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HLA Matched Unrelated or Non-genotype Identical Related Donor Transplantation for Chronic Granulomatous Disease

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ELIGIBILITY CHECKLIST

PATIENT ID_____   PATIENT NAME _____

The following must be answered "YES" for a patient to be eligible to participate in this study:

<u>YES</u>   <u>NO</u>

____   ____ Chronic Granulomatous Disease (CGD) patients as documented by an abnormal NBT assay in a male patient and/or abnormal NADPH enzyme mutation confirmed by genetic analysis with abnormal NBT.

____   ____ Patient must not have an HLA genotype identical donor.

____   ____ Patient has a 5/6 or 6/6 HLA matched unrelated donor or a 5/6 or 6/6 HLA phenotype matched related donor.

____   ____ Patient has had at least one serious infection characteristic of those manifested in patients with CGD.

____

____   ____ Patients must not have active infection. An active infection may include the following: 1) clinical findings consistent with an infection such as fever, cavitary organ lesions, osteomyelitis 2) progression of presumed infection based upon findings of diagnostic imaging [two or more studies at least 1 month a part]

____   ____ Patient has echocardiogram shortening fraction $\geq 28\%$.

____   ____ Patient has DLCO $\geq 50\%$ predicted or FEV1 $\geq 50\%$ predicted.

____   ____ Patient has no cumulative organ dysfunction that in the estimation of the treating physicians will diminish the patient's likelihood to survive this procedure.

____   ____ Negative pregnancy test for post-pubertal female patients.

The following must be answered NO for a patient to be eligible to enroll in this study:

_____   _____ Active or uncontrolled infection (e.g. lung infection, cavitary organ lesions, osteomyelitis).

_____   _____ Markedly elevated C reactive protein or sedimentation rate relative to patient's baseline.

_____   _____Invasive bone or bone marrow disease.

_____   _____Lack of potential hematologic blood product donors in the past (related to McLeod phenotype).

1. **Objectives**
   1.1.   PRIMARY: To estimate the engraftment rate for patients with CGD using busulfan, cyclophosphamide, fludarabine and Alemtuzumab (Campath 1H) as conditioning therapy for SCT from 5/6 or 6/6 HLA matched unrelated or 5/6 or 6/6 HLA phenotype matched related donors.
   1.2.   SECONDARY:
     1.2.1.   To estimate the likelihood of complete donor chimerism for patients with CGD using busulfan, cyclophosphamide, fludarabine and Alemtuzumab (Campath 1H) as conditioning therapy for SCT from 5/6 or 6/6 HLA matched unrelated or 5/6 or 6/6 HLA phenotype matched related donors.
     1.2.2.   To estimate the risk for acute GVHD and regimen related morbidity/mortality for patients with CGD following SCT from 5/6 or 6/6 HLA matched unrelated or 5/6 or 6/6 HLA phenotype matched related donors.
     1.2.3.   To estimate the risk for chronic GVHD and regimen related morbidity/mortality for patients with CGD following SCT from 5/6 or 6/6 HLA matched unrelated or 5/6 or 6/6 HLA phenotype matched related donors.
   1.3.   EXPLORATORY:
     1.3.1.   To explore the ability to treat patients with Chronic Granulomatous Disease (CGD) refractory to conventional therapy and without a HLA matched sibling donor by performing stem cell transplantation (SCT) from 5/6 or 6/6 HLA matched unrelated or 5/6 or 6/6 HLA phenotype matched related donors.
     1.3.2.   To examine the potential for reversal of organ toxicity (e.g. lung, liver, intestine) following engraftment and stable normal neutrophil function.

2. **Background**
   2.1.   Chronic Granulomatous Disease (CGD) is a life threatening primary immunodeficiency caused by the abnormal function of any of four components of the nicotinamide adenine dinucleotide phosphate (NADPH) oxidase system present in the phagocitic cells. The estimated incidence of CGD in the United States is between 1/200,000 – 1 /255,000 live births.(1)

   2.2.   The most common (accounting for 70% of CGD occurrences) and severe form of CGD is caused by an X-linked recessive mutation of the NADPH oxidase protein subunit gp91[phox]. This mutation as well as 3 additional mutations affecting other component proteins of the NADPH oxidase system result in marked susceptibility to life-threatening infections, primarily severe recurrent bacterial and fungal infections. These infections are the direct result of an inability of phagocytic leukocytes (e.g. neutrophils, eosinophils, monocytes), to generate superoxide and its metabolites, hydrogen peroxide and hypochlorous acid, necessary for intracellular killing and activation of anti-microbicidal enzymes.(2) CGD patients have exaggerated susceptibility to infection with catalase-positive pathogens (e.g. Staphylococcus aureus, Aspergillus species, Nocardia species, Candida species, Gram-negative enteric bacilli). These infections present primarily as pneumonia (75-80%), abscess formation (64-70%, [subcutaneous, liver, lung, perirectal, and brain]), adenitis (32-59%) or osteomyelitis (14 – 27%), among others.(1) Absent effective intracellular killing necessitates the prolonged use of antibiotics and ultimately the appearance of resistant strains. This

illustrates the complicated task of providing adequate antimicrobial prophylaxis and treatment to CGD patients.(3)

2.3.   In spite of our efforts, the morbidity and mortality for this patient population continues to be extremely high, primarily among patients with the X-linked form of the disease who appear to have a more severe clinical phenotype.   These patients often die in the first or second decade of life.   Morbidity and mortality often follow bouts of severe pneumonia with attendant complications (e.g. dissemination to chest wall, vertebrae and central nervous system).

2.4.   Continuous prophylactic antibiotic therapy with trimethropin-sulfamethoxasole or itraconazole is not sufficient to prevent recurrent infections in this patient population. In our experience, prophylactic administration of antifungal agents has failed to prevent infection. Once infected these agents are often inadequate to the task. As eradication of invasive fungal infections relies primarily on aggressive antibiotic therapy, CGD affected children repeatedly spend weeks or months in the hospital receiving therapy with multiple antibiotics.

2.5.   A landmark in the medical care of CGD patients was the introduction during the last decade of interferon gamma therapy as standard of care for this patient population. The mechanism by which interferon therapy decreases the number of infections is unclear but in spite of this therapy about 50% of children still experience severe recurrent infections. (1;4;5)

2.6.   Morbidity that accompanies prolonged hospitalizations and aggressive antibiotic therapy may be worsened by the need for surgical resection of necrotic and infected tissue. Granuloma of the lung or liver, abscess, empyema, osteomyelitis, etc. complicate the management of infections. Due to the frequency and severity of lung infections and the need for surgical intervention, patients are often left with severe pulmonary insufficiency, oxygen dependency or inability to participate in activities of daily life (e.g. school). In addition, this aggressive care can lead to irreparable secondary organ damage (e.g. renal failure, deafness, oxygen dependency, pancreatitis), further compromising the already poor quality of life, and perpetuating an endless need for chronic care.

3. **Stem Cell Transplantation – Rationale**
3.1.   Therapy for CGD has been largely symptomatic as noted above. While symptomatic treatment may adequately control specific infections, over time CGD patients invariably develop progressive tissue injury, such as to the lung or liver, from chronic infections. The treatment itself can be injurious, for example persistent administration of renal toxic antibiotics may compromise renal function. More than half of CGD patients die before the end of the third decade of life and for most of the others the quality of life is diminished by the chronic disease manifestations.(6)

3.2.   Neutrophil dysfunction that arises from the defect that underlies CGD can be compensated by granulocyte transfusions. These have been reported as life-saving in circumstances of severe infections, poorly responsive to drug therapy and

unapproachable surgically. Clearly the benefit of this approach is due to the administration of functioning neutrophils capable of effective microbial phagocytosis. Unfortunately, granulocyte transfusions are complicated by fever and respiratory distress, and with the invariable development of leukoagglutinins poor granulocyte recovery. White blood cell transfusions are never an effective maintenance strategy.(1)

3.3.   Based upon patterns of X-inactivation in X-linked carriers of CGD, both affected and unaffected neutrophils are produced. Unaffected or normal neutrophil production does not show a proliferative advantage over abnormal neutrophils. Typically, CGD carriers with as few as 10% normal phagocytes do not develop serious infections.(1)This suggests that susceptibility to infection in CGD can be prevented by a relatively small portion of normal cells admixed with a larger number of abnormal cells.

3.4.   Clinical manifestations of CGD are limited to the consequences of abnormal neutrophil function. Implicit therefore is that CGD patients should benefit substantially, and perhaps be cured, if normal neutrophil production can be established. This is the premise underlying the role for stem cell transplantation in CGD.(1)

3.5.   There are now adequate data to substantiate stem cell transplantation as best practice for CGD patients with HLA matched related donors.

3.5.1.   The European Group for Blood and Marrow Transplantation (EBMT) and the European Society for Immunodeficiencies (ESID) reported on the outcome for 27 patients with CGD undergoing allogeneic stem cell transplantation between 1985 and 2000.(7) Patients (23 males) ranged from 0.8 to 38.7 years of age, median age 7 years, at transplantation. All patients had at least one prior episode of invasive infection; 9 patients had therapy-refractory infection (n=8 fungus; n=1 mycobacterium) at transplantation. Of the remaining 18 patients, 7 evinced signs of active inflammation or organ sequelae from prior illness. Donors were HLA matched siblings for 25 patients (5/25 CGD carriers) and matched unrelated donors for 2 patients. Busulfan (16 – 20 mg/kg total dose) combined with cyclophosphamide, fludarabine, or melphalan comprised the conditioning regimen for 23 patients; 4 patients received less than fully myeloablative therapy. All patients received cyclosporine; some received either methotrexate or prednisone in addition, as GVHD preventive therapy.

Twenty-two/23 patients transplanted after myeloablative therapy engrafted, while 2/4 patients transplanted using submyeloablative therapy failed to engraft. The median time to granulocyte recovery was 18 days (9 to 40 days). For the 24 engrafted patients, NBT assay reached donor levels (5 pts transplanted from CGD carriers exhibited a mosaic pattern of NBT positivity).

Four/27 patients have died and deaths were exclusive to the group of 9 patients who underwent transplant with therapy-refractory infection (n=2 from progressive infection; n=1 carotid hemorrhage; n=1 inflammatory pulmonary

reaction and skin GVHD). It is noteworthy, however, among this same therapy-refractory cohort, 4 patients actively infected with Aspergillus survived and eradicated their infection after engraftment. Overall 81%, 22/27, patients are alive, 21/22 maintain complete donor myeloid chimerism, and all are without clinical or laboratory findings of CGD. Seven/27 patients experienced grade II-IV GVHD, and 3 patients have developed chronic GVHD. Two patients who received stem cells from HLA matched but unrelated donors (MUD) are alive and well.

Beyond the excellent survival and the ability to eradicate infection in therapy-refractory patients, improvements were documented in lung function (lessened restrictive pulmonary function, discontinued supplemental oxygen, resolution of clubbing), resolution of colitis, and increased growth rate.
3.5.2.   In a study conducted at the NIH, 10 patients with CGD underwent peripheral blood stem cell transplantation (PBSCT) from matched siblings.(8) All patients were male, from 5 – 36 y/o, median 15 years; CGD in 8 of these was caused by the X-linked mutation. Seeking to avoid transplant related morbidity/mortality, nonmyeloablative transplant conditioning therapy was employed: fludarabine 25 mg/m^2 days –5 to –1 and cyclophosphamide 60 mg/kg days –7 and -6. GVHD preventive therapy included cyclosporine and T-cell depletion. Donors underwent leukapharesis after 4 days G-CSF priming and were collected on days 5 and 6. Manipulation of these products included CD34+ selection using the Isolex 300i system and treatment with T-cell monoclonal antibodies (CD2, CD6, and CD7). Other manipulations included T-cell 'add-back' to provide the recipient with 1 X 10^5/kg CD3+ T-cells, and periodic infusions of donor T-cells post PBSCT to promote full donor engraftment.

As anticipated conditioning regimen related toxicity was negligible. Neutrophil recovery occurred at the median d+10 (range 6-22 days) and thrombocytopenia (< 20,000/mm^3) occurred in only 4 patients, resolving by d+11 at the latest. Unfortunately, 1 patient failed to engraft and died from complications related to a second transplant; and 1 patient developed late graft failure. At 17 months median follow up post PBSCT, 6/8 patients maintain complete (n=4) or almost complete myeloid and lymphoid chimerism, while 2 patients are stable mixed chimeras. Two of these 8 patients died, 1 from complications of GVHD (3 patients developed > grade II acute GVHD) and 1 from bacterial sepsis. It is noteworthy that the onset of acute GVHD followed donor T-cell infusions.

Although hematopoietic recovery ensued rapidly post PBSCT, complete donor chimerism, when it occurred, required weeks to months to establish. Furthermore it was necessary to administer multiple donor T-cell infusions to promote donor chimerism and for several patients this process instigated GVHD. Others have observed incomplete chimerism following subablative conditioning. Thus the benefit of this approach is not compelling when compared to the EBMT/ESID report.

3.5.3. While fewer patients with CGD have been transplanted from donors other than HLA matched siblings, reports indicate that the outcome is no less satisfactory.(7;9-11) Seven of eight such patients were reported to have engrafted following fully ablative conditioning, and all engrafted patients remained alive.

3.6. A number of reports find that the overall survival is comparable between matched sibling and matched unrelated donor transplants.(12;13)Comparison between patient groups is problematic; however our results for transplantation of children with aplastic anemia find the 4-year event free survival very similar for patients transplanted from matched sibling donors (92%) verus patients transplanted form matched unrelated donor (88%). There is no reason to believe that results should be inferior in the setting of matched unrelated donor transplantation for CGD.

3.7. Subablative or nonmyeloablative transplant conditioning therapy may permit more rapid hematopoietic recovery with lessened immediate transplant morbidity. This approach has been advantageous when transplantation has been performed in older patients or patients with significant pretransplant morbidity; engraftment has been satisfactory and is regularly accompanied by complete donor chimerism. However these results have been limited to patients with hematopoietic malignancy, for whom prior treatment has induced immunosuppression and reduced marrow reserve. These circumstances are not characteristic for patients with CGD. Indeed several reports document the failure to engraft or maintain donor engraftment for CGD patients treated with subablative conditioning regimens.(7;8)

3.8. Busulfan combined with cyclophosphamide has been used most widely and with considerable success to condition CGD patients for SCT.(7) The regimen that is proposed in this study, which adds fludarabine to busulfan and cyclophosphamide, has been used effectively to treat patients with a variety of malignant and nonmalignant diseases.(14)(personal communication Lawson, Amrolia) In the past, busulfan has presented problems related to unpredictable absorption. With the availability of an intravenous formulation of busulfan and the ability to assay plasma busulfan concentration, problems related to under or over dosing have been overcome.

3.9. Additionally, patients will be treated with Campath (Alemtuzumab) an antibody directed against the CD52 antigen, which is present on most lymphocytes. This agent has shown efficacy as an immunosuppressive promoting donor engraftment and prevention of GVHD.(15)

4. **Drug profiles** (see Appendix 1)
  4.1. Busulfan
  4.2. Fludarabine
  4.3. Cyclophosphamide
  4.4. Alemtuzumab
  4.5. Cyclosporine
  4.6. Methotrexate

5. **Eligibility**

5.1.   CGD patients as documented by an abnormal NBT assay in a male patient and/or abnormal NADPH enzyme mutation confirmed by genetic analysis with abnormal NBT.

5.2.   Patients must not have an HLA genotype identical donor

5.3.   Patients must have a 5/6 or 6/6 HLA matched unrelated donor or a 5/6 or 6/6 HLA phenotype matched related donor.

5.4.   Patients must have had at least one serious infection characteristic of those manifested in patients with CGD.

5.5.   Patients must not have active infection. An active infection may include the following: 1) clinical findings consistent with an infection such as fever, cavitary organ lesions, osteomyelitis 2) progression of presumed infection based upon findings of diagnostic imaging [two or more studies at least 1 month a part]

5.6.   No cumulative organ dysfunction that in the estimation of the treating physicians will diminish the patient's likelihood to survive this procedure.

5.7.   Negative pregnancy test for post-pubertal female patients.

5.8.   Echocardiogram shortening fraction $\geq$ 28%.

5.9.   DLCO $\geq$ 50% predicted or FEV1 $\geq$ 50% predicted.

6. Exclusion criteria

6.1.   Active or uncontrolled infection (e.g. lung infection, cavitary organ lesions, osteomyelitis).

6.2.   Markedly elevated C reactive protein or sedimentation rate relative to patient's baseline.

6.3.   Invasive bone or bone marrow disease.

6.4.   Lack of potential hematologic blood product donors in the past (related to McLeod phenotype).

7. Treatment Plan

Protocol day	Treatment
-10	Anti-Convulsant therapy begins
-9	Busulfan (see below regarding dosing)
-8	Busulfan
-7	Busulfan
-6	Busulfan
-5	Alemtuzumab (see below regarding dosing)
	Fludarabine 30 mg/m^2
	Cyclophosphamide 50mg/kg +MESNA
-4	Alemtuzumab
	Fludarabine 30 mg/m^2
	Cyclophosphamide 50mg/kg+MESNA
	Anti-Convulsant therapy ends
-3	Alemtuzumab
	Fludarabine 30 mg/m^2
	Cyclophosphamide 50 mg/kg + MESNA
-2	Alemtuzumab

	Fludarabine 30 mg/m²
	Cyclosporine
	Cyclophosphamide 50mg/kg +MESNA
-1	REST
0	Stem cell infusion
+1	Mtx
+3	Mtx
+6	Mtx
+11	Mtx

7.1.   Busulfan: The initial dose of busulfan is 1 mg/kg. Dosing is based upon actual weight unless actual weight exceeds ideal weight by 30% for which adjusted weight will be calculated as the ideal body weight plus 25%. Busulfan is administered intravenously every 6 hours for 16 doses. Blood samples will be obtained with the first and ninth dose and busulfan plasma levels will be determined. Using appropriate pharmacokinetic modeling, the busulfan AUC will be estimated. The dose of busulfan will be modified to maintain a busulfan AUC 900 – 1200 μmol/min/L.

7.2.   All patients require anticonvulsant therapy while receiving busulfan. The clinical pharmacist must review orders for anticonvulsants. Blood anticonvulsant levels should dictate dosing.

7.3.   Fludarabine: The dose of fludarabine is 30 mg/m².

7.4.   Cyclophosphamide: The planned dose of cyclophosphamide is 50 mg/kg but the dose will be based upon the ideal body weight or the actual body weight, whichever is less. For patients whose actual body weight exceeds the ideal body weight by more than 25%, dose adjustment will be required. These adjustments will be made in conjunction with the clinical pharmacist. Hemorrhagic cystitis may develop at this dose level of cyclophosphamide. Adequate urine flow is essential and the exact hydration and MESNA doses should be followed.

7.5.   Alemtuzumab: The dose of Alemtuzumab will be based upon recipient actual weight. Alemtuzumab 3 mg will be administered to recipients less than 15 kg; Alemtuzumab 5 mg to recipients > 15 kg to 30 kg; Alemtuzumab 10mg to recipients > 30 kg. Alemtuzumab is administered on four successive days. Prior to Alemtuzumab patients will receive a premedication as appropriate to prevent allergic reaction.

7.6.   Stem Cell: Either bone marrow, cord blood, or peripheral blood stem cells may be used for stem cell transplantation. It is desired to infuse: for bone marrow, nucleated cells $\geq$ 4 X 10^8/kg recipient weight; for cord blood $\geq$ 3 X 10^7/kg nucleated cells; for peripheral blood stem cells $\geq$ 1 X 10^7/kg CD34+ cells.

7.7.   Graft versus Host Disease Preventive Therapy: Cyclosporine will be administered beginning day -2. Initial dose will 5 mg/kg infused over 24 hours. Cyclosporine dose is based upon the actual body weight unless the actual body weight exceeds the ideal body weight by 30%; then the ideal body weight will be used. Modifications in cyclosporine dose will be determined by the measured blood concentration (cyclosporine levels will be maintained within the acceptable therapeutic range). For patients without GVHD, cyclosporine will be discontinued by 6 months. Methotrexate will be administered on day +1, +3, +6, and +11 at 5 mg/m² IV.

8. **Supportive care**

8.1. <u>Standard operating procedures</u> will be followed regarding the prevention or management of CMV infection, EBV infection, fever and neutropenia, fungus infection, GVHD therapy, blood transfusion support

9. **Studies to be obtained prior to transplantation**

9.1. Serologic panel: hepatitis A, B, C; HIV 1 & 2, HTLV 1 & 2, CMV, herpes simplex, herpes zoster, syphilis,

9.2. Diagnostic imaging studies as appropriate to evaluate sites of previous or current infection

9.3. ECHO and EKG

9.4. 24 hour creatinine clearance or glomerular filtration rate

9.5. Liver function tests: ALT, AST, alkaline phosphatase, GGT, total, direct and indirect bilirubin, albumin, triglycerides

9.6. CBC, sedimentation rate, prothrombin time, partial thromboplastin time, fibrinogen, blood type

9.7. Electrolytes, BUN, creatinine, magnesium, calcium, phosphorus, uric acid

9.8. Urinalyis

9.9. Amylase and lipase

9.10. Quantitative immunoglobulins

9.11. Pulmonary function tests where age appropriate

10. **Studies to be obtained day –8 to day +100**

10.1. At least weekly from day -8 to day +28 and thereafter as clinically indicated: CBC, AST, ALT, alkaline phosphatase, GGT, total and direct and indirect bilirubin, albumin, electrolytes, calcium, magnesium, phosphorus, uric acid, triglycerides, urinanalysis.

10.2. CMV antigenemia beginning day +14 and weekly after the ANC has reached 500 or as indicated until day +120 or cyclosporine and other GVHD therapy is discontinued. Thereafter CMV antigenemia test is performed as clinically indicated. CMV PCR may used in place of CMV antigenemia if ANC < 500 or as indicated.

10.3. Cyclosporine assay weekly until dose is tapered and then perform assay as indicated.

10.4. Chimerism estimation by FISH for sex chromosome or short tandem repeat (STR) for sex match between donor and recipient is to be initiated between day +10 and +21 and repeated as clinically indicated. Either blood or bone marrow are suitable for assay. Chimerism determination should be performed 4 weeks, 8 weeks, and 12 weeks post transplant and as clinically indicated.

10.5. NBT assay or other assay for neutrophil function will be performed at approximately day+28 and then as clinically indicated.

10.6. Beyond day +120 studies obtained are as clinically indicated.

11. **Evaluation of toxicity**

11.1. Definition of a Toxic Event. A toxic event is defined as grade III or grade IV toxicity by the NCI Common Toxicity Criteria Version 3.0 (http://ctep.info.nih.gov/reporting/ctc.html). Fever and hematologic toxicity are excluded as these are expected side effects of this therapy. Severe hematologic toxicity resulting from cytopenias will be managed in the same way as that which may result from conventional conditioning regimen.

12. **Risks and potential hazards**

12.1.   Nonengraftment. A major risk of this protocol is nonengraftment. Since the preparative regimen is ablative, we would anticipate the risk for non-engraftment would be 5%. This estimate is based upon extensive experience with the busulfan, cyclophosphamide, Alemtuzumab regiment used to treat patients with various hematolgic diseases. To reduce concern for the risks of engraftment failure, we will obtain, when possible, 'back-up' marrow from the patient for cryopreservation before conditioning is initiated.

12.2.   Incomplete engraftment. Even if engraftment occurs it may be incomplete, producing a state of mixed chimerism. Complete donor chimerism may become established progressively or it may be favored and accelerated by infusion of additional donor T lymphocytes. While mixed chimerism would likely be an acceptable outcome for treatment of the CGD, we will monitor chimerism and functional neutrophil assays to determine the impact of mixed chimerism on clinical course of CGD. Thus all patients will be monitored for engraftment at regular intervals post transplant. If chimerism is less than complete and the clinical course unsatisfactory, e.g. persistent or recurrent infections, patients will be afforded the option of Donor lymphocyte Infusion (DLI) infusion as 'per' the CAGT SOP, in which increasing doses of lymphocytes are administered at predetermined intervals if mixed chimerism persists. It is also possible that patients may also be given additional CD34+ selected donor stem cells.

12.3.   GVHD. Acute GVHD leads to a skin rash, liver dysfunction, and enteritis. GVHD prophylaxis in most types of patients reduces the incidence of grade II-IV GVHD to about 35% with commonly used regimens of today. Chronic GVHD is a multiorgan autoimmune disease usually requiring therapy with steroids and other immunosuppressives. It is seen in about 30% of adults undergoing transplant and a lower percentage of children. Chronic GVHD usually may end after several months or may lead to organ failure (liver failure is common) or infectious death. GVHD will be prevented in this study by the use of Alemtuzumab, which should persist over the transplant period to produce depletion of infused donor T, cells and by administration of cyclosporine and methotrexate. If GVHD occurs it will be treated by steroid administration. Patients whose GvHD is steroid resistant will be offered entry to Phase I/II protocols for treatment of steroid resistant GvHD.

12.4.   Additional Risks. The degree of immune suppression of marrow transplant patients leads to an increased risk of opportunistic infections, especially those due to CMV, HSV, EBV, Pneumocystis, and other viruses and fungi. Prophylaxis is given where possible (Acyclovir, Bactrim, antifungal and antibacterial mouth rinses, etc.), and treatment for suspected agents is initiated very promptly, usually before the infection is confirmed.

CGD clinical course may recur. The administration of blood products carries the risk of blood born infections such as hepatitis viruses, CMV, and HTLV-III. Blood products are screened for these agents but testing is not totally effective in excluding contaminated blood products.

Other complications of an unexpected nature may be seen. In fact, most every patient presents with some new or very rare complication during his transplant. Patients are made aware of this possibility.

Rarely, patients with CGD have experienced acute inflammatory reactions accompanied by severe skin rash at the time of donor engraftment. These reactions have been associated with administration of white cell growth factors (GCSF and GMCSF), and for this reason, these agents will not be used routinely to promote rapid engraftment.

13. **Statistical considerations**

13.1.   Patients with CGD who do not have a HLA genotype identical sibling donor are eligible for this nonrandomized phase II study to examine the efficacy of allogeneic stem cell transplantation from unrelated 5/6 or 6/6 HLA matched or phenotype 5/6 or 6/6 HLA matched donors. The preparatory regimen is busulfan, cyclophosphamide, fludarabine, and Alemtuzumab IH. We expect to enroll 10 to 15 patients over a period of 3 to 4 years. Safety monitoring will be conducted using Wald's sequential probability ratio test to monitor the rate of engraftment. The primary endpoint, or neutrophil engraftment is determined at or before day 28 and defined as the first day of 3 consecutive days for which the absolute neutrophil count exceeds 500/uL. Donor chimerism will be assessed for each patient at 120 days post transplant (+120). Complete donor is defined as ≥90% donor cells. Acute GVHD is assessed between day 0 and day 100 while chronic GVHD is assessed between day 100 and day 365. We expect 90% of the patients to engraft with an incidence of acute GVHD of 15%-20% grade I, and 5%-10% grade II or greater.

13.2.   The table, below, lists the total number of engraftment failures (including deaths or lost grafts prior to day +120) observed on day +120 necessary to trigger a suspension of accrual until a thorough reevaluation of the protocol has been completed. Additionally, if at any time two deaths prior to day +120 are observed accrual will be suspended and a thorough analysis conducted. These estimates provide a 10% chance of erroneously concluding that the true rate of engraftment is 90% or greater when in fact it is less than 80%. They also allow for a 10% chance of mistakenly concluding that the true engraftment rate is less than 80% when in fact it is at least 90%.

Engraftment Failure
Safety Monitoring Guidelines

Patients Entered (n)	Engraftment Failures
1	-
2	-
3	-
4	4
5	4
6	4
7	4
8	4
9	5
10	5
11	5
12	5
13	5
14	5
15	5

13.3.   GVHD will be monitored so that patient entry will be suspended until the study investigators have thoroughly reevaluated the protocol when either: (a) the sum of the overall grades assigned to each entered patient exceeds 21, or (b) two patients develop grade III or IV GVHD. Although the literature suggests the incidence of grade I or II GVHD to be 25%, it is not uncommon for all patients in a series to exhibit grade I GVHD.   By using a sum of 21 as the reevaluation trigger; a sample acceptable distribution of GVHD might include 10 grade I, 4 grade II, and 1 grade 3 GVHD patients. These criteria also monitor against unacceptable levels of extreme GVHD which exceed the expected 3% incidence.

13.4.   Kaplan-Meier estimates and associated confidence intervals of disease free survival, time to engraftment failure, and survival will be developed for all patients registered on the study.

13.5.   The other objective of the protocol, to evaluate the reversal of organ dysfunction is strictly observational in nature. The patient group is expected to exhibit a variety of organ dysfunctionality at the time of transplant making it nearly impossible to predict the types of dysfunction much less develop criteria for reversal of the dysfunction. Improvement can only be determined in relationship to the pre-transplant findings (e.g. lung function as assayed by pulmonary function tests). Results will be reported descriptively, and no statistical analyses will be undertaken.

13.6.   One year post transplant (day 365) should be considered the off-study date.

14. **Data Collection and Adverse Event Reporting:**

14.1.   Any questions regarding patients on this study should be addressed to Dr. Krance (Ph. 832-824-4693).

14.2.   Register all patients with the research coordinator at 832-824-4881.

14.3.   Drug Toxicity and/or adverse reactions
     Adverse events will be collected according to SOP J02.05.XX, J02.06.XX and J02.78.XX.

14.4   Relapse and survival data will be collected until 1 year post transplant.

14.5   Data Collection:
     The following forms will be used to collect data
          Prestudy
          Adverse events
          Response
          Relapse / progression
          Off study
          Death.

15. **Informed consent**
15.1.   All patents and/or their legal guardian must sign a document of informed consent consistent with local institutional and Federal guidelines stating that they are aware of the investigational nature of this protocol and of the possible side effects of treatment. Further, patients must be informed that no efficacy of this therapy is guaranteed, and that unforeseen toxicities may occur. Patients have the right to withdraw from this protocol at any time. No patient will be accepted for treatment without such a document signed by him or his legal guardian. Full confidentiality of patients and patient records will be provided according to institutional guidelines. The costs associated with the transplant will be billed to the third party payors. The patients or their families will not receive any payment for participation in this study.

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Appendix 1
Drug Profiles

FLUDARABINE

Therapeutic Classification: Purine antimetabolite.

Pharmaceutical Data: In vials of 50 mg.

Solution Preparation: Reconstituted with 2 ml of sterile water for injection. The resulting solution will contain 25 mg/ml. For infusion (maximum concentration of 10mg/ml) intravenously in 100 ml of standard intravenous piggy back fluid (dextrose 5%, or normal saline) over 30 minutes.

Stability and Storage Requirements: Prior to mixing: Store under refrigeration 2 to 8 degrees Celsius. After mixing: Stable for 16 days at room temperature, but needs to be used in 8 hours because of the lack of antibacterial preservatives.

Routes of Administration: IV infusion.

Usual Dosage Range: 25-30 mg/m^2 in a single dose each day for 5 days every 4 weeks.

Side effects: Myelosuppression, exacerbation of hemolytic anemia Prolonged immunosuppression, opportunistic infection and rare Neurotoxicity.

Special Precautions: Increased myelosuppression in patients with Creatinine clearances of less than 50ml/min.

Mechanism of Action: A purine antimetabolite modified with fluorine and monophosphate to resist deamination by adenosine deaminase and to increased solubility. Dephosphorylation followed by cellular incorporation and conversion to active triphospahte, which is a competitive inhibitor of DNA synthesis.

Antitumor Data: The drug has greater activity to T-cells than B-cells, but clinical activity is observed in B-cell malignancies.

Human Pharmacology: Fludarabine can only be given via intravenous route. Renal excretion accounts for 23%. Half-life is 10 hours.

BUSULFAN

Therapeutic Classification: Bifunctional alkylating agent

Pharmaceutical Data: Busulfan (Busulfex Injection® Orphan Medical) is supplied as a sterile solution in single-use ampules containing 60 mg at a concentration of 6mg/ml. It is provided as a mixture of demethylacetamide (DMA) and polyethylene glycol 400 (PEG400).

Solution Preparation: Busulfan solution for injection must be diluted with either 0.9% Sodium Chloride Injection (NS) or 5% Dextrose Injection (D5W) prior to administration. The diluent quantity must be 10 times the *volume* of busulfan, ensuring that the final concentration is $\geq$ 0.5 mg/ml. Sample calculation for a 50 kg patient: (50 kg) x (0.8 mg/kg of busulfan) = 40 mg = 6.7 ml. 6.7 ml of busulfan + 67 ml of NS = 74 ml total volume. Final concentration: 0.54 mg/ml.

Stability and Storage Requirements: After dilution with NS or D5W, busulfan is stable at room temperature (25 degrees Celsius) for 8 hours. The infusion must be completed within that time. Prior to mixing: Store under refrigeration (2 to 8 degrees Celsius). Busulfan for injection is stable at 4 ° for at least 12 months.

Route of Administration: Busulfan should be administered intravenously via a central venous catheter as a two-hour infusion.

Usual Dosage Range: 0.8-1 mg/kg/dose given every 6 hours for a total of 16 doses. For patients less than 4 years of age a dose of 1 mg/kg/dose will be used and for patients > 4 years the starting dose will be 0.8 mg/kg/dose. Doses are based on actual body weight, unless the patient's weight is greater than 30% of ideal body weight, then dosing will be based on adjusted weight of ideal plus 25%. Busulfan pharmacokinetics will be performed on all patients with dose adjustment as appropriate.

Pharmacokinetics: Doses will be adjusted to achieve the desired plasma area under the curve (AUC) of 900 – 1200 ☐mo⫲min/L. Doses will be adjusted as necessary pending the results of the first dose pharmacokinetics. For patients whose AUC

values are greater than 5% outside the acceptable AUC range, the dose will be adjusted to achieve a target AUC of 1125 $\mu mol \cdot min/L$ (midpoint of acceptable range) not to exceed a maximum dose of 1.6 mg/kg per dose of busulfan.

Side effects: Myelosuppression, neurotoxicity (manifesting as seizures), mild to moderate nausea and vomiting, mild to moderate tachycardia, skin hyperpigmentation, sterility, and rarely hepatotoxicity (hepatic veno-occlusive disease) and pulmonary toxicity (interstitial fibrosis).

Special Precautions: Increased toxicity in obese patients unless dose is adjusted appropriately. Generalized seizures have been reported after use of high dose busulfan. All patients will be treated with phenytoin 5 mg/kg/dose (IV or PO) q 6 hr beginning on day -10 for 24 hr until completion of busulfan. A trough phenytoin level should be obtained 6 hours after the fourth dose. The therapeutic range for this drug is 10-20 μg/ml. If the level is below 10 μg/ml, two additional doses of 5 mg/kg/dose should be administered q 6 hours. If the level is below 5 μg/ml, an additional four doses of phenytoin should be administered q 6 hours over the next 24 hours. If the phenytoin level is within the therapeutic range, maintenance therapy should begin at a dose of 5 mg/kg/day (IV or p.o.) in two divided doses until day -4. Patients experiencing seizure activity should be evaluated neurologically and treated as clinically indicated.

Mechanism of Action: Busulfan is a bifunctional alkylating agent in which two labile methanesulfonate groups are attached to opposite ends of a four-carbon alkyl chain. In aqueous media, busulfan hydrolyzes to release the methanesulfonate groups. This produces reactive carbonium ions that can alkylate DNA. DNA damage is thought to be responsible for much of the cytotoxicity of busulfan.

Human Pharmacology: Busulfan can be administered orally or intravenously. Busulfan achieves levels in cerebrospinal fluid similar to plasma levels. Busulfan is predominantly metabolized by conjugation with glutathione, both spontaneously and by glutathione S-transferase (GST) catalysis. This conjugate undergoes further extensive oxidative metabolism in the liver. Approximately 30% of busulfan and metabolites can be recovered in the urine within 48 hours after administration.

CAMPATH 1H

Source and Pharmacology: CAMPATH-1H is a humanized antilymphocyte monoclonal antibody. The Campath-1 antigen in humans (CD52) is predominantly expressed on peripheral blood lymphocytes, monocytes, and macrophages. CAMPATH-1H causes lysis of lymphocytes by fixing to CD52, a highly expressed, non-modulating antigen on the surface of lymphocytes. It mediates the lysis of lymphocytes via complement and antibody dependent cell mediated cytotoxicity mechanisms. CAMPATH-1H rapidly reaches a peak concentration after IV administration. The half-life has been thought to be approximately 60 hours, although recent data suggest a more prolonged persistence. The route of elimination is not known, but is probably via uptake and metabolism by the reticulo-endothelial system of the liver and spleen.

Dose: The dose has been chosen based on the plasma concentrations required for activity, and on the previous extensive published human experience using this dose for lymphocyte depletion

Formulation and Stability: CAMPATH-1H is supplied as a purified preparation diluted in phosphate buffered saline (PBS) with 0.05 mmol EDTA. Tween 80 is added to a concentration of 0.01%. The final product is clear, colorless isotonic solution free of visible particulate matter. The glass ampules will contain 30 mg of antibody in 3 mL of strile PBS at a concentration of 10 mg/mL. It should be stored, protected from light, in a refrigerator at between 2° and 8° C. After dilution in D5W or NS, the resulting solution is stable for 24 hours at room temperature. However, the product contains no preservative and must therefore be used within 8 hours. Intravenous CAMPATH-1H will be diluted in 100cc of 0.9% normal saline or 5% Dextrose and

administered intravenously over 2 hours. CAMPATH-1H must be filtered with a sterile, low-protein binding, 5 m filter prior to dilution.

Supplier: Investigational. L&I Partners, LP

Toxicity: CAMPATH-1H is a potent lymphocyte depleting agent. WHO Grade 3 and 4 neutropenia and thrombocytopenia emerge on treatment in approximately 10-20% of patients. Infections resulting from CAMPATH-1H induced immunosuppression are the major type of adverse event occurring outside the CAMPATH-1H administration period. The most common infections being mucocutaneous herpes simplex and candidiasis.

The majority of other adverse events seen in CAMPATH-1H trials can be categorized as being administration-related and of short duration. There is usually a first dose effect consisting of cytokine release type phenomena including hypotension, rigors, fever, shortness of breath, chills, rashes, etc. These side effects can be significantly ameliorated or avoided by premedicating the patients with methylprednisolone or hydrocortisone.

Route of Administration: Intravenous

Methotrexate

Therapeutic classification: An antimetabolite and antifolate agent with antineoplastic and Immunosuppressant activities.

Pharmaceutical Data: Methotrexate injection is a sterile isotonic liquid, preservative free, supplied in a single dose vial containing 25 mg/mL of methotrexate in the following packaging strengths: 2 mL (50 mg), 4 mL (100mg), 8 mL (200 mg), and 10 mL(250 mg). Methotrexate injection, preservative free, is also supplied in a sterile 1 gram single dose vial of lyophilized powder containing 1 gram methotrexate as the base.

Solution Preparation: Single dose vial containing 25 mg/mL: If desired the solution may be further diluted immediately prior to use with an appropriate sterile preservative free medium such as 5% dextrose solution or sodium chloride injection.
Single dose vial of 1 gram lyophilized powder: Reconstitution should be with an appropriate sterile preservative free medium such as 5% dextrose solution, or sodium chloride injection. The 1 gram vial should be reconstituted with 19.4 mL to a concentration of 50 mg/mL. When high doses of methotrexate are administered by IV infusion, the total dose is diluted in 5% dextrose solution.

Stability and Storage: Store at room temperature 20°-25°C (68°-77°F); protect from light. Use immediately after diluting or reconstituting drug.

Route of Administration: Intravenous (IV).

Usual dosage range: Methotrexate may be given by IV on Days 1, 3, 6 and 11. Dosing is either 5mg/m2/day for all four doses or 15mg/m2 on day 1 and 10mg/m2 for three remaining doses. Consider reducing dose by 50% or holding dose for Grade 3 mucositis or weight gain > 5 kg (>5% baseline for children); consider leucovorin rescue. Hold dose for Grade 4 mucositis, doubling of serum creatinine from baseline, or weight gain >10 kg (>10% baseline for children) or as clinically indicated.

Side Effects: Methotrexate is a medication used to try to prevent GVHD. It causes damage to cells, and therefore can affect many different tissues of your body. It may cause or can worsen the mouth sores or inflammation of the mouth. Methotrexate may slow down the recovery of blood cells after transplantation. Methotrexate can cause kidney damage or worsen kidney function. If kidney damage does occur, the methotrexate dose may be reduced or the medication may not be given at all. Methotrexate can interfere with the body's defense system (the immune system) causing more infections (especially viral infections and pneumonia) for several months after stem cell transplant.

Mechanism of action: Methotrexate binds to and inhibits the enzyme dihydrofolate reductase, resulting in inhibition of purine nucleotide and thymidylate synthesis and, subsequently, inhibition of DNA and RNA syntheses. Methotrexate also

exhibits potent immunosuppressant activity although the mechanism(s) of actions is unclear.

CYCLOPHOSPHAMIDE (Cytoxan)

Source and Pharmacology: Cyclophosphamide is an alkylating agent related to nitrogen mustard.

Cyclophosphamide is inactive until it is metabolized by P-450 isoenzymes (CYP2B6, CYP2C9 and CYP3A4) in the liver to active compounds. The initial product is 4-hydroxycyclophosphamide (4-HC) which is in equilibrium with aldophosphamide which spontaneously releases acrolein to produce phosphoramide mustard. Phosphoramide mustard, which is an active bifunctional alkylating species, is 10 times more potent in vitro than is 4-HC and has been shown to produce interstrand DNA cross-link analogous to those produced by mechlorethamine. Approximately 70% of a dose of cyclophosphamide is excreted in the urine as the inactive carboxyphosphamide and 5-25% as unchanged drug. The plasma halflife ranges from 4.1 to 16 hours after IV administration.

Toxicity:

	Common Happens to 21-100 children out of every 100	Occasional Happens to 5-20 children out of every 100	Rare Happens to < 5 children out of every 100
Immediate: Within 1-2 days of receiving drug	Anorexia, nausea & vomiting (acute and delayed)	Abdominal discomfort, Diarrhea	Transient blurred vision, nasal stuffiness with rapid administration, arrhythmias (rapid infusion), skin rash, anaphylaxis, SIADH
Prompt: Within 2-3 weeks, prior to the next course	Leukopenia, alopecia, Immune suppression	Thrombocytopenia, Anemia, Hemorrhagic cystitis (L),	Cardiac toxicity with high dose (acute — CHF hemorrhagic myocarditis, myocardial necrosis) (L), hyperpigmentation, nail changes, impaired wound healing, infection secondary to immune suppression
Delayed: Any time later during therapy, excluding the above conditions	Gonadal dysfunction: azoospermia or oligospermia (prolonged or permanent)[1] (L)	Amenorrhea[1]	Gonadal dysfunction : ovarian failure[1] (L) Interstitial pneumonitis, pulmonary fibrosis[2](L),
Late: Any time after completion of treatment			Secondary malignancy (ALL, ANLL, AML), bladder carcinoma (long term use > 2 years), bladder fibrosis
Unknown Frequency and Timing:	Fetal toxicities and teratogenic effects of cyclophosphamide (alone or in combination with other antineoplastic agents) have been noted in humans. Toxicities include: chromosomal abnormalities, multiple anomalies, pancytopenia, and low birth weight. Cyclophosphamide is excreted into breast milk. Cyclophosphamide is contraindicated during breast feeding because of reported cases of neutropenia in breast fed infants and the potential for serious adverse effects.		

[1] *Dependent on dose, age, gender and degree of pubertal development at time of treatment*
[2] *Risk increased with pulmonary irradiation and higher doses.*
(L) Toxicity may also occur later.

Formulation and Stability:

Cyclophosphamide for Injection is available as powder for injection or lyophilized powder for injection in 500 mg, 1 gram and 2 gram vials. The powder for injection contains 82 mg sodium bicarbonate/100 mg cyclophosphamide and the lyophilized powder for injection contains 75 mg mannitol/100 mg cyclophosphamide. Storage at or below 25ºC (77ºF) is recommended. The product will withstand brief exposures to temperatures up to 30º C (86ºF).

Guidelines for Administration:

Cyclophosphamide for Injection: Reconstitute with sterile water or Bacteriostatic water for injection (paraben preserved only) to a concentration of 20 mg/ml. Solutions reconstituted with preservative should be used within 24 hours if stored at room temperature or within 6 days if stored under refrigeration. If administered as undiluted drug at the 20 mg/ml concentration, reconstitute with normal saline (NS) only to avoid a hypotonic solution.

Cyclophosphamide may be further diluted in dextrose or saline containing solutions for IV use.

Supplier: Commercially available from various manufacturers.

CYCLOSPORINE A (cyclosporine, CYA, Sandimmune®, Neoral®, Gengraf®):

Source and Pharmacology: Cyclosporine (CSA) is a lipophilic fungal peptide consisting of 11 amino acids. CSA is a potent immunosuppressive agent which prolongs survival of allogeneic transplants involving skin, heart, kidney, pancreas, bone marrow, small intestine, and lung. Current evidence suggests that cyclosporine selectively inhibits the transcription of IL-2; the action of which stimulates the proliferation of activated T-lymphocytes. CSA has been shown in vitro to be a potent inhibitor of P-glycoprotein, which has been postulated to be a factor in multi-drug resistance to various antineoplastic agents. The terminal half-life of CSA is approximately 19 hours (range 10-27 hours). Ninety-nine percent of CSA is metabolized. Elimination is primarily biliary with approximately 6% excreted in the urine. In the circulation, CSA is mainly bound to high, low, or very low density lipoproteins and to chylomicrons. Only a small fraction circulates unbound. The volume of distribution varies from 3.5 L/kg to 13 L/kg with higher concentrations of drug found in the liver, lymphocytes, kidney, heart, lung, pancreas, fat, neural and muscle cells. CSA clearance rates have been shown to be higher in pediatric patients and for patients < 25 years old.

The absorption of cyclosporine from the gastrointestinal tract is incomplete and variable, exhibiting large intra- and inter-patient variability. Drugs that stimulate or inhibit hepatic p-450 enzymes will alter clearance of CSA and close attention to potential drug interactions is crucial.

Toxicity:

	Common Happens to 21-100 children out of every 100	Occasional Happens to 5-20 children out of every 100	Rare Happens to <5 children out of every 100
Immediate: Within 1-2 days of receiving drug	Hypertension (L), immunosuppression (L)	Headache (L), nausea and vomiting, diarrhea	Anaphylaxis, angioedema, seizure (L), arrhythmia, chest pain, fever, facial flushing
Prompt: Within 2-3 weeks, prior to the next course	Tremor (L), renal dysfunction (acute with decrease in GFR, impaired urinary concentrating ability, and sodium retention)	Hypomagnesemia (L), hyperlipidemia (L)	Confusion (L), somnolence (L), insomnia, depression (L), anxiety, dizziness, rash, urticaria, acne, hyperkalemia, encephalopathy, hemolytic-uremic syndrome, cardiac failure, MI, leukopenia (L), anemia, thrombocytopenia, increased creatinine, infections, hyperbilirubinemia (L), hepatic insufficiency
Delayed: Any time later during therapy, excluding the above conditions	Hirsutism (L)	Gingival hyperplasia (L)	Tinnitus, vestibular disorder, cholelithiasis, cataracts (L), gynecomastia, chronic renal dysfunction
Late: Any time after completion of treatment			Lymphoproliferative disorders, skin malignancies
Unknown Frequency and Timing:	Fetal toxic effects of cyclosporine have been noted in animals. Of the reported outcomes of 116 pregnancies in women receiving cyclosporine during pregnancy, 90% of whom were transplant patients, and most of whom received cyclosporine throughout the entire gestational period, the only consistent patterns of abnormality were premature birth (gestational period of 28 to 36 weeks) and low birth weight for gestational age. Cyclosporine is excreted in human milk, nursing should be avoided.		

(L) Toxicity may also occur later.

Formulation and Stability:
IV formulation:
Cyclosporine (Sandimmune®) is available as a (50 mg/mL) 5mL ampule use containing 650 mg polyoxyethylated castor oil (cremophor) and 32.9% alcohol. Store at temperatures below 30°C (86°F) and protected from light.

Oral formulations:
Cyclosporine (Sandimmune®) 25 mg, and 100 mg capsule, 100 mg/mL oral solution. Inactive ingredients include: Sandimmune® Capsule = Dehydrated alcohol, sorbitol, glycerol, corn oil, gelatin, polyoxyethylated glycolysed glycerides; Sandimmune® Solution = Olive oil, dehydrated alcohol, polyoxyethylated glycolysed glycerides.

Cyclosporine USP modified (microemulsion) (Neoral®, Gengraf®) 25 mg, 100 mg capsule, 100 mg/mL oral solution. Inactive ingredients include: Neoral® capsule and solution = dehydrated alcohol, corn oil, polyoxyl 40 hydrogenated castor oil, tocopherol, gelatin, and propylene glycol. Gengraf® Capsule and solution = dehydrated alcohol, gelatin, polyethylene glycol, polyoxyl 35 castor oil, polysorbate 80, propylene glycol, and sorbitan monooleate.
Store capsules in the original unit-dose container at controlled room temperature 15°-30°C (59°-86°F).

Store Oral solutions in the original container at controlled room temperature 68°-77°F (20°-25°C). Do not store in the refrigerator. Once opened, the contents must be used within two months. At temperatures below 68°F (20°C) the solution may gel; light flocculation or the formation of a light sediment may also occur. There is no impact on product performance or dosing using the syringe provided. Allow to warm to room temperature 77°F (25°C) to reverse these changes.
NOTE: Sandimmune®, Neoral® and Gengraf® ARE NOT BIOEQUIVALENT. Liquid formulations of each trade name are equivalent to capsules of that same trade name. Conversion from one trade name product to another is generally done at a 1:1 ratio, but requires close monitoring. Conversions from IV to PO are usually done at a 1:3 ratio, but should be monitored closely. Adjusting emulsion products to the same trough concentration as other oral products results in greater total exposure to the drug.

Dilute I.V. concentrate 1 mL (50 mg) of cyclosporine injection in 20 mL-100 mL 0.9% Sodium Chloride Injection or 5% Dextrose Injection (0.5- 2.5 mg/mL). Diluted infusion solutions are stable for 24 hours at room temperature under fluorescent light.

The Cremophor®EL (polyoxyethylated castor oil) contained in the concentrate for intravenous infusion can cause phthalate stripping from PVC. It is strongly recommended that glass bottles and non-PVC tubing be used to minimize patient exposure to DEHP.
Monitor closely for an acute allergic reaction for the first 30 minutes and at frequent intervals thereafter. Note: cyclosporine absorbs into plastics and can give falsely high serum or blood concentrations if blood samples are collected from the same line through which cyclosporine was administered.

Oral: Administer at a consistent time of day and at consistent intervals with regard to meals. Do not use plastic or styrofoam cups. If diluted with juice or milk, use a glass container and rinse with additional diluent, and then consume to ensure that complete dose has been taken. Do not use water or cleaning agents on the dosing syringe. To improve palatability, mix Sandimmune® with milk, chocolate milk or orange juice; mix Neoral® or Gengraf® with orange juice or apple juice but NOT milk. After mixed, have patient consume immediately. DO NOT MIX GRAPEFRUIT JUICE with any CSA product.

Supplier: Commercially available from various manufacturers.