation of the preparative regimen.

Any deviation from the protocol-specified regimen must be reported to the BMT CTN DCC protocol coordinator.

TABLE 2.4.2: PREPARATIVE AND TREATMENT REGIMEN

Days –70 → –10	Hydroxyurea 30 mg/kg po daily
	Hgb S fraction < 35% prior to administration of Thymoglobulin. If Hbg S > 35%, automated exchange transfusion (RBC apheresis) is the preferred strategy to lower Hgb S with a goal of 20% and the maximum Hgb S fraction not to exceed 35%.
Day –9***	Thymoglobulin 0.5 mg/kg IV with pre-meds
Day –8***	Thymoglobulin 2 mg/kg IV qd with pre-meds
Day –7***	Thymoglobulin 2 mg/kg IV qd with pre-meds Thiotepa 5 mg/kg IV q 12 h (10 mg/kg total)
Days –6, –5	Fludarabine 30 mg/m^2 IV over 30-60 minutes, then Cyclophosphamide (CY) 14.5 mg/kg IV over 1-2 hours*
Days –4 → –2	Fludarabine 30 mg/m^2 IV over 30-60 minutes
Day –1	TBI 200 cGy with gonadal shielding**
Day 0	Non-T-cell depleted bone marrow
Days +3, +4	Cyclophosphamide 50 mg/kg IV Mesna 40 mg/kg IV*
Day +5	Begin sirolimus (section 2.4.5.1)**, mycophenolate mofetil (MMF) 15 mg/kg pot id with maximum daily dose 3 gm/d
Day +35	Discontinue MMF

*Uroprophylaxis may be altered per institutional preference (see below)
**See Section 2.4.2.10 and section 4.4.6 regarding planned protocol change in TBI dose if safety monitoring guideline for 100-day graft failure is passed in the first 12 evaluable participants per stratum.
***If using corticosteroids as premedication, discontinue before Day -1.

The following are suggested dose adjustment formulas:

2.4.2.1 Recommended Ideal Body Weight Calculation for Children Age 5-17 Years:

$$IBW = (Height\ (cm)^2 \times 1.65)/1000$$

2.4.2.2 Recommended Ideal Body Weight Calculation for Adults Age ≥ 18 Years:

IBW (females) = (Height (cm) ÷ 2.54 – 60) x 2.3 kg + 45.5 kg IBW (males) = (Height (cm) ÷ 2.54 – 60) x 2.3 kg + 50 kg

2.4.2.3 Adjusted Ideal Body Weight (AIBW) (all ages): AIBW = adjusted ideal body weight is estimated as follows: AIBW = IBW + [(0.25) x (ABW - IBW)]

(ABW = actual body weight)

2.4.2.4 Patients Receiving Iron Chelation Therapy before BMT

Iron chelation therapy will be discontinued before commencing hydroxyurea on Day -70. Iron chelation therapy or a program of phlebotomy may be resumed after neutrophil and red cell recovery at the discretion of the transplant center.

2.4.2.5 Hydroxyurea

Hydroxyurea (30 mg/kg/day) will be commenced as a single daily oral dose on Day -70 and continued through Day -10. Hydroxyurea will be adjusted to the ideal body weight (IBW) in children and adults weighing > 125% IBW. A CBC will be obtained weekly and the dose will be reduced if the ANC<1500 or platelet <100,000. HU will be discontinued on Day -10.

2.4.2.6 Thymoglobulin

Thymoglobulin (rATG) will be infused through a 0.22 micron filter with pre-medications to include: Oral or intravenous (IV) acetaminophen and diphenhydramine. An anaphylaxis kit will be kept at bedside during Thymoglobulin administration. The dose will be 0.5 mg/kg IV on Day -9 over 6 hours and 2 mg/kg IV on Days -8 and -7 over 4 hours. Solumedrol should be administered per institutional standard to prevent infusional toxicity. An intradermal skin test prior to dose of Thymoglobulin may be performed per institutional practice.

2.4.2.7 Thiotepa

Thiotepa 5 mg/kg will be administered IV every 12 hours over 2 hours or per institutional guidelines on Day -7 (administer 2 doses to total 10 mg/kg). Thiotepa will be adjusted to the ideal body weight (IBW) in children weighing > 125% IBW.

2.4.2.8 Fludarabine

Fludarabine 30 mg/m^2/day will be administered over 30-60 minutes intravenous infusion on Days –6 through –2 for a total dose of 150 mg/m^2. Fludarabine will be dosed according to the recipient's actual body weight. Patients who have an estimated or measured creatinine clearance < 70 mL/min/1.73 m^2 are excluded from enrollment, thus there will be no adjustment in the fludarabine dose for renal insufficiency.

2.4.2.9 Pre-transplantation Cyclophosphamide

Hydration prior to cyclophosphamide may be given according to institutional standards. A **recommended** approach is as follows: Patients are instructed to increase fluids overnight before cyclophosphamide administration. Hydration with normal saline at 3 mL/kg/hr IV will be started 2 hours prior to cyclophosphamide, then the rate will be reduced to 2 mL/kg/hr for 1 hour pre-cyclophosphamide and continued at 2 mL/kg/hr for 8 hours post-cyclophosphamide.

Mesna may be administered per institutional standards. Mesna dose will be based on the cyclophosphamide dose being given. The total daily dose of mesna must be equal to 80% of the total daily dose of Cy. A suggested approach is as follows: divided doses IV 30 min pre-Cy and at 3, 6, and 8 hours post-Cy.

Cyclophosphamide 14.5 mg/kg/day will be administered as a 1-2 hour intravenous infusion on Days –6 and –5. Cyclophosphamide will be dosed according to the recipient's ideal body weight

(IBW), unless the patient weighs more than 125% of IBW, in which case the drug will be dosed according to the adjusted IBW (AIBW; see Section 2.4.2.3 for formulas). Uroprotection can be administered according to institutional guidelines. Mesna is recommended to accompany pre-transplantation Cy but is not required.

Cyclophosphamide will be dosed according to the recipient's ideal body weight (IBW), unless actual body weight is less than IBW, in which case use actual body weight. If the patient weighs greater than125% of IBW, cyclophosphamide will be dosed according to the adjusted IBW (AIBW).

2.4.2.10 Total Body Irradiation

200 cGy TBI will be administered in a single fraction on Day –1. Radiation sources and dose rates will be defined by the institution. TBI may be delivered from either linear accelerator or Cobalt sources. Graft may be infused on the same day as TBI administration as long as there is 4-6 hours between administration of TBI and infusion of bone marrow.

If graft failure rate exceeds that specified in the 100-day safety monitoring guideline for graft rejection and notice is provided to all centers by the DCC, then future participants will be administered 400 cGy TBI in a single fraction on Day -1.
For either the 200cGy TBI or 400cGy TBI, gonadal shielding will be used in all male patients. Gonadal shielding will not be used in females.

2.4.3 Bone marrow transplantation and graft information

On Day 0, patients will receive unprocessed bone marrow unless there is a major ABO incompatibility, in which case red blood cells will be depleted from the donor marrow using institutional practices. Institutional practices will determine if there will be processing for minor ABO incompatibilities. Donor bone marrow will be harvested to minimum yield of **2.5×10^8** with a target yield of 4×10^8 nucleated cells per kilogram of recipient's IBW or actual BW (ABW), whichever BW is lower. We recommend taking no more than 10 mL per aspirate. In addition to calculating the total nucleated cell dose /kg, flow cytometry will need to be done on the sample to determine the content of $CD34^+$cells. The use of cryopreserved marrow is not permitted.

2.4.4 Post-Transplantation cyclophosphamide

Cyclophosphamide 50mg/kg will be given as an IV infusion over 1-2 hours (depending on volume) on Day 3 post-transplant (between 60 and 72 hours after marrow infusion) and on Day 4 post-transplant (24 hours after Day 3 cyclophosphamide).

Corticosteroids may not be used as anti-emetic agent and should not be administered until 24 hours after the completion of post-transplant cyclophosphamide unless used for adrenal support or during a medical emergency (e.g., treatment of anaphylaxis).

2.4.5 GVHD prophylaxis

On Day 5, patients will begin prophylaxis with Sirolimus and Mycophenolate Mofetil (MMF).

2.4.5.1 Sirolimus

Sirolimus for participants ≥18.00 years old: A one-time sirolimus loading dose, 6 mg PO, is given on Day 5, at least 24 hours after Cyclophosphamide completion. Sirolimus is then continued at a maintenance dose (start 2 mg PO QD), with dose adjustments to maintain a trough of **5 – 15 ng/mL** as measured by HPLC or immunoassay**.** There is no planned taper. Sirolimus prophylaxis is discontinued after the last dose on Day 365 or may be continued if there is GVHD. Sirolimus troughs should be measured weekly at a minimum.

Sirolimus for participants < 18.00 years old: Sirolimus dosing is based on actual body weight; however, an adjusted body weight may be used if the actual weight is > 50% greater than IBW. A one-time sirolimus loading dose, 3 mg/m2 PO with the dose not to exceed 6 mg, is given on Day 5, at least 24 hours after Cyclophosphamide completion. Sirolimus is then continued at a maintenance dose (start 1 mg/m2 PO QD, maximum 2 mg PO QD), with dose adjustments to maintain a trough of **3 – 12 ng/mL** as measured by HPLC or immunoassay. There is no planned taper. Sirolimus prophylaxis is discontinued after the last dose on Day 365 or may be continued if there is GVHD.

2.4.5.2 Mycophenolate Mofetil

MMF will be given at a dose of 15 mg/kg PO TID (based upon actual body weight) with the maximum total daily dose not to exceed 3 grams (1 g PO TID). MMF prophylaxis will begin on Day 5 post-transplant and will be discontinued after the last dose on Day 35 or may be continued if active GVHD is present.

2.4.6 Encephaloduroarteriosynangiosis (EDAS)

If the local team elects Encephaloduroarteriosynangiosis (EDAS) to be performed prior to transplant as adjunctive therapy for secondary stroke prevention, then it is strongly recommended to delay the initiation of pre-transplant hydroxyurea treatment at least 6 months due to the minimum time expected for the revascularization to benefit from the engrafted vessel. Since hydroxyurea is a myelosuppressive agent, treatment with hydroxyurea may impede healing of the new engrafted vessels after the revascularization procedure.

2.4.7 Additional Supportive Care

Ursodiol should begin at time of hydroxyurea conditioning (Day -70) in all patients. Supportive care for VOD will follow institutional practice. There is a significant risk of intracranial hemorrhage after HCT in patients with SCD and stroke. For this reason and because of inconsistent efficacy, heparin and low molecular weight heparin (LMWH) are not permitted for VOD/SOS prophylaxis. [48,49]

2.4.8 Infection Prophylaxis

Participants will receive infection prophylaxis and nutritional support according to institutional guidelines. Infection prophylaxis should include, but is not limited to, agents or strategies (e.g., PCR screening and preemptive therapy) to reduce the risk of bacterial, herpes simplex, CMV, HHV-6, EBV, Pneumocystis jiroveci, and fungal infections. Penicillin prophylaxis or its equivalent should be administered per institutional standard due to asplenism that occurs insickle cell disease.

2.4.9 EBV Monitoring

It is recommended that patients will have EBV DNA quantitative PCR testing on peripheral blood every two weeks from Day 14 to Day 100. In the event of persistent EBV virema or signs/symptoms consistent with EBV-related post-transplant lymphoproliferative disease (PTLD, adenopathy, fever, etc.) therapy with rituximab is recommended per local institutional standard.

2.4.10 Transfusion Support

The hemoglobin level must be maintained between 9.0 and 11.0 g/dL and platelet count 50,000/mm^3 after transplantation until transfusion independent to minimize the risk of neurological adverse events. Irradiated blood products should be administered universally, and Cytomegalovirus (CMV) negative or leuko-filtered blood products are recommended for CMV sero-negative recipients.

2.4.11 Indwelling Central Venous Catheter

Placement of a double or triple lumen central venous catheter will be required for the transplantation procedure and administration of IV medications and transfusion of blood products. This catheter may be removed and replaced as clinically indicated. However, the graft MUST be infused through a central line.

2.4.12 Prevention of post-BMT Neurological Events and Posterior Reversible Encephalopathy Syndrome (PRES)

Prophylaxis against seizures is mandatory in all recipients and should be commenced at the start of thymoglobulin conditioning. Suitable drugs for prophylaxis should be administered according to institution guidelines. Seizure prophylaxis should be continued for 365 days after transplant or until sirolimus is discontinued, whichever is later. Serum magnesium level should be maintained > 1.5 mg/dL during the period of treatment to reduce the risk of seizures.

Hypertension should be strictly controlled to prevent central nervous system (CNS) toxicity. Blood pressure should be monitored closely and both systolic and diastolic hypertension should be treated promptly to maintain blood pressure at the patient's pre-transplant baseline. Explicit orders must be written to intervene if systolic or diastolic blood pressure exceeds 10% over baseline. Detailed guidelines for management of Posterior Reversible Encephalopathy Syndrome (PRES) are included in Appendix I.

2.4.13 Pre-infusion Medication and Hydration Regimen

The pre-medication and hydration regimen prior to blood products transfusion and transplantation will be given following institutional guidelines.

2.4.14 Contraception Practices

Females of child bearing potential (to include all female participants > 10 years of age, unless postmenopausal for a minimum of 1 year before the screening visit or surgically sterilized), must agree to practice two (2) effective methods of contraception at the same time, or agree to practice true abstinence when this is in line with the preferred and usual lifestyle of the subject, from the time of signing of informed consent through 12 months post-transplant. Periodic abstinence (e.g., calendar, ovulation, symptothermal, post-ovulation methods) and withdrawal are not acceptable methods of contraception.

Male subjects (even if surgically sterilized) must agree to one of the following: practice effective barrier contraception, or practice true abstinence when this is in line with the preferred and usual lifestyle of the subject, from the time of signing of informed consent through 12 months post-transplant. Periodic abstinence and withdrawal are not acceptable methods of contraception.

2.4.15 Management of Slow Engraftment and Graft Failure

Slow engraftment or graft failure shall be managed according to institutional practices and may include the administration of colony stimulating factors and prophylactic antibiotics.

Graft failure following BMT in patients with sickle cell disease is usually associated with autologous reconstitution of the bone marrow with host hematopoiesis. It is associated with a steady decline in donor chimerism, increasing representation of hemoglobin S (in the absence of ongoing RBC transfusion therapy) and clinical manifestations of sickle cell disease. A second transplant or donor cellular infusion should not be considered unless the patient has < 5% donor chimerism.

2.5 Study Drug Information

2.5.1 Fludarabine

Fludarabine phosphate is commercially available.

Fludarabine phosphate is purine antimetabolite that, after administration, undergoes rapid conversion in plasma to the nucleoside 2-fluoro ara-A (F-araA). F-araA subsequently enters cells where it is phosphorylated to F-araATP and the monophosphate F-araAMP. Once activated, F-araATP inhibits DNA polymerase and ribonucleotide reductase. The monophosphate F-araAMP, once incorporated into DNA, is an effective DNA chain terminator.

Fludarabine monophosphate, 50 mg/vial, is reconstituted with 2 ml of sterile water, resulting in a 25mg/ml solution. The desired dose is further diluted to concentrations of 0.04-1 mg/ml in normal saline or 5% dextrose (50-100ml) for injection and will be administered by IV infusion over 30 minutes or longer.

Following IV administration, the drug is metabolized to 2-F-araA and widely distributed in tissues. 2-F-araA is excreted primarily in urine and has a terminal elimination half-life of 7 to 12 hours.

Clinical toxicities of fludarabine monophosphate include myelosuppression, primarily lymphopenia and granulocytopenia, alopecia, rash, dermatitis, nausea, vomiting, anorexia, stomatitis, diarrhea, somnolence, fatigue, peripheral neuropathy, mental status changes, cortical blindness, hepatocellular toxicity with elevation in serum transaminase, and interstitial pneumonitis. These effects are reversible when the drug is discontinued.

Fludarabine may cause dose-related CNS toxicity, including the posterior reversible encephalopathy syndrome (PRES), acute toxic leukoencephalopathy (ATL) and other leukoencephalopathy conditions (OLE). PRES is usually reversible with supportive care. Typical clinical features include seizures, persistent headache, and vision changes, accompanied by variable mental status alterations. Patients with ATL can present with cognitive dysfunction, decreased levels of consciousness, and vision changes. Other leukoencephalopathy (OLE) includes patients who behave similar to the ATL group, but with less prominent deep white matter changes on MRI. Both ATL and OLE are less likely to be reversible[50].

Fludarabine will be administered by IV infusion over 30 minutes in a dose of 30 mg/m^2/day on Days -6 to -2.

2.5.2 Cyclophosphamide (Cytoxan®)

Cyclophosphamide is commercially available.

Cyclophosphamide is an alkylating agent which prevents cell division primarily by cross-linking DNA strands. Cyclophosphamide is cell cycle non-specific.

Cyclophosphamide for injection is available in 2000 mg vials which are reconstituted with 100 ml sterile water for injection. The concentration of the reconstituted product is 20 mg/ml. The calculated dose will be diluted further in 250-500 ml of Dextrose 5% in water. Each dose will be infused over 1-2 hr (depending on the total volume).

Clinical toxicities of cyclophosphamide include alopecia, nausea and vomiting, headache and dizziness, hemorrhagic cystitis, cardiotoxicity, immunosuppression, myelosuppression, pulmonary fibrosis, increased hepatic enzymes and syndrome of inappropriate anti-diuretic hormone (SIADH).

2.5.3 Mesna (sodium-2-mercapto ethane sulphonate)

Mesna is a prophylactic agent used to prevent hemorrhagic cystitis induced by the oxasophosphorines (cyclophosphamide and ifosphamide). It has no intrinsic cytotoxicity and no antagonistic effects on chemotherapy. Mesna binds with acrolein, the urotoxic metabolite produced by the oxasophosphorines, to produce a non-toxic thioether and slows the rate of acrolein formation by combining with 4-hydroxy metabolites of oxasophosphorines.

Mesna is available in 200 mg, 400 mg and 1000 mg vials containing a 100 mg/ml solution. Each dose of mesna will be diluted further in 50 ml of normal saline to be infused over 15 minutes (or

as per institutional standards). Mesna dose will be based on the cyclophosphamide dose being given. The total daily dose of mesna is equal to 80% of the total daily dose of cyclophosphamide.

At the doses used for uroprotection mesna is virtually non-toxic. However, adverse effects which may be attributable to mesna include nausea and vomiting, diarrhea, abdominal pain, altered taste, rash, urticaria, headache, joint or limb pain, hypotension and fatigue.

2.5.4 Thiotepa

Antineoplastic and alkylating agent, that reacts with DNA phosphate groups to produce cross-linking of DNA strands leading to inhibition of DNA, RNA, and protein synthesis; mechanism of action has not been explored as thoroughly as the other alkylating agents, it is presumed that the aziridine rings open and react as nitrogen mustard; reactivity is enhanced at a lower pH. No dosage adjustment provided in manufacturer's labeling. For renal or hepatic impairment, caution and dose adjustment may be required. Extensively hepatic; major metabolite (active): TEPA. Half-life elimination: Terminal (dose-dependent clearance): ~2 hours; excretion: Urine (as metabolites and unchanged drug)

Solution Reconstituted, Injection: Generic: 15 mg (1 ea); I.V.: Administer as a rapid injection. Infusion times may be longer for high-dose (unlabeled use) treatment.

The most common adverse reactions include: Fertility effects: May be mutagenic and teratogenic; Myelosuppression, secondary malignancies: Potentially carcinogenic; myelodysplastic syndrome and acute myeloid leukemia (AML) have been reported; Use with caution in participants with hepatic and renal impairment; dosage reduction recommended. It can cause significant skin toxicity due to lipid solubility, thus bathing three times daily during its administration through 24 hours after the last dose is strongly recommended.

2.5.5 Sirolimus (rapamycin, Rapamune®)

Sirolimus is an immunosuppressant that inhibits cytokine-stimulated T-cell activation and proliferation, and also inhibits antibody formation. The mean bioavailability of sirolimus after administration of the tablet is ~27% higher than the oral solution. Sirolimus oral tablets are not bioequivalent to the oral solution. Clinical equivalence has been demonstrated at the 2mg dose level; however, it is not known if higher doses are clinically equivalent on an mg to mg basis.

2.5.5.1 Sirolimus oral solution

Sirolimus oral solution (1 mg/mL) should be stored protected from light and refrigerated at 2°C to 8°C (36°F to 46°F). For dilution, the appropriate dose should be measured using an amber oral syringe, then added to a glass or plastic container that holds at least 60 mL. Before taking the dose, it should be diluted with water or orange juice then taken immediately; it should not be diluted with grapefruit juice. The syringe should be discarded after one use. Sirolimus oral solution provided in bottles may develop a slight haze when refrigerated, which does not affect product quality; allow the product to stand at room temperature and shake gently until the haze disappears.

2.5.5.2 Sirolimus tablets

Sirolimus tablets are available in 1mg and 2mg tablets that cannot be crushed or broken. Sirolimus tablets should be stored at 20° to 25° C (68°–77°F), protected from light. The most common adverse reactions of sirolimus are peripheral edema, hypertriglyceridemia, hypercholesterolemia, hypertension, increased creatinine, constipation, abdominal pain, nausea, diarrhea, headache, fever, urinary tract infection, anemia, thrombocytopenia, arthralgia, pain. Adverse reactions that have resulted in rates of sirolimus discontinuation >5% were increased creatinine, hypertriglyceridemia, and thrombotic thrombocytopenic purpura (TTP) / thrombotic microangiopathy (TMA). Sirolimus toxicities are summarized:

2.5.6 Summary of Sirolimus Toxicities

	Common (>20%)	**Occasional (5-20%)**	**Rare (<5%)**
Immediate (within 1-2 days)	Headache (L), hypertension (L), immunosuppression (L), fever, nausea, diarrhea, constipation	Chest pain, insomnia, dyspepsia, vomiting, *dyspnea*	Hypotension, asthma, cough, flu-like syndrome, tachycardia, anorexia, *hypersensitivity reactions*
Prompt (within 2-3 weeks)	Tremor (L), renal dysfunction, pain (abdominal, back, arthralgias), *hyperlipidemia* [c] *(hypercholesterolemia, hypertriglyceridemia),* hyperglycemia, edema including *peripheral edema*, anemia	Elevated LFT's (with elevated sirolimus levels) [a], stomatitis, infections (including UTI, URI), mild *thrombocytopenia*, *leukopenia*, electrolyte disturbances (hyper/hypokalemia [L], hypophosphatemia, hypomagnesemia [L]), rash, hives, pruritus, *delayed wound healing or dehiscence (L), proteinuria, TTP/HUS/TMA* [b] especially with concurrent CNI	Pleural and pericardial effusions, *pulmonary toxicity* (non-infectious pneumonitis, BOOP, pulmonary fibrosis), thrombosis, myalgias
Delayed (any time later during therapy, excluding above conditions)	Acne		Kidney disease, CHF, ascites, arthrosis, bone necrosis, osteoporosis
Late (any time after completion of treatment)			Lymphoproliferative disorders, skin malignancies
Unknown frequency and timing	Embryo/fetotoxic; unknown whether excreted in human milk		

(L): Toxicity may also occur later.

[a] Significant transaminitis, generally without sequelae, may occur. Sirolimus has been associated with higher rates of veno-occlusive disease after myeloablative conditioning.

[b] Incidence 3% to < 20% in a trial of kidney transplantation. In allogeneic BMT, increase in TMA from 4.2% with

tacrolimus or cyclosporine alone, versus 10.8% with tacrolimus/sirolimus combination was noted.[66]
[c] Lipid-lowering agent may be required; consider if fasting serum triglycerides are > 2.5 x ULN and recommend starting if > 800 mg/dL.

Sirolimus is associated with a high risk for VOD after myeloablative conditioning, but this risk is lower in patients after RIC HCT, such as the regimen that will be utilized in this study. There is a still a possibility of VOD/SOS after RIC conditioning, however as illustrated in a sub-group of patients treated by second RIC HCT after a first HCT[51].

Drug interactions: Sirolimus is known to be a substrate for both cytochrome CYP3A4 and P-glycoprotein. Agents that may increase sirolimus levels include tri-azole drugs (especially voriconazole and posaconazole*), amiodarone, calcium channel blockers, macrolide antibiotics (but not azithromycin), micafungin, gastrointestinal prokinetic agents (cisapride, metoclopramide), cimetidine, cyclosporine, grapefruit juice, and HIV protease inhibitors. Agents that may decrease sirolimus levels include anticonvulsants (carbamezepine, phenobarbital, phenytoin), rifamycins, St. John's Wort.

Dose adjustments: The sirolimus dose is adjusted to maintain a serum trough level of 5 –15 ng/mL. Changes in levels due to altered bioavailability should be apparent within 24-48 hours. For sirolimus without CNI as in this study, a 20-25% reduction of sirolimus dose is recommended for trough levels >15 – 18 ng/mL, and a 20-25% increase is recommended for trough levels < 5 ng/mL. Renal failure does not affect the excretion of sirolimus. Excretion is reduced in liver failure; impaired hepatic function should prompt consideration of reduction in sirolimus maintenance doses, but no dose adjustment of the loading dose is necessary.

Due to extreme interactions with voriconazole and posaconazole, these drugs are relatively contraindicated during sirolimus therapy. Sirolimus dose is to be reduced by 90% when voriconazole is initiated and should also be significantly reduced with posaconazole.

2.5.7 Mycophenolate Mofetil (Cellcept®)

Mycophenolate Mofetil is an ester prodrug of the active immunosuppressant mycophenolic acid (MPA). This active metabolite is a noncompetitive, reversible inhibitor of inosine monophosphate dehydrogenase (IMPDH). There are no pharmacokinetic interactions with ganciclovir, cotrimoxazole, oral contraceptives and cyclosporine.

Side effect profiles include diarrhea, leukopenia, sepsis, allergic reactions, and vomiting. There is also an increase in certain types of infection mainly from the herpes virus family (CMV, HSV & VZV) and Candida.

2.5.8 Rabbit antithymocyte globulin (rATG)

Thymoglobulin® [Anti-thymocyte Globulin (Rabbit)] is a purified, pasteurized, gamma immune globulin, obtained by immunization of rabbits with human thymocytes. This immunosuppressive product contains cytotoxic antibodies directed against antigens expressed on human T-lymphocytes. This drug is commonly used to treat graft rejection in kidney transplantation. It is also commonly used in bone marrow transplantation as part of the conditioning regimen to avoid graft failure and to prevent graft-versus-host disease.

Thymoglobulin is a sterile, freeze-dried product for intravenous administration after reconstitution with Sterile Water for Injection, USP (SWFI). Each 10 mL vial contains 25 mg anti-thymocyte globulin (rabbit) as well as 50 mg glycine, 50 mg mannitol, and 10 mg sodium chloride. After reconstitution with 5 mL SWFI, each vial of reconstituted product contains approximately 5 mg/mL of Thymoglobulin, of which >90% is rabbit gamma immune globulin (IgG). The reconstituted solution has a pH of 7.0 ± 0.4. Human red blood cells are used in the manufacturing process to deplete cross-reactive antibodies to non-T-cell antigens. The manufacturing process is validated to remove or inactivate potential exogenous viruses. All human red blood cells arefrom US registered or FDA licensed blood banks. A viral inactivation step (pasteurization, i.e., heat treatment of active ingredient at 60°C/10 hr) is performed for each lot. Each Thymoglobulin lot is released following potency testing (lymphocytotoxicity and E-rosette inhibition assays), and cross-reactive antibody testing (hemagglutination, platelet agglutination, anti-human serum protein antibody, antiglomerular.)

Adverse side effects include immunodeficiency, infusion related toxicities such as hypertension, chills, rigors, tachycardia, capillary leak syndrome, hyperglycemia, cytopenias, transient hepatitis, anaphylaxis, serum sickness, myalgias, sensory changes including hearing loss, headaches, renal toxicity, dyspnea and bronchial spasm, fevers. The drug is potentially teratogenic and is unknown if it can be passed to children in breastfeeding.

rATG–rabbit must be infused through a 0.22 micro filter with premedication per institutional standard. If corticosteroids are used, please discontinue prior to Day -1.
Note: Keep anaphylaxis kit at bedside during Thymoglobulin administration.

2.5.9 Hydroxyurea

Administration can cause anemia, neutropenia, thrombocytopenia, nausea, vomiting, mouth sores, rash, painful and difficult urination, kidney damage, headache, dizziness, jaundice, hallucinations, seizures, diarrhea, temporarily increased levels of liver enzymes, darkening of the skin, and nail changes.

2.5.10 Corticosteroids

Corticosteroids may not be used as an anti-emetic agent and should not be administered until 24 hours after the completion of post-transplantation cyclophosphamide, unless used for adrenal support or during a medical emergency (e.g. treatment of anaphylaxis). Corticosteroids may be used as a pre-medication during Thymoglobulin infusion and discontinued immediately thereafter.

2.5.11 Total Body Irradiation

Total body irradiation will be administered (200 cGy). This dose is a fraction of the irradiation delivered in typical myeloablative conditioning regimens, thus modulated toxicity is anticipated. Nonetheless, irradiation can cause short-term and long-term toxicities, particularly in children. The long-term toxicities include a risk of malignancy secondary to the exposure and a risk of infertility.

- Testicular shielding will be administered to all male patients.
  The testis is one of the most radiosensitive tissues, and even low doses of radiation can

cause impairment of function. In adult men, doses as low as 10 cGy can cause damage to spermatogonia[52]. At single fraction doses of 200 to 300 cGy there is overt damage to spermatocytes in adult men and return to pre-irradiation sperm concentrations and germinal cell numbers may take up to 30 months[53]. Fractionated doses are more toxic to spermatogenesis, and complete sterilization may occur if the fractionated dose exceeds 100 – 200 cGy. However, in the pre-pubertal state, the impact of radiotherapy is mitigated. In one study of 12 pre-pubertal boys who received 2400 cGy testicular irradiation to treat ALL, it was observed that testosterone levels were normal in all 12 and basal LH was normal in 9 and elevated in 3[54]. Nonetheless, to preserve fertility and Leydig cell function in post- pubertal males, we propose to administer testicular shielding to all males during the single fraction of TBI. This shielding will have no impact on the immunosuppressive effect of the radiotherapy.

- There will be no gonadal shielding in female patients.
  The effects of radiotherapy on ovarian function and fertility are dose- and age-dependent. However, ovarian doses of less than 400 cGy do not result in permanent ovarian dysfunction and the calculated LD50 of the human oocyte dose is approximately 400 cGy[55] [56] [57]. Thus, ovarian shielding to preserve ovarian function will not be necessary for the dose of TBI administered in this investigation; moreover, shielding in this setting could reduce the immunosuppressive effects of TBI if intra-abdominal lymph nodes were inadvertently shielded.

CHAPTER 3

STUDY ENDPOINTS

3.1 Primary Endpoint

The primary endpoint is event-free survival (EFS) at 2 years post-transplant estimated within each stratum. EFS is defined as survival without a qualifying event. Primary graft rejection, secondary graft rejection, second infusion of hematopoietic cells or death will count as events for this endpoint. A secondary endpoint will assess EFS at 1 year post-transplant.

3.2 Secondary Endpoints

All endpoints are stratum-specific.

3.2.1 Overall Survival

Overall survival will be described post-enrollment and 1 and 2 years post-HaploBMT. Death from any cause will be the event and patients will be censored at the date of last contact or two years post-HaploBMT whichever comes first. Start time will either be the date of enrollment or HaploBMT for the two objectives.

3.2.2 Event Free Survival

Event-free survival is defined as survival without a qualifying event.

3.2.3 Engraftment and Graft Rejection

3.2.3.1 Engraftment

Engraftment is defined as having greater than or equal to 5% donor cells post-transplant, from any molecular chimerism assessment (e.g., unsorted, myeloid, or T-cell) on a peripheral blood or bone marrow aspirate sample.

3.2.3.2 Primary Graft Failure

Primary graft failure is defined as never achieving ≥ 5% donor whole blood or myeloid chimerism (myeloid is preferable) assessed by bone marrow or peripheral blood chimerism assays by day +42 post-transplant. Second infusion of stem cells is also considered indicative of primary graft failure by day +42 post-transplant.

3.2.3.3 Secondary Graft Failure

Secondary graft failure is defined as < 5% donor whole blood or myeloid chimerism (myeloid is preferable) in peripheral blood or bone marrow beyond day +42 post-transplant in patients with prior documentation of hematopoietic recovery with ≥5% donor cells by day +42 post-transplant. Second infusion of stem cells is also considered indicative of secondary graft failure.

3.2.3.4 Second Transplant or Infusion of Hematopoietic Cells

Infusion of a second stem cell product will be considered graft rejection, and counted toward primary or, depending on timing of the second infusion, secondary graft rejection.

3.2.4 Lineage Specific Chimerism following HCT for SCD

Current methods for measuring hematopoietic chimerism are based on DNA polymorphisms that distinguish recipient from donor. Pyrosequencing of lineage-specific mRNA directly measures functional reconstitution of donor cells and provides valuable information that can affect clinical decisions in patients with nonmalignant diseases following allogeneic transplant. Peripheral blood samples on post-transplant days 28, 42, 100 and 180 days, 365 and 730 will be assayed into CD3+CD56- (T-lymphocytes) and CD15+, CD33+ or CD45+ (granulocytes) per institutional standard. Genomic DNA extracted from peripheral blood will be analyzed for variable number of tandem repeats (VNTR) to detect donor engraftment in mononuclear cells and CD3+ lymphocytes. Percent donor chimerism will be analyzed per institutional standard.

3.2.5 Transplant-related Complications

3.2.5.1 Grade II-IV and Grade III-IV Acute GVHD

Incidence of grade II-IV and III-IV acute GVHD will be graded according to the NIH consensus criteria (refer to Appendix D).

3.2.5.2 Chronic GVHD

Incidence and severity of chronic GVHD will be scored according to the NIH consensus criteria. (refer to Appendix E).

3.2.5.3 Neutrophil Recovery

Time to neutrophil recovery is defined as the first of 3 measurements on different days when the patient has an absolute neutrophil count of ≥500/μL after conditioning.

3.2.5.4 Platelet Recovery

Time to platelet recovery is defined as the first day of a minimum of 3 measurements on different days that the patient has achieved a platelet count > 50,000/μL AND did not receive a platelet transfusion in the previous 7 days. The exception is the case in which a subject is given a platelet transfusion specifically to achieve a platelet threshold to allow an elective invasive procedure, such as a central catheter removal.

3.2.5.5 Veno-occlusive Disease

Veno-occlusive disease (VOD) will be diagnosed and defined in accordance with either the modified Seattle or Baltimore criteria as outlined in Appendix J.

3.2.5.6 Idiopathic Pneumonia Syndrome (IPS)

IPS will be defined evidence of widespread alveolar injury and in which infectious etiologies and cardiac dysfunction, acute renal failure, or iatrogenic fluid overload have been excluded.[58] Refer to Appendix K for complete IPS criteria. Diagnosis via bronchoscopy is strongly encouraged.

3.2.5.7 CNS Toxicity

CNS toxicity will be defined as seizures, CNS hemorrhage, or Posterior reversible leukoencephalopathy syndrome (PRES or RPLS). PRES is defined as an increased diffusion coefficient in areas of T2 hyperintensities on diffusion-weighted imaging in the context of clinical symptoms or physical findings including headache, seizures, visual disturbances, and altered level of consciousness. Please refer to Appendix I for additional details and recommended guidelines regarding PRES prophylaxis and management

3.3 Infection

Significant infections will be recorded including but not limited to bacterial or fungal sepsis, CMV reactivation with/without clinical disease, adenovirus infection, EBV PTLD, or other significant viral reactivations or community-acquired viral mold infections and invasive fungal infections.

3.4 Sickle Vasculopathy

Evaluate if sickle vasculopathy is halted by successful transplantation as determined by comparing brain MRI pre- and 2 years post-haploBMT. Cerebral MRI is required for all pediatric patients to assess progression of CNS disease. For adults with where the indication for the transplant is a stroke, cerebral MRI is required after transplant to assess progression of CNS disease.

3.5 Disease Recurrence

Disease recurrence is defined as the return of sickle erythropoiesis (in the absence of RBC transfusion, Hb S level > 70%), or primary or secondary graft rejection, as defined above, or second infusion of hematopoietic cells.

3.6 Patient-reported HRQoL Outcomes including Pain and Fatigue Domains in the Adult Stratum

Using Patient Reported Outcome Measurement Information System (PROMIS) modules as the standardized assessment tool, patient reported outcomes in two specific quality of life domains, pain and fatigue, will be obtained from patients in the adult stratum at baseline (prior to the initiation of hydroxyurea), 1 year and 2 years post-transplant. Only English- and Spanish-speaking adult stratum patients are eligible to participate in the HRQoL component of this trial.

3.7 Pain Intensity

Change in pain intensity between the baseline (prior to the initiation of hydroxyurea), one-year, and two-year assessments for English-speaking patients ≥ 15.00 years of age at time of enrollment.

Based on estimates of daily pain in adults with SCD[59], we anticipate that patients will report pain in pain diaries with a mean pain intensity of 3.9 on a scale of 10 at baseline. We anticipate that adult patients will demonstrate a clinically significant decline in mean pain intensity to a level of 1.3 units on a scale of 10 two years later. While our endpoint is the change between the baseline and two-year assessments, we will also examine pain at one year, in order to better understand the trajectory of changes during the two year follow up period.

3.8 Disease-related Complications

3.8.1 Stroke

An overt stroke is defined as a focal neurologic event and neurologic deficit lasting more than 24 hours with neuroimaging changes. Patients with new MRI lesions and ongoing neurologic injury to the brain that does not result in focal motor impairment are referred to as having silent cerebral infarcts. These lesions are defined as a MRI signal abnormality measuring at least 3 mm visible on two views on T-2 weighted images. Both forms of neurologic injury that develop as a new event post-transplant will be considered a disease related complication.

3.8.2 Occurrence of Sickle Related Events

In addition to stroke as outlined above, participants will be followed for the entire 2 year duration of their time on study for the recurrence of SCD related complications. These SCD related complications are henceforth referred to as SCD events of special interest (SCD-EOSI). For all participants, the following specific, SCD related events are expected events of special interest (SCD-EOSI):

- pulmonary hypertension
- significant cerebrovascular events, including:
  - stroke
  - transient ischemic attack
  - seizure
- renal function compromise, including:
  - proteinuria
  - increased creatinine grades ≥2 per CTCAE version 4.0
- avascular necrosis of the hip or shoulder
- leg ulcers
- acute chest syndrome (ACS) requiring hospitalization
- painful vaso-occlusive crises (VOC) requiring hospitalization or parenteral opioid drugs in the outpatient setting (self-reported events without clinical documentation should not be included)

While these events do not require expedited reporting via completion of the Adverse Events forms, each occurrence of any SCD event of special interest (SCD-EOSI) must be reported on the SCD-

EOSI Form at each regularly scheduled follow up interval. A single event occurrence is defined as a new onset of any of the above listed SCD-EOSIs after screening evaluations are completed. A single occurrence resolves once the SCD-EOSI returns to the participant's baseline. Therefore, multiple occurrences of the same or several SCD-EOSIs may be reported in one reporting interval. The completion of the SCD-EOSI Form is required and further detailed in the Data Management Handbook.

3.9 Lung function (PFTs)

A growing body of evidence indicates the pulmonary toxicities associated with SCD are progressive in nature. A complete set of pulmonary function tests (PFTs) will be obtained at baseline (prior to the initiation of hydroxyurea), 1 year and 2 years post-transplant. In addition to assessing changes in FEV1 at 1 and 2 years post BMT as compared with baseline, we will also assess the proportion of participants in whom evidence of restrictive lung disease is found. For the purposes of this protocol, restrictive lung disease is defined as TLC below the 5th percentile adjusted for age, gender, race and height. In the interest of good clinical practice, PFTs should be obtained more frequently if clinically indicated, in the setting of cGVHD and/or in accordance with institutional standard of care mandates. A 6 minute walk test should accompany any significant decrease (>15% drop in predicted value of FEV1 or FEV1/FVC to determine if alternation in PFTs is associated with significant DOE).

3.10 TRJV (Cardiac Function)

Tricuspid regurgitant jet velocity (TRJV) is a marker for the severity and progression of SCD. Changes in TRJV from baseline (prior to the initiation of hydroxyurea) to 1 year and 2 years post-transplant will be measured.

3.11 6-minute Walk Distance (6MWD)

The 6MWD, a common assessment tool utilized in this disease population, will be administered at baseline (prior to the initiation of hydroxyurea), 1 year and 2 years post-transplant, with all testing to be administered under standardized procedures and in the same clinical setting. This endpoint will measure absolute change from baseline, with increased distance identified as positive change.

3.12 Hematological Outcomes

Hematological outcomes including Hgb, reticulocyte count, %HbS, LDH, bilirubin, and last date of red blood cell transfusion will be measured pre-transplant and on post-transplant days 28, 100 and 180 days, 365 and 730.

3.13 Proportion of Participants Still on Immunosuppression 2 years after BMT

Proportion of participants still receiving immunosuppressive therapy because of GVHD or concern about graft rejection will be determined.

CHAPTER 4

PATIENT ENROLLMENT AND EVALUATION

4.1 Screening Procedures

Prospective participants at participating institutions will be identified and recruited by the site investigator or their designee. The investigator or designee at the enrolling site will conduct a donor evaluation for prospective enrollees. If a suitable haploidentical donor is identified, written informed consent and assent for study enrollment review will be obtained and eligibility determined.

The participating center will register the participant using the BMT CTN Advantage Electronic Data Capture (AdvantageEDC) System. An authorized user at the center will complete the initial screening by entering patient demographics, the Segment 0 registration (date of informed consent), and the HLA form.

4.1.1 Participant Entry and Registration

If the participant is determined to be eligible for enrollment, an authorized user at the center will complete the Segment A enrollment form (inclusion/exclusion criteria). The Segment A form must be completed no more than 30 days prior to initiation of the hydroxyurea conditioning regimen. The date of completion of the Segment A enrollment form is the date of study enrollment. The Segment A enrollment successful screen **must** be obtained prior to initiating the protocol-specified dose of hydroxyurea.

4.1.2 Pain and Priapism Adjudication

For only the pediatric stratum, the protocol chairs will review a completed questionnaire for potential participants referred for multiple pain episodes or multiple priapism episodes. In addition to the responses to the questionnaire, children meeting either of the two criteria (pain or priapism) will be considered eligible; 1) acute severe vaso-occlusive pain episodes requiring hospitalization and recalcitrant to maximum medical therapy and 2) recurrent priapism (episodes lasting at least 4 hours at least twice in the last 12 months or 3 times in the last 24 months) recalcitrant to medical treatment or unable to use hydroxyurea due to SCD phenotype.

After the protocol chairs review the questionnaire responses and the two eligibility criteria, participants will be designated as eligible, not eligible, or not evaluable based on the available information.

4.2 Participant Assessments

4.2.1 Follow-up Schedule

The Follow-up Schedule for scheduled study visits is outlined in Table 4.2.1. A detailed description of each of the forms and the procedures required for forms completion and submission can be found in the Data Management Handbook and User's Guide.

TABLE 4.2.1: FOLLOW-UP SCHEDULE

Study Visit	Target Day
Pre-Enrollment	
Pre-Enrollment	≤ 180 days prior to Segment A Enrollment AND/OR ≤ 30 days prior to Segment A Enrollment as specified in section 4.2.2
Pre-transplant	
Pre-HU	Within 30 days Post Segment A Enrollment, Prior to start of Hydroxyurea
Pre-Thymoglobulin	≤ 30 days prior to start of Thymoglobulin
Post-transplant	
1 week	7 ± 3 days
2 week	14 ± 3 days
3 week	21 ± 3 days
4 week	28 ± 3 days
5 week	35 ± 3 days
6 week	42 ± 3 days
7 week	49 ± 3 days
8 week	56 ± 3 days
9 week	63 ± 3 days
10 week	70 ± 3 days
11 week	77 ± 3 days
12 week	84 ± 3 days
13 week	91 ± 3 days
100 day	100 ± 3 days
6 month	180 ± 28 days
One year	365 ± 28 days
Day 540	540 ± 28 days
Two years	730 ± 28 days

4.2.2 Pre-enrollment Evaluations

The following observations are required within the following time frames prior to transplant.

4.2.2.1 Evaluations Required Prior to Enrollment

The following evaluations are required ≤ 180 days prior to enrollment into Segment A unless otherwise noted below (considered standard of care evaluations for transplant patients). Observations prior to enrollment will be done according to institutional guidelines:

1. CBC with differential, reticulocyte count, serum creatinine, creatinine clearance, direct/conjugated bilirubin, alkaline phosphatase, ALT, and AST, serum ferritin, magnesium.
2. Solid Phase Immunoassay for HLA antibody specificity testing to confirm absence of anti-donor HLA antibodies (see Section 2.2.2 for definition of a positive anti-donor HLA antibody; preferably ≤ 30 days before Segment A enrollment and must be ≤ 100 days before Segment A enrollment).
3. Baseline EKG.
4. A 6-minute walk distance test.
5. Baseline 2-D transthoracic echocardiography and/or MUGA for left ventricular ejection fraction (LVEF), left ventricular shortening fraction, and presence or absence of tricuspid regurgitation (TRJV).
    a. If tricuspid regurgitation present, measure TRJV as a risk factor for early mortality and pulmonary hypertension.
6. Liver MRI for estimation of hepatic iron content is required for participants who are currently receiving ≥8 packed red blood cell transfusions per year for ≥1 year or have received ≥20 packed red blood cell transfusions (lifetime cumulative). Liver MRI must be performed using a validated methodology per institutional preference (T2* or R2* or by ferriscan [R2 MRI]).
    a. If hepatic iron content ≥ 10 mg Fe/g liver dry weight, a Gastroenterology/hepatology consultation with a liver biopsy is required to document the absence of cirrhosis, bridging fibrosis1[1], and active hepatitis.[2]
7. A cardiac (T2*) MRI imaging when clinically indicated. Given the uncommon event of asymptomatic cardiac hemosiderosis in SCD, patients should be considered forevaluation with cardiac MRI on a case-by-case basis and per institutional preference if liver iron concentration (LIC) is > 22 mg of iron per gram of dried tissue.[60,61]
8. Pulmonary function testing: Spirometry and lung volumes, including FEV1, FVC, and DLCO. Record oxygen saturation by pulse oximetry.
    a. Patients unable to perform DLCO due to young age or other inability must record absence or presence of dyspnea at rest and O2 saturation level.
9. HLA typing by allele-level methodology at HLA -A, -B, -C, and DRB1 loci, ABO and Rh typing. Confirmatory donor typing must be completed ≤ 100 days prior to enrollment in Segment A.

[1] The absence of bridging fibrosis will be determined using the histological grading and staging scale as described by Ishak and colleagues (1995)[1,2,3].

[2] If a liver biopsy is performed ≤180 prior to enrollment in Segment A, a Liver MRI is not required to be repeated prior to enrollment.

4.2.3 The following evaluations are required ≤ 30 days prior to enrollment into Segment A (considered standard of care evaluations for transplant patients). Observations prior to enrollment will be done according to institutional guidelines:

1. History, physical examination, height, and weight.
2. Lansky / Karnofsky performance status (Appendix F).
3. β-HCG serum pregnancy test for females of childbearing potential; to include all female participants > 10 years of age unless post-menopausal for a minimum of 1 year before the screening visit or surgically sterilized.
4. CMV antibody test, hepatitis panel (HepA Ab, HepB sAb, HepB sAg, HepB Core Ab, HepC Ab), herpes simplex, syphilis, HIV and HTLV1 I/II antibody, varicella zoster virus antibody, and EBV serostatus.
5. Hemoglobin evaluation by HPLC, electrophoresis, IEF, or gene test for quantification of: Hb F, Hb S, Hb A levels per institutional standards. In chronically transfused patients or those transfused within the last 3 months prior to enrollment, SCD genotype must be confirmed by one of the following:
    a. Prior laboratory determination of SCD genotype by referring providers, **OR**
    b. Confirmation test by HPLC or Hb electrophoresis after positive newborn screening, **OR**
    c. DNA based test from peripheral blood for globin chain analysis

4.2.3.1 Evaluations Required Post-Segment A Enrollment but Pre-Transplant:

The following evaluations are required post-Segment A enrollment, but prior to initiation of Hydroxyurea (Day -70)[1]:

1. Baseline health-related quality of life (HRQoL) assessments (PROMIS pain and fatigue domains) (Appendix G) for English- and Spanish-speaking patients in the adult stratum. PROMIS modules should be administered on a single day ≥ 7 days and ≤ 28 days from initiating the 28 day electronic pain diary.
2. 28-day pain diary for English-speaking patients ≥ 15.00 years of age at time of enrollment (twice daily for 28 days prior to starting HU). For patients who were on HU pre-enrollment, patients may complete pain diary while on protocol dose of HU.
3. Review and documentation of SCD Events of Special Interest (SCD-EOSI) at baseline (see Section 3.8.2)

[1] Refer to Section 2.4.6 for recommendations regarding Encephaloduroarteriosynangiosis (EDAS) performed prior to transplant as adjunctive therapy for secondary stroke prevention.

4.2.4 The following evaluations are required ≤ 30 days prior to initiation of Thymoglobulin (Day -9) and are considered standard of care evaluations in transplant patients:

1. Lansky / Karnofsky performance status (Appendix F).
2. CBC with differential, reticulocyte count, serum creatinine, direct/conjugated bilirubin, alkaline phosphatase, ALT, and AST, serum ferritin, magnesium. A CBC will be obtained weekly during hydroxyurea (Day -70 until Day -10) and the dose will be reduced if the ANC<1500 or platelet <100,000.
3. Radionuclide Glomerular Filtration Rate (GFR) or creatinine clearance (CrCl) (although GFR or CrCl is permissible, GFR is preferred)
4. Chest x-ray or chest CT scan (contrast not required for CT chest)
5. β-HCG serum pregnancy test for females of childbearing potential; to include all female participants > 10 years of age unless post-menopausal for a minimum of 1 year before the screening visit or surgically sterilized.
6. MRI/MRA of the brain to assess for radiologic evidence of cerebral vasculopathy.
    a. Cerebral MRI/MRA is required for all pediatric patients enrolled under any of the neurological criteria prior to transplant to assess status of CNS disease. For children ages 5.00 to 14.99 years of age with overt stroke ischemia, if there is clinical or radiologic evidence of a recent cerebral infarct, participants will be deferred for at least 6 months with repeat cerebral MRI/MRA to ensure stabilization of the neurologic event prior to proceeding to transplantation. Cerebral MRI images will be uploaded for central MRI review. Additional information is located on the BMT CTN website on the 1507 private study page.
    b. For adults where the indication for the transplant is a stroke, cerebral MRI/MRA is required prior to transplant to assess progression of CNS disease.
7. Optional Immune Reconstitution Research Samples as described in Appendix C:
    a. Children < 15: 24-40 mL into three-four 10 mL Vacutainer tubes, containing sodium heparin anticoagulant collected prior to initiating conditioning (Thymoglobulin).
    b. Adults >15: 40 mL into four 10 mL Vacutainer tubes, containing sodium heparin anticoagulant collected prior to initiating conditioning (Thymoglobulin).
    c. Donors: 20 mL into two 10 mL Vacutainer tubes, containing sodium heparin anticoagulant collected prior to donation.

4.2.5 Transplant Evaluations

Total nucleated cell and CD34+ cell content of the infused marrow product on Day 0.

4.2.6 Post-Transplant Evaluations

The following assessments should be conducted at the specified time points post-transplant:

1. History, physical exam, height, and weight weekly until Day 28, then at Day

100, six months, one year and two years post-transplant.

2. Lansky / Karnofsky performance status (Appendix E) weekly until Day 28, then at Day 100, six months, one year and two years post-transplant.
3. History and physical exam to assess GVHD and other morbidity weekly until Day 100 post-transplant, then at six months, one year, 18 months, and two years post-transplant. GVHD evaluation and grading as outlined in the BMT CTN Technical MOP.
4. Quality of life assessment (at 1 year and two years post-transplant) for English- and Spanish-speaking patients in the adult stratum using the PROMIS pain and fatigue domains (Appendix G). PROMIS modules should be administered on a single day ≥ 7 days and ≤ 28 days from initiating the 28-day electronic pain diary.
5. Pain diary twice daily over a 28-day period for English-speaking patients ≥ 15.00 years of age at time of enrollment (Year 1: day 337 to 365; Year 2: day 702 to 730).
6. CBC three times a week from Day 0 until neutrophil recovery. Platelet count three times a week from Day 0 until platelet recovery. Thereafter, CBC and platelet count twiceweekly until Day 28, then weekly until 12 weeks, then six months, one year and two years post- transplant.
7. Weekly reticulocyte counts beginning on Day 28 until Day 100, or until > 30,000/µL on two different days, then at six months, one year and two years post-transplant.
8. Quantitative hemoglobin evaluation (Hb F, Hb A, Hb A2, and Hb S) and last date of red blood cell transfusion on Day 28, 100, 6 months, one year, and two years post-transplant.
9. Creatinine, bilirubin, alkaline phosphatase, ALT, AST, magnesium twice weekly until Day 28, then weekly until 12 weeks, and then at Day 100, six months, one year and two years post-transplant.
10. Serum ferritin at 1 year and two years post-transplant.
11. Radionuclide Glomerular Filtration Rate (GFR) or creatinine clearance (CrCl) (although either GFR or CrCl is permissible, GFR is preferred); urine protein (or microalbumin) to creatinine ratio at 1 year and 2 years post-transplant.
12. Heparinized peripheral blood sample for post-transplant chimerism assay by molecular methods collected at Day 28, Day 42[1], and at Day 100, 6 months and 1 and 2 years. All patients must have chimerism assay between day 21 and day 42. For patients with primary graft failure (i.e. anytime within day 42 reporting period) a separate measurement is not required. For patients with mixed chimerism (tested between day 21 – 42), follow up chimerism assays per local institutional standard is recommended to document sustained engraftment or graft failure. Secondary graft rejection is based on two separate measurements obtained a **minimum of 1 week and a maximum of 2 weeks apart**.

[1]The absence of bridging fibrosis will be determined using the histological grading and staging scale as described by Ishak and colleagues (1995)[1,2,3].

13. Fractionated chimerism examining the myeloid and lymphoid fractions at Day 28, Day 42[1], and at Day 100, 6 months and 1 and 2 years (bone marrow aspirate preferred, particularly at Day 100). If a center is unable to perform the bone marrow aspiration procedure as standard of care, peripheral blood is acceptable. Secondary graft rejection is based on two separate measurements obtained a **minimum of 1 week and a maximum of 2 weeks apart**.
14. Pulmonary function testing with FEV1, FVC, DLCO, and oxygen saturation by pulse oximetry at 1 and 2 years post-transplant.
    a. Patients unable to perform DLCO, lung volumes or spirometry due to young age or other inability must record absence or presence of dyspnea at rest and O_2 saturation level.
15. For adults where the indication for the transplant is echocardiographic finding of tricuspid valve regurgitant jet velocity (TRJV) ≥ 2.7 m/sec, at a minimum surveillance with 2-D transthoracic echocardiography for assessment of LVEF, left ventricular shortening fraction, presence or absence of tricuspid regurgitation at one- and two-years post-transplant is recommended. As some institutional standards may not require surveillance for left ventricular function or assessment of TRJV if the last velocity is < 2.7 m/sec, echocardiogram assessments post-transplant may be performed per institutional standards.
16. A 6-minute walk distance test at one- and two-years post-transplant.
17. MRI/MRA of the brain to assess for radiologic evidence of cerebral vasculopathy will be obtained at 1- and 2-years post-transplant. Cerebral MRI is required for all pediatric patients 1- and 2-years post-transplant to assess status of CNS disease. For adults where the indication for the transplant is a stroke, cerebral MRI is required 1- and 2-years post- transplant to assess status of CNS disease. Cerebral MRI images will be uploaded for central MRI review. Refer to the BMT CTN 1507 private study page for details.
18. Liver MRI is recommended when clinically indicated and per institutional preference. Patients with transfusional iron overload (defined as liver iron concentration ≥ 10 mg Fe/g liver dry weight) should be evaluated by a validated methodology (T2* or R2* or by ferriscan [R2 MRI]) for hepatic iron concentration per institutional preference at one year and two years post-transplant.
19. A cardiac (T2*) MRI imaging when clinically indicated. Given the uncommon event of asymptomatic cardiac hemosiderosis in SCD, patients should be considered for evaluation with cardiac MRI on a case-by-case basis and per institutional preference if liver iron concentration (LIC) is > 22 mg of iron per gram of dried tissue (1- and 2-years post- transplant).
20. Review and documentation of SCD Events of Special Interest (SCD-EOSI) at 6 months, 1 year, Day 540, and 2-years post-transplant (refer to Section 3.8.2)
21. HbS percent beginning at Day 100, 1- and 2-years post-transplant for patients who are transfusion independent.

22. Optional Immune Reconstitution Research Samples at Days 28, 60, 100, 180, and 1 year post-transplant as described in Appendix C:
    a. Children < 15: 24-40 mL into three-four 10 mL Vacutainer tubes, containing sodium heparin anticoagulant.
    b. Adults >15: 40 mL into four 10 mL Vacutainer tubes, containing sodium heparin anticoagulant.

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[1] In the event that the chimerism assessment at day 28 reflects ≥5% donor hematopoietic cells by whole blood or myeloid chimerism (myeloid is preferable) in peripheral blood or bone marrow, the day 42 chimerism does not need to be completed

TABLE 4.2.2A: SCHEDULE OF EVALUATIONS PRIOR TO TRANSPLANT[1]

	Pre-Enrollment		Pre-Transplant	
Assessments	**≤180 days prior to Enrollment**	**≤30 days prior to Enrollment**	**Pre-HU (Baseline)[2]**	**Prior to Thymoglobulin[3]**
CBC with differential and blood chemistries[4]	X			X[8]
Renal Function Assessments[5]	X			X
Anti-donor HLA Antibody Testing	X			
EKG	X			
6-minute walk distance test	X			
Echocardiogram or MUGA[6]	X			
Liver MRI	X			
Liver biopsy[7]	X			
Cardiac MRI imaging	X			
Pulmonary Function Assessments *(per section 4.2.2.1)*	X			
Confirmatory HLA typing *(per section 4.2.2.1)*	X			
History, physical exam, height, weight		X		
Karnofsky/Lansky Performance Score		X		X
Pregnancy Test		X		X
Infectious disease titers *(per section 4.2.2.1)*		X		
Hemoglobin evaluation and date of last RBC transfusion		X		
QOL assessment (PROMIS)			X	
28-Day Pain Diary[9]			X	
Evaluation of Sickle Cell Disease Events of Special Interest			X	
Chest X-ray or chest CT scan				X
Cerebral MRI/MRA *(Per section 4.2.2.2)*				X
Optional blood samples[10]				X

[1]Assessments summarized in this table should be performed per section 4.2.2.1 and 4.2.2.2 of the protocol as applicable.
[2] Baseline assessments (pre-HU) should be performed ≤ 30-day after enrollment in Segment A, and prior to initiation of hydroxyurea, unless otherwise noted.
[3] Pre-transplant conditioning assessments should be performed ≤ 30-day prior to initiation of Thymoglobulin (these assessments can be completed during administration of hydroxyurea).

[4] Blood Chemistries performed per sections 4.2.2.1 and 4.2.2.2 include reticulocyte count, direct/conjugated bilirubin, alkaline phosphatase, ALT, AST, serum ferritin, and magnesium.

[5] Serum Creatinine and Creatinine Clearance required <180 days prior to enrollment. Radionuclide Glomerular Filtration Rate (GFR) or creatinine clearance (CrCl) required prior to initiation of thymoglobulin (although GFR or CrCl is permissible, GFR is preferred). Refer to protocol sections 4.2.2.1 and 4.2.2.2.

[6] Refer to section 4.2.2.1 for 2-D transthoracic echocardiography requirements.

[7] If hepatic iron content ≥ 10 mg Fe/g, a gastroenterology/hepatology consultation with liver biopsy is required to document absence of cirrhosis, bridging fibrosis, and active hepatitis.

[8] A CBC will be obtained weekly during hydroxyurea (Day -70 until Day -10) and the dose will be reduced if the ANC <1500 or platelet <100,000. CBC

[9] Pain diary at baseline is completed twice daily over 28 days prior to starting hydroxyurea. For patients who were on HU pre-enrollment, patients may complete pain diary while on protocol dose of HU.

[10] Only required for patients and donors that consent to provide option research samples.

TABLE 4.2.2B: SCHEDULE OF EVALUATIONS FOLLOWING TRANSPLANT[1]

		Days Post-HCT																		
Assessments	**0**	**7**	**14**	**21**	**28**	**35**	**42**	**49**	**56**	**60**	**63**	**70**	**77**	**84**	**91**	**100**	**180**	**365**	**540**	**730**
Total nucleated cell and CD34+ cell content of the infused marrow product	X																			
History, physical exam, height, weight		X	X	X	X											X	X	X		X
Karnofsky/Lansky Performance Score		X	X	X	X											X	X	X		X
GVHD and morbidity assessments		X	X	X	X	X	X	X	X		X	X	X	X	X	X	X	X	X	X
QOL assessment (PROMIS)[2]																		X		X
28-Day Pain Diary[3]																		X		X
CBC with differential[4]		X	X	X	X	X	X	X	X		X	X	X	X			X	X		X
Reticulocyte *(per section 4.2.4)*					X	X	X	X	X		X	X	X	X	X	X	X	X		X
Hemoglobin evaluation and date of last RBC transfusion					X											X	X	X		X
Blood Chemistries[5]		X	X	X	X	X	X	X	X		X	X	X	X		X	X	X		X
Serum ferritin																		X		X
Renal Function Assessments[6]																		X		X
Chimerism *(per section 4.2.4)*					X		X									X	X	X		X
Pulmonary Function Assessment *(per section 4.2.4)*																		X		X
Echocardiogram or MUGA *(per section 4.2.4)*																		X		X
6-minute walk distance test																		X		X
Cerebral MRI/MRA *(Per section 4.2.4)*																		X		X
Liver MRI[7]																		X		X
Evaluation of Sickle Cell Disease Events of Special Interest																	X	X	X	X
Optional blood samples[8]					X					X						X	X	X		

[1] Assessments summarized in this table should be performed per section 4.2.4 of the protocol.
[2] Should be administered to English- and Spanish-speaking patients in the adult stratum on a single day ≥ 7 days and ≤ 28 days from initiating the 28-day electronic pain diary.
[3] Pain diary over a 28-day period for English-speaking patients ≥15.00 from Day 337 to 365, and Day 702 to 730.
[4] CBC three times a week from Day 0 until neutrophil recovery. Platelet count three times a week from Day 0 until platelet recovery. Thereafter, CBC and platelet count twice weekly until Day 28, then weekly until 12 weeks, then six months, 1 year and 2 years post-transplant.
[5] Blood Chemistries include serum creatinine, bilirubin, alkaline phosphatase, ALT, AST, magnesium twice weekly until Day 28, then weekly until 12 weeks, and then at six months, one year and two years post-transplant.

[6] Measured Radionuclide Glomerular Filtration Rate (GFR) or eGFR with creatinine clearance (CrCl) is permissible. However, measured GFR is preferred. Report urine albumin-to-creatinine ratio.
[7] May be performed per institutional preference and as clinically indicated.
[8] Only required for patients and donors that consent to provide optional research samples.

4.3 Data Reporting and Study Monitoring

This study must be approved by an appropriate institutional review committee as defined by Federal Regulatory Guidelines (Ref. Federal Register Vol. 46, 17, January 27, 1981, part 56) and the Office for Protection from Research Risks Reports: Protection of Human Subjects (Code of Federal Regulations 45 CFR 46.

4.3.1 Reporting Participant Deaths

Death information must be entered into AdvantageEDC^{SM} within 24 hours of knowledge of the participant's death. If the cause of death is unknown at that time, it need not be recorded at that time. However, once the cause of death is determined, the form must be updated in AdvantageEDC^{SM}. In addition, death must be reported via the expedited reporting requirements in AvantageEDC^{SM} (see 4.2.3 below).

4.3.2 Adverse Event Reporting

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

Expectedness: An adverse event can be Expected or Unexpected

- **Expected adverse events** are those that have been previously identified as resulting from administration of the agent. For the purposes of this study, an adverse event is considered expected when it appears in the current adverse event list, the Investigator's Brochure, or is included in the informed consent document as a potential risk.
- **Unexpected adverse events** are those that vary in nature, intensity or frequency from information in the current adverse event list, the Investigator's Brochure, or when it is not included in the informed consent document as a potential risk.

Serious Adverse Event: A serious adverse event (SAE), as defined by per 21 CFR 312.32, is any adverse event that results in one of the following outcomes, regardless of causality and expectedness:

- Results in death.
- **Is life-threatening.** Life-threatening means that the person was at immediate risk of death from the reaction as it occurred, i.e., it does not include a reaction which hypothetically might have caused death had it occurred in a more severe form.
- **Requires or prolongs inpatient hospitalization** (i.e., the event required at least a 24-hour hospitalization or prolonged a hospitalization beyond the expected length of stay). Hospitalization admissions and/or surgical operations scheduled to occur during the study period, but planned prior to study entry are not considered SAEs if the illness or disease existed before the person was enrolled in the trial, provided that it did not deteriorate in an unexpected manner during the trial (e.g., surgery performed earlier than planned).

- **Results in persistent or significant disability/incapacity.** Disability is defined as a substantial disruption of a person's ability to conduct normal life functions.
- **Is a congenital anomaly or birth defect; or**
- **Is an important medical event** when, based upon appropriate medical judgment, it may jeopardize the participant and require medical or surgical intervention to prevent one of the outcomes listed above. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home; blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse.

Medical and scientific judgment should be exercised in deciding whether expected reporting is also appropriate in situations other than those listed above. For example, important medical events may not be immediately life threatening or result in death or hospitalization but may jeopardize the subject or may require intervention to prevent one of the outcomes listed in the definition above (e.g., suspected transmission of an infectious agent by a medicinal product is considered a Serious Adverse Event). Any event is considered a Serious Adverse Event if it is associated with clinical signs or symptoms judged by the investigator to have a significant clinical impact.

4.3.3 Required Adverse Event Reporting:

Adverse event reporting will be consistent with BMT CTN procedures (BMT CTN Administrative Manual of Procedures, Chapter 6). It is BMT CTN policy that AEs must be reported even if the investigator is unsure whether a relationship exists between the adverse event and the use of study treatment. Unexpected serious adverse events will be reported through the expedited AE reporting system in AdvantageEDC. Unexpected, Grade 4-5 SAEs must be reported within 24 hours of knowledge of the event. All other unexpected SAEs must be reported within three business days of knowledge of the event. Events entered in AdvantageEDC will be reported using NCI's Common Terminology Criteria for Adverse Events (CTCAE) Version 4.0. Expected AEs will be reported using NCI's Common Terminology Criteria for Adverse Events (CTCAE) Version 4.0 at regular intervals as defined on the Form Submission Schedule (e.g. Re-Admission/Hospitalization, Infection, GVHD, and Toxicity Forms). Any expected life-threatening SAE not collected on another study form must be reported through the expedited AE reporting system via AdvantageEDC.

The NHLBI Data and Safety Monitoring Board will receive summary reports of all adverse experiences at least twice yearly.

In addition to the standard BMT CTN SAE reporting, all deaths, central nervous system (CNS) events, diagnosis of posterior reversible encephalopathy syndrome (PRES)/reversible posterior leukoencephalopathy syndrome (RPLS), and/or events requiring advanced care interventions or admission/transfer to an intensive care unit (ICU) are required to be reported via the expedited AE reporting system in AdvantageEDC for this study. All event reporting is to be expedited. Grade 4-5 events should be reported within 24 hours of knowledge of the event. All other events should be reported within three business days of the event.

For the purposes of this protocol, examples of "ICU" or "advanced care intervention" requiring expedited AE reporting are one or more of the following:

- The patient is transferred to an advanced care unit *or* the ICU* (see footnote below)
- Continuous Renal Replacement Therapy (CRRT) by dialysis (CAVHD) or filtration (CAVH)
- Non-invasive positive pressure ventilation: continuous positive airway pressure (CPAP), bi-level positive airway pressure (BIPAP), intermittent positive pressure ventilation (IPPV)
- Endotracheal intubation with mechanical ventilation
- Pressor support
- Organ failure – acute renal failure, acute liver failure, acute respiratory distress syndrome, respiratory failure, or multi-organ failure

4.3.4 * Elective transfer to an advanced care/ICU unit for a scheduled intervention does not need to be reported via the expedited AE reporting system in AdvantageEDC.

4.3.4.1 Sickle Cell Disease Related Events of Special Interest Reporting:

The following sickle cell disease related events of special interest (SCD-EOSI) are expected events for all participants:

- pulmonary hypertension
- significant cerebrovascular events, including:
  - stroke
  - transient ischemic attack
  - seizure
- renal function compromise, including:
  - proteinuria (absent or present, and if present, date of onset)
  - increased creatinine grades ≥2 per CTCAE version 4.0
- avascular necrosis of the hip or shoulder
- leg ulcers
- acute chest syndrome (ACS) requiring hospitalization
- painful vaso-occlusive crises (VOC) requiring hospitalization or parenteral opioid drugs in the outpatient setting (self-reported events without clinical documentation should not be included.)

While these events may not require expedited reporting via completion of the Adverse Events forms, each occurrence of any SCD-EOSI must be reported on the SCD Events of Special Interest Form at each regularly scheduled follow up interval. A single event occurrence is defined as a new onset of any of the above listed SCD-EOSIs after screening evaluations are completed. A single occurrence resolves once the SCD-EOSI returns to the participant's baseline. Therefore, multiple occurrences of the same or several SCD-EOSIs may be reported in one reporting interval.

4.4 Data Monitoring

4.4.1 Criteria for Forms Submission

Forms that are not entered into AdvantageEDC^{SM} within the specified time will be considered delinquent. A missing form will continue to be requested either until the form is entered into the AdvantageEDC^{SM} and integrated into the Data and Coordinating Center's (DCC) master database, or until an exception is granted and entered into the Missing Form Exception File.

4.4.2 Center for International Blood and Marrow Transplant Research (CIBMTR) Data Reporting

Centers participating in BMT CTN trials must register pre- and post-transplant outcomes on all consecutive hematopoietic stem cell transplants done at their institution during their time of participation to the Center for International Blood and Marrow Transplant Research (CIBMTR). Registration is done using procedures and forms of the Stem Cell Transplant Outcomes Database (SCTOD). (Note: Federal legislation requires submission of these forms for all US allotransplant recipients.) Enrollment on BMT CTN #1507 must be indicated on the SCTOD pre-transplant registration form. Additionally, CIBMTR pre- and post-transplant Report Forms must also be submitted for all patients enrolled on this trial. CIBMTR forms will be submitted directly to the CIBMTR at the times specified on the Form Submission Schedule.

4.4.3 Internal Data Monitoring

It is the responsibility of the site PI to ensure the validity of all participant data as well as the safety of all participants.

4.4.4 Data Monitoring and Auditing

The Data Safety and Monitoring Board (DSMB) for the BMT CTN will convene as per standard procedure to review serious toxicities and adverse events for the purpose of determining whether the trial should be modified or stopped. Triggers for referral to the DSMB are described in the Stopping Rules Criteria of section 5.3.

4.4.5 Records

Any publication or presentation will refer to patients by a unique patient number and not by name to assure patient confidentiality.

4.4.6 Archiving Records

Archival of participant study charts/records should be conducted in accordance with the guidelines provided in the Data Management Handbook.

4.4.7 Planned Protocol Amendments

If the monitoring of the safety endpoints described in Section 5.3 results in a trigger for consultation with the DSMB, the DSMB may make recommendations about whether or not to continue accrual.

In addition, if the rate of 100-day graft failure passes the stopping guideline in the 12 evaluable participants in a stratum (see section 5.3.1), the conditioning regimen will change from TBI 200cGy to TBI 400cGy in the affected stratum. Accrual to the strata under the original TBI dose of 200cGy will be stopped. The goal will be to enroll 40 more participants for the stratum that implements the changed regimen. Safety monitoring for all safety endpoints in that stratum will be restarted only using participants enrolled after the new conditioning regimen is implemented.

4.4.8 On-study Date

Participants will be in the screening phase from date of consent to date of enrollment onto Segment 0 or notice of non-eligibility. Participants will be on-study from the date of enrollment onto Segment A. All patients enrolled in Segment A will be followed for 2 years post-transplant regardless of the treatment received.

CHAPTER 5

STATISTICAL CONSIDERATIONS

5.1 Study Design

This study is designed as a Phase II multi-center trial to determine the feasibility of achieving a high rate of event-free survival at 2 years post-transplant using pre-conditioning HU with a conditioning regimen that consists of a combination of rATG/CY/Flu/TT with post-grafting high-dose cyclophosphamide in patients with severe SCD who have HLA-haploidentical donors. EFS is defined as survival without a qualifying event (see Section 3.1). This is a single arm study in which participants will be enrolled into one of two strata. The first stratum will be restricted to children who have stroke and 40 children will be transplanted in this stratum. The second stratum will consist of adult patients with severe sickle cell disease and 40 participants will be transplanted in this stratum.

Safety monitoring will occur on a monthly basis for several safety endpoints. If the safety criterion for graft rejection is met in either stratum, and if the DSMB and sponsor agree that accrual to a stratum should stop, then accrual to that stratum will be halted. For that stratum, participants already accrued will continue to be followed per protocol. Further, with DSMB and sponsor approval, an additional 40 participants will be transplanted in the strata but will use a different conditioning regimen. Safety monitoring for all safety endpoints will be reset for the stratum that changes conditioning regimen (see Section 5.3 for further details).

All analyses are within-stratum, and each stratum will follow it is own timeline of completion for the primary analysis for the stratum If a new conditioning regimen is implemented for any strata, then for that stratum, descriptive statistics or listings will be provided using data from participants enrolled under the original dose of TBI; the stratum-specific analyses will be limited to the participants enrolled under the new conditioning regimen.

5.1.1 Accrual

It is estimated that it will require four years to complete the accrual with 10 patients transplanted in each stratum per year. If accrual to any arm is restarted based on the planned change in conditioning regimen, then it is estimated accrual in that strata will take four years from the time when the change in conditioning regimen is implemented.

5.1.2 Study Duration

Participants will be followed for the 28-day baseline pain diary period, 70 day conditioning period and for two years post-transplant for primary and secondary endpoints.

5.1.3 Randomization and Blinding

This is a single arm trial with no randomization and no blinding.

5.1.4 Primary Endpoint

The primary objective of this clinical trial is to estimate two-year post-transplant event-free survival in each age stratum. EFS is defined as survival without a qualifying event. Events contributing to this endpoint are as below:

i. Primary graft rejection (Primary graft failure is defined as never achieving ≥ 5% donor whole blood or myeloid chimerism (myeloid is preferable) assessed by bone marrow or peripheral blood chimerism assays by day +42 post-transplant. Second infusion of stem cells is also considered indicative of primary graft failure by day +42 post-transplant).
ii. Secondary graft rejection (Secondary graft failure is defined as < 5% donor whole blood or myeloid chimerism (myeloid is preferable) in peripheral blood or bone marrow beyond day +42 post-transplant in patients with prior documentation of hematopoietic recovery with >5% donor cells by day +42 post-transplant. Second infusion of stem cells is also considered indicative of secondary graft failure).
iii. Second infusion of hematopoietic cells (Infusion of a second stem cell product will be considered graft rejection, and counted toward primary or, depending on timing of the second infusion, secondary graft rejection).
iv. Death (from any cause by 2 years post-transplant)

The EFS probability estimates will be based on the Kaplan-Meier product limit estimator using Greenwood's formula as the variance estimate. For the primary analysis, time to event will be calculated as the minimum of the time from study start of HU conditioning to the first event, loss to follow-up, or two years post- transplant. Reasons for not getting a transplant or not starting conditioning will be described.

A secondary endpoint will assess EFS at 1 year post-transplant. Further analyses include EFS from time of transplant and will restrict analyses to those receiving a transplant.

5.2 Sample Size Considerations

The sample size is 40 transplanted participants per each of the two stratum in this trial. Ninety-five percent confidence intervals were calculated for varying probabilities based on the sample size. The primary endpoint, event-free survival (EFS), is considered for sample size determination. Table 5.2 provides confidence intervals for a variety of observed proportions based on the binomial proportion. Of particular interest is where the EFS probability is 80%. If the observed EFS probability is 80%, the width of the confidence interval is 24.8. The tabulated percentages and confidence intervals for observed probabilities above and below 80% are meant to represent other plausible EFS percentages.

TABLE 5.2A: CONFIDENCE INTERVAL LENGTHS AND POSSIBLE CONFIDENCE INTERVALS FOR VARIOUS OBSERVED EVENT-FREE SURVIVAL PROBABILITIES

40 participants enrolled and transplanted				
N (per stratum)	Event-free survival %	Length of 95% Confidence Interval	Possible Confidence Intervals	
40	90	18.6	80.7	99.3
40	85	22.1	73.9	96.1
40	80	24.8	67.6	92.4
40	75	26.8	61.6	88.4

The EFS probability estimate will be based on the Kaplan-Meier product limit estimator using Greenwood's formula as the variance estimate. In the absence of censoring, the Kaplan-Meier estimate reduces to the simple binomial proportion. If the linear confidence intervals extend beyond 0 or 100, log-log confidence intervals may be used.

It is estimated that there may be more than 40 participants enrolled to reach 40 transplanted participants. For example, there may be 53 participants enrolled with 15% censored. The following table shows how the binomial confidence intervals would change based on 45 participants who were not censored for the primary endpoint.

TABLE 5.2B: CONFIDENCE INTERVAL LENGTHS AND POSSIBLE CONFIDENCE INTERVALS FOR VARIOUS OBSERVED EVENT-FREE SURVIVAL PROBABILITIES

45 participants not censored for primary endpoint				
N (per stratum)	Event-free survival %	Length of 95% Confidence Interval	Possible Confidence Intervals	
45	88.9	18.4	79.7	98.1
45	84.4	21.1	73.9	95.0
45	80.0	23.4	68.3	91.7
45	75.6	25.1	63.0	88.1

5.3 Interim Analysis and Stopping Guidelines

There will be no formal interim analyses for this trial.

Monitoring of key safety endpoints will be conducted monthly, and if rates significantly exceed pre-set thresholds, the NHLBI will be notified in order that the DSMB can be advised. Policies and composition of the DSMB are described in the BMT CTN's Administrative Manual of Procedures. The stopping guidelines serve as a trigger for consultation with the DSMB for additional review.

The key safety endpoint of 100-day graft failure will be monitored, with a planned protocol modification of the conditioning regimen planned if rates exceed 15% in the first 12 evaluable participants in a stratum. After the first 12 evaluable participants, if the stopping boundary has not been crossed, then the graft failure rate will continue to be monitored as a safety guideline for consultation with the DSMB but it is unlikely the conditioning regimen will change and accrual be restarted.

Within each stratum, the additional key safety endpoints to be monitored are the following rates:

- Acute, grade III-IV GVHD by 100 days post-transplant
- Overall mortality by 180 days post initiation of hydroxyurea
- Severe chronic GVHD by 18 months post-transplant

A truncated version of the sequential probability ratio test (SPRT) will be used for monitoring, as described in greater detail below and in Appendix H. This sequential testing procedure conserves type I error across all of the planned examinations for a treatment arm for a given stopping rule and stratum. Two versions of the SPRT will be used. For graft failure, a binomial SPRT will be used, and a plot will display the number of evaluable patients versus the number of observed events. Other safety endpoints will be monitored using an extension to the SPRT for censored exponential data. This extension will plot the observed time on study versus the number of observed events (see Appendix H). The continuation region for both versions of the SPRT is defined by two parallel lines. Only the upper boundary will be used for monitoring to protect against excessive rates. If the graph crosses the upper boundary, the SPRT rejects the null hypothesis, and concludes that there are more events than predicted. Otherwise, the SPRT continues until the monitoring period is complete.

The usual measures of performance of an SPRT are the error probabilities α and β of rejecting H_0 when $\theta = \theta_0$ and of accepting H_1 when $\theta = \theta_1$, respectively, and the expected sample size $E(N|\theta_i)$.

5.3.1 Monitoring Guideline: 100-day Graft Failure Rate within a Stratum

The monitoring of graft failure is a key safety endpoint for which a protocol modification for a change in the dose level of TBI is pre-planned in the event the boundary is crossed within the first 12 evaluable participants.

This monitoring procedure assumes a binary distribution for graft failure during the first 100 days post-transplant. If the participant has been transplanted but has had primary or secondary graft rejection by day 100, the participant is assumed to be a graft failure at day 100. Chimerism from samples collected within the D100 visit window will be used for evaluation of graft failure at Day 100. This monitoring guideline uses an SPRT contrasting a null hypothesis of 15% versus an alternative of 35% 100-day graft failure which results in decision boundaries with a common slope of 0.240 and an upper intercept of 2.59, with nominal type I and type II errors of 5% and 10%, respectively. Table 5.3.1 summarizes the stopping rule.

5.3.2 Table 5.3.1: Stopping Rule for 100-Day Graft Failure

Number of evaluable participants	Stop if graft failure occurs in
4-5	4
6-10	5
11-14	6
15-18	7
19-22	8
23-26	9
27-30	10
31-34	11
35-39	12

5.3.2.1 Modified Conditioning Regimen

While participants are enrolled under the 200 cGy dose of TBI, if the stopping boundary is crossed before the end of the first 12 evaluable participants, then a planned protocol modification may be discussed by the DSMB. If a decision is made to stop accrual under the initial conditioning regimen, then sites will be notified to stop accrual in that stratum. If the DSMB and sponsor choose to implement the modified conditioning regimen, accrual will be restarted using the modified conditioning regimen (TBI 400 cGy). An additional 40 patients will be enrolled with the new conditioning regimen and the first 12 patients receiving the modified regimen will be monitored for incidence of 100-day graft failure. Safety monitoring for all endpoints for participants in that stratum will be restarted.

If the safety boundary has not been triggered in the first 12 evaluable participants in each stratum and the DSMB recommends continuing accrual, then safety monitoring for 100-day graft failure will continue using the SPRT. Alternatively, if the stopping boundary is crossed after the first 12 evaluable participants, then DSMB will be consulted but it is anticipated the recommendation will be to terminate accrual in that stratum and not restart with the new regimen, given the logistical difficulty of completing enrollment of an additional 40 participants in a timely manner.

5.3.3 Monitoring Guideline: Grade III-IV Acute GVHD by Day 100 Post Transplant within a Stratum

The incidence of acute grade III-IV GVHD is approximately 15-17% after HLA-identical sibling BMT.[62, 63, 64, 65] Each month, the null hypothesis that the 100-day acute GVHD rate is 20% is tested.

This monitoring procedure assumes a censored exponential distribution for the time until acute grade III-IV GVHD during the 100 days post-transplant, and censor's follow-up time after 100 days. Only events that occur after transplant and on or before the participant has been followed for 100 days post-transplant are counted. Total time on study is computed as time from transplant to acute grade III-IV GVHD, or to 100 days, whichever comes first, summed for all transplanted participants on study. This monitoring guideline uses an SPRT contrasting a null hypothesis of 20% versus an alternative of 40% 100-day acute grade III-IV GVHD which results in decision

boundaries with a common slope of 1.27 and an upper intercept of 2.94, with nominal type I and type II errors of 7% and 20%, respectively.

The actual operating characteristics of this truncated test, shown in Table 5.3.2, were determined in a simulation study that assumed uniform accrual and transplant of 40 individuals per strata over a 4-year time period with monitoring occurring monthly after the first transplant for 52 months.

TABLE 5.3.2: OPERATING CHARACTERISTICS OF SEQUENTIAL TESTING PROCEDURE FOR 100-DAY ACUTE GVHD FROM A SIMULATION STUDY WITH 100,000 REPLICATIONS

Acute GVHD at 100 Days Post-Transplant

True 100-Day Post-Transplant Rate	**20%**	**25%**	**30%**	**35%**	**40%**
Probability Reject Null	0.05	0.17	0.37	0.62	0.82
Mean Month Stopped Post-Transplant	50.5	47.5	42.1	35.0	27.9
Mean # Endpoints in 100 days	7.8	9.2	9.8	9.6	8.8
Mean # Participants Transplanted	39.0	36.8	33.1	28.2	22.9

5.3.4 Monitoring Guideline: Overall Mortality by Day 180 Post Conditioning within a Stratum

Overall mortality (OM) is defined as death with or without disease recurrence. The monitoring procedure assumes a censored exponential distribution for the time until death during the first 180 days, and censor's follow-up time after 180 days. Deaths that occur on or before the participant has been followed for 180 days are counted as events. Total time on study is computed as time from start of hydroxyurea (HU) at day -70 pre-BMT to death, loss to follow-up, or to 180 days, whichever comes first, summed for all participants on study. This monitoring guideline uses an SPRT contrasting a null hypothesis of 10% versus an alternative of 30% 180-day mortality, which results in decision boundaries with a common slope of 0.42 and an upper intercept of 2.05, with nominal type I and type II errors of 7% and 15%, respectively.

The actual operating characteristics of this truncated test, shown in Table 5.3.3, were determined in a simulation study that assumed uniform accrual of 40 individuals per stratum over a 4-year time period with monitoring occurring monthly for 54 months.

TABLE 5.3.3A: OPERATING CHARACTERISTICS OF SEQUENTIAL TESTING PROCEDURE FOR DAY 180 POST CONDITIONING OM FROM A SIMULATION STUDY WITH 100,000 REPLICATIONS

Overall Mortality at Day 180 post HU Conditioning, N=40

True Rate at Day 180 post HU initiation	**10%**	**15%**	**20%**	**25%**	**30%**
Probability Reject Null	0.05	0.20	0.47	0.73	0.90
Mean Month post HU Conditioning Stopped	52.5	47.9	40.1	31.4	24.0

True Rate at Day 180 post HU initiation	10%	15%	20%	25%	30%
Mean # Endpoints	3.9	5.3	6.0	5.9	5.4
Mean # Participants Enrolled	39.0	36.0	30.9	25.0	19.7

The following table shows the simulated results for 53 people accrued per stratum over a 4-year time period with monitoring occurring monthly for 54 months.

TABLE 5.3.3B: OPERATING CHARACTERISTICS OF SEQUENTIAL TESTING PROCEDURE FOR DAY 180 POST CONDITIONING OM FROM A SIMULATION STUDY WITH 100,000 REPLICATIONS

Overall Mortality at Day 180 post HU Conditioning, N=53

True Rate at Day 180 post HU initiation	10%	15%	20%	25%	30%
Probability Reject Null	0.05	0.23	0.54	0.81	0.95
Mean Month post HU Conditioning Stopped	52.3	46.7	37.2	27.3	19.9
Mean # Endpoints	5.1	6.9	7.3	6.7	5.7
Mean # Participants Enrolled	51.5	46.5	38.1	29.1	21.9

5.3.5 Monitoring Guideline: 18-Month Severe Chronic GVHD Within a Stratum

The incidence of severe chronic GVHD (defined as NIH severe chronic GVHD) isapproximately 5-20% after HLA-identical sibling BMT.[62,66,67] The working hypothesis is that the overall toxicity of mini-haploBMT is not significantly greater than HLA-identical sibling BMT after myeloablative conditioning.

This monitoring procedure assumes a censored exponential distribution for the time until severe cGVHD during the eighteen months post-transplant, and censors follow-up time after eighteen months. Onlyevents that occur after transplant and on or before the participant has been followed for 18 months post-transplant are counted. Total time on study is computed as time from transplant to severe cGVHD, or to eighteen months, whichever comes first, summed for all transplanted participants on study. This monitoring guideline uses an SPRT contrasting a null hypothesis 20% versus an alternative hypothesis of 40% 18-month severe cGVHD which results in decision boundaries with a common slope of 0.23 and an upper intercept of 2.87, with nominal type I and type II errors of 8% and 14%, respectively.

The actual operating characteristics of this truncated test, shown in Table 5.3.4, were determined in a simulation study that assumed uniform accrual and transplant of 40 individuals per strata over a 4-year time period with monitoring occurring monthly after the first transplant for 66 months.

TABLE 5.3.4: OPERATING CHARACTERISTICS OF SEQUENTIAL TESTING PROCEDURE FOR 18-MONTH POST TRANSPLANT SEVERE CGVHD FROM A SIMULATION STUDY WITH 100,000 REPLICATIONS

True 18-Month Post-Transplant Rate	**20%**	**25%**	**30%**	**35%**	**40%**
Probability Reject Null	0.05	0.17	0.38	0.63	0.83
True 18-Month Post-Transplant Rate	**20%**	**25%**	**30%**	**35%**	**40%**
Mean Month Post-Transplant Stopped	64.0	59.9	52.8	43.7	35.1
Mean # Endpoints in 18 Months	7.8	9.1	9.8	9.5	8.7
Mean # Participants Transplanted	39.1	37.5	34.7	30.8	26.6

5.4 Demographic and Baseline Characteristics

Demographic and baseline characteristics will be summarized for all participants. Characteristics to be examined may include: age, gender, race/ethnicity, Karnofsky/Lansky performance status, number and duration of prior transfusions, TRJV, disease risk characteristics, disease symptoms, donor characteristics, HLA match, serum bilirubin level, serum creatinine level, pulmonary function tests, cardiac function, splenic function, hemoglobin electrophoresis, cerebral MRI, serum ferritin, QOL, and other baseline characteristics.

5.5 Analysis of Secondary Endpoints

All secondary endpoints will be reported within each stratum. The primary analysis of the secondary endpoints will be limited to the participants enrolled under the TBI conditioning regimens in effect at the close of the study. The data from participants enrolled under the initial conditioning regimens that were subsequently modified as part of the planned protocol amendment will be provided using descriptive statistics or lists only. If the endpoint definitions indicate the endpoint is only for adults, the analysis will be restricted to those participants.

5.5.1 Overall Survival

Overall survival will be estimated using the Kaplan-Meier estimator. Death from any cause will be the event and patients will be censored at the date of last contact or two years from the analysis start day, whichever comes first. Start day will either be the date of enrollment or date of HaploBMT for the two objectives. A point estimate and confidence interval will be provided for the rate of OS at one year and two years for both analyses.

5.5.2 One-year Event-free Survival

For those who start HU conditioning, EFS from start of HU conditioning will be estimated using the Kaplan-Meier estimator. A point estimate and confidence interval will be provided for the rate of EFS at one year since the start of HU conditioning.

For those with a transplant, EFS from day of transplant will be estimated using the Kaplan-

Meier estimator. A point estimate and confidence interval will be provided for the rate of EFS at one and two years post-transplant.

5.5.3 Primary or Secondary Graft Rejection

Primary graft failure will be summarized by the frequency and proportion of participants experiencing primary graft failure at Day 42 post-transplant.
Secondary graft rejection will be estimated using the cumulative incidence function, treating death as a competing risk. A point estimate and confidence interval will be provided for the cumulative incidence at one year and two years post-transplant for secondary graft rejection.

5.5.4 Disease Recurrence

To assess the incidence of disease recurrence from enrollment, a cumulative incidence curve will be estimated, treating death as a competing risk. A point estimate and confidence interval will be provided for the cumulative incidence at one year and two years post conditioning, and post-transplant, respectively.

5.5.5 Neutrophil and Platelet Recovery

The incidence of neutrophil recovery from transplant will be estimated using the cumulative incidence function, treating death or second transplant as a competing risk. A point estimate and confidence interval will be provided for the cumulative incidences at Day 42 post-transplant.

The incidence of platelet recovery from transplant will be estimated using the cumulative incidence function, treating death or second transplant as a competing risk. A point estimate and confidence interval will be provided for the cumulative incidence at Day 100 post-transplant.

5.5.6 Lineage Specific Chimerism following HCT for SCD

Lineage specific chimerism taking into account the genotype of the donor will be described. A point estimate and confidence interval will be provided for the mean percent donor chimerism at the time points specified in the Schedule of Evaluations Table. The proportions with mixed (5-95%), full (>95%), or low (<5%) will be tabulated and described.

5.5.7 Grade II-IV and Grade III-IV Acute GVHD

Rates of acute GVHD will be tabulated. Cumulative incidence of acute grade II-IV GVHD will be estimated using the cumulative incidence function, treating death and second transplant prior to acute GVHD as a competing risk. A point estimate and confidence interval will be provided for the cumulative incidence at day 100 post-transplant. Time to GVHD will be calculated from transplant date.

Cumulative incidence of acute grade III-IV GVHD will be estimated using the cumulative incidence function, treating death and second transplant prior to acute GVHD as a competing risk. A point estimate and confidence interval will be provided for the cumulative incidence at day 100

post-transplant.

5.5.8 Chronic GVHD

Rates of chronic GVHD will be tabulated by severity. Cumulative incidence of chronic GVHD will be estimated using the cumulative incidence function, treating death and second transplant prior to chronic GVHD as a competing risk. A point estimate and confidence interval will be provided for the cumulative incidence at 6 months, one year, and two years post-transplant. Analysis of severe, chronic GVHD will be similar. Time to GVHD will be calculated from transplant date.

5.5.9 Complications and Events

The frequency of the following complications and events will be tabulated, and the proportion of participants experiencing each at baseline and by 1 year and 2 years will be estimated, with confidence intervals provided for each estimate:

- Idiopathic pneumonia syndrome (IPS)
- Veno-occlusive disease
- Various CNS toxicities
- Stroke
- Infection – bacterial/fungal sepsis, CMV reactivation with/without clinical disease, adenovirus, EBV PTLD, adenovirus, invasive fungal infection, CMV specified, viral mold infections and invasive fungal infections
- Proportion of participants still on immunosuppression for GVHD

The number of significant infections and number of participants experiencing infections will be tabulated by type of infection, severity, and time period post-transplant.

In addition, the proportion of participants still on immunosuppression at 2 years after transplant will be estimated and a confidence interval will be provided.

5.5.10 Quality of Life and Pain Intensity

Quality of life will be described using the PROMIS pain and fatigue domains. Descriptive statistics and confidence intervals for the change from baseline will be reported for each domain for English- and Spanish-speaking patients in the adult stratum. Exploratory models may be used to investigate the change over time adjusted for participant covariates.

The electronic pain diary is administered twice daily for 28 consecutive days for English-speaking patients ≥ 15.00 years of age at the time of enrollment. An average value will be computed for each day on which the diary shows two entries. If the diary reflects only a single entry for a day, then the single value will be used, and if a patient did not enter a value on a given day, that non- adherence will be tallied separately. The mean pain intensity from the diary will be the sum of the reported daily pain intensities averaged over the number of days on which at least one pain intensity was reported. Descriptive statistics and

confidence intervals for the change from baseline will be reported. Exploratory models may be used to investigate the change over time adjusted for participant covariates.

5.5.11 Other Secondary Endpoints

The following outcomes will be analyzed for transplanted participants:

- Hematological outcomes
- Renal function
- Sickle vasculopathy as measured by brain MRI
- Lung function (PFT), including FEV1 and restrictive lung disease
- Cardiac Function (TRJV)
- 6 minute walk distance
- Other SCD-EOSI (defined in Section 3.8.2)

For binary outcomes, proportions and confidence intervals will be provided at each time point. For continuous outcomes, results will be summarized with descriptive statistics and confidence intervals will be provided where appropriate.

5.6 Safety Analysis

AEs and toxicities will be reported and summarized, and descriptive statistics will be provided.
Infections will be reported as described in section 5.5.9.
Hospitalizations will be tabulated and summarized.

APPENDIX A
HUMAN SUBJECTS

Appendix A
HUMAN SUBJECTS

1. SUBJECT CONSENT

A conference will be held with the patient and family to discuss this study and alternative treatments available for the treatment of the underlying disease. The Principal Investigator or another designated qualified staff member will conduct the conference. All potential risks associated with the use of conditioning regimen, GVHD prophylaxis medications, and all study treatments and interventions should be discussed as objectively as possible.

The BMT CTN will provide a template of the consent form to each center. Each center will customize the template according to their local requirements and submit it for review by their Institutional Review Board (IRB) of record. The DCC will verify the adequacy of the consent forms prior to submission to the IRB. Each center must provide evidence of IRB approval.

The consent document should be reviewed with the patient and family prior to proceeding to transplantation. Informed consent from the patient and the patient's parent/guardian will be obtained using a form approved by the Institution's IRB of record prior to enrolling the patient.

2. CONFIDENTIALITY

Confidentiality will be maintained by individual names being masked and assigned a patient identifier code. The code linking the patient's identity with the ID code will be kept separately at the center. The ID code will be transmitted to the BMT CTN Data and Coordinating Center upon enrollment.

3. PARTICIPATION OF WOMEN AND MINORITIES AND OTHER POPULATIONS

Women and ethnic minorities and other populations will be included in this study. Accrual of women and minorities at each center will be monitored to determine whether their rates of enrollment are reflective of the distribution of potentially eligible women and minorities expected from data reported to the CIBMTR. Centers will be notified if their rates differ significantly from those expected and asked to develop appropriate recruitment strategies.

It is expected that the number of minority patients on this study will be large given the disease being studied.

Appendix B
INFORMED CONSENTS

INFORMED CONSENT TO PARTICIPATE IN RESEARCH

[Insert site logo and/or address]

YOUR NAME: _____

Study Title: **Reduced Intensity Conditioning for Haploidentical Bone Marrow Transplantation in Patients with Symptomatic Sickle Cell Disease**

Protocol: BMT CTN # 1507

Principal Investigator: [Insert site PI]

Co-Investigators: [Insert site co-I]

Study Coordinators: [Insert site study coordinator/s]

[Insert site department/facility name, address, and phone number]

National Heart, Lung, and Blood Institute (NHLBI) of the National Institutes of Health (the NIH), Bethesda, Maryland

CONSENT FOR AN ADULT OR CHILD TO BE A SUBJECT IN CLINICAL RESEARCH AND AUTHORIZATION TO PERMIT THE USE AND DISCLOSURE OF IDENTIFIABLE MEDICAL INFORMATION (PROTECTED HEALTH INFORMATION) FOR RESEARCH PURPOSES.

1. INTRODUCTION

We invite you to join this clinical trial. A clinical trial is a research study. You're being asked to join this study because:

- You have **Severe Sickle Cell Disease (SCD)**
- You're between 5 - 45 years old
- An **allogeneic transplant** is a treatment option for you
- You have a donor (parent, brother, sister, or other family member) who is a half match

SCD is sometimes treated with an allogeneic transplant (also known as a bone marrow transplant). An allogeneic transplant may use a matched or mismatched donor.

We're doing this study to learn if a new type of allogeneic transplant called a **haploidentical transplant** is safe and effective to treat SCD. This study will use a parent, brother, sister or other family member who is a mismatched donor that has a half match with you.

If you agree to be in the study it will be for 2 years. The study will include 80 people (40 children and 40 young adults) at 30 – 40 transplant centers.

Definitions of **bolded terms** are in the next section.

This Consent Form will tell you about the purpose of the study, possible risks and benefits, other options available to you, and your rights as a participant in the study. Please take your time to make your decision.

Everyone who takes part in research at [insert facility name] should know that:

- Being in any research study is voluntary.
- You may or may not benefit from being in the study. Knowledge we gain from this study may benefit others.
- If you join the study, you can quit the study at any time.
- If you decide to quit the study, it will not affect your care at [insert name of facility or institution].
- Please ask the study staff questions about anything that you do not understand, or if you would like to have more information.
- You can ask questions now or any time during the study.
- Please take the time you need to talk about the study with your doctor, study staff, and your family and friends. It's your decision to be in the study. If you decide to join, please sign and date the end of this Consent Form.

You and your doctor will discuss other treatment choices if you don't want to participate in this study.

2. STUDY BACKGROUND

The National Institutes of Health (NIH), through the Blood and Marrow Transplant Clinical Trials Network (BMT CTN), are providing staff support and money for this research study. The BMT CTN and the NIH will make decisions about how to manage the study.

SEVERE SICKLE CELL DISEASE AND ALLOGENEIC TRANSPLANT

Severe Sickle Cell Disease (SCD) is a blood disorder you're born with. Red blood cells carry oxygen to different parts of your body. SCD is caused by a change in **hemoglobin**, the protein that helps red blood cells carry oxygen in the body. The change blocks the blood flow in small vessels which can cause severe (serious) pain. It can also damage your lungs, brain, kidneys, and other organs. A person with SCD lives about 30 years shorter than a person without SCD.

An **allogeneic transplant**, or bone marrow transplant, uses healthy blood-making cells from a donor to replace unhealthy ones. It requires donors and patients to have matched **human leukocyte antigens (HLA)**. HLA is a protein marker found on most cells in your body. HLA markers are inherited (given to you from your mother or father). You get half of your HLA markers from your mother and half from your father.

A **haploidentical transplant** is an allogeneic transplant where the donor matches exactly half of your HLA. Parents are always a half-match for their biological children and children are always a half-match for their biological parents. Biological siblings (brothers or sisters) have a 50% (1 out of 2) chance of being a half-match for each other.

In this study, we want to learn if a **haploidentical transplant** is effective and safe to treat SCD. This study will use a parent or other half-matched family member donor. This treatment may stop the disease and many of its health problems. But there is also a chance that it may not work, and the disease can come back.

ALLOGENEIC TRANSPLANT – REDUCED INTENSITY TRANSPLANT

There are 2 main steps with allogeneic transplant. First, to prepare your body, we use chemotherapy and radiation to destroy the abnormal blood cells. This step is called the **conditioning regimen**. Then, when the conditioning regimen is done, you're given the donor cells for your transplant.

In this study, your doctor will use lower doses of chemotherapy and radiation than are normally used for allogeneic transplant. This is called a **reduced intensity transplant**.

3. STUDY PURPOSE

We're doing this study to learn if a **haploidentical transplant** is safe and effective to treat SCD. A haploidentical transplant uses a half or partially matched donor. ("Haplo" means half.) This study will use a

parent or other family member who is a half tissue match.

You're being asked to join this study because you:

- Are between 5 – 14 years old and had one or more of the following:
  - Prior or silent stroke
    - Overt stroke –a permanent injury to the brain associated with physical manifestations that is associated with future overt strokes or silent strokes
    - Silent stroke –a permanent injury to the brain associated with decreased thinking abilities that might be as severe as an overt stroke and is associated with future silent stroke or overt strokes.
  - Serious pain episodes requiring hospitalization not fully managed by medical therapy.
  - One episode of life-threatening acute chest syndrome episode resulting in intensive care admission.
  - Increased blood pressure in the arteries of your lungs (pulmonary hypertension) or in the rest of your body (systemic hypertension) that can make it difficult to breathe.
  - Recurrent priapism (prolonged penile erection for at least 4 hours) occurring twice in 12 months or 3 times in 24 months while on hydroxyurea if indicated.
- Are between 15 – 45 years old and had 1 or more of the following:
  - Stroke
  - 2 or more episodes of acute chest syndrome in the last 2 years while getting other treatment such as hydroxyurea
  - 3 or more episodes of serious pain crises in the last 2 years while getting other treatment such as hydroxyurea
  - Regular red blood cell (RBC) transfusions (8 or more transfusion events during the last year to prevent sickle-related health problems)
  - Fast blood flow in your heart and in the wrong direction. This may mean you may have a higher risk of dying.
- Have a donor (parent, brother, sister, or other family member) who is a half match
  You can't be in this study if you have one or more of the following:
  - A brother or sister who closely matches more than half of your HLA
  - Liver cirrhosis (damage to the liver which can lead to scarring and liver failure)
  - A very serious bacterial, fungal, or viral infection in the past 6 weeks
  - HIV infection
  - Already had a bone marrow transplant
  - Joined a study where you took a study drug or used a medical device.
  - A serious autoimmune disorder (where your own immune system attacks your body) like lupus or scleroderma
  - Are currently pregnant or breast feeding
  - Do not agree to use contraception or sexual abstinence for 12 months after the bone

marrow transplant

4. RIGHT TO ASK QUESTIONS AND/OR WITHDRAW

You have the right to ask questions about the study at any time. If you have questions about your rights as a participant or you want to leave the study, please contact:

[Insert contact info]

Being in this study is voluntary. You can choose to not be in this study or leave this study at any time.

If you choose not to take part or to leave this study, it will not affect your regular medical care in any way.

Your study doctor and study staff will be available to answer any questions that you may have about taking part or leaving this study.

5. STUDY TREATMENTS AND TESTS

If you join this study, we'll check your health before, during, and for 2 years after your treatment.

A. BEFORE YOUR TREATMENT

You'll have several check-ups and tests before your transplant. Some of these tests are given to all patients who get a transplant. Others are for this study only.

Some of the tests will be done up to 6 months before you enroll and some of the tests will be done later, about a month before enrolling on the study and then again before starting the drug called hydroxyurea, or before starting the drug called ATG (Thymoglobulin). The tests that are given to all transplant patients (not just patients in this study) and include:

- Medical history, including past and present use of medications. **Note:** Incomplete information could have a serious effect on your health and safety.
- Physical exam, height and weight
- Performance score to see how well you can do certain activities like going to work and caring for yourself
- Urine test to check for proteins and infections
- Radiology test or a 24-hour urine collection to learn how well your kidneys work
- 1 tablespoon of blood to check for:
  - Blood counts including the number of platelets
  - Number of elements and minerals in your blood
  - Different kinds of hemoglobin proteins in your red blood cells
  - Infections with hepatitis, herpes simplex, syphilis, HIV, Human T-Lymphotropic Virus (HTL), chicken pox, and shingles that are active now or happened in the past

- 1 teaspoon of blood for:
- Pregnancy test, if you're female and able to get pregnant

The total amount of blood needed for these tests will be a little over 3 teaspoons.

- Optional research samples
  - If you agree, we will also collect about 4-8 teaspoons of blood for research.

Other tests for this study include:

- Pain diary: Your doctor and/or research team will provide the instructions you will need to complete the pain dairy. If you speak English and are 15 or older, self-documentation of pain will occur twice daily for 28 days via an electronic application.
- Questions about your health and quality of life if you speak English or Spanish and are 15 years or older (to find out how healthy you feel you are and how well you can do your everyday activities): a survey to measure health outcomes from the patient perspective. Your doctor and/or the research team will provide the instructions and materials you will need to complete the survey at each time point it is due.
- Electrocardiogram (ECG and EKG) to take pictures of your heart and see how well it's working
- Chest X-ray
- Lung function tests to see how well your lungs work (including a test to see how far you can walk in 6 minutes)
- Brain scan (MRA or MRI) to show a picture of your brain and its blood vessels
- MRI of your liver if you've had a lot of red blood cell transfusions
- Liver biopsy if needed (we'll take a small sample of your liver if your liver MRI shows high iron) to see if there is damage from iron
- Cardiac scan (MRI) if needed
- HLA typing and blood test for rejecting the donor bone marrow
- Data regarding your clinical situation, including follow-up 2 years after your transplant, may be obtained from the CIBMTR, which captures information on all US transplants.

B. DURING YOUR TREATMENT

Getting Your Catheter:

A surgeon will place a **central venous catheter** in your body, most likely in your chest. A catheter is a thin, hollow tube that allows medicines, blood transfusions, and blood draws to be done painlessly and to avoid repeated needle sticks. It's also called the "central line".

The catheter will need to be cleaned every day to avoid blood clots and infection. You and your caregiver will be given instructions on how to take care of the catheter.

The surgeon will talk with you before the surgery about the risks related to having a catheter.

Preparing for Transplant:

To prepare your body for transplant, we use chemotherapy and radiation to destroy or stop the

abnormal blood cells from being made. This step is called the "conditioning regimen".

The chemotherapy (chemo) drugs also help the donor cells engraft. "Engraft" means that the cells start to grow and make new cells and show up in your blood.

See **Table 1: Schedule of Treatments Before and After Transplant** for a list of the chemo drugs you'll get and how often you'll take them. The chemo drugs and radiation before transplant include:

1. Hydroxyurea: This treatment will start in the hospital (or clinic) and continue at home.
2. Rabbit anti-thymoglobulin (rATG)
3. Thiotepa
4. Fludarabine
5. Cyclophosphamide
6. Total body irradiation (TBI): A single dose will be given while you are put in front of a machine.

Transplant Day:

On transplant day (Day 0), you'll get your donor cells through your catheter. Your blood pressure, heart and respiration rates, and temperature will be taken before and during the transplant.

The study doctor may also give you medicines before the transplant to help with any side effects or discomfort. Some of these side effects and discomforts include:

- Complications with catheter like blood clots and infection
- Slow recovery of blood counts
- New stem cells aren't growing at all (graft failure) which means you continue to have your disease
- **Graft-Versus-Host Disease (GVHD)** (described below)
- Damage to the vital organs in your body
- Serious infections
- Disease comes back (this happens when your body rejects the new stem cells)
- Death
- Reproductive risks (ability to have or father children after the transplant)

  See Section 6: Risks and Discomforts for more information.

C. AFTER YOUR TRANSPLANT

Graft Versus Host Disease (GVHD) Prevention Drugs:

Graft Versus Host Disease (GVHD) happens when the donor cells see your body as foreign (or different) and attack it. It can be a very serious side effect of transplant.

We'll give you 3 drugs to help prevent GVHD after your transplant. See **Table 1: Schedule of Treatments Before and After Transplant** for a list of the GVHD prevention drugs you'll get and how often you'll take them:

1. Cyclophosphamide: You'll get cyclophosphamide to prevent GVHD and also to help the donor cells engraft. "Engraft" means that the cells start to grow and make new cells and show up in your blood.
2. MESNA: Because there is a risk of irritation and damage to the bladder, MESNA will be given before and after cyclophosphamide to help protect the bladder.
3. Mycophenolate mofetil (MMF): Your doctor may keep giving MMF to you after 35 days, if you have GVHD.
4. Sirolimus: The study doctor may change the dose based on how your body reacts to the drug. Also, your doctor may keep giving it to you after 1 year, if you have GVHD.

TABLE 1: SCHEDULE OF TREATMENTS BEFORE AND AFTER TRANSPLANT

Treatments	Days -70 to -10	Days -9, -8	Da y -7	Days -6, -5	Days -4 to -2	Da y -1	Day 0 (Transplant day)	Days +3, +4	Days +5 to +35	Days +5 to +365 / 1 year
Hydroxyurea, by mouth	X									
Rabbit anti-thymocyte globulin (rATG), by IV		X	X							
Thiotepa, by IV 2 times in one day			X							
Cyclophosphamide, by IV 2 times daily				X						
Fludarabine, by IV				X	X					
Total body irradiation (TBI), 1 dose standing in front of machine						X				
Transplant day (donor marrow)							X			
Cyclophosphamide, by IV								X		
Mesna, by IV before and after cyclophosphamide								X		
Mycophenolate mofetil (MMF), by mouth or injection 3 times daily									X	
Sirolimus, by mouth										X

After-Transplant Tests:

1 to 100 days after your transplant (Day 1 – 100)

- Weekly GVHD tests
- Weekly Medical history, physical exam, height and weight (until Day 28)
- Weekly Performance score to see how well you can do certain activities like going to work and caring for yourself (until Day 28)
- 1 Tablespoon of blood to check for:
    - Blood counts including the number of platelets (two times per week until Day 28, weekly until Day 84)
    - Number of elements and minerals in your blood (beginning on Day 28 until Day 100) 28 days after your transplant (Day 28)
- Blood or bone marrow sample to find the amount of donor cells in your body (chimerism test) done at Day 28 and Day 42 (if needed)
- Weekly Medical history, physical exam, height and weight
- Optional blood samples

60 days after your transplant (Day 60)

- Optional blood samples

100 days after your transplant (Day 100)

- Medical history, physical exam, height and weight
- Hemoglobin (blood) test to measure how much sickle hemoglobin is in your blood
- Performance score to see how well you can do certain activities like going to work and caring for yourself
- Blood or bone marrow sample to find the amount of donor cells in your body (chimerism test)
- Optional blood samples

6 months after your transplant (Day 180)

- Performance score to see how well you can do certain activities like going to work and caring for yourself
- Hemoglobin (blood) test to measure how much sickle hemoglobin is in your blood
- GVHD tests
- Blood sample to find the amount of donor cells in your body (chimerism test)
- Blood counts including the number of platelets
- Weekly Medical history, physical exam, height and weight
- Optional blood samples

18 months after your transplant (Day 540)

- GVHD tests

1 and 2 year(s) after your transplant (Day 365 and Day 730)

- Medical history and physical exam, including height and weight
- GVHD tests
- Immune system test to see how well your immune system is working
- Hemoglobin (blood) test to measure how much sickle hemoglobin is in your blood
- Blood counts including the number of platelets
- 24-hour urine collection to learn how well your kidneys work
- Lung function test and oxygen level
- 6 minute walk distance test
- Echocardiogram (echo) to take pictures of your heart and see how well it's working if your doctor determines this is needed
- Cardiac MRI if needed
- Liver MRI for those with iron overload
- Brain scan to show a picture of your brain and its blood vessels
- Questions about your health and quality of life if you speak English or Spanish and are 15 or older
- Blood sample to find the amount of donor cells in your body (chimerism test)
- Twice daily pain diaries if you are 15 or older (Days 337-365 and Days 702-730)
- Optional blood samples (at Day 365)

Most of these tests will be done as part of your regular medical care after transplant. The tests for this study include lung function test, echocardiogram (if needed), questions about your health and quality of life, brain scan, and optional blood samples.

Table 2: Timeline of Tests after Your Transplant lists the tests you'll have after your bone marrow transplant.

TABLE 2: TIMELINE OF TESTS AFTER YOUR TRANSPLANT

Tests	Days After Transplant							
	Day 1- 100 (weekly)	**+42**	**+60**	**+100**	**+180**	**+365**	**+540**	**+730**
Tests for GVHD	☐				☐	☐	☐	☐
Test required by the hospital, including blood and urine tests	☐	☐		☐	☐	☐		☐
Hemoglobin (blood) test				☐	☐	☐		☐
Tests to see how much of the donor cells are in your body (chimerism)	☐ (Day 28)	☐ (if needed)		☐	☐	☐		☐
Quality of life surveys if you speak English or Spanish and are 15 or older						☐		☐
Performance score to see how well you can do certain activities like going to work and caring for yourself	☐ (weekly until Day 28)			☐	☐	☐		☐
Immune system test						☐		☐

Tests	**Days <u>After</u> Transplant**							
	Day 1- 100 (weekly)	**+42**	**+60**	**+100**	**+180**	**+365**	**+540**	**+730**
Lung function test (including a test to see how far you can walk in 6 minutes)						☐		☐
Echocardiography test to take pictures of your heart and see how well it's working (if needed)						☐		☐
Brain scan						☐		☐
Pain diaries over 28 days if you speak English and are 15 or older						☐		☐
Cardiac MRI (if needed)						☐		☐
Liver MRI (for those with iron overload						☐		☐
Optional blood samples	☐ (Day 28)		☐	☐	☐	☐		

6. RISKS AND DISCOMFORTS

You may have side effects while on the study. Side effects can range from mild to serious. Your health care team may give you medicines to help with certain side effects like nausea (feeling sick to your stomach). In some cases, side effects can last a long time or may never go away.

A. RISKS OF MEDICINES

Table 3: Risks and Side Effects shows how side effects are grouped together. The 3 groups are based on how often patients get each side effect.

TABLE 3: RISKS AND SIDE EFFECTS

Likely	What it means: This type of side effect is expected in more than 20% of patients. This means that 21 or more patients out of 100 might get this side effect.
Less Likely	What it means: This type of side effect is expected in 20% of patients or fewer. This means that 20 patients or fewer out of 100 might get this side effect.
Rare, but Serious	What it means: This type of side effect is expected in fewer than 2% of patients. This means that 1 or 2 patients (or fewer) out of 100 might get this side effect. It doesn't happen very often but is serious when it does.

Risks of Conditioning Regimen Medicines

The risks of the chemotherapy drugs you get as part of the conditioning regimen are listed below.

HYDROXYUREA – CHEMOTHERAPY DRUG

Likely (May happen in more than 20% of patients)	**Less Likely** (May happen in fewer than 20% of patients)	**Rare, but Serious** (May happen in fewer than 2% of patients)
• Nausea (feeling sick to your stomach) • Throwing up • Diarrhea • Loss of appetite • Low number of red blood cells (anemia) • Low number of white blood cells • Low number of blood platelets • Mouth sores • Throat inflammation (sore, red, swollen throat), going down to the stomach • Bleeding • Infection	• Dizziness • Headaches • Changes in behavior • Confusion • Seeing or hearing things that are not really there • Feeling drowsy • Abnormal liver tests • Yellow tint to skin or eyes (jaundice) • Darkening skin • Pain and difficulty with urination • Kidney damage • Seizures • Finger and toe nail changes	• Hives • Skin rash • Sudden high fever

FLUDARABINE (FLUDARA®) – CHEMOTHERAPY DRUG

Likely (May happen in more than 20% of patients)	**Less Likely** (May happen in fewer than 20% of patients)	**Rare, but Serious** (May happen in fewer than 2% of patients)
• Low number of red blood cells (anemia) • Low number of white blood cells • Low number of blood platelets • Feeling tired • Nausea (feeling sick to your stomach) • Throwing up (vomiting) • Weak immune system • Pneumonia • Infection • Bleeding • Pain • Electrolyte imbalance	• Diarrhea • Mouth sores • Skin rash • Fever • Swelling of hands and feet • Numbness and tingling in hands and/or feet • Loss of appetite	• Changes in vision • Feeling nervous or anxious • Confusion • Cough • Difficulty breathing • Feeling weak • Severe brain injury which can lead to death • Kidney damage that could require dialysis • Coma • New (secondary) cancers

RABBIT ANTI-THYMOCYTE GLOBULIN (R-ATG, THYMOGLOBULIN®) – CHEMOTHERAPY DRUG

Likely (May happen in more than 20% of patients)	**Less Likely** (May happen in fewer than 20% of patients)	**Rare, but Serious** (May happen in fewer than 2% of patients)
• Fever • Chills • Low number of red blood cells (anemia) • Low number of white blood cells • Low number of blood platelets • Weak immune system • Bleeding • Infection • Skin rash • Joint ache and pain	• Nausea (feeling sick to your stomach) • Throwing up (vomiting) • Diarrhea • Headache • Sweating • Back pain • Severe itching • Feeling tired • Loss of appetite Serum sickness with: • Severe skin rash • Mouth sores • Vaginal sores, if female • Pain and swelling in joints • Kidney damage	• Stomach (belly) pain • Feeling dizzy • High blood pressure • Blisters • Muscle pain • Herpes simplex infection • Throat inflammation (sore, red, swollen throat) • Kidney failure Severe allergic reaction which may cause: • Life-threatening drop in blood pressure • Wheezing • Difficulty breathing • Severe hives

THIOTEPA – CHEMOTHERAPY DRUG

Likely (May happen in more than 20% of patients)	**Less Likely** (May happen in fewer than 20% of patients)	**Rare, but Serious** (May happen in fewer than 2% of patients)
• Low number of red blood cells (anemia) • Low number of white blood cells • Low number of blood platelets • Bleeding • Infection • Diarrhea • Nausea (feeling sick to your stomach) • Throwing up (vomiting) • Mouth and throat sores • Liver damage • Temporary hair loss • Loss of appetite • Infertility (inability to have children)	• Change in vision • Swelling in lower legs and feet • Abnormal liver tests • Skin rash • Skin darkening	• Red face • Painful and difficult urination • Kidney damage • Yellow tint to skin and eyes (jaundice) • Headache • Feeling dizzy • Seeing or hearing things that are not really there • Confusion • Disorientation • Seizures • Temporarily increased levels of liver enzymes • Finger or toenail changes • New (secondary) cancers

TOTAL BODY IRRADIATION (TBI) – RADIATION THERAPY

Likely (May happen in more than 20% of patients)	**Less Likely** (May happen in fewer than 20% of patients)	**Rare, but Serious** (May happen in fewer than 2% of patients)
• Nausea (feeling sick to your stomach) • Throwing up (vomiting) • Diarrhea • Stomach (belly) pain • Feeling tired • Low number of red blood cells (anemia) • Low number of white blood cells • Low number of blood platelets • Bleeding • Infection • Temporary hair loss • Cataracts • Slow growth in children • Infertility (inability to have children) • Thyroid problems or diabetes • Mouth sores	• Lung inflammation and pneumonia • Red skin color • Abnormal liver tests	• Pain and swelling under the chin • Difficulty swallowing • Back pain • New (secondary) cancers • Lung damage • Kidney damage

Risks of Drugs Used to Prevent GVHD

You will get medicines to help prevent GVHD after your transplant. The side effects of the GVHD drugs are listed below. These side effects usually stop when you're done taking the medicines.

CYCLOPHOSPHAMIDE (CYTOXAN®) – GVHD PREVENTION DRUG

Likely (May happen in more than 20% of patients)	**Less Likely** (May happen in fewer than 20% of patients)	**Rare, but Serious** (May happen in fewer than 2% of patients)
• Low number of white blood cells • Low number of blood platelets • Bleeding • Infection • Blood in urine • Weak immune system • Temporary hair loss • Nausea (feeling sick to your stomach) • Throwing up (vomiting) • Headache • Dizziness • Loss of appetite • Sores in mouth or on lips • Diarrhea • Menstrual periods stop in females • Low sperm count in males	• Low number of red blood cells (anemia) • Temporary darkening of finger and toenail beds • Acne • Feeling tired • Damage to unborn baby or miscarriage • Birth defects	• Lung scars, with cough and shortness of breath • Severe heart muscle injury and death • New (secondary) cancers

MESNA – BLADDER DAMAGE PREVENTION DRUG

Likely (May happen in more than 20% of patients)	**Less Likely** (May happen in fewer than 20% of patients)	**Rare, but Serious** (May happen in fewer than 2% of patients)
• Nausea (feeling sick to your stomach) • Throwing up (vomiting) • Diarrhea • Stomach (belly) pain • Feeling tired • Headache • Pain in joints • Pain in arms and hands, legs and feet • Change in the way things taste • Skin rash • Hives	• Low blood pressure • Feeling dizzy or faint	

SIROLIMUS – GVHD PREVENTION DRUG

Likely (May happen in more than 20% of patients)	**Less Likely** (May happen in fewer than 20% of patients)	**Rare, but Serious** (May happen in fewer than 2% of patients)
• Nausea (feeling sick to your stomach) • Throwing up (vomiting) • Diarrhea and/or constipation • High blood pressure • Tremor (shaking) • Infection • Fever • Feeling weak • Feeling tired • Abnormal kidney tests • Low number of red blood cells (anemia) • Headache and/or back pain • Joint or muscle pain • Stomach or belly pain • High levels of sugar in the blood • High cholesterol • Swelling in feet and lower legs • Weight gain • Acne	• Chest pain • Insomnia (unable to sleep) • Low level of magnesium, potassium and/or phosphate • Abnormal liver tests • Skin rash or hives • Upset stomach (heartburn) • Throat inflammation (red, sore, swollen throat), going down to stomach • Shortness of breath • Low number of white blood cells • Low number of blood platelets • Bleeding • Infections • Slow wound healing • Unwanted hair growth	• Low blood pressure • Lung problems, including asthma • Loss of appetite • Serious infections • Fast heartbeat • Heart damage • Blood clots • Kidney failure • Bone thinning (osteoporosis) or loss (necrosis) which may cause broken bones • New (secondary) skin cancers

MYCOPHENOLATE MOFETIL (MMF, CELLCEPT®) – GVHD PREVENTION DRUG

Likely (May happen in more than 20% of patients)	Less Likely (May happen in fewer than 20% of patients)	Rare, but Serious (May happen in fewer than 2% of patients)
• Birth control pills may not work • Birth defects • Damage to unborn baby or miscarriage • Nausea (feeling sick to your stomach) • Throwing up (vomiting) • Diarrhea • Stomach (belly) pain • Headache • Low number of white blood cells • Infection • Swelling of hands, feet, or lower legs	• Low number of red blood cells (anemia) • Skin rash • Insomnia (unable to sleep) • Feeling dizzy • Hand shaking (tremors)	• Difficulty breathing • Abnormal bruising • Fast heartbeat • Feeling very tired • Feeling weak • Blood in stool • Blood in vomit • New (secondary) cancers • Progressive multifocal leukoencephalopathy (a disease that damages the brain and may lead to death)

Risks and Toxicities Related to Transplant

The following problems may happen because of your transplant. These risks may happen with all allogeneic transplants, whether they are part of a study or not. The risks include:

Complications with Central Venous Catheter:

The most common problems with central venous catheters are: 1) blood clots in the catheter and 2) infection. Your doctor will inject medicines into the catheter to dissolve a clot or treat an infection. If the medicines don't work, the catheter might be replaced with a new one.

Slow Recovery of Blood Counts:

The red blood cells, white blood cells, and platelets can be slow to recover after a transplant. Until your blood counts recover, you'll be at risk for bleeding and infection, and you'll need blood and platelet transfusions.

Graft Failure:

We'll check your blood often to see how well the stem cells (the "graft") are growing inside your body. If the graft isn't growing well or at all (graft failure), we'll give you medicines to help the stem cells grow. You may also need a second transplant.

Past experience suggests that there can be a 10 - 15% chance of graft failure. If your graft fails, you'll stop being in this study and receive the standard care for SCD.

Graft-Versus-Host Disease (GVHD):

GVHD happens when the donor cells see your body as foreign and attack it. In most cases, GVHD can be successfully treated. Sometimes GVHD is serious or difficult to treat and may lead to death. You'll be watched closely for GVHD and given drugs to help prevent and treat it.

Acute GVHD, which can happen 0 – 3 months after transplant, may cause skin rash, nausea, vomiting, diarrhea, abdominal pain, problems with your liver, and an increased risk of infection.

Chronic GVHD, which can happen 3 months or later after transplant, may produce skin rashes, hair loss, thickened dry skin, dry eyes, dry mouth, liver disease, weight loss, diarrhea, and an increased risk of infection.

To confirm the diagnosis of acute or chronic GVHD, your doctor may do a biopsy of your skin, gut, or liver. A biopsy is a small sample of your tissue to look at under the microscope.

Damage to the Vital Organs in Your Body:

The transplant may cause problems with your organs such as the heart, lungs, liver, gut, kidneys, bladder, or brain. The kidneys and liver are most likely to be damaged.

Some patients will experience serious lung problems from an infection, from chronic GVHD or from the chemotherapy and radiation.

Serious Infections:

Full and complete recovery of your immune system may take several months. During this time, there is an increased risk of infection. We'll give you drugs to reduce the chance of infection, but these treatments don't always work. If you get an infection, you may have to stay in the hospital longer or be re-hospitalized. Most infections can be treated, but some result in death.

Recurrence of Disease and Graft Rejection:

There is a risk that the new marrow will fail to "take" and won't grow after your transplant. This is called **graft rejection**. If this happens, your own marrow will recover, and sickle cell disease will come back. Pain crises or other complications might also return. Very rarely, other complications from graft rejection can happen including serious infection and the need for a blood transfusion.

Damage to Central Nervous System (CNS):

The transplant may affect your central nervous system (CNS). Your CNS includes your brain and spine. We'll watch you closely after your transplant for any side effects to your brain or spine.

Risk of Death:

Side effects of an allogeneic transplant can be very serious and possibly lead to death. Death can occur either as a result of the medications you receive to prepare your body for transplant or from a complication of transplant. We'll do everything we can to make the transplant as safe as possible for you.

Reproductive Risks:

The drugs used in this research study may damage your reproductive organs and affect your ability to have children. We don't know the exact risk of sterility (inability to have children) caused by taking the study drugs.

You should talk to a specialist about your options for fertility preservation. Your options may include storing your sperm or eggs, or tissue from your ovary or testes. We'll refer you to a fertility preservation center before your transplant.

The treatments in this study have not been proven to be safe at any stage of pregnancy, including when the sperm enters the egg (conception).

- **If you're female**:
  - Your menstrual cycle might become irregular or permanently stop if you've gone through puberty.
  - You must use 2 effective methods of birth control if you're sexually active (having sexual intercourse with a male partner) during your transplant. You must continue to use 2 effective birth control methods or refrain from all acts of vaginal sex (abstinence) until you're finished with your GVHD prevention medicines or GVHD treatment or 12 months after your transplant (whichever comes last).

Examples of effective birth control include:

1. Consistent use of birth control pills
2. Injectable birth control methods (Depo-Provera, Norplant)
3. Tubal sterilization or male partner who has undergone a vasectomy
4. Placement of an IUD (intrauterine device)
5. Use a diaphragm with contraceptive jelly every time you have sex and/or
6. Use condoms with contraceptive foam every time you have sex.

Tell your doctor right away if you become pregnant during the study.

- **If you're male:**
  - If you're sexually active (having sexual intercourse with a female partner), you or your partner must use 2 effective methods of birth control during your transplant. You or your partner must continue to use 2 effective birth control methods or refrain from all acts of vaginal sex (abstinence) until you are finished with your GVHD prevention medicines or GVHD treatment or 12 months after your transplant (whichever comes last).

Examples of effective birth control include:

1. Consistent use of birth control pills
2. Injectable birth control methods (Depo-Provera, Norplant)
3. Tubal sterilization or male partner who has undergone a vasectomy
4. Placement of an IUD (intrauterine device)
5. Use a diaphragm with contraceptive jelly every time you have sex and/or
6. Use condoms with contraceptive foam every time you have sex.

Tell your doctor right away if your partner becomes pregnant during the study.

Check with your doctor to understand more about these risks.

OTHER RISKS

Quality of Life Surveys:

There are a few risks from completing the quality of life surveys. Some of the questions or topics may upset you. You may feel emotional or that your privacy is lost. Talk to your doctor about your privacy concerns. We can put you in touch with a counselor or trained support specialist, if needed.

Risks of Blood Draws:

There are no major risks with blood draws. Having your blood drawn can be uncomfortable and may cause a bruise. In rare cases, a blood draw can cause fainting. Only trained people will draw your blood.

Unforeseen Risks:

Other new risks might appear at any time during the study. These risks might be different from what is listed in this Consent Form. There may be some unknown or unanticipated discomforts or risks associated with this treatment in addition to those specified above, but every precaution will be taken to assure your personal safety and to minimize discomforts.

Other Treatments or Medicines:

Some medicines react with each other, so it's important to tell the study doctor or staff about any other drugs, treatments, or medicines you're taking. This includes non-prescription or over-the-counter medicines, vitamins, and herbal treatments.

It's also important that you tell the study staff about any changes to your medicines while you're in the study.

For more information about risks and side effects, ask your study doctor.

7. OTHER TREATMENTS

It's optional to join this study. If you choose not to join, you may still receive an allogeneic transplant to treat your disease. The treatment you'd receive could be very similar to what you'd receive in this study.

Your study doctor will talk with you about your options. Your other choices may include:

- Other types of transplant that use different doses of medicines (check with your doctor)
- Other clinical trials that use mismatched related (parent, brother, sister or other family member) donors, unrelated donors or other stem cell sources (check with your doctor)

- Standard care for SCD (such as hydroxyurea or blood transfusions)
- Another experimental treatment that is not a transplant

Every treatment option has risks and benefits. Your study doctor will discuss the options, including the risks and benefits, with you.

If you decide not to join this study, your medical care will not be affected in any way.

8. POSSIBLE BENEFITS

We don't know if the treatments in this study will make your SCD better. If the transplant works well, you may not have any more symptoms of SCD such as serious pain.

The study results will help doctors know what works best to treat SCD. This knowledge could help SCD patients in the future.

9. NEW INFORMATION AVAILABLE DURING THE STUDY

The study doctors could learn new information about the risks and benefits of allogeneic transplant and the study medicines while the study is going on. If this happens, they'll tell you about it. Your doctor may decide to stop your participation. You may decide that you don't want to continue in the study.

If you decide to stop being in the study, your doctor will discuss other treatment options with you.

10. PRIVACY, CONFIDENTIALITY AND USE OF INFORMATION

Your privacy is very important to us. The study doctors will make every effort to protect it. The study doctors have a privacy permit to help protect your records if there is a court case. However, some of your medical information may be given out if required by law. If this should happen, the study doctors will do their best to make sure that any information that goes out to others will not identify who you are.

If information from this study is published or presented at scientific meetings, your name and other personal information will not be used. Your study number is not related to your name, social security number or medical record number at [insert facility name].

Information about your transplant from your original medical records may be seen or sent to national and international transplant registries, including:

- The Center for International Blood and Marrow Transplant Research (CIBMTR)
- The National Marrow Donor Program (NMDP)

- The Food and Drug Administration (FDA)
- The National Institutes of Health (NIH), which include the National Heart, Lung, and Blood Institute (NHLBI) and the National Cancer Institute (NCI)
- Data and Coordinating Center of the Blood and Marrow Transplant Clinical Trials Network (BMT CTN)
- Data Warehouse Consultants (DWC) which is an agent of Emory University that will manage the pain diary. DWC will have access to your phone number, email address, IP Address and data that is entered in the pain dairy application.
- Researchers and staff members at Vanderbilt University for central review of brain MRI/MRA images.
- Dr. Catherine Bollard and laboratory staff at Children's National Medical Center
- Other authorized study organizations

We'll not identify you by name in any publications or reports that come from these organizations or groups.

11. ENDING YOUR PARTICIPATION

The study doctor or the study sponsor may stop the study at any time. We may ask you to leave the study if you don't follow directions or if you suffer from side-effects of the treatment. If you are asked to leave the study, the reasons will be discussed with you.

Possible reasons to end your participation in this study include:

- You don't meet the study requirements.
- You need a medical treatment not allowed in this study.
- The study doctor decides that it would be harmful to you to stay in the study.
- You're having serious side-effects.
- You become pregnant.
- You can't keep appointments or take study drugs as directed.
- The study is stopped for any reason.

12. PHYSICAL INJURY AS A RESULT OF PARTICIPATION

It's important that you tell your study doctor or study staff if you feel that you've been hurt or injured because of taking part in this study.

You'll get medical treatment if you're injured as a result of taking part in this study. You and/or your health plan will be charged for this treatment. This study will not pay for this medical treatment.

In case of injury resulting from this study, you don't lose any of your legal rights to seek payment by signing this form.

13. COMPENSATION OR PAYMENT

If you speak English and are 15 years or older, you will be asked to complete a pain diary when first enrolled and then 1 and 2 years later. If you complete the pain diary twice daily, you will receive a $50 gift card. This compensation will be the same for all three of the 28-day reporting periods. The potential compensation for completing the pain diary at all of these time points is $150 ($50 for each 28-day reporting period).

14. COSTS & REIMBURSEMENTS

Most of the visits for this study are standard care for SCD patients who have an allogeneic transplant and will be billed to your insurance company.

You and/or your health insurance company will need to pay for some or all of the costs of standard treatment in this study.

You will not be charged for the collection of optional samples.

15. ETHICAL REVIEW

The ethical aspects of this research study have been reviewed and approved by [name of IRB].

16. FOR MORE INFORMATION

If you'd like more information about this study, or if you have any problems while you're participating in this study, you can contact the study doctor or staff.

They may be contacted at the telephone numbers listed here:

[Insert name and contact details]

A description of this clinical trial will also be available on http://www.ClinicalTrials.gov, as required by U.S. Law. This web site will not include information that can identify you. At most, the web site will include a summary of the results. You can search this web site at any time.

17. INDEPENDENT CONTACT

If you wish to speak to someone not directly involved in the study, or have any complaints or questions about your rights as a research participant, you may contact:

[Insert appropriate contact details]

18. OPTIONAL BLOOD SAMPLES FOR TRIAL-RELATED RESEARCH STUDIES

This section of the Consent Form is about providing optional blood samples for use in additional research.

You can choose to give blood samples for optional trial-specific studies if you want to. You can still be a part of the main study even if you say "no" to giving optional blood samples for these studies. Please mark your choice at the end of this section.

Researchers are trying to learn more about how the human body recovers in SCD patients after transplant. This research is meant to gain knowledge that may help people in the future and make transplants even more successful.

If you agree to provide optional blood samples, here is what will happen:

- We'll take the sample from your catheter or by a vein in your arm. We will collect 40mL (about 8 teaspoons) if you are 15 years or older, or 24-40 mL (about 4-8 teaspoons) if you are 5 - 14 years old. We'll collect this sample when you have your check-up before treatment starts and 28, 60, 100, 180, and 365 days after your transplant.
    - These blood samples will be shipped on the day of collection to Children's Research Institute laboratory for an important study related to this trial.
    - The samples will be labeled with unique codes that do not contain information that could identify you. A link to this code does exist. The link is stored at the Data and Coordinating Center for the Blood and Marrow Transplant Clinical Trials Network (BMT CTN DCC). The staff at the Children's Research Institute Laboratory where your samples are being stored do not have a link to this code.
    - When the Children's Research Institute Laboratory has completed their research to learn about immune system recovery in sickle cell disease after transplant, leftover research samples will be transferred and stored at the National Marrow Donor Program (NMDP) Biorepository for approved research studies by other investigators. However, these laboratory investigators will not be able to trace the sample back to you.

- Your research samples will continue to be stored at the NMDP Biorepository until they are used up for research. They will be kept unless you happen to change your mind and request to have your samples destroyed by withdrawing from the study or the Sponsor requests use of stored samples to be discontinued. If you stop being in the primary study before it is finished, upon your written request to [Insert site investigator] any remaining research samples you have given will be discarded when you tell us that you want to stop being in the study. Results we get before you stop being in the study will be kept.
- Your name and other information that could directly identify you (such as address or social security number) will not be used. Researchers have a duty to protect your privacy and to keep your information confidential.

Genetic Studies

DNA from your stored blood samples might be used in future genetic studies. We would like to test your DNA (or genes) to learn if some genes predict who will have serious complications of sickle cell disease. DNA is inherited information like a blueprint about the structure and functions of human body traits that make up the color of our hair and eyes and may affect the way our bodies respond to things that happen outside the body such as smoking, an illness, or infections. We are interested in the possibility that there are genes besides the sickle hemoglobin mutation that predict the development of other complications due to sickle cell disease.

Genome-Wide Association Studies

DNA from your stored blood samples might be used in genome-wide association (GWA) studies for a future project either done or supported by the National Institutes of Health (NIH). Genome-wide association studies are a way for scientists to find genes that have a role in human disease or treatment. Each study can look at hundreds of thousands of genetic changes at the same time.

If your coded samples are used in such a study, the researcher is required to add your test results and sample information into a shared, public research database. This public database is called the NIH Genotype and Phenotype Database and it is managed by the National Center for Biotechnology Information (NCBI). The NCBI will never have any information that would identify you, or link you to your information or research samples, although the results of genetic studies could theoretically include identifying information about you.

How can I find out about the results of the research?

You will not have any direct health benefits from providing your specimens for future research. It will probably take a long time for the research performed to be used to produce health-related information that we will know how to interpret accurately. For this reason, and because we will not know who the individual sample donors are, we will not be able to give you individual results from studies that may be conducted using the specimens. Knowledge from future research studies

is likely to yield information that is more widely or generally applicable and not specific to an individual.

Benefits

The research that may be done with your blood samples is not designed specifically to help you. The benefits of research using blood samples include learning more about how the immune system recovers in SCD patients after transplant.

Risks

There is a small risk of an infection or fainting from the blood draw.

A possible risk is the loss of confidentiality about your medical information. We will do our best to make sure that your personal information is kept private. The chance that this information will be given to someone else is very small.

Some general things to think about when letting us collect your blood samples for research are:

- The choice to let us collect your blood samples is up to you. No matter what you decide to do, it will not affect your care.
- If you decide now that your blood samples can be collected for research, you can change your mind at any time. Just contact your study doctor in writing and let him or her know that you do not want us to collect anymore of your blood samples for research. His/her mailing address is on the first page of this form. Then any further optional blood samples needed for this research will not be collected. However, samples that have already been collected cannot be taken back or destroyed.
- People who do research on these blood samples may need to know more about your health. While the study doctor or others involved in running this study may give the researchers reports about your health, they will not give them your name, address, phone number, or any other information that will let the researchers know who you are.
- Your blood will be used only for research and will not be sold. The research done with your blood may help to develop new products in the future. You will not get paid for any samples or for any products that may be developed from current or future research.
- Reports about research done with your blood will not be given to you or your doctor. These reports will not be put in your health record. The research will not have an effect on your care.

Risks of Genetic Testing

In the course of these studies, we may find new genes that are inherited and predict the development of sickle cell disease related illnesses. Once we have obtained your DNA (or genes) from the white blood cells, we will put the DNA in tubes. These tubes will be labeled with a code and will have no markings to link the tube with you specifically. If we learn anything of importance to our research from this testing, we may publish the results in a medical journal. However, you

will not be identified in the article as the patient who provided the blood sample for our testing.

In rare instances, it is possible that we could find out information about a specific gene that could affect you or other members of your family in terms of insurability, employability, or paternity. We will do everything possible to ensure that your identity and confidentiality will not be breached. As previously mentioned, the code linking your identifying information to the sample will be kept secure by the BMT CTN DCC staff in a password protected file in a secure location.

Making Your Choice

Please read each sentence below and think about your choice. After reading each sentence, please indicate your choice below. If you have any questions, please talk to your doctor or nurse, or call our research review board at [telephone number].

No matter what you decide to do, it will not affect your care.

You can change your mind at any time about allowing us to use your samples for research. However, samples that have already been collected cannot be taken back or destroyed.

Statement of Consent for Optional Blood Samples for Trial-Related Research Studies

The purpose of collecting optional blood samples, the procedures involved, and the risks and benefits have been explained to me. I have asked all the questions I have at this time and I have been told whom to contact if I have more questions. I have been told that I will be given a signed copy of this consent form to keep. I understand that I do not have to allow the collection and storage of my blood samples for study-specific research. If I decide to not let you collect research samples now or in the future, it will not affect my medical care in any way.

I voluntarily agree that optional blood samples may be collected and that my blood samples can be sent to the Children's Research Institute Laboratory for research to learn about immune system recovery in sickle cell disease patients after transplant. Any leftover blood sample will be stored at the NMDP Biorepository for future research.

❑ I do agree to give blood samples for study-specific research.

❑ I do <u>not</u> agree to give blood samples for study-specific research.

❑ I do agree to give leftover blood samples for future research which may include DNA genetic studies.

❑ I do not agree to give leftover blood samples for future research which may include DNA genetic studies.

_____ _____

Signature Date

HEALTH INSURANCE PORTABILITY AND ACCOUNTABILITY ACT 1 (HIPAA[1]) AUTHORIZATION TO USE AND DISCLOSE INDIVIDUAL HEALTH INFORMATION FOR RESEARCH PURPOSES

A. Purpose

As a research participant, I authorize the Principal Investigators and the researcher's staff to use and disclose my individual health information for the purpose of conducting the research study:

(List PI and research staff names)

B. Individual Health Information to be Used or Disclosed

My individual health information that may be used or disclosed to do this research includes:

- Demographic information (for example: date of birth, sex, weight).
- Medical history (for example: diagnosis, complications with prior treatment).
- Findings from physical exams.
- Laboratory test results obtained at the time of work up and after treatment (for example: blood tests, biopsy results).

C. Parties Who May Disclose My Individual Health Information

The researcher and the researcher's staff may collect my individual health information from:

(List hospitals, clinics or providers from which health care information can be requested).

D. Parties Who May Receive or Use My Individual Health Information

The individual health information disclosed by parties listed in item c and information disclosed by me during the course of the research may be received and used by the following parties:

- Principal Investigator and the researcher's staff (List names)

[1] HIPAA is the Health Insurance Portability and Accountability Act of 1996, a federal law related to privacy of health information

- Study Sponsors
  National Heart, Lung, and Blood Institute (NHLBI) and the National Cancer Institute (NCI), both of the National Institutes of Health (NIH); Blood and Marrow Transplant Clinical Trials Network (BMT CTN)

- U.S. government agencies that are responsible for overseeing research such as the Food and Drug Administration (FDA) and the Office of Human Research Protections (OHRP)

- U.S. government agencies that are responsible for overseeing public health concerns such as the Centers for Disease Control (CDC) and federal, state and local health departments.

- Data Warehouse Consultants (DWC) which is an agent of Emory University that will manage the pain diary. DWC will have access to your phone number, email address, IP Address and data that is entered in the pain dairy application.

- Dr. Catherine Bollard and laboratory staff at Children's National Medical Center

E. Right to Refuse to Sign this Authorization

I do not have to sign this authorization. If I decide not to sign the authorization, I will not be allowed to participate in this study or receive any treatment related to research that is provided through the study.

My decision not to sign this authorization will not affect any other treatment, payment, or enrollment in health plans or eligibility for benefits.

F. Right to Revoke

I can change my mind and withdraw this authorization at any time by sending a written notice to the Principal Investigator to inform the researcher of my decision.

If I withdraw this authorization, the researcher may only use and disclose the protected health information already collected for this research study. No further health information about me will be collected by or disclosed to the researcher for this study.

G. Potential for Re-disclosure

My individual health information disclosed under this authorization may be subject to re-disclosure outside the research study and no longer protected.

Examples include potential disclosures for law enforcement purposes, mandated reporting or abuse or neglect, judicial proceedings, health oversight activities and public health measures.

H. This authorization does not have an expiration date.

Principal Investigator(s)

Name: Phone:

Address: Fax:

Email:

- I've read and understood this Consent Form. The nature and purpose of the research study has been explained to me.
- I've had the chance to ask questions and understand the answers I've been given. I understand that I may ask questions at any time during the study.
- I freely agree to be a participant in the study.
- I understand that I may not directly benefit from taking part in the study.
- I understand that, while information gained during the study may be published, I won't be identified, and my personal results will stay confidential.
- I've had the chance to discuss my participation in this research study with a family member or friend.
- I understand that I can leave this study at any time and doing so will not affect my current care or prevent me from receiving future treatment.
- I understand that I'll be given a copy of this signed consent form.

_____ _____
Participant or Parent/Guardian Name Date

_____ _____
Participant or Parent/Guardian Signature Date

_____ _____
Parent/Guardian Signature Date

I certify that I have provided a verbal explanation of the details of the research study, including the procedures and risks. I believe the participant has understood the information provided.

_____ _____
Counseling Physician Date

_____ _____
Signature of Counseling Physician Date

ASSENT TO PARTICIPATE IN RESEARCH

Reduced Intensity Conditioning for Haploidentical Bone Marrow Transplantation in Patients with Symptomatic Sickle Cell Disease

Study Title: Reduced Intensity Conditioning for Haploidentical Bone Marrow Transplantation in Patients with Symptomatic Sickle Cell Disease

Protocol: BMT CTN 1507

A. Why am I here?

We are inviting you to join our study because you have severe Sickle Cell Disease (SCD) or because you've had a stroke from the disease. You are now getting blood transfusions, hydroxyurea or other medicines. There is another treatment for sickle cell disease called bone marrow transplant (BMT).

SCD is a blood disease. In SCD, the red blood cells, which are normally shaped like a donut, become sickle shaped. When this happens, they can get stuck in your blood vessels. This can cause pain and damage in different body parts.

BMT uses blood making cells from another person to replace your cells that are not healthy. Donor is the name for a person who gives some of their cells for a transplant. In order for them to give you their cells, their DNA has to match yours.

B. WHY ARE YOU DOING THIS STUDY?

We know that a transplant can cure SCD but we don't know if it works when the donor is not a perfect match. We are doing this study to see if a transplant with a donor who is related to you, but is not a perfect match, is safe and if it makes you better.

C. WHAT WILL HAPPEN TO ME?

If you say you want to be in the study, we will ask you to:

- Have check-ups with the study doctors
- Give some blood (about 3 teaspoons)

- If you agree, we will collect extra blood (about 4 teaspoons) at 5 different times. Your blood samples will be used for research about transplant in patients with sickle cell disease. Your samples will be sent to a lab for an important study. All research samples will be tied to a number and researchers testing your samples will not be able to identify you.

We will watch you carefully for fevers, any sign of infection or other problems. The study will be done over 2 years.

Before your transplant, you will get a small bendable tube put in your chest in the operating room (you will be asleep for this). The small tube makes it easier for you to get your medicines. It will also make it easier for drawing blood for tests because you will not be poked.

We will give you medicines that will help make the cells from your donor grow in your body. These medicines might make you feel sick. You might throw up, lose your hair, or get sores in your mouth.

You will get cells from your donor. This is your transplant. Your new cells will come from your donor's bone marrow. The cells may make new and healthy cells in your body. Because your donor is not a perfect match, you will also get medicines after the transplant to stop the donor cells from attacking your body. This is a problem called graft-versus-host disease (GVHD).

GVHD happens when the donor cells attack your body. It can give you diarrhea, a skin rash, make you feel sick and throw up, or make you not feel hungry. Your doctors will give you medicines to try to make sure you don’t get GVHD.

You will stay in the hospital for several days before your transplant and for about 4 weeks after your transplant. After you go home, you will need to go back to see your doctor often.

It is possible that your disease will come back. If this happens, your doctor will find another way to treat you.

D. WILL IT HURT?

For your transplant, we will put a small tube in your chest. It might hurt a little and you might bleed a little. Your doctor and nurses will make sure you feel as little pain as possible. If you get mouth sores or if you get graft-versus-host disease, this can also hurt, but your doctor will give medicine to help with the pain. These problems usually will get better after a while.

When you have your blood taken with a needle, it may feel like a pinch. It will hurt for a minute and sometimes the place where the needle went might be red and sore. You might get a little bruise from the needle, but it goes away in a few days.

The medicines you get might also make you sick. You might feel sick to your stomach or throw up. You might feel tired and your body might hurt. But your doctor will give you other medicine to help you feel better. Also, you might lose your hair. But it will grow back after you are done taking the medicines.

E. WILL THE STUDY HELP ME?

We don't know if the study will help you or not. What we learn from it might help other people in the future.

F. WHAT IF I HAVE QUESTIONS?

You can ask any questions that you have about the study. If you forget to ask a question and think of it later, you can call me [insert office number]. You can also ask your question the next time you see me.

You can call the study office at any time to ask questions about the study.

G. DO I HAVE TO BE IN THIS STUDY?

If you don't want to be in the study, you need to tell us and your parent or guardian. Your doctor will not be angry or upset if you don't want to join.

Whether you are in the study or not, you will still need to have treatment for SCD. There might be other studies for sickle cell disease you can join, or a different kind of transplant

You can say yes now and change your mind at any time. Your doctor will not be angry if you change your mind.

Please talk this over with your parent or guardian before you decide if you want to be in the study. We will also ask your parents or guardian to give their permission for you to join this study.

Writing your name on this page means that you agree to be in the study and know what will happen to you. If you decide to quit the study, all you have to do is tell the person in charge.

You and your parent or guardian will get a copy of this form after you sign it.

Signature of Participant	Date
Printed Name of Participant	
Signature of Researcher	Date
Printed Name of Researcher	

Donor Informed Consent to Participate in Research

Reduced Intensity Conditioning for Haploidentical Bone Marrow Transplantation in Patients with Symptomatic Sickle Cell Disease

Your Name: _____

Study Title: Reduced Intensity Conditioning for Haploidentical Bone Marrow Transplantation in Patients with Symptomatic Sickle Cell Disease

Protocol: BMT CTN #1507

Co-Investigator: Catherine Bollard MBChB, MD
Professor of Pediatrics
The George Washington University
Director – Center for Cancer and Immunology Research
Children's National Hospital
111 Michigan Ave NW Washington, DC 20010
(O) 202-476-4776
cbollard@childrensnational.org

Principal Investigator: *[Insert local PI information]*

Sponsor: The National Institutes of Health (NIH) is sponsoring this study by providing financial support through the Blood and Marrow Transplant Clinical Trials Network (BMT CTN).

1. INTRODUCTION

We invite you to provide blood samples for research. You're being asked to join because you're a bone marrow donor for a family member who is going to receive a haploidentical (half-matched) transplant in the main study, BMT CTN 1507.

This consent form is about a research study to learn how patients' immune system recovers after they get cells from a donor (transplant). In order to do this study, we will need extra blood samples from you.

It's your choice to give blood samples. Even if you say 'no' to giving samples for this research

study, your family member can still receive a transplant from you as part of the main study. If you agree to give blood samples, we will collect them at the time of your bone marrow donation.

- These blood samples will be shipped on the day of collection to Children's Research Institute laboratory for an important study related to this trial.
- The samples will be labeled with unique codes that do not contain information that could identify you. A link to this code does exist. The link is stored at the Data and Coordinating Center for the Blood and Marrow Transplant Clinical Trials Network (BMT CTN DCC). The staff at the Children's Research Institute Laboratory where your samples are being stored do not have a link to this code.
- When the Children's Research Institute Laboratory has completed their research to learn about immune system recovery in sickle cell disease after transplant, leftover research samples will be transferred and stored at the National Marrow Donor Program (NMDP) Biorepository for approved research studies by other investigators. However, these laboratory investigators will not be able to trace the sample back to you.
- Your research samples will continue to be stored at the NMDP Biorepository until they are used up for research. They will be kept unless you happen to change your mind and request to have your samples destroyed by withdrawing from the study or the Sponsor requests use of stored samples to be discontinued. If you stop being in the primary study before it is finished, upon your written request to [Insert site investigator] any remaining research samples you have given will be discarded when you tell us that you want to stop being in the study. Results we get before you stop being in the study will be kept.
- Your name and other information that could directly identify you (such as address or social security number) will not be used. Researchers have a duty to protect your privacy and to keep your information confidential.

Genetic Studies

DNA from your stored blood samples might be used in future genetic studies. We would like to test your DNA (or genes) to learn if some genes predict who will have serious complications of sickle cell disease. DNA is inherited information like a blueprint about the structure and functions of human body traits that make up the color of our hair and eyes and may affect the way our bodies respond to things that happen outside the body such as smoking, an illness, or infections. We are interested in the possibility that there are genes besides the sickle hemoglobin mutation that predict the development of other complications due to sickle cell disease.

Genome-Wide Association Studies

DNA from your stored blood samples might be used in genome-wide association (GWA) studies for a future project either done or supported by the National Institutes of Health (NIH). Genome-wide association studies are a way for scientists to find genes that have a role in human disease or

treatment. Each study can look at hundreds of thousands of genetic changes at the same time.

If your coded samples are used in such a study, the researcher is required to add your test results and sample information into a shared, public research database. This public database is called the NIH Genotype and Phenotype Database and it is managed by the National Center for Biotechnology Information (NCBI). The NCBI will never have any information that would identify you, or link you to your information or research samples, although the results of genetic studies could theoretically include identifying information about you.

How can I find out about the results of the research?

You will not have any direct health benefits from providing your specimens for future research. It will probably take a long time for the research performed to be used to produce health-related information that we will know how to interpret accurately. For this reason, and because we will not know who the individual sample donors are, we will not be able to give you individual results from studies that may be conducted using the specimens. Knowledge from future research studies is likely to yield information that is more widely or generally applicable and not specific to an individual.This Consent Form will tell you about the purpose of the samples for research, the possible risks and benefits, other options available to you, and your rights as a research participant.

Everyone who takes part in research at [insert facility name] should know that:

- Being in any research study is voluntary.
- You will not directly benefit from being in the study. Knowledge we gain from this study may benefit others.
- If you give blood samples for research, you can change your mind at any time.
- If you decide to quit the study, it will not affect your care or the care of your family member at [insert name of facility or institution].
- Please ask the study staff questions about anything that you do not understand, or if you would like to have more information.
- You can ask questions now or any time during the study.
- Please take the time you need to talk about the study with your doctor, study staff, and your family and friends. It is your decision to provide blood samples for research. If you decide to join, please sign and date the end of the Consent Form.

The National Institutes of Health (NIH), through the Blood and Marrow Transplant Clinical Trials Network (BMT CTN), are giving staff support and money for this research study.

The BMT CTN will lead the research study and, along with the NIH, will make decisions about how to manage the study.

2. STUDY PURPOSE

We are collecting extra blood samples because we want to learn more about how the immune system recovers in SCD patients that have received a haploidentical donor bone marrow transplant

3. RIGHT TO ASK QUESTIONS AND/OR WITHDRAW

You have the right to ask questions about the study at any time. If you have questions about your rights as a study participant or you want to leave the study, please contact:

[insert contact info]

Giving blood samples for research is voluntary. You can choose not to give samples or change your mind at any time.

If you choose not to take part or change your mind, it will not affect your donation process or the treatment of your family member in the main study in any way.

If you change your mind, any unused blood samples will be destroyed. However, samples and information that have already been used for research cannot be taken back or destroyed.

Your study doctor and study staff will be available to answer any questions that you may have.

4. STUDY TREATMENTS AND TESTS

If you agree to give blood samples, here is what will happen:

a.) We will collect an extra blood sample at the time of your bone marrow donation. The amount of blood collected from you will be 20 mL (about 4 teaspoons).

b.) The blood samples will be sent to the Children's Research Institute laboratory for processing. All samples will be given a unique bar code that cannot be linked to you by the researchers testing your samples.

5. RISKS AND DISCOMFORTS

There are no major risks to having your blood drawn. It can be uncomfortable to have your blood taken and it can sometimes leave a bruise. You might faint, but this is unlikely to happen. Only trained people will take your blood.

Risks of Genetic Testing

In the course of these studies, we may find new genes that are inherited and predict the development of sickle cell disease related illnesses. Once we have obtained your DNA (or genes) from the white blood cells, we will put the DNA in tubes. These tubes will be labeled with a code and will have no markings to link the tube with you specifically. If we learn anything of importance to our research from this testing, we may publish the results in a medical journal. However, you will not be identified in the article as the patient who provided the blood sample for our testing.

In rare instances, it is possible that we could find out information about a specific gene that could affect you or other members of your family in terms of insurability, employability, or paternity. We will do everything possible to ensure that your identity and confidentiality will

not be breached. As previously mentioned, the code linking your identifying information to the sample will be kept secure by the BMT CTN DCC staff in a password protected file in a secure location.

6. POSSIBLE BENEFITS

You will not directly benefit from taking part in this study. The information from this study will help doctors learn more about how well transplant recipients do with a haploidentical (half- matched) donor.

This information could help other people with SCD who may need a transplant in the future.

7. PRIVACY, CONFIDENTIALITY AND USE OF INFORMATION

Your privacy is very important to us. The study doctors will make every effort to protect it. The study doctors have a privacy permit to help protect your records if there is a court case. However, some of your medical information may be given out if required by law. If this should happen, the study doctors will do their best to make sure that any information that goes out to others will not identify who you are.

[Name of Transplant Center] and the organizations listed below will not disclose your participation by any means of communication to any person or organization, except by your written request, or permission, or unless required by federal, state or local laws, or regulatory agencies.

- The National Institutes of Health (NIH), which include the National Heart, Lung, and Blood Institute (NHLBI) and the National Cancer Institute (NCI)
- U.S. government agencies that are responsible for overseeing research such as The Food and Drug Administration (FDA) and the Office of Human Research Protection (OHRP)
- U.S. government agencies that are responsible for overseeing public health concerns such as the Centers for Disease Control (CDC) and federal, state, and local health departments
- The Data and Safety Monitoring Board (DSMB), not part of [Institution]
- Institutional Review Boards (IRBs) responsible for this study
- Blood and Marrow Transplant Clinical Trials Network Data and Coordinating Center (BMT CTN DCC), including:
    - The Center for International Blood and Marrow Transplant Research (CIBMTR)
    - The National Marrow Donor Program (NMDP)
    - Emmes, who is coordinating the studies of the BMT CTN
- Dr. Catherine Bollard and laboratory staff at Children's National Medical Center

- Study investigators

Individuals authorized by the organizations above will have access to your research and medical information. They may use this information for inspections or audits to study the outcomes of your treatment. In agreeing to participate, you consent to these inspections. You also consent to allow authorized individuals to copy parts of your records, if required by these organizations.

We may give out your personal information if required by law. If information from this study is published or presented at scientific meetings, your name and other personal information will not be used.

A description of this clinical trial will be available on *http://www.ClinicalTrials.gov*, as required by U.S. Law. This Web site will not include information that can identify you. At most, the Web site will include a summary of the results. You can search this Web site at any time.

For questions about access to your medical records, please contact [name] at [number].

8. ENDING YOUR PARTICIPATION

The study doctor or the study sponsor may stop the study at any time, and we may ask you to leave the study. We may ask you to leave the study if you do not follow directions or if you suffer from side effects of the blood draw. The study sponsor may decide to end the study at any time. If we ask you to leave the study, the reasons will be discussed with you.

Possible reasons to end your participation in this study include:

- You do not meet the study requirements.
- You become unable to donate bone marrow to your family member.
- The study is stopped for any reason.

9. PHYSICAL INJURY AS A RESULT OF PARTICIPATION

It's important that you tell your doctor, [investigator's name(s)], or study staff if you feel that you have been injured because you provided blood samples for research. You can tell the doctor in person or call him/her at [telephone number].

You will get all available medical treatment if you are injured as a result of providing blood samples for research.

You, your health plan, or your family member's health plan will be charged for this treatment for injury. The study will not pay for medical treatment.

In case of injury resulting from providing blood samples for this study, you do not lose any of your legal rights to seek payment by signing this form.

10. PAYMENT AND STUDY COSTS

You will not be paid for your participation in the research study or for providing blood samples for research.

You will not be compensated or reimbursed for any extra costs (for example, travel and meals) from taking part in this study.

The visit at which this sample will be collected are standard for bone marrow donors and will be billed to your family member's insurance company.

You will not be charged for the collection of these optional samples or for the research tests done with these samples. The costs of shipping your blood samples will be paid by the BMT CTN.

For questions about your costs, financial responsibilities, and/or medical insurance coverage for your transplant and this study, please contact /Center/ Financial Counselor at /Number.

11. FOR MORE STUDY INFORMATION

If you need more information about providing blood samples for research, or if you have problems while you are participating in this study, you can contact the study doctor or his/her staff. They can be reached at the telephone numbers listed here:

[Insert name and contact details].

12. CONTACT SOMEONE ABOUT YOUR RIGHTS

If you wish to speak to someone not directly involved in the study, if you have any complaints about the study, or would like more information about your rights as a research participant, you may contact:

[Insert appropriate contact details].

The ethical aspects of this research study have been reviewed and approved b[name of IRB].

For more information about your rights when providing blood samples for research, call the [name of center] Institutional Review Board (a group of people who review the research to protect your rights) at [telephone number].

HEALTH INSURANCE PORTABILITY AND ACCOUNTABILITY ACT (HIPAA)[1] AUTHORIZATION TO USE AND DISCLOSE RESEARCH PURPOSES:

A. PURPOSE:

As a research participant, I authorize the Principal Investigators and the researcher's staff to use and disclose my individual health information for the purpose of conducting the research study, *Reduced Intensity Conditioning for Haploidentical Bone Marrow Transplantation in Patients with Symptomatic Sickle Cell Disease*.

B. INDIVIDUAL HEALTH INFORMATION TO BE USED OR DISCLOSED:

My individual health information that may be used or disclosed to do this research includes:

- Demographic information (for example: date of birth, sex, weight)
- Medical history
- Findings from physical exams
- Laboratory test results obtained at the time of work up

C. PARTIES WHO MAY DISCLOSE MY INDIVIDUAL HEALTH INFORMATION:

The researcher and the researcher's staff may collect my individual health information from: [*List hospitals, clinics or providers from which health care information can be requested*]

D. PARTIES WHO MAY RECEIVE OR USE MY INDIVIDUAL HEALTH INFORMATION:

The individual health information disclosed by parties listed in item c and information disclosed by me during the course of the research may be received and used by the following parties:

- Principal Investigators and the researcher's staff:
  - Dr. Robert Brodsky, Co-Principal Investigator
  - Dr. Michael DeBaun, Co-Principal Investigator
  - Dr. Adetola Kassim, Co-Principal Investigator
  - Dr. Mark Walters, Co-Principal Investigator
- Dr. Catherine Bollard and laboratory staff at Children's National Medical Center
- Study Sponsors:
  - National Heart, Lung, and Blood Institute (NHLBI) and the National Cancer Institute (NCI), both of the National Institutes of Health (NIH),
  - Blood and Marrow Transplant Clinical Trials Network (BMT CTN), data coordinating center
- U.S. government agencies that are responsible for overseeing research such as the Food and Drug Administration (FDA) and the Office of Human Research Protections (OHRP)
- U.S. government agencies that are responsible for overseeing public health concerns such as the Centers for Disease Control (CDC) and federal, state, and local health departments.
- The Data and Safety Monitoring Board (DSMB), not part of [Institution]
- Institutional Review Boards (IRBs) responsible for this study

E. RIGHT TO REFUSE TO SIGN THIS AUTHORIZATION:

I do not have to sign this Authorization. If I decide not to sign the Authorization, I will not be allowed to participate in this study or receive any treatment related to research that is provided through the study.

My decision not to sign this authorization will not affect any other treatment, payment, or enrollment in health plans or eligibility for benefits.

F. RIGHT TO REVOKE:

I can change my mind and withdraw this authorization at any time by sending a written notice to the Principal Investigator to inform the researcher of my decision.

If I withdraw this authorization, the researcher may only use and disclose the protected health information already collected for this research study. No further health information about me will be collected by or disclosed to the researcher for this study.

G. POTENTIAL FOR RE-DISCLOSURE:

My individual health information disclosed under this authorization may be subject to re-disclosure outside the research study and no longer protected.

Examples include potential disclosures for law enforcement purposes, mandated reporting for abuse or neglect, judicial proceedings, health oversight activities and public health measures.

H. THIS AUTHORIZATION DOES NOT HAVE AN EXPIRATION DATE.

TITLE: BMT CTN 1507: Reduced Intensity Conditioning for Haploidentical Bone Marrow Transplantation in Patients with Symptomatic Sickle Cell Disease

Principal Investigator: Name: Phone:

Address 1: Fax:

Address 2: Email:

For donors under 18, consent must be provided by the Legally Authorized Representative and Donor Assent is required (see **Assent Section** on the next page).

- I have had the chance to ask questions and understand the answers I have been given. I understand that I may ask questions at any time during the study.
- I freely agree to be a participant in the study.

[1] HIPAA is the Health Insurance Portability and Accountability Act of 1996, a federal law related to privacy of health information.

- I understand that I will not directly benefit from taking part in the study.
- I understand that, while information gained during the study may be published, I will not be identified, and my personal results will stay confidential.
- I have had the chance to discuss my participation in this research study with a family member or friend.
- I understand that I can leave this study at any time and doing so will not affect my family member's current care or prevent my family member from receiving future treatment.
- I understand that I will be given a copy of this signed consent form.

Participant Name	Date
Participant's Signature (if 18 years or older)	Date

Statement of Consent for Future Use of Optional Ancillary Study Blood Samples for DNA Genetic Testing Studies

Making Your Choice

Please read each sentence below and think about your choice. After reading each sentence, please indicate your choice below. If you have any questions, please talk to your doctor or nurse, or call our research review board at [telephone number].

No matter what you decide to do, it will not affect your care.

I voluntarily agree that any leftover blood sample stored at the NMDP Biorepository can be used for future research.

- ❑ I do agree to give leftover blood samples for future research which may include DNA genetic studies.
- ❑ I do not agree to give leftover blood samples for future research which may include DNA genetic studies.

Signature	Date

I certify that I have provided a verbal explanation of the details of the research study, including the procedures and risks. I believe the participant has understood the information provided.

Name of Counseling Physician/Staff	Date
Signature of Counseling Physician/Staff	Date
Name of Interpreter	Date
Signature of Interpreter	Date

PEDIATRIC DONOR ASSENT TO PARTICIPATE IN RESEARCH

Study Title: Reduced Intensity Conditioning for Haploidentical Bone Marrow Transplantation in Patients with Symptomatic Sickle Cell Disease

Protocol: **BMT CTN 1507**

A. Why am I here?

If you give us your permission, we would like to have an extra sample of your blood. We would collect the extra sample at the same time you donate your bone marrow.

B. Why are you doing this study?

We are collecting blood samples from donors, like you, to learn more about SCD patients do after their transplants. Your name will not be on the samples.

C. What will happen to me if I join the study?

If you say you want to be in the study, we will ask you for:

- **1 extra blood sample:** We'll collect about 4 teaspoons of blood 1 time.

We will use a small needle to collect the blood from a vein in your arm.

D. Will the blood draw hurt?

When we collect your blood from a vein in your arm, it may feel like a pinch. It will hurt for a minute and the place where the needle went might be red and sore. You might get a little bruise from the needle but it goes away in a few days. We will collect the extra sample at the same time as you have other blood tests done.

E. What if I have questions?

You can ask any questions that you have about this study. If you forget to ask a question and think of it later, you can call me at: [insert office number].

You can also ask your question the next time you see me. You can call the study office at any time to ask questions about the study.

F. How will you use my health information and blood samples?

Your blood samples will be used for a study about transplant in patients with sickle cell disease.

G. Where will my blood samples be sent?

Your blood samples will be sent to a laboratory at Children's Research Institute.

All research samples will be tied to a number. This number will not be linked to your nameor other identifying information.

H. Will the study help me?

This study will not help you or your family member, but it may help other SCD patients who have a transplant in the future.

I. Will I be paid to be in the study?

No, you will not be paid to be in the study. It will not cost you anything to be in the study.

J. Do I have to be in this study?

If you don't want to be in this study, you need to tell us and your parent or guardian.

Your doctor will not be angry or upset if you do not want to join. You can still give bone marrow to your family member who needs it. They will still get the exact same care.

You can say yes now and change your mind at any time.

Please talk this over with your parents before you decide if you want to give an extra blood sample for research. We will also ask your parents to give their permission for you to give an extra sample for research.

TITLE: BMT CTN 1507: Reduced Intensity Conditioning for Haploidentical Bone Marrow Transplantation in Patients with Symptomatic Sickle Cell Disease

Principal Investigator:

Name: Phone:

Address 1: Fax:

Address 2: Email:

Writing your name on this page means that you agree to give an extra blood sample and know what will happen.

If you want to quit the study, all you have to do is tell the person in

charge. You and your parent or guardian will get a copy of this

form after you sign it.

Signature of Child	Date
Signature of Person Conducting Assent	Date

APPENDIX C

IMMUNE RECONSTITION ANCILLARY STUDY PROCEDURES

Appendix C
IMMUNE RECONSTITUTION ANCILLARY STUDY PROCEDURES

Study patients and associated haploidentical bone marrow donors will be provided the opportunity to participate in an optional correlative laboratory study associated with the BMT CTN 1507 trial. The overall objective of the protocol-defined correlative laboratory study is to examine in greater detail how the immune system recovers in SCD patients that have received a haploidentical donor bone marrow transplant.

1. Background:

Recent data presented at the 2016 BMT Tandem Meeting showed that in 8 sickle cell patients receiving ex-vivo T-cell deplete transplant that sustained engraftment was seen and the average Day +30 donor NK chimerism was 90%[68]. Early predominant donor NK chimerism may thus be an important part of engraftment and may serve as an early marker of success. However, the role of NK cell chimerism in a post-transplant cyclophosphamide based T-cell depletion approach for sickle cell disease, as proposed for the BMT CTN 1507 trial, is unknown; this correlative laboratory study hopes to address this gap in knowledge.

As T cell depletion in an important part of GVHD prevention in a mismatched setting such as haploidentical transplantation it is important to confirm that viral T cell immunity is not negatively impacted. Performing T cell depletion of donor lymphocytes **ex-vivo** often leads to delayed viral-specific immunity (up to one year) leaving the patient susceptible to life threating viral infection. CMV, EBV and Adenovirus infections/reactivations are particularly problematic, with adenovirus alone affecting > 80% of pediatric patients after T cell depleted haploidentical transplant and is associated with significant morbidity and a high mortality rate if the patient develops viral disease[69]. However, in a previously published study from the Johns Hopkins group after haploidentical transplant for sickle cell disease, 3 of 14 (21%) of patients had CMV reactivation but no patient developed CMV disease, and one of these patients had EBV reactivation treated effectively with a single dose Rituximab[70]. In the preliminary experience of reduced intensity haploidentical transplantation for SCD for the BMT CTN 1507 trial, 7 of 12 patients had viral reactivation (CMV, EBV, VZV, HHV6) but no disease. We hypothesize that with a post-transplant cyclophosphamide approach that viral-specific T cells transferred with the allograft remain relatively intact given that alloreactive T cells rather than viral-specific T cells are most active at the day
+3 and +4 time-points of cyclophosphamide infusion and are preferentially depleted and accounts for the absence of viral disease. We plan to test this hypothesis by evaluating patients post-transplant for the presence of viral-specific T cells in their peripheral blood.

2. Immune Reconstitution Study Aims:

a. To determine if predominantly donor Day 28, NK cell chimerism (>90%) is more predictive of sustained engraftment than Day 28 T cell chimerism following haploidentical donor BMT for sickle cell disease

b. To determine at what time-point viral specific T cells are detectable after haploidentical BMT using a post-transplant cyclophosphamide approach for sickle cell disease.

3. Study Procedures

Study patients will be presented the option to participate in this important correlative laboratory study at the time of informed consent. For those patients agreeing to provide the optional research blood samples, their haploidentical bone marrow donor will also be asked to participate in this research study.

Peripheral blood samples will be collected from consenting patients within 30 days prior to transplant conditioning and on Days 28, 60, 100, 180 and 365 post-transplant. A single, pre-bone marrow donation peripheral blood sample will be collected from associated haploidentical donors consenting to providean optional research sample only if the patient agrees to participate in this research study.

- **Adult Patients:** 40 mL peripheral blood collected in four 10 mL sodium heparin tubes.
- **Pediatric Patients (5.00 – 14.99 years old):** 24-40 mL peripheral blood collected in 3-4, 10 mL sodium heparin tubes based on a 3 mL/kg sample collection guidance.
- **Haploidentical Bone Marrow Donors:** 20 mL peripheral blood collected in two 10 mL sodium heparin tubes.

Table 1: Requirements for Immune Reconstitution Correlative Study samples based on Donor and Patient consent

Scenario	Sample Requirement
Patient AND Donor consent to provide optional immune reconstitution samples	Patient and Donor samples should be provided at specified timepoints
Patient consents to provide optional immune reconstitution samples and Donor does NOT consent to provide optional immune reconstitution samples	Patient samples should be provided at specified timepoints; Donor samples should NOT be provided at specified timepoints
Patient does NOT consent to provide optional immune reconstitution samples and Donor does NOT consent to provide optional immune reconstitution samples	No Patient or Donor samples should be provided at specified timepoints
Patient does NOT consent to provide optional immune reconstitution samples and Donor consents to provide optional immune reconstitution samples	No Patient or Donor samples should be provided at specified timepoints. *Note: if patient does not consent to provide samples, it is not required to approach the Donor with the optional research sample consent form.*

The blood tubes will be shipped on the day of collection to the Children's Research Institute project laboratory by Priority Overnight FedEx courier. Please see the protocol-specific Research Sample Information Guide for detailed sample labeling, GlobalTrace, and same-day sample shipping procedures

Table 2: Optional – Immune Reconstitution Correlative Study Research Samples					
Subjects	**Sample Type**	**Sample Collection Time Points**	**Sample Collection Summary**	**Shipping Specifications**	**Shipping and TestingLocation**
Patients	Peripheral Blood (sodium heparin) **40 mL** (adults) **24-40 mL** (pediatric age 5-14)	**Pre-HSCT Conditioning** Within 30 days prior to transplant conditioning (Thymoglobulin) **Post-transplant** Day 28, 60, 100, 180, 365	Collect the peripheral blood sample and place into four (3-4 for children) 10 mL Vacutainer tubes, containing sodium heparin anticoagulant. Gently mix sample by inversion 8-10 times to mix sample well with anticoagulant.	Blood sample tubes will be shipped at ambient temperature, on the day of collection, to project laboratory by priority overnight FedEx® delivery.	Children's Research Institute Laboratory
Haploidentical Donors	Peripheral Blood (sodium heparin) **20 mL**	Prior to Bone Marrow Collection	Collect the peripheral blood sample and place into two 10 mL Vacutainer tubes, containing sodium heparin anticoagulant. Gently mix sample by inversion 8-10 times to mix sample well with anticoagulant.	Blood sample tubes will be shipped at ambient temperature, on the day of collection, to project laboratory by priority overnight FedEx® delivery.	Children's Research Institute Laboratory

APPENDIX D

CONSENSUS CRITERIA FOR GRADING OF SEVERITY OF ACUTE GVHD

Appendix D
CONSENSUS CRITERIA FOR GRADING OF SEVERITY OF ACUTE GVHD

TABLE 1: ACUTE GVHD GRADING CRITERIA

Stage	Skin	Liver (Bilirubin)	GI Tract (Stool Output/d)
0	No GVHD rash	<2 mg/dL	Adult: <500 mL/d Child: <10 mL kg/d
1	Maculopapular rash <25% BSA	2–3 mg/dL	Adult: 500–999 mL/d Child: 10–19.9 mL/kg/d or persistent nausea, vomiting, or anorexia, with a positive upper GI biopsy
2	Maculopapular rash 25%–50% BSA	3.1–6 mg/dL	Adult: 1000–1500 mL/d Child: 20–30 mL/kg/d
3	Maculopapular rash >50% BSA	6.1–15 mg/dL	Adult: >1500 mL/d Child: >30 mL/kg/d
4	Generalized erythroderma with bullous formation	>15 mg/dL	Severe abdominal pain with or without ileus

Overall clinical grade: grade 0, no stage 1–4

of any organ; 1, stage 1–2 skin rash and no

liver or GI involvement;

2, stage 3 skin rash, or stage 1 liver involvement, or stage 1 GI involvement;

3, stage 0–3 skin rash, with stage 2–3 liver involvement, or stage 2–3

GI involvement; 4, stage 4 skin rash, liver, or GI involvement.

- Grade 0 indicates no clinical evidence of disease.
- Grade I indicates rash on less than 50% of skin and has no gut or liver involvement.
- Grade II indicates rash covering more than 50% of skin, bilirubin level of 2-3 mg/dL, diarrhea of 10-15 mL/kg/d, or persistent nausea.
- Grade III or IV indicates generalized erythroderma with bullous formation, bilirubin level of more than 3 mg/dL, or diarrhea of more than 16 mL/kg/d.

APPENDIX E

NIH CONSENSUS CRITERIA FOR DIAGNOSIS AND GRADING OF SEVERITY OF CHRONIC GVHD

Appendix E
NIH CONSENSUS CRITERIA FOR DIAGNOSIS AND GRADING OF SEVERITY OF CHRONIC GVHD

TABLE 1: SIGNS AND SYMPTOMS OF CHRONIC GVHD

Organ or site	Diagnostic (Sufficient to Establish the Diagnosis of Chronic GVHD)	Distinctive (Seen in Chronic GVHD, but Insufficient Alone to Establish a Diagnosis of Chronic GVHD)	Other features	Common (Seen with Both Acute and Chronic GVHD)
Skin	Poikiloderma	Depigmentation	Sweat impairment	Erythema
	Lichen planus-like features		Ichthyosis	Maculopapular rash
	Sclerotic features		Keratosis pilaris	Pruritus
	Morphea-like features		Hypopigmentation	
	Lichen sclerosus-like features		Hyperpigmentation	
Nails		Dystrophy		
		Longitudinal ridging, splitting, or brittle features		
		Onycholysis		
		Pterygium unguis		
		Nail loss (usually symmetric; affects most nails)†		
Scalp and body hair		New onset of scarring or nonscarring scalp alopecia (after recovery from chemoradiotherapy) Scaling, papulosquamous lesions	Thinning scalp hair, typically patchy, coarse, or dull (not explained by endocrine or other causes)	
			Premature gray hair	
Mouth	Lichen-type features	Xerostomia		Gingivitis

Organ or site	Diagnostic (Sufficient to Establish the Diagnosis of Chronic GVHD)	Distinctive (Seen in Chronic GVHD, but Insufficient Alone to Establish a Diagnosis of Chronic GVHD)	Other features	Common (Seen with Both Acute and Chronic GVHD)
	Hyperkeratotic plaques	Mucocele		Mucositis
	Restriction of mouth opening from sclerosis	Mucosal atrophy Pseudomembranes † Ulcers †		Erythema Pain
Eyes		New onset dry, gritty, or painful eyes‡	Photophobia	
		Cicatricial conjunctivitis Keratoconjunctivitis sicca ‡ Confluent areas of punctate keratopathy	Periorbital hyperpigmentation	
			Blepharitis (erythema of the eyelids with edema)	
Genitalia	Lichen planus-like features	Erosions†		
	Vaginal scarring or stenosis	Fissures†		
		Ulcers†		
GI tract	Esophageal web Strictures or stenosis in the upper to mid third of the esophagus †		Exocrine pancreatic insufficiency	Anorexia Nausea Vomiting Diarrhea
				Weight loss
				Failure to thrive (infants and children)
Liver				Total bilirubin, alkaline phosphatase >2 × upper limit of normal†

Organ or site	Diagnostic (Sufficient to Establish the Diagnosis of Chronic GVHD)	Distinctive (Seen in Chronic GVHD, but Insufficient Alone to Establish a Diagnosis of Chronic GVHD)	Other features	Common (Seen with Both Acute and Chronic GVHD)
				ALT or AST >2 × upper limit of normal
Lung	Bronchiolitis obliterans diagnosed with lung biopsy	Bronchiolitis obliterans diagnosed with PFTs and radiology‡		BOOP
Muscles, fascia, joints	Fasciitis Joint stiffness or contractures secondary to sclerosis	Myositis or Polymositis	Edema Muscle cramps Arthralgia or arthritis	
Hematopoietic and immune			Thrombocytopenia	
			Eosinophilia	
			Lymphopenia	
			Hypo- or hypergammaglobulinemia	
			Autoantibodies (AIHA and ITP)	
Other			Pericardial or pleural effusions	
			Ascites	
			Peripheral neuropathy	
			Nephrotic syndrome	
			Myasthenia gravis	
			Cardiac conduction abnormality or cardiomyopathy	

Can be acknowledged as part of the chronic GVHD symptomatology if the diagnosis is confirmed
† In all cases, infection, drug effects, malignancy, or other causes must be excluded
‡ Diagnosis of chronic GVHD requires biopsy or radiology confirmation (or Schirmer test for eyes).

The Working Group recommends that the diagnosis of chronic GVHD require at least 1 diagnostic manifestation of chronic GVHD or at least 1 distinctive manifestation, with the diagnosis confirmed by pertinent biopsy, laboratory tests, or radiology in the same or another organ. As in acute GVHD, infection and other causes may confound or complicate the differential diagnosis of chronic GVHD (e.g., nail dystrophies associated with onychomycosis, herpes simplex, or *Candida albicans* infections of the oral cavity; drug toxicity) and must be excluded. Diagnostic and distinctive manifestations of chronic GVHD can be found in the skin and appendages, mouth, eyes, female genitalia, esophagus, lungs, and connective tissues. Biopsy or other testing is always encouraged and often valuable to confirm the presence of chronic GVHD, but it is not always feasible and is not mandatory if the patient has at least 1 of the diagnostic findings of chronic GVHD (Table 1). Please note that an in-depth discussion of recommended terminology for histopathologic interpretation may be found in a forthcoming histopathology working group report. A biopsy read as "consistent with" or "unequivocal" chronic GVHD will be considered sufficient to establish the diagnosis of chronic GVHD if accompanied by at least 1 distinctive clinical manifestation.

Characteristics that establish the diagnosis of chronic GVHD might not serve as the most appropriate parameters for assessing the severity of chronic GVHD. Valid and reliable diagnostic criteria might not be sufficiently sensitive to change to be useful as treatment- response criteria. Conversely, a sensitive measure of chronic GVHD response might not necessarily serve as an appropriate diagnostic and scoring tool.

A. Organ-specific manifestations of chronic GVHD

In all cases, drug reaction, infection, recurrent or new malignancy, and other causes must be excluded. Diagnostic clinical or laboratory features sufficient for the diagnosis of chronic GVHD are italicized in the sections below.

B. Skin

Diagnostic manifestations include *poikiloderma* (e.g., atrophic and pigmentary changes), *lichen planus-like eruption* (e.g., erythematous/violaceous flat-topped papules or plaques with or without surface reticulations or a silvery or shinyappearance on direct light), *deep sclerotic features* (e.g., smooth, waxy, indurated skin—"thickened or tight skin," caused by deep and diffuse sclerosis over a wide area), *morphea-like* superficial scleroticfeatures (e.g., localized patchy areas of moveable smooth or shiny skin with a leathery-like consistency, often with dyspigmentation), or *lichen sclerosus-like lesions* (e.g., discrete to coalescent gray to white moveable papules or plaques, often with follicular plugs, with a shiny appearance and leathery consistency). Severe sclerotic features characterized by thickened, tight, and fragile skin are often associated with poor wound healing, inadequate lymphatic drainage, and skin ulcers from minor trauma.

A distinctive feature for chronic GVHD (not seen in acute GVHD, but not sufficiently unique to be considered diagnostic of chronic GVHD) is depigmentation. However, depigmentation would contribute to the diagnosis of chronic GVHD in combination with

biopsy or laboratory confirmation of GVHD in skin or another organ. Sweat impairment and intolerance to temperature change from loss of sweat glands are seen in chronic GVHD. Other common, nondistinctive skin manifestations found with both acute and chronic GVHD include erythema, maculopapular rash, and pruritus.

C. Nails

Dystrophy consisting of longitudinal ridging, nail splitting or brittleness, onycholysis, pterygium unguis, and nail loss (usually symmetric and affecting most nails) are distinctive signs of chronic GVHD but are not sufficient for diagnosis.

D. Hair

Distinctive features of chronic GVHD include new scarring and nonscarring scalp alopecia (after recovery from chemotherapy or radiotherapy) and loss of body hair. Other characteristics seen with chronic GVHD include premature graying, thinning, or brittleness, but these findings are not diagnostic.

E. Mouth

Diagnostic features of oral chronic GVHD include *lichen planus-like changes* (whitelines and lacy-appearing lesions of the buccal mucosa, tongue, palate, or lips), *hyperkeratotic plaques* (leukoplakia), or *decreased oral range of motion in patients with sclerotic features of skin GVHD*. Distinctive features of chronic GVHD include xerostomia (dryness), mucoceles, mucosal atrophy, pseudomembranes, and ulcers (infectious pathogens such as yeast or herpesvirus; secondary malignancy must be excluded). Manifestations common to both acute and chronic GVHD include gingivitis, mucositis, erythema, and pain.

F. Eyes

Distinctive manifestations of chronic GVHD include new onset of dry, gritty, or painful eyes; cicatricial conjunctivitis; keratoconjunctivitis sicca; and confluent areas of punctate keratopathy. Other features include photophobia, periorbital hyperpigmentation, difficulty in opening the eyes in the morning because of mucoid secretions, and blepharitis (erythema of the eye lids with edema). New ocular sicca documented by low Schirmer test values with a mean value of both eyes ≤5 mm at 5 minutes or a new onset of keratoconjunctivitis sicca by slit-lamp examination with mean values of 6 to 10 mm on the Schirmer test is sufficient for the diagnosis of chronic GVHD if accompanied by distinctive manifestations in at least 1 other organ.

G. Genitalia

Diagnostic features for the genitalia include *lichen planus-like features* and *vaginal scarring or stenosis* (often associated with oral GVHD).

H. Gastrointestinal Tract

Diagnostic features for the gastrointestinal (GI) tract include *esophageal web*, *stricture*, or *concentric rings* documented by endoscopy or barium contrast radiograph. Chronic GVHD may be associated with pancreatic exocrine insufficiency, which often improves with enzyme supplementation. Manifestations common to both acute and chronic GVHD (as well as other causes, such as drug side effects, motility disorders, infections, or malabsorption) include anorexia, nausea, vomiting, diarrhea, weight loss, and failure to thrive. Wasting syndrome may be a manifestation of chronic GVHD but is often multifactorial (e.g., decreased caloric intake, poor absorption, increased resting energy expenditures, and hypercatabolism). Endoscopic findings of mucosal edema and erythema or focal erosions with histologic changes of apoptotic epithelial cells and crypt cell dropout may be seen but are not considered diagnostic of chronic GVHD unless the patient also has distinctive features in a non-GI system. Patients with unresolved acute GVHD may have more severe intestinal mucosal lesions, including ulcers and mucosal sloughing.

I. Liver

Hepatic acute and chronic GVHD typically presents as cholestasis, with increased bilirubin or alkaline phosphatase, but it may also present as acute hepatitis. Because of many possible alternative diagnoses, liver biopsy is required to confirm GVHD involvement of the liver. Note that because of the histologic similarity between acute and chronic liver GVHD, the diagnosis of chronic GVHD cannot be made on the basis of liver biopsy alone but requires a distinctive manifestation in at least 1 other organ system.

J. Lungs

The only diagnostic manifestation of chronic GVHD is biopsy-proven *BO*. BO diagnosed via pulmonary function and radiologic testing requires at least 1 other distinctive manifestation in a separate organ system to establish the diagnosis of chronic GVHD. BO is characterized by the new onset of an obstructive lung defect. Clinical manifestations may include dyspnea on exertion, cough, or wheezing. Some patients may be asymptomatic early in the disease process. Pneumothorax, pneumomediastinum, and subcutaneous emphysema are rare and often represent advanced disease. Restrictive pulmonary function abnormalities secondary to advanced sclerosis of the chest wall are attributable to skin GVHD. BO is clinically diagnosed when all of the following criteria are met:

1. Forced expiratory volume in 1 second/forced vital capacity ratio <0.7 and forced expiratory volume in 1 second <75% of predicted.
2. Evidence of air trapping or small airway thickening or bronchiectasis on high- resolution chest computed tomography (with inspiratory and expiratory cuts), residual volume >120%, or pathologic confirmation of constrictive bronchiolitis.
3. Absence of infection in the respiratory tract, documented with investigations directed by clinical symptoms, such as radiologic studies (radiographs or computed tomographic scans) or microbiologic cultures (sinus aspiration, upper respiratory tract viral screen, sputum culture, or bronchoalveolar lavage).

4. BO-organizing pneumonia not due to infections may represent a manifestation of either acute or chronic GVHD and is considered a common feature.

K. Musculoskeletal System

Diagnostic features include *fascial involvement* often affecting the forearms or legs and often associated with sclerosis of the overlying skin and subcutaneous tissue. Fascial involvement may develop without overlying sclerotic changes of the skin and can result in *joint stiffness or contractures* when present near joints. *Fasciitis* is detected on examination by stiffness, a restricted range of motion (e.g., often decreased dorsal wrist flexion or inability to assume a Buddha prayer posture), edema of the extremities with or without erythema (early sign), peau d'orange (edematous skin with prominent pores resembling the surface of an orange), or *joint contractures* (late complications). Clinical myositis with tender muscles and increased muscle enzymes is a distinctive but nondiagnostic manifestation of chronic GVHD. Myositis may present as proximal myopathy, but this complication is rare and does not explain the frequent complaints of severe cramps. Evaluation of myositis involves electromyography and measurement of creatine phosphokinase or aldolase. Arthralgia and arthritis are uncommon and are occasionally associated with the presence of autoantibodies.

L. Hematopoietic and Immune Systems

Abnormalities are common in chronic GVHD but cannot be used to establish the diagnosis of chronic GVHD. Cytopenias may result from stromal damage or autoimmune processes. Lymphopenia (≤500/μL), eosinophilia (≥500/μL), hypogammaglobulinemia, or hypergammaglobulinemia may be present. Autoantibodies may develop with autoimmune hemolytic anemia and idiopathic thrombocytopenic purpura. Thrombocytopenia (<100 000/μL) at the time of chronic GVHD diagnosis has been associated with a poor prognosis.

M. Other Findings

Serositis (pericardial or pleural effusions or ascites), peripheral neuropathy, myasthenia gravis, nephrotic syndrome, and cardiac involvement have been attributed to chronic GVHD, but these manifestations are rare. For these manifestations, chronic GVHD is often a diagnosis of exclusion.

Differential diagnosis between acute and chronic GVHD

The Working Group recognized 2 main categories of GVHD, each with 2 subcategories (Table 2). The broad category of acute GVHD includes (1) classic acute GVHD (maculopapular rash, nausea, vomiting, anorexia, profuse diarrhea, ileus, or cholestatic hepatitis) occurring within 100 days after transplantation or DLI (without diagnostic or distinctive signs of chronic GVHD) and (2) persistent, recurrent, or late acute GVHD: features of classic acute GVHD without diagnostic or distinctive manifestations of chronic GVHD occurring beyond 100 days of transplantation or DLI (often seen after withdrawal of immune suppression). The broad category of chronic GVHD includes (1) classic chronic GVHD without features characteristic of acute GVHD and (2) an overlap syndrome in which features of chronic and acute GVHD appear together. In the absence

of histologic or clinical signs or symptoms of chronic GVHD, the persistence, recurrence, or new onset of characteristic skin, GI tract, or liver abnormalities should be classified as acute GVHD regardless of the time after transplantation. With appropriate stratification, patients with persistent, recurrent, or late acute GVHD or overlap syndrome can be included in clinical trials with patients who have chronic GVHD.

TABLE 2: CATEGORIES OF ACUTE AND CHRONIC GVHD

Category	Time of Symptoms after HCT or DLI	Presence of Acute GVHD Features	Presence of Chronic GVHD Features
Acute GVHD			
Classic acute GVHD	≤100 d	Yes	No
Persistent, recurrent, or late-onset acute GVHD	>100 d	Yes	No
Chronic GVHD			
Classic chronic GVHD	No time limit	No	Yes
Overlap syndrome	No time limit	Yes	Yes

GVHD indicates graft-versus-host disease; HCT, hematopoietic cell transplantation; DLI, Donor lymphocyte infusion

APPENDIX F
LANSKY/KARNOFSKY PERFORMANCE STATUS SCALES

Appendix F
LANSKY/KARNOFSKY PERFORMANCE STATUS SCALES

LANSKY SCALE FOR PARTICIPANTS < 15.00 YEARS

Percentage	
100	Fully Active
90	Minor restriction in physically strenuous play
80	Restricted in strenuous play, tires more easily, otherwise active
70	Both greater restrictions of, and less time spent in, active play
60	Ambulatory up to 50% of time, limited active play with assistance/supervision
50	Considerable assistance required for any active play; fully able to engage in quiet play
40	Able to initiate quiet activities
30	Needs considerable assistance for quiet activity
20	Limited to very passive activity initiated by others (e.g., TV)
10	Completely disabled, not even passive play
0	Dead

KARNOFSKY SCALE FOR PARTICIPANTS ≥ 15.00 YEARS

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

APPENDIX G

QUALITY OF LIFE ASSESSMENT PROMIS PAIN & FATIGUE DOMAINS AND ELECTRONIC PAIN DIARY

Appendix G
QUALITY OF LIFE ASSESSMENT PROMIS PAIN & FATIGUE DOMAINS

Using PROMIS modules regarding pain and fatigue as the standardized assessment tool, QoL measures will be administered to English- and Spanish-speaking patients in the adult stratum ***prior to the initiation of hydroxyurea, at 1 year post-transplant, and 2 years post-transplant*** to participants in each stratum. All QoL assessments will be completed during the routine clinic visit associated with the corresponding study time point. PROMIS modules should be administered on a single day ≥ 7 days and ≤ 28 days from initiating the 28 day electronic pain diary.

A. ELECTRONIC PAIN DIARY

Subjects who speak English and are 15 years or older will be asked to use a web-based diary to report pain. The questions are generally related to description of pain and the impact it has on different aspects of life. The pain diary will be completed twice a day for a period of 4 weeks prior to the transplant, at 1 year, and 2 years after the transplant. The pain diary can be accessed via the internet using your personal computer, tablet or smart phone. Subjects will receive automated reminders to report pain at least twice a day, which will be set based on their stated preference after joining the study. Additionally, study staff may follow up with the subject after multiple days of missed pain diary entries. The study will not provide any smartphones or electronic devices or provide any compensation for phone and internet plans/bills as part of this study. If patients are admitted to the hospital during one of these 4-week periods, information about pain and related information from the medical record. *The electronic diary is NOT a means of communicating with healthcare providers or for seeking healthcare attention. It is ONLY a reporting tool for this study. If subjects have any concerns, they MUST contact their healthcare provider or go to the Emergency Room. They must not wait for study staff to contact them.*

APPENDIX H

DERIVATION OF A SEQUENTIAL TEST STATISTIC FOR CENSORED EXPONENTIAL DATA

Appendix H
DERIVATION OF A SEQUENTIAL TEST STATISTIC FOR CENSORED EXPONENTIAL DATA

Background – The Sequential Probability Ratio Test

Let $f(.,\theta)$ be the density function for random variable X. According to Neyman and Pearson, the most powerful test of $H_0:\theta=\theta_o$ versus $H_1:\theta=\theta_1$ decides in favor of H_1 or H_0 if $L_n > c_\alpha$ or $L_n < c_\alpha$, respectively, where $L_n = \prod_i^n f(x_i;\theta_1)/f(x_i;\theta_0)$ is the likelihood ratio, and c_α is determined to have the size α. When the sample size is not fixed in advance, further improvement is possible by using Wald's Sequential Probability Ratio Test (SPRT). The SPRT continues to sample as long as $B < L_n < A$ for some constant $B < 1 < A$, stops sampling and decides in favor of H_1 as soon as $L_n > A$, and stops sampling and decides in favor of H_0 as soon as $L_n < B$.

The usual measures of performance of such a procedure are the error probabilities and β of rejecting H_0 when $\theta=\theta_0$, and of accepting H_0 when $\theta=\theta_1$, respectively, and the expected sample size $E(N \mid \theta_j) \equiv E_j(N)$. Wald and Wolfowitz showed that among all tests, sequential or not, for which $\Pr_0(\text{reject } H_0) \le \alpha$ and $\Pr_1(\text{reject } H_0) \le \beta$, and for which $E_j(N)$ are finite, j=0,1, the SPRT with error probabilities and β minimizes $E_0(N)$ and $E_1(N)$. If, in addition, the $x1, x2, \ldots$ are independent and identically distributed (i.i.d.) with density function $f(x,\theta)$, with monotone likelihood ratio in $\tau(x)$, then any SPRT for testing θ_0 against $\theta_1 (>\theta_0)$ has non-decreasing power function.

For the SPRT with error probabilities α and β, the SPRT boundaries are given approximately by $A = (1-\beta)/\alpha$ and $B = \beta/(1-\alpha)$. The operating characteristics of the SPRT are given by $O(\theta,\alpha,\beta,\theta_0,\theta_1) = (A^{h(\theta)}-1)/(A^{h(\theta)}-B^{h(\theta)})$ where $h(\theta)$ is the non-trivial solution to the equation $\int (f(x;\theta_1)/f(x,\theta_2))^{h(\theta)} f(x;\theta)dx = 1$.

The formula $E(N;\theta) = [[(1-O(\theta)]\log A + O(\theta)\log B]/E(z;\theta)$ provides the average sample number for an arbitrary θ. The sample size distribution is very highly skewed, $Var(N) \approx [E(N)]^2$. Thus we will consider a truncated test with maximum sample size of N_0 and simulate to obtain the operating characteristics of the test.

Derivation of the SPRT for Censored Exponential Survival Times

Suppose that we wish to construct a sequential test for the composite null hypothesis that the rate of overall mortality at an early time point t is less than or equal to p_0 versus the alternative hypothesis that it is greater than or equal to p_0. Let us assume that the survival times, $T_1, T_2, ..., T_n$, are i.i.d. with exponential density function $f(T,\theta) = \theta e^{-\theta T}$. Although an exponential model may not fit well for overall mortality, it usually provides a reasonable model over a short time frame for modeling toxicity, so in all discussion below we assume that exponential survival times are censored at time point t. In the exponential parameterization, a t-day survival rate of p_0 translates into a mean survival of μ_0=-t/ln(1-p_0) (rate parameter θ_0 = -ln(1-p_0)/t).

The SPRT is derived with reference to a simple null and alternative hypothesis for the rate parameter, in this case, $H_0 : \theta = \theta_o$ versus $H_1 : \theta = \theta_1$. The log-likelihood ratio for the exponential in the presence of censoring is $\log\prod_i^n f(x_i;\theta_1) - \log\prod_i^n f(x_i,\theta_0) = d(\log(\theta_1) - \log(\theta_0)) - (\theta_1 - \theta_0)\sum_i^n T_i$, where d is the number of events. The SPRT can be represented graphically when plotting the number of deaths (d) on the y axis against the total time on study $\sum_i^n T_i$ on the x axis. The continuation region in terms of d is bounded by two parallel lines given by

$$\left\lceil \frac{\log(B)}{(\log\theta_1 - \log\theta_0)} + \frac{(\theta_1 - \theta_0)}{(\log\theta_1 - \log\theta_0)} \sum_i^n T_i \right\rceil < d < \left\lceil \frac{\log(A)}{(\log\theta_1 - \log\theta_0)} + \frac{(\theta_1 - \theta_0)}{(\log\theta_1 - \log\theta_0)} \sum_i^n T_i \right\rceil$$

with common slope $(\theta_1 - \theta_0)/(\log\theta_1 - \log\theta_0)$, and intercepts $\log A/(\ln\theta_1 - \ln\theta_0)$ and $\log B/(\ln\theta_1 - \ln\theta_0)$, for the upper and lower bounds, respectively. For monitoring purposes, at an interim analysis calendar time point s, suppose that d(s) events have occurred and that the total time on study is $\sum T_i(s)$. The cumulative number of events d(s) is plotted on the y axis against the total time on study, $\sum_i^n T_i(s)$. When this graph crosses the upper boundary, the null

hypothesis is

rejected. In practice, monitoring will be scheduled monthly after the start of enrollment to the study.

A truncated version of the SPRT can be obtained by specifying a maximum sample size. We truncate the SPRT by declaring that if the test has failed to terminate after the maximum sample size, that the null hypothesis will be accepted. Since the probability that the untruncated SPRT would reject the null at the maximum sample size is negligible, it makes little difference how the final boundary value is selected, and this rule is chosen for simplicity. The operating characteristics of this proposed truncated SPRT for censored exponential data can be estimated by simulation.

APPENDIX I
PRES GUIDELINES

Appendix I
GUIDELINES FOR PREVENTION AND MANAGEMENT OF PRES

Mission Statement

To describe the pathophysiology, clinical diagnosis, prevention and management of Posterior reversible encephalopathy syndrome (PRES). PRES is a disorder of reversible subcortical vasogenic brain edema with a constellation of acute neurological symptoms and brain imaging findings of vasogenic edema predominantly involving the bilateral parieto-occipital regions. Patients with sickle cell disease are particularly susceptible to PRES.[71,72]

Pathophysiology

PRES results from endothelial injury related to abrupt blood pressure changes or direct effects of cytokines on the endothelium, which leads to the breakdown of the blood–brain barrier and subsequent brain edema. With early diagnosis and appropriate management, PRES is reversible, both radiographically and clinically, and generally has a favorable prognosis.

Clinical Presentation

Clinical presentation of PRES include[73,74,75,76,77,78] seizure (60–75%), encephalopathy (50–80%), headache (50%), visual disturbances (33%), focal neurological deficit (10–15%), and Status epilepticus (5–15%). Symptoms typically occur in the setting of renal failure, blood pressure fluctuations, cytotoxic drugs, autoimmune disorders, and pre-eclampsia or eclampsia.[76] Calcineurin inhibitor (CNI)-induced PRES may occur with elevated blood pressure within days to weeks of CNI initiation and typically occurs without elevated medication levels.[79] Clinical and radiographic recovery occur in 75–90% of patients with a mean time to full clinical recovery range of 2–8 days, although some patients can take several weeks to achieve full recovery.[79,80,81,82] Concomitant GVHD with the use of steroids is an important risk factor for PRES.[83]

Imaging

Brain imaging is useful to confirm the diagnosis of PRES and to exclude alternative diagnoses.[76,77,82,84] Although vasogenic edema can be visualized using non-contrast CT in some patients, brain MRI (particularly T2-weighted sequences such as fluid-attenuated inversion recovery [FLAIR]) is much more sensitive.[82,84] Brain imaging usually reveals vasogenic edema in the parieto-occipital regions of both cerebral hemispheres. The subcortical white matter is always affected, and the cortex is also often involved. The edema is usually asymmetric, but almost always bilateral. Three primary descriptive variations exist in approximately 70% of patients: a dominant parieto-occipital pattern, holo-hemispheric watershed pattern, and superior frontal sulcus pattern. Neither the pattern nor the severity of brain edema, is associated with the type or severity of clinical presentation.[76,82]

Differential Diagnosis

The symptoms and signs are non-specific, thus necessitating brain imaging with the primary intent to exclude alternative diagnoses. However, the diagnosis of PRES is not solely radiological; the clinical context and the judgment of the clinician are crucial to making the correct diagnosis. Differential diagnoses to be considered include infectious encephalitis, vasculitis, post-transplant lymphoproliferative disorder and progressive multifocal leucoencephalopathy.

Risk factors in HCT predisposing to PRES

Incidence of PRES in allo-HCT is 7-9% with greater risks with myeloablative than with non-myeloablative regimens (16% vs 3%). PRES commonly occurs in the first month following HCT and is associated with increased HLA mismatch and with acute GVHD. Low –level neurotoxicity such as tremors, anxiety, and psychiatric dysfunction has been observed in 10%-40% of patients receive CNIs. Cyclosporine can induce endothelial injury/dysfunction leading to enhanced vasoconstrictive effects, increased sympathetic activation, and coagulation effects. Blood levels of Cyclosporine do not appear to correlate with severe neurotoxicity or PRES. Immune challenge from the transplant such as rejection and GVHD, effects of chemotherapy, and sepsis may all contribute to the risk of PRES. Discontinuation or switch of CNIs usually results in clinical improvement.

Unique Risk for PRES in patients with SCD undergoing HCT

Patients with SCD have impaired dynamic cerebrovascular autoregulation with decreased ability to buffer the transfer of blood pressure surges to cerebral tissue[85] as well as reduced cerebrovascular reserve capacity or vasodilatory capacity.[86] This may place them at unique risk for developing PRES. PRES has been reported in patients with SCD following severe acute chest syndrome, blood transfusion, hyper-transfusion with rapid increase in hemoglobin, recent use ofcorticosteroids, hypomagnesaemia, and in the absence of any precipitating factors.[87,88,89,90,91]

TABLE I

Medians and 90th Percentiles of Systolic and Diastolic Blood Pressure for Sickle Cell Anemia Subjects in the Cooperative Study of Sickle Cell Disease (CSSCD)

Age (Years)		2–3	4–5	6–7	8–9	10–11	12–13	14–15	16–17	18–24	25–34	35–44
Females N		257	97	72	68	57	71	89	81	227	199	66
SYS	Median	90	95	96	96	104	106	110	110	110	110	110
BP	90th	100	110	110	110	110	118	120	122	122	125	130
DIA	Median	52	60	60	60	60	62	70	70	64	68	70
BP	90th	62	70	70	70	74	74	80	78	80	80	84
Males N		276	111	78	66	75	61	75	53	179	166	41
SYS	Median	90	95	100	100	100	110	108	112	112	114	110
BP	90th	104	110	108	116	112	120	120	128	130	130	132
DIA	Median	54	60	60	60	60	64	64	70	68	70	70
BP	90th	66	68	68	70	70	72	78	80	80	80	84

SYS BP = systolic blood pressure; DIA BP = diastolic blood pressure. **Pegelow, C et al Am J Med 102 p171-177)**

The prevalence of seizures in children with SCD is 10 times that of the general population.[92,93,94] The observation of neurological complications in 30% of patients including intracranial hemorrhages in 38% of those with a previous history of stroke in an early series of patients with SCD undergoing HCT led to the universal adoption of measures for the prevention of PRES.[71] Measures for prevention of PRES include extended duration of anticonvulsant prophylaxis, intensified antihypertensive management and aggressive platelet support.[71]

Prevention of PRES in HCT for SCD

Prevention of PRES requires careful attention to the following measures (Table 2):

a. **Control of Blood pressure.** Blood pressure in patients with SCD has been reported to be lower than published standards for age, sex, and race-matched controls.[95,96,97] Decreased survival has been observed for patients with SCD whose systolic or diastolic pressures were above the 90th percentile for HbSS subjects.[97] Pressures above the 90th percentile for HbSS may overlap levels considered normal in non-SCD patients. Blood pressures may be elevated with fluid infusions or use of medications such as corticosteroids or CNIs. Supportive care orders must indicate the importance of keeping BP within 10% above the median for age for HbSS patients as described by Pegelow et al (Table I) or the baseline BP for the patient, whichever is lower. Mean arterial pressure (MAP) should be maintained at <70mmHg. Close monitoring and aggressive management with anti-hypertensive agents will be required to prevent PRES.

b. **Maintenance of adequate platelet count**. Thrombocytopenia and coagulopathy may be associated with increased risk of PRES-related ICH.[71],[98] It is therefore recommended to keep platelet count> 50, 000/µL.[71]

c. **Maintenance of euvolemic state**.

Large fluid shifts should be avoided.[98] Close attention must be paid to fluid balance since fluid overload with weight gain associated with increased blood pressure and consequently, increased risk of PRES.

Table 2. Measures for Prevention of PRES

Measure	Action
Control of Blood pressure	Physician to be notified and PRN anti- hypertensives to be administered if systolic or diastolic BP exceeds 10% above median for age in HbSS patients or > 10% baseline for patient, whichever is lower.
Maintenance of adequate platelet count.	Transfuse to keep Platelets> 50, 000/µL
Maintenance of euvolemic state	Avoid rapid fluid shifts. Maintain weight as close to baseline as possible.
Maintenance of adequate magnesium level	Maintain serum magnesium level ≥ 1.8mg/dL (0.75mmol/L) when lab normal range (1.7–2.4 mg/dL or 0.7–1 mmol/L)
Prevention of seizures	Institute anticonvulsant therapy before Busulfan and continue through the duration of administration of any calcineurin inhibitor.

c. Maintenance of adequate level of serum Magnesium. Severe hypomagnesemia can present with clinical and radiological features similar to PRES.[99,100] Magnesium sulphate is considered the drug of choice in the treatment of PRES associated with Eclampsia.[101] It is therefore recommended that patients receive magnesium supplementation in order maintain patient Maintain serum magnesium level ≥ 1.5mg/dL (0.75mmol/L) or ≥ 1.8 mg/dLwhen lab normal range (1.7–

2.4 mg/dL or 0.7–1 mmol/L). Maintenance of mild hypermagnesemia with serum Magnesium 2- 3mg/dL may be advisable, but maybe difficult to achieve because of side effects of P.O Magnesium. Bioavailability of magnesium in the aspartate, citrate, lactate, diglycinate and chloride forms is higher than magnesium oxide or sulfate.[102,103,104,105,106,107]

e. Treatment and management of PRES

The management of PRES consists of eliminating the precipitating cause, control of blood pressure and the institution of comprehensive supportive measures (Table 3). If PRES is caused by a specific medication, this medication should be discontinued. Failure to do so can perpetuate the syndrome. Alternative immunosuppression might be considered, but corticosteroids should be avoided. If steroids are added, careful consideration must be given to aggressive management of blood pressure. While it is important to treat the hypertension, the initial goal in treating patients with severe hypertension is to reduce blood pressure by 25% within the first few hours.[3] Pronounced fluctuations of blood pressure should be avoided, and continuous infusions of intravenous drugs might be required. Excessive or rapid blood pressure reduction could provoke cerebral ischemia. Seizures are treated with antiepileptic medications. Other underlying disorders, such as sepsis, and flare-ups of autoimmune disorders, should be treated.

Table 3. Measures for Treatment and Management of PRES	
Measure	**Action**
Supportive care	Immediately admit to ICU and initiate management as below while evaluating cause for neurological manifestations. High index of suspicion and early diagnosis are key. Obtain MRI with FLAIR. Monitor neurological status closely.
Removal of drug precipitating PRES	Caution is advised in adding steroids because of the risk of hypertension and progression of PRES.
Control of Blood pressure	Institute intravenous medications to control Blood pressure. Labetalol is drug of choice. Decrease BP by 25% over 6 hours. Gradual reduction of BP thereafter.
Maintenance of adequate platelet count.	Transfuse to keep Platelets> 50, 000/µL
Maintenance of euvolemic state	Avoid rapid fluid shifts. Maintain weight as close to baseline as possible. Close attention to intake and output. Avoid hypertransfusion. Keep Hb<12gm/dL.
Maintenance of adequate magnesium level	Correct hypomagnesemia. Maintain serum magnesium level ≥ 1.8mg/dL (0.75mmol/L) when lab normal range (1.7–2.4 mg/dL or 0.7–1 mmol/L)
Table 3. Measures for Treatment and Management of PRES	

Measure	Action
Treatment of seizures	Institute anticonvulsant therapy immediately.
Management of concurrent illness	Treat sepsis, or fluid overload.
Rehabilitation	Consider early introduction of physical and occupational therapy as appropriate

f. Prognosis

Despite its name, PRES is not always fully reversible. Early diagnosis and prompt management is crucial since the most severe forms of the PRES result in death, or chronic neurological sequelae.[108] Severe neurological injury and death can be attributed to intracranial hemorrhage, posterior fossa edema with brainstem compression, acute hydrocephalus[109], or marked diffuse cerebral edema and increased global intracranial pressure. Persistent neurological sequelae are reported in 10–20% of patients with PRES.[72,73,74,75,76,77,78]

APPENDIX J
VENO-OCCLUSIVE DISEASE CRITERIA

Appendix J
VENO OCCLUSIVE DISEASE CRITERIA

Veno occlusive disease will be defined using either the Baltimore or modified Seattle criteria as detailed in table I.1 below. This table is adapted from Dignan FL et al. *Br J Haematol*.[110]

TABLE I1. MODIFIED SEATTLE CRITERIA AND BALTIMORE CRITERIA

Modified Seattle Criteria
Two of the following symptoms must be present within 20 days of transplant:
• Bilirubin > 34.2 µmol/L (2 mg/dL) • Hepatomegaly or right upper quadrant pain • Weight gain (>2% from pre-transplant weight)
Baltimore Criteria
Bilirubin > 34.2 µmol/L (2 mg/dL) within 21 days of transplant and 2 of the following symptoms:
• Hepatomegaly • Ascites • Weight gain (>5% from pre-transplant weight)

APPENDIX K

IPS CRITERIA

Appendix K
IDIOPATHIC PNEUMONIA SYNDROME (IPS) CRITERIA

IDIOPATHIC PNEUMONIA SYNDROME

I: Evidence of widespread alveolar injury:

a. Multilobar infiltrates on routine chest radiographs or computed tomography

b. Symptoms and signs of pneumonia (cough, dyspnea, tachypnea, rales)

c. Evidence of abnormal pulmonary physiology

1. Increased alveolar to arterial oxygen difference

2. New or increased restrictive pulmonary function

test abnormality II: Absence of active lower respiratory

tract infection based upon:

a. Bronchoalveolar lavage negative for significant bacterial pathogens including acid-fast bacilli, Nocardia, and Legionella species

b. Bronchoalveolar lavage negative for pathogenic nonbacterial microorganisms:

1. Routine culture for viruses and fungi

2. Shell vial culture for CMV and respiratory RSV

3. Cytology for CMV inclusions, fungi, and Pneumocystis jiroveci (carinii)

4. Direct fluorescence staining with antibodies against CMV, RSV, HSV, VZV, influenza virus, parainfluenza virus, adenovirus, and other organisms

c. Other organisms/tests to also consider:

1. Polymerase chain reaction for human metapneumovirus, rhinovirus, coronavirus, and HHV6

2. Polymerase chain reaction for Chlamydia, Mycoplasma, and Aspergillus species

3. Serum galactomannan ELISA for Aspergillus species

d. Transbronchial biopsy if condition of the patient permits

III: Absence of cardiac dysfunction, acute renal failure, or iatrogenic fluid overload as etiology for pulmonary dysfunction

APPENDIX L

HYDROXYUREA PERIOD DOSE MODIFICATIONS RECOMMENDATIONS

Appendix L
HYDROXYUREA PERIOD DOSE MODIFICATION RECOMMENDATIONS

Hydroxyurea (30 mg/kg/day) will be commenced as a single daily oral dose on Day -70 and continued through Day -10. Hydroxyurea will be adjusted to the ideal body weight (IBW) in children and adults weighing > 125% IBW. A CBC will be obtained weekly and the dose will be reduced if the ANC<1500 or platelet <100,000. HU will be discontinued on Day -10.

1. Patients with side effects to hydroxyurea such as headache, mild gastrointestinal symptoms, including abdominal discomfort or nausea, dermatologic changes, including skin hyperpigmentation or darkening of the nails (melanonychia), are sporadic and not dose-dependent.

   Recommendations: Patients with nausea or gastrointestinal complaints should be advised to take hydroxyurea at bedtime and using single dosing. If symptoms persist, suggest dose reduction by 2.5 -5 mg/kg/day decrements.

2. If a patient on protocol dose of hydroxyurea therapy is admitted or treated for an acute pain episode, hydroxyurea should not be held unless there is evidence of hematological toxicity. Table 1 list suggested hydroxyurea dose modifications based on published studies.

Table 1. Hematological toxicity thresholds requiring hydroxyurea dose modifications

Lab parameter	Value	Action[1]
Neutrophils	absolute neutrophil count (ANC) < 1.5 $\times 10^9$/L (1000/µL	Hold hydroxyurea
Hemoglobin (Hb)	Decline in Hb >20% from the baseline, or Hb <5.0 g/dL, with low reticulocytes	Hold hydroxyurea
Reticulocytes	$<80\times10^9$/L (80,000/µL) unless the hemoglobin concentration is >8.0 gm/dL	Hold hydroxyurea
Platelets	$<100\times10^9$/L (100,000/µL)	Hold hydroxyurea
Non- hematologic toxicity	Increase in serum creatinine by	Non- hematologic toxicity

[1] If toxicity is identified, hydroxyurea will be held, and when the laboratory abnormality resolved, hydroxyurea will be resumed at the previous dose. If toxicity lasted for more than 2 weeks or reoccurred after the medication was resumed, the dose was permanently decreased by 2.5 mg/kg/day.

a. In patients who develop unexpected serious medical complications or complicated vaso-occlusive pain episode (i.e., sickle cell pain episode complicated by acute chest syndrome, stroke, pneumonia, acute kidney injury, bacteremia/sepsis or splenic sequestration), hydroxyurea will be held and resumed at treating physician's discretion.
b. For adults prescribed hydroxyurea in 500 mg capsules, the hydroxyurea dose will be rounded up if dose calculation is between doses of 500 mg. If not tolerated, then the dose may be titrated down as indicated above[7].
c. Any modifications to the preparative regimen or drug stoppage should be reported to the BMT CTN DCC protocol coordinator for review and approval by the Protocol Chairs/Officer prior to continuation of the protocol.

APPENDIX M
REFERENCES

Appendix M
REFERENCES

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