GM-CSF for Reversal of immunopAralysis in pediatriC sEpsis-induced MODS (GRACE)
CPCCRN Protocol Number 078

Collaborative Pediatric Critical Care Research Network
Eunice Kennedy Shriver National Institute for Child Health and Human Development (NICHD)

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This protocol is CPCCRN Protocol Number 078, and has been authored by Mark Hall, M.D., Nationwide Children's Hospital, for implementation with the CPCCRN investigators.

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PROTOCOL TITLE:

GM-CSF for Reversal of immunopAralysis in pediatriC sEpsis-induced MODS

Short Title: GRACE
CPCCRN Protocol Number: 078

Lead Investigator and Author:
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I confirm that I have read this protocol, I understand it, and I will conduct the study according to the protocol. I will also work consistently with the ethical principles that have their origin in the Declaration of Helsinki and will adhere to the Ethical and Regulatory Considerations as stated. I confirm that if I or any of my staff are members of the Institutional Review Board, we will abstain from voting on this protocol, its future renewals, and its future amendments.

Principal Investigator Name: _____

Principal Investigator Signature: _____

Date: _____

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Abstract

Despite advances in pediatric sepsis care, children who develop sepsis-induced multiple organ dysfunction syndrome (MODS) face mortality rates as high as 20-50%. Multiple pathophysiological phenotypes exist in this population, with severe innate immune suppression being among the most common. Impairment of innate immune function is characterized by a reduced capacity of whole blood to produce the pro-inflammatory cytokine tumor necrosis factor (TNFα) upon ex vivo stimulation with bacterial lipopolysaccharide (LPS). When this reduction is severe (TNFα response $<$ 200 pg/ml) it is termed immunoparalysis. Immunoparalysis is associated with high risks for nosocomial infection, prolonged organ dysfunction, and death. CPCCRN has developed the capacity to perform highly standardized, generalizable, functional immune monitoring that is suitable for multi-center studies including interventional trials of subject-specific immunomodulation. Our pediatric preliminary data, along with several small adult studies, suggest that granulocyte-macrophage colony-stimulating factor (GM-CSF) can reverse immunoparalysis with the potential for improving clinical outcomes. GM-CSF is FDA-approved and has a low side-effect profile, but its role in the treatment of children with sepsis-induced MODS is unclear. While a dose of 125 mcg/m^2/day for 7 days appears to be associated with reversal of immunoparalysis in small single-center studies, the optimal route of delivery (intravenous vs subcutaneous) is unknown. Further, the impact of reversal of

immunoparalysis on clinical outcomes has never been tested in the setting of a large randomized controlled trial (RCT) in children with sepsis-induced MODS.

This study represents a necessary step in the development of a multi-center placebo-controlled, double-blind RCT of GM-CSF for the reversal of immunoparalysis in this population. In GRACE we will perform an open-label multi-center interventional trial to identify an adequate dose of GM-CSF in two different routes of delivery in children with sepsis-induced MODS, with reversal of immunoparalysis as the primary outcome variable. This will include the collection and analysis of unique pharmacokinetic data that will further inform future GM-CSF dosing strategies. We will also gather feasibility and effect magnitude and variation data that will be crucial for developing a future RCT to evaluate whether GM-CSF treatment reduces sepsis-induced MODS.

1 Study Summary

This study is an open-label, multi-center, interventional trial in which children with sepsis-induced MODS undergo surveillance immune function testing beginning on Day 2 of MODS. Those children who demonstrate immunoparalysis (TNFα response < 200 pg/ml) will receive a 7-day course of GM-CSF at a dose of 125 or 250 mcg/m^2/day by either the intravenous (IV) or subcutaneous (SQ) route.

The goal of the study is to establish the dose and route of delivery that results in resolution of immunoparalysis (TNFα response ≥ 200 pg/ml) by the morning after the 3rd scheduled dose with persistent resolution of immunoparalysis on the morning after the 7th scheduled dose. Resolution of immunoparalysis in 8 out of the first 10 subjects in a study treatment arm represents a successful dose and route. The goal of this study will be achieved through the following Specific Aims:

Specific Aim 1. Establish the immunologic efficacy of GM-CSF administered by the IV and SQ routes in children with immunoparalysis in the setting of sepsis-induced MODS.

Specific Aim 2. Estimate the pharmacokinetic parameters by the IV and SQ GM-CSF administered in pediatric sepsis-induced MODS.

Specific Aim 3. Demonstrate the feasibility of screening, enrollment, drug delivery, and sample collection for a multi-center immunostimulation trial in children with sepsis-induced MODS.

2 Rationale and Background

2.1 Sepsis, MODS, and innate immune suppression

Severe sepsis/septic shock remain a major source of pediatric morbidity and mortality worldwide, with the highest rates of adverse outcomes seen in children who develop failure of two or more organs. Children with sepsis-induced multiple organ dysfunction syndrome (MODS) represent a heterogeneous group of patients who have been shown to have several distinct phenotypes of underlying pathophysiology [1]. One common phenotype is that related to an exaggerated compensatory anti-inflammatory response. Impairment of the innate immune system is common and measurable in pediatric sepsis. Innate immune cells such as monocytes and neutrophils serve critical functions including migration to sites of infection, phagocytosis of pathogens, promotion of microbial killing, antigen presentation, and production of immunomodulatory cytokines. *Reductions* in pro-inflammatory cytokine production capacity have been shown to occur commonly following the onset of sepsis in adults [2, 3, 4, 5, 6], often carrying associations with nosocomial infection and death. We have shown similar associations between innate immune suppression and adverse outcomes in children with multiple organ dysfunction syndrome and sepsis. [7, 8]

2.2 Whole Blood *ex vivo* LPS-induced TNFα production capacity

Stimulation of whole blood with lipopolysaccharide (LPS) allows for quantification of the innate immune systems responsiveness to a challenge. LPS stimulation should result in rapid and robust production of the proinflammatory cytokine tumor necrosis factor (TNFα). Severe reductions in the TNFα response have been associated with increased risks for infection and death [7, 8, 9, 10, 11, 12]. To assure consistency between investigators, CPCCRN uses a consistent LPS type with rigorous quality control procedures, small blood volumes appropriate for pediatric studies, a four-hour incubation period (suitable for same-day testing), and TNFα quantitation on a highly automated, Good Laboratory Practices instrument. The *Immulite 1000* (Siemens, Deerfield, IL) is an automated chemiluminometer that is used to measure hormones and other analytes in clinical laboratory studies. We use this instrument to perform cytokine measurements, under a Research Use Agreement, using commercially available test units that are used clinically in Europe for patient management. The intra-assay coefficient of variation for the TNFα assay on the *Immulite* is less than 10%. By using the *Immulite* we avoid the need to perform highly operator-dependent assays such as enzyme-linked immunosorbent assays

(ELISAs).

2.3 Reversibility of innate immune suppression through GM-CSF therapy

GM-CSF is an endogenous, immunostimulating cytokine produced primarily by TH1 lymphocytes. It is available in recombinant human form (sargramostim, Leukine; Bayer Healthcare Pharmaceuticals, Montville, NJ) and has been FDA-approved for bone marrow reconstitution following bone marrow transplantation (BMT) since 1991. It has a long track record of safe use in acutely and critically ill patients, including children, with a low incidence of adverse events[13, 14, 15]. Rapid IV administration of GM-CSF (over ≤ 2 hours) has been associated with respiratory distress, peripheral edema (11% incidence with GM-CSF vs 7% incidence with placebo) and pericardial effusion (4% vs 1%), but this appears to be related to the rate of infusion[16]. These side effects have not been associated with infusion durations of > 2 hours or via the SQ route.

Monocyte hyporesponsiveness has been shown to be reversible *in vitro* through co-culture with GM-CSF [17, 18, 19, 20, 21]. GM-CSF has been used in four small published immunomodulation studies in critically ill adults, summarized in Table 1 on the facing page. The doses used were substantially lower than the FDA-approved used for bone marrow reconstitution (250 g/m^2/day). In all four adult studies, immune recovery was prompt (within 3 days of initiation of GM-CSF therapy) though it is unclear if longer durations of therapy were necessary to prevent relapse of innate immune depression. There were no serious adverse events ascribed to GM-CSF in any of the studies. GM-CSF therapy did not result in increased systemic inflammation as measured by plasma levels of the pro-inflammatory cytokines interleukin (IL)-6 or IL-8.

2.4 Immunoparalysis and MAS

Sepsis induced multiple organ dysfunction syndrome (MODS) is a poorly understood syndrome for which treatment is directed by organ failure instead of inflammation biomarker responses. CPCCRN investigators [1] have identified several distinct sepsis phenotypes. Another phenotype of pediatric sepsis-induced MODS is one analogous to macrophage activation syndrome (MAS), characterized by high serum ferritin levels ($\geq$2,000 ng/ml). The MAS phenotype is relatively uncommon (<10% of sepsis-induced MODS that we have studied), but represents a potential contraindication to receipt of GM-CSF therapy. We will therefore screen for and exclude children with serum ferritin levels $\geq$ 2,000 ng/ml.

Author, Year, [Ref]	Patient Population	Trial Design	GM-CSF Regimen	Patients Dosed with GM-CSF	Result
Presneill et al, 2002 [23]	Septic adults	Placebo-controlled RCT	3 g/kg/day (~90g/m2/day) IV x 5 days	10	Increased neutrophil function and oxygenation
Nierhaus et al, 2003 [24]	Septic adults	Uncontrolled case series	5 g/kg/day (~200g/m2/day) SQ x 3 days	9	Increased monocyte HLA-DR expression, TNF production capacity, and 66% survival
Rosenbloom et al, 2005 [25]	Septic adults	Placebo controlled RCT	125 g/m2 (~4g/kg) IV over 3 days	18	Increased monocyte HLA-DR expression, faster resolution of infection
Meisel et al, 2009 [26]	Septic adults	Placebo controlled RCT	4 g/kg/day (~125 g/m2/day) SQ x 8 days	19	Increased monocyte HLA-DR expression, TNF production capacity, improved illness severity

ALI: acute lung injury, RCT: randomized, controlled trial, SQ: subcutaneous.

Table 1: Clinical Evidence for the Use of GM-CSF for Immunomodulation in Critically Ill Adults

3 Subject Eligibility, Accrual and Study Duration

3.1 Eligibility criteria

Inclusion criteria are:

- $\geq$ 40 weeks gestational age to $<$ 18 years; AND
- Onset of $\geq$ 2 new organ dysfunctions (compared to pre-sepsis baseline) as measured by the Proulx criteria; AND
- Documented or suspected infection as the MODS inciting event.

Exclusion criteria are:

- Weight <5kg; OR
- Limitation of care order at the time of screening; OR
- Patients at high risk for brain death; OR
- Active (or planned within 7 days) immunosuppressive treatment; OR
- Known primary immunodeficiency disorder; OR
- Diagnosis of myeloid leukemia, myelodysplasia, or autoimmune thrombocytopenia; OR
- Known allergy to GM-CSF; OR
- Documented hyperferritinemia (serum ferritin $\geq$ 2,000 ng/ml) during current sepsis event; OR
- Contraindication to SQ injection (ECMO); OR
- Burns where >5% of the total body surface area is affected; OR
- Renal replacement therapy at the time of screening; OR
- On ECMO or anticipated to require ECMO; OR
- Known pregnancy; OR
- Inability to collect and ship sample for immune testing on MODS Day 2; OR
- Previous enrollment in the GRACE study.

These criteria were chosen in order to identify children with early, sepsis-induced MODS and to exclude children for whom GM-CSF is unlikely to be beneficial (children with limitation of care orders), those for whom the effect of GM-CSF may be altered by the receipt of active immunosuppression, those with known contraindications to GM-CSF, and those with concurrent secondary hemophagocytic lymphohistiocytosis (HLH) or macrophage activation syndrome (MAS).

3.2 Subject Accrual and Study Duration

This study will require 20 to 30 evaluable subjects. We define an evaluable subject as a subject who receives all 7 doses of GM-CSF (with an exception made when WBC > 50K cells/mm^3), and from whom both Drug Day 4 and Drug Day 8 immune testing samples were obtained. We anticipate that approximately one third of enrolled subjects will qualify to receive GM-CSF, and anticipate that half of the subjects receiving GM-CSF will be evaluable due to post-enrollment exclusions described below. This will require enrollment of up to 180 subjects.

The CPCCRN network enrolled over 400 subjects with multiple organ failure in two years in a previous study. We estimate that each of the eight participating hospitals will have 8 to 10 eligible patients per year. Study completion will require up to three years of subject accrual.

4 Study Design and Procedures

Sepsis-induced MODS remains an important source of morbidity and mortality in the PICU. We have demonstrated that failure of the immune system is a common feature of this disease state and is associated with high risks for the development of prolonged organ failure, nosocomial infection, and death. Using our small blood-volume immune function monitoring approach, our preliminary data suggest that immunoparalysis is reversible with the potential for improved clinical outcomes in this setting. The study is a prospective project designed to measure the effect of treatment with GM-CSF on innate immune function when given by the IV or SQ routes in children with demonstrated immunoparalysis in the setting of sepsis-induced MODS. The study workflow is summarized in Figure 1 on the next page.

4.1 Screening and Enrollment

All patients admitted to the pediatric or cardiac ICU at CPCCRN sites will be evaluated for study eligibility.

Patients who meet inclusion criteria will be entered into the data capture system and exclusion criteria will be recorded in that system. If the patient is eligible (no exclusion criteria are present) then the legal guardian(s) will be approached and offered the opportunity for their child to participate in the GRACE study. We will perform immune function screening beginning on Day 2 of sepsis-induced MODS (MODS Day 2).

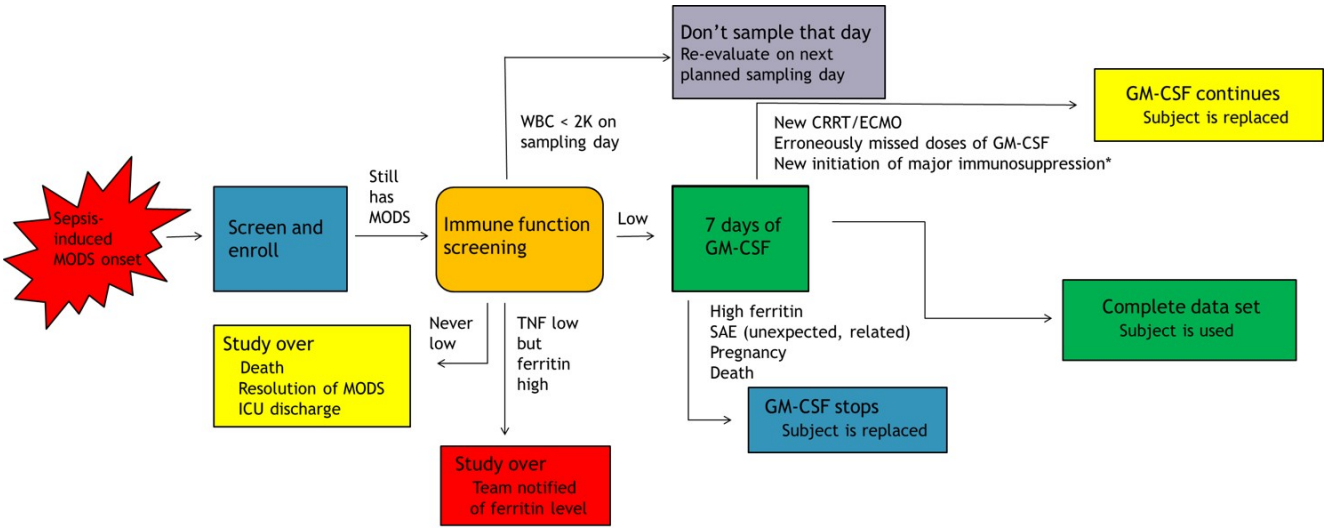

Figure 1: Diagram of the GRACE Study Workflow

4.2 Assessment of Innate Immune Function

Once a subject is enrolled on the GRACE study, the subject will undergo a 1.5 ml blood draw for measurement of whole blood LPS-induced TNFα production capacity (TNFα response), serum ferritin, plasma IL-6 and IL-8 levels. Enrolled subjects who demonstrate severe leukopenia (WBC $<$ 2K) at the time of planned screening sampling will not undergo sampling on that day, but will be re-sampled at the time of the next planned screening day provided their WBC count has increased to $\geq$ 2K. Blood samples will be stimulated on-site within one hour of sample collection using highly standardized LPS stimulation kits provided by the Immune Surveillance Laboratory (ISL) at the Research Institute at Nationwide Children's Hospital. Aliquots of 50 μL of whole blood will be stimulated in duplicate with LPS as well as two controls, consuming approximately 200 μL. The LPS incubation period is four hours. Samples are immediately centrifuged and all supernatants wil be overnight-shipped on dry ice to the ISL where TNFα production capacity and serum ferritin levels will be measured the next morning (MODS Day 3). If a subject's TNFα production capacity is $<$ 200 pg/ml and the serum ferritin is $<$ 2,000 ng/ml, the subject has documented immunoparalysis without MAS, and is eligible to be considered for GM-CSF administration. If immune function is normal, but MODS remains present, the subject will have repeat testing twice a week until resolution of MODS, MODS duration $>$ 20 days, discharge from the ICU, or death. Repeat testing would be on days 6 ± 1, 9 ± 1, 13 ± 1, 16 ± 1, and 20 ± 1. If test results indicate that the subject has

documented immunoparalysis without MAS, the subject is eligible to be considered for GM-CSF administration. Subjects who have improved to single-organ dysfunction on the first day of GM-CSF administration will also undergo measurement of their TNFα production capacity prior to receiving their first dose of GM-CSF, but these samples will not be analyzed in real-time. Rather, they will be batch-shipped with the later immune function samples.

Subjects who receive GM-CSF will undergo repeat innate immune function and ferritin testing the morning after their 3rd and 7th scheduled doses of GM-CSF to monitor the effect of GM-CSF on immunoparalysis associated with pediatric sepsis-induced MODS, as well as to reassess serum ferritin. Immune function testing is summarized in Figure 2.

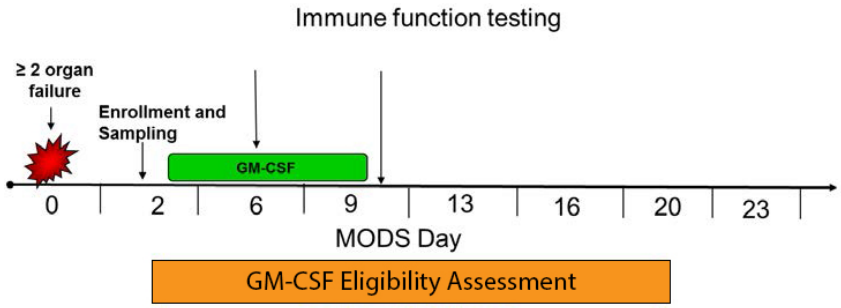

Figure 2: Schematic of the GRACE Study

If the serum ferritin is $\geq$ 2,000 ng/ml, the subject ends further study participation permanently, and the site will be immediately notified so that site-specific confirmation and management of hyperferritinemia can take place.

The Nationwide Children's site is able to assess subject eligibility on the same day as LPS stimulation (MODS Day 2), but GM-CSF administration will be delayed until the next morning (MODS Day 3) in order to remain consistent with the dosing schedule at other CPCCRN sites.

4.2.1 Eligibility for Ongoing Immune Function Screening

If the TNF response is $>$ 200 pg/ml and the ferritin level is $<$ 2,000 ng/ml on the initial screening sample, the subject will undergo repeat immune function screening according

to the schedule described in section 4.2 above, unless any of the following occur:

Immune function screening will be temporarily paused in the event of the following:

- WBC count $<$ 2K on the planned screening day
- Initiation of CRRT or ECMO
- Immune function screening will resume on the next planned sampling day once the WBC count is $\geq$ 2K or the subject comes off CRRT or ECMO provided that the subject remains in the ICU, still has MODS, and has no other exclusions.

Immune function screening will be permanently discontinued in the event of the following:

- Documentation (either through study-specific or clinical laboratory findings) of serum ferritin level $\geq$ 2,000 ng/ml
- Confirmation of a diagnosis other than sepsis
- Diagnosis of pregnancy
- Initiation of new major immunosuppressive therapy (See Manual of Operations for list)

4.3 Eligibility to Receive Initial GM-CSF Dose

Subjects eligible to be considered for initial administration of GM-CSF will receive study drug if the following additional requirements are met:

- No diagnosis of pregnancy.
- The presence of dysfunction of at least one organ system
- Subject remains in ICU.
- WBC $\leq$ 50K cells/mm^3.
- CRRT is not being used and there are no immediate plans to start.
- No new immunosuppressive drugs are being given or planned in the next 7 days.
- Not on ECMO or have immediate plans to start ECMO.

The WBC measurement is required prior to GM-CSF, and must be the most recent measurement prior to drug administration. The sample must be no earlier than 6 hours prior to drug administration.

4.4 Eligibility to Receive Subsequent GM-CSF Doses

Subjects will continue to receive GM-CSF for up to 7 days but must continually qualify by meeting the following criteria:

- Not pregnant (pregnancy is a permanent disqualification for drug; stop screening)
- Subject remains in hospital (ICU not required).
- WBC $\leq$ 50K cells/mm^3.
- Absence of documentation (through clinical laboratory findings) of serum ferritin level $\geq$ 2,000 ng/ml.

The WBC measurement is required prior to GM-CSF, and must be the most recent measurement prior to drug administration. The sample must be no earlier than 6 hours prior to drug administration. If the WBC exceeds 50K cells/mm^3, GM-CSF should be withheld on this study day. The subject may be re-assessed for GM-CSF on the next day (up to Drug Day 7).

Repeat sampling for testing innate immune function and ferritin levels is conducted on Drug Day 4. On Drug Day 5, GM-CSF should not be administered until verification that the ferritin remains below 2,000 ng/ml. If the ferritin measurement from Drug Day 4 indicates hyperferritinemia ($\geq$ 2,000 ng/ml), all subsequent study drug administration will stop, and the site will be immediately notified so that the hyperferritinemia can be confirmed and managed according to site clinical protocols.

On each day, it must be verified that if there have been serious adverse events that are related or probably related to GM-CSF, the treating physician authorizes the safety of continued GM-CSF administration.

4.5 Assignment of Treatment Arms

Study drug will be administered in 4 sequential treatment arms (125 mcg/m^2/day IV, 250 mcg/m^2/day IV, 125 mcg/m^2/day SQ, 250 mcg/m^2/day SQ) dependent on the success of an arm. Enrollment will begin in the 125 mcg IV arm. If 125 mcg IV does not prove successful, we will move to the 250 mcg IV dose arm. If an IV arm is successful, we will open a SQ arm at the same treatment dose level that the IV arm completed. If we begin with 125 mcg SQ arm, then we will move to the 250 SQ arm if the 125 SQ arm is not successful.

A subject is considered to have failed to achieve resolution of immunoparalysis if the TNFα response is $<$ 200 pg/ml on Drug Days 4 or 8. If a 3rd subject in a 125 mcg dose arm fails to respond to study drug, that arm is considered to have failed, and no further subjects will be enrolled in that arm. Enrollment will proceed to the 250 mcg arm for the respective route (IV or SC). In the 250 mcg arms, we will enroll fully to ten evaluable subjects even if these arms have 3 or more subjects who do not respond. This is to fully understand the PK of the drug at this FDA approved dose.

A successful dose and route is defined as the resolution of immunoparalysis in 8 out of the first 10 subjects in a treatment arm. Enrollment will not stop early in a study treatment arm due to achieving 8 successes. Instead we will continue to enroll until that study treatment arm is complete and we have confirmed that the arm has a sufficient number (ten) of evaluable subjects who have received study drug.

4.6 Achieving Enrollment Balance

Corticosteroid use and subject age represent potentially important variables to consider since it is not clear how they will impact the effect of GM-CSF on immune function. We will make good-faith efforts to enroll some subjects with and without corticosteroid use, and we will make good-faith efforts to achieve a similar distribution of ages in our final IV and SQ arms. The study investigator may choose not to approach the legal guardian(s) of otherwise eligible patients if the age or steroid bins (defined below) for that subject are already filled. However, it is left to the discretion of the study investigator to override these requirements if they cause undue delays in enrollment.

Corticosteroid use. Corticosteroid use will be assessed at the time of assessment for enrollment eligibility and subjects will be classified by whether or not they are using corticosteroids at that time. For each treatment arm, we will enroll to allow for a 60%/40%, 50%/50%, or 40%/60% split of corticosteroid exposure at time of screening. If the bin to which a screened subject belongs (using corticosteroids vs. not using corticosteroids) already has six evaluable subjects, then the bin is closed and the subject will not be enrolled in the study. When a new dose/route arm is initiated the bins are reset to zero.

Age. Subjects in the IV treatment arm(s) will be recruited without regard to age (other than that stipulated in the eligibility criteria). However, in order to ensure that subjects in the final SQ arm have a similar age distribution to the final IV arm, subjects will enroll into one of two age bins. The first bin will be for subjects no older than the median age

from the final IV arm. The second age bin will be for older subjects. We will require a 60%/40%, 50%/50%, or 40%/60% split between the two age bins analogously to the corticosteroid use bins.

4.7 Subject Replacement

In the event that a subject newly develops an exclusion criterion after GM-CSF treatment has begun, i.e. the discovery of an allergy to GM-CSF, the following approach to drug delivery and replacement in the study treatment arm will be taken:

- **Ferritin $\geq$ 2,000 ng/ml:** If a subject develops a serum ferritin level $\geq$ 2,000 ng/ml as measured on the post-3rd dose sample, GM-CSF will be discontinued and the study team will be notified so that they may confirm the ferritin level at their institution per the treating team. This subject **will be replaced** in the study treatment arm.
- **Initiation of CRRT or ECMO:** The initiation of CRRT or ECMO may affect the PK/PD of GM-CSF. If a subject is newly placed on CRRT or ECMO while receiving GM-CSF, GM-CSF treatment will continue and PK and immune function testing will continue per protocol, however the subject's data will not be used for determination of treatment success. This subject **will be replaced** in the study treatment arm.
- **Serious AE:** If a subject's GM-CSF is discontinued as the result of an SAE that is perceived to be drug-related, the subject **will be replaced** in the study treatment arm.
- **Pregnancy:** If pregnancy is diagnosed in a subject who is receiving GM-CSF, the study drug will be stopped and the subject **will be replaced** in the study treatment arm.
- **Drug interruption:** If a subject who is assigned to study drug does not receive all 7 doses of GM-CSF (except for WBC > 50,000), the subject **will be replaced** in the study treatment arm.
- **Initiation of major immunosuppressive therapy:** If a subject who is receiving GM-CSF is prescribed new immunosuppressive therapy (See Manual of Operations for list) study participation will be concluded and the subject **will be replaced** in the study treatment arm. GM-CSF therapy can continue at the discretion of the clinical team. If GM-CSF is **continued**, Day 4 and Day 8 samples should still be collected.

- **WBC > 50,000:** If one or more doses of GM-CSF are held due to a subject's WBC count being > 50,000 cell/mm^3, sampling will continue on what would otherwise have been the morning after the 3rd and 7th scheduled doses. Samples will be collected and data will be analyzed per protocol and the subject **will be included** in the study treatment arm.
- **Confirmed diagnosis other than sepsis:** If a subject is later determined definitively to have a diagnosis other than sepsis that lead to sepsis-like symptoms, this subject **will be replaced** in the study treatment arm.
- **Missing/unusable immune function sample:** On Drug Day 4 or Drug Day 8 immune function testing samples are collected for a subject. If a subject's sample from either of these days was not obtained or was obtained but found to be unusable, this subject **will be replaced** in the study treatment arm.
- **Hospital Discharge:** If a subject who is receiving GM-CSF is discharged from the hospital prior to receiving all doses, this subject **will be replaced** in the study treatment arm.
- **Study Withdrawal:** If a subject who is receiving GM-CSF is withdrawn from the study, this subject **will be replaced** in the study treatment arm.

4.8 GM-CSF Pharmacokinetics

Pharmacokinetic (PK) data will be collected from subjects who receive study drug to evaluate drug clearance, volume of drug distribution, and therapeutically beneficial concentrations. The half-life of GM-CSF is sufficiently short (2-5 hours) that steady state pharmacokinetics (PK) are never reached. We will obtain GM-CSF levels from serum of enrolled subjects around the 3rd and 7th scheduled doses of drug. For these evaluations, 6-8 timed blood samples of 1 mL each for determination of serum GM-CSF concentration will be obtained from an existing intravenous or intra-arterial line at pre-specific time points around those doses. PK samples will only be collected if subjects have an indwelling vascular catheter that permits painless blood draws.

4.9 Other Sampling and Shipping

The remaining plasma from blood samples obtained for immune function monitoring (approximately 1.3 ml blood per sample) and post-drug samples will be collected, aliquoted, and stored for later analyses of the relationships between biomarkers of coagulation (e.g.,

PAI-1, thrombomodulin, angiopoietin-2) and immune function over time. Plasma will also be analyzed for levels of the cytokines IL-6 and IL-8 in order to assess the relationships between GM-CSF therapy and systemic inflammation over time. Endotracheal aspirates will be collected on immune function screening days and on each of the 7 days of GM-CSF dosing for analyses of the relationships between immune function and the airway microbiome in the setting of immunomodulatory drug therapy.

Screening samples and Drug Day 4 samples will be overnight shipped to the ISL as noted in Section 4.2. All other samples will be stored at the site and batch-shipped upon completion of each subject's participation in the GRACE study.

4.10 Withdrawal from Study

Parents may withdraw their child from participation in this study at any time, including discontinuation of data collection and sampling on Day 8 after a subject has received all 7 scheduled doses. However, investigators will continue to follow the subject for the occurrence of adverse events occurring in the hospital after the start of the first dose of GM-CSF on Drug Day 1 through Drug Day 21 or hospital discharge, whichever occurs first.

Discontinuation of study drug does not constitute withdrawal from the study.

4.11 Summary

Patients will be screened by eligibility criteria, tested to determine the presence of immunoparalysis without the presence of MAS, and if they then qualify, will receive GM-CSF in order to observe the effect of GM-CSF on immunoparalysis. For subjects who receive study drug, Table 2 on the next page summarizes drug administration, sampling, and shipping of specimens to Nationwide.

5 GM-CSF Administration

5.1 IV Infusion Arms

The first dose of the study drug will be infused over a minimum of 6 hours on the day after the qualifying immune testing sample. The infusion may be interrupted if needed, but must run over a total of no more than 12 hours. GM-CSF may be given via a peripheral or central IV catheter. The first administered dose of GM-CSF will be initiated within 4 hours after receipt of notification of qualification for drug. If the 4 hours elapse, GM-CSF

		Drug Study Day									
	Schedule of Events	1	2	3	4	5	6	7	8	9-21/ hospital discharge	22 - hospital discharge
	GM-CSF administration	X	X	X	X	X	X	X			
	AE monitoring	X	X	X	X	X	X	X	X	X	
	AE resolution monitoring	X	X	X	X	X	X	X	X	X	X
	PK sampling			X				X			
	1.5 mL blood sample prior to GM-CSF (4AM- noon) immune function/ferritin testing				X				X		
	Sample shipment to Nationwide Initial immune function/ferritin testing				X						
	Tracheal aspirates	X	X	X	X	X	X	X	X		
	Batch shipment to Nationwide PK samples, tracheal aspirates, and final immune function samples	X	X	X	X	X	X	X	X		

Table 2: Summary of Study Events

should still be started as soon as possible. Subjects who have initiated the study drug treatment course will need to continually qualify for subsequent doses as indicated under the protocol section 4.4 for subsequent GM-CSF doses. All subsequent IV doses will be initiated between 4 AM and noon. If the window is missed, GM-CSF should still be given as soon as possible.

5.2 Subcutaneous Arms

GM-CSF will be given SQ once daily for 7 days with the first dose given within 4 hours after receipt of notification of qualification for drug. If the 4 hours elapse, GM-CSF should still be started as soon as possible. Subsequent SQ doses will be given between 4 AM and noon.

6 Statistical Analysis

Statistical analyses and comparisons for this study will be primarily descriptive. The cohort will be characterized with simple statistical summaries, i.e. frequencies, means, standard deviations, medians, and interquartile ranges. An arm will be considered

successful if 8 of the first 10 completing subjects in the arm have a good immune response, i.e. TNFα > 200 pg/mL on Drug Days 4 and 8. Figures will be created to illustrate the distribution over the treatment course of TNFα, IL-6, and IL-8. Pharmacokinetic modeling of GM-CSF will be conducted using non-linear mixed models as implemented in NONMEM (version VI, ICON Development Solutions, Ellicott City, MD). All other inferential analyses that may be conducted will be considered exploratory and will be reported as such.

7 Data Collection

Baseline data collection will include demographic and clinical variables such as date of birth, sex, race, ethnicity, and medical history. Data collection regarding the current hospitalization will include aspects of clinical treatment and assessments such as organ dysfunction, surgical interventions, use of monitoring devices, presence of co-morbidities, microbiology reports, labs, medications used, severity of illness, and timing of events.

8 Data Management

8.1 Clinical Site Data Management

The investigators and study staff are responsible for maintaining a comprehensive and centralized filing system containing all study-related documentation. Study worksheets are to be completed in a neat, legible manner to ensure accurate interpretation of data. Any corrections or changes on the worksheets when made, the original entry should be crossed out using a single line, and must be dated and initialed by the individual making the change. The original entry will not be erased or overwritten. The site will maintain an Essential Documents Binder, which may be in paper or electronic form. Copies of all informed consent documents will be kept on file and be available for site monitoring inspection (on site or remote).

8.2 Electronic Data Capture System

An electronic data capture system will be used that allows sites to enter data via a secure web interface. An electronic discrepancy management system will be used to notify sites of inconsistent or erroneous data entry, which will be corrected by the clinical site. An audit trail of all data corrections will be maintained.

9 Data Coordinating Center

9.1 IT Infrastructure

The Data Coordinating Center (DCC) in the Department of Pediatrics at the University of Utah School of Medicine provides data coordination and management services for a variety of national research networks. Anchoring these services is a state-of-the-art, energy efficient data center completed in 2013. The data center facility supports more than 1400 users around the world and provides a secure, reliable, enterprise-wide infrastructure for delivering critical DCC systems and services. The data center was built using high industry standards and energy efficient cooling solutions. The data center is cooled by Rittal's LCP inline cooling technology, providing efficiency, redundancy and modularity. Cooling is based upon a hot/cold aisle design that allows for even air distribution with minimal hot spots. The data center electrical power system contains a redundant Mitsubishi uninterruptible power system (UPS) with a diesel backup generator. The data center is protected with a FM200 fire suppression system, early warning smoke detectors and a heat detection warning system to act as a secondary system to the smoke detectors. Security guards are on-site conducting access control and rounds 24/7/365. Entry into the data center is restricted by card access and layered security measures and controls. The data center and external building access points are monitored with video surveillance.

The data center uses a large scale VMware server virtualization environment consisting of more than 200 virtual servers. This provides key advantages:

- high availability – in the event of hardware failure, virtual servers automatically go back online in a seamless process.
- flexible infrastructure – disk storage, memory and processor capacity can be increased or reallocated at any time.
- rapid deployment – servers can be provisioned on-demand with minimal waiting on hardware of software.

The data center optimizes its storage resources by implementing a networked storage system to support its virtualized environment. The data center currently manages over 50 terabytes of data. The storage solution consists of Dell's EqualLogic PS Series Storage system for providing a virtualized storage area network (SAN). This technology provides excellent performance and complete redundancy.

Production servers running critical applications are clustered and configured for failover

events. Servers are backed up with encryption through a dedicated backup server that connects across an internal 10 gigabit network to a tape drive. DCC storage area networking (SAN) applications, clusters, and switch-to-switch links are also on a 10 gigabit network. Incremental backups occur hourly Monday through Friday from 6 am to 6 pm. Incremental backups also are performed each night with full system backups occurring every Friday. Tapes are stored in a fireproof safe inside the data center facility, and full backups are taken off site on a weekly basis to an off-site commercial storage facility.

In the event of catastrophic failure, such as a fire in the server facility, daily backups would probably survive because of the fire suppression system and fireproof safe, but there would be obvious delay in re-establishing data center function because the servers will not survive such a disaster. Total destruction of the data center facility could cause the loss of up to one week's data. In future investments, the data center is making co-location, disaster recovery and business continuity solutions a top priority.

DCC information systems are available 24 hours a day, 7 days a week to all users unless a scheduled maintenance interruption is required. If this occurs, we notify all users of the relevant systems, and data entry can be deferred until after the interruption is over. Critical systems availability has exceeded 99.9% for the past five years, and there has been no unscheduled downtime in over eight years.

9.2 Security and Confidentiality

The data center coordinates the network infrastructure and security with the Health Sciences Campus (HSC) information systems at the University of Utah. This provides us with effective firewall hardware, automatic network intrusion detection, and the expertise of dedicated security experts working at the University. Communication over public networks is encrypted with virtual point-to-point sessions using transport layer security (TLS) or virtual private network (VPN) technologies, both of which provide at least 128 bit encryption. All of our Web-based systems use the TLS protocol to transmit data securely over the Internet. Direct access to data center machines is only available while physically located inside our offices, or via a VPN client.

All network traffic is monitored for intrusion attempts, security scans are regularly run against our servers, and our IT staff is notified of intrusion alerts. Users are required to change their passwords every 90 days, and workstations time out after 5 minutes of inactivity. All files are protected at group and user levels; database security is handled in a similar manner with group-level access to databases, tables, and views in Microsoft

SQL Server. Finally, all laptop computers in use in the School of Medicine are whole-disk encrypted.

The data center uses control center tools to continuously monitor systems and failure alerts. Environmental and network systems are also monitored to ensure up time. Highly trained system administrators on staff are available to respond in high risk emergency events.

All personnel involved with the DCC have signed confidentiality agreements concerning data encountered in the course of their daily work. All personnel (including administrative staff) have received Human Subjects Protection and Health Information Portability and Accountability Act (HIPAA) education. We require all users to sign specific agreements concerning security, confidentiality, and use of our information systems, before access is provided.

10 Protection of Human Subjects

10.1 Institutional Review Board (IRB) Approval

The CPCCRN uses the University of Utah as its central IRB, and this study will be reviewed and approved by the central IRB prior to enrollment of human subjects. The CPCCRN institutions are responsible for fulfilling non-IRB human subjects protection program activities such as handling conflicts of interest, radiation safety, and other functions. The DCC will not permit subject enrollment without documentation that a site has completed its non-IRB activities concerning protection of human subjects.

10.2 Informed Consent

Parental Permission

Subjects who are eligible for this study are under 18 years of age, and written permission from parents or legal guardians will be required for participation. After determining that a subject is eligible, the site investigator or designee will approach the parent or legal guardian to offer participation for their child in the study. The parent or legal guardian will be informed about the objectives of the study and the potential risks and benefits of participation. Subjects will only be enrolled if their parent or guardian provides permission for their child to participate.

Child Assent

Subjects who are eligible for this study are often acutely ill, and child assent may not be possible at the time of study enrollment. However, as the illness resolves, issues about assent become applicable. Children who are capable of giving assent and who are alert and competent, will be asked, following an age-appropriate discussion of risks and benefits, to give assent to the study or further study procedures. Assent will be waived if the child is too young, has a severely reduced mental age, decreased level of consciousness, psychological problems, or other legitimate reasons as judged by the Institutional Review Board at each site. Assent will not be required for subjects who become able to assent after study procedures are complete, as follow up will not be required for safety purposes.

Subject Consent

Subjects who are eligible for this study are under 18 years of age at the time of screening. If a subject attains the age of 18 years during the study period, then informed consent becomes applicable. If this occurs, 18-year-old subjects who are alert and competent and capable of giving consent will be asked, following an appropriate discussion of risks and benefits, to give consent to the study for further study procedures. Subject consent will be waived if the subject has a severely reduced mental age, decreased level of consciousness, psychological problems, or other legitimate reasons as judged by the Institutional Review Board at each site. Consent will not be required for subjects who become able to consent after study procedures are complete, as follow up will not be required for safety purposes.

10.3 Potential Risks

10.3.1 GM-CSF risk

The drug GM-CSF has a long track record of safe use in adults and children. The incidence of adverse events such as fever, chills, bone pain, dyspnea, tachycardia, and hemodynamic instability was no different between GM-CSF and placebo-treated groups in controlled adult BMT studies. Rapid IV administration of GM-CSF (over $\leq$ 2 hours) has been associated with peripheral edema (11% incidence with GM-CSF vs 7% incidence with placebo) and pericardial effusion (4% vs 1%), but this appears to be related to the rate of infusion. We are unaware of these side effects being reported with slow IV infusion or SQ administration. Among the published small randomized controlled trials in critically ill adults and neonates, there were no GM-CSF related adverse events reported and no evidence that GM-CSF treatment resulted in increased systemic inflammation, though this risk is unknown in the critically ill pediatric population. A risk of hypersensitivity/allergic

reaction exists with any drug.

10.3.2 Phlebotomy risks

Another potential risk to enrolled subjects in this study is anemia. Any blood sampling can contribute to a low red blood cell count, or anemia. While many children with critical illness go on to require a blood transfusion, we will keep the total blood drawn for research purposes to < 3 ml/kg over the entire study period. This volume of blood loss should not significantly increase the risk of an enrolled subject to require a blood transfusion. Some children enrolled in this study may need to undergo additional venipuncture for the purposes of study sample collection, particularly for later time points when central catheters may have been removed. We will try to coordinate blood sampling with previously ordered phlebotomy but if we are unable we may have to perform venipuncture. While venipuncture can cause pain, bruising, bleeding, and rarely, infection. These risks are generally viewed as modest. Guidance will be provided to sites for truncation of sampling and/or reduction of blood sample volume such that subjects >5kg may participate in the GRACE study without exceeding the 3ml/kg total blood draw limit.

10.3.3 Loss of Confidentiality

There is a minimal risk of loss of confidentiality for data collected in this study.

10.4 Protections Against Potential Risks

There are several elements of this study that will help minimize risks to subjects. The exclusion criteria eliminate certain populations that may be at higher risk of complications. Most importantly, patients will be hospitalized in intensive care units and monitored closely for potential complications and adverse events as a part of usual clinical care. These intensive care units are all staffed by board-certified pediatric intensivists and highly experienced ICU nurses.

10.4.1 GM-CSF risk

The dose of GM-CSF being tested in this submission is at or below the doses used for bone marrow reconstitution following transplantation. Also, the rate of infusion chosen for this submission (over 6 - 12 hours) is extremely conservative and should eliminate infusion-rate-dependent side effects.

10.4.2 Phlebotomy risks

The risk of anemia is minimized by minimizing the volume of blood samples taken and by excluding infants. The risk of phlebotomy is minimized by making use of indwelling catheters when present and by timing sampling with previously scheduled venipuncture if able.

10.4.3 Loss of Confidentiality

The minimal risk of loss of privacy is mitigated by the substantial data management resources at the the data coordinating center and a process to verify that identifying information is not inappropriately uploaded.

10.5 Potential Benefits

The overall goal of this research program is to improve outcomes from pediatric sepsis-induced MODS through reduction of nosocomial infection risk, speeding resolution of organ failure, and thereby reducing mortality through optimization of immune function. Therefore, if GM-CSF is effective, children enrolled in this study could benefit from this treatment. Knowledge gained from this study may help other children with sepsis-induced MODS and other forms of critical illness in the future, as these data will inform the design of larger, multi-center trails of GM-CSF in these settings. No incentives will be offered for study participation.

11 Data and Safety Monitoring Plan

11.1 Data Safety Monitoring Board (DSMB)

The study will have a Data Safety Monitoring Board (DSMB). The DSMB will have a charter, will approve the protocol prior to implementation, and will review interim data as applicable. The purpose of the DSMB is to advise the Principal Investigator(s) regarding the continuing safety of study subjects and the continuing validity and scientific merit of the study. The DSMB is responsible for monitoring accrual of study subjects, adherence to the study protocol, assessments of data quality, performance of individual Clinical Center, review of adverse events, and other subject safety issues.

11.2 Adverse Event Reporting

11.2.1 Definitions, Relatedness, Severity and Expectedness

Definition: An adverse event (AE) is any untoward medical occurrence experienced by a subject. An event constitutes a disease, a set of related signs or symptoms, or a single sign or symptom. On each study day, the site investigators will evaluate adverse events. The nature of each experience, date and time (where appropriate) of onset, outcome, course, and relationship to treatment should be established.

Relatedness: The suspected relationship between study interventions and any adverse event will be determined by the site investigator using the following criteria. *Relatedness may **not** be assessed by a research coordinator, and must be assessed by an investigator.*

Not Related: The event is clearly related to other factors, such as the subject's clinical state, therapeutic interventions, or concomitant drugs administered to the subject.

Possibly Related: The event follows compatible temporal sequence from the time of beginning the assigned study intervention, but could have been produced by other factors such as the subject's clinical state, therapeutic interventions, or concomitant drugs administered to the subject.

Probably Related: The event follows a reasonable temporal sequence from the time of beginning the assigned study intervention, and *cannot be reasonably explained* by other factors such as the subject's clinical state, therapeutic interventions, or concomitant drugs administered to the subject.

Seriousness: The seriousness of clinical adverse events and laboratory abnormalities will be recorded by the site investigator and categorized. A serious adverse event (SAE) is an adverse event that:

- results in death; or
- is life-threatening (the patient was, in the view of the site investigator, in immediate danger of death from the event as it occurred); or
- requires inpatient hospitalization or prolongs an existing hospitalization; or
- results in persistent or significant disability or incapacity; or
- results in congenital anomaly/birth defect; or
- any other event that, based upon appropriate medical judgment, may jeopardize the subject's health and may require medical or surgical intervention to prevent one of the other outcomes listed in this definition.

Expectedness of the Event: All adverse events, including serious adverse events, will be evaluated as to whether their occurrence was expected or unexpected. Expected events include known side effects from administration of GM-CSF as well as complications from pediatric sepsis-induced MODS. Adverse events occurring in the hospital after the start of the first dose of GM-CSF on Drug Day 1 and on or prior to Drug Day 21 will be recorded. Serious adverse events will be monitored for final status until hospital discharge.

Treatment or Action Taken: For each adverse event, the site investigator will record whether an intervention was required:

- Intervention: Surgery or procedure
- Other Treatment: Medication initiation, change, or discontinuation
- None: No action taken

Outcome of Event: Finally, the site investigator will record the clinical outcome of each adverse event as follows:

- Death
- Recovered and the patient returned to baseline status
- Recovered with permanent sequelae
- Symptoms continue

11.2.2 Data Collection Procedures for Adverse Events

After initiation of GM-CSF, all adverse events, whether anticipated or unanticipated, will be recorded according to the date of first occurrence, severity, and their duration, as well as any treatment prescribed. Any medical condition that exists prior to initiation of GM-CSF which remains unchanged or improves, will not be recorded as an adverse event at subsequent evaluations. However, worsening of a medical condition that was present prior to initiation of GM-CSF will be considered a new adverse event and reported.

Abnormal laboratory values that are clinically significant will be recorded as adverse events and the site investigator will assess the severity and relationship to the study. Laboratory values that are abnormal at the time of randomization and that do not worsen will not be recorded as adverse events.

Adverse events will be coded using the MedDRA coding vocabulary. Coding will be done centrally at the Data Coordinating Center because this requires specific training.

11.2.3 Unanticipated Problems

Unanticipated problems are defined as incidents, experiences, or outcomes that are unexpected, related to participation in the study, and suggest that the research places subjects at a greater risk of harm than was previously known or recognized. The site investigator will report unanticipated problems to the Data Coordinating Center within 24 hours. A detailed completed report will be required to be sent to the Data Coordinating Center within 3 working days of the event. After receipt of the complete report, the Data Coordinating Center will report these unanticipated problems to the National Institute for Child Health and Human Development (NICHD) Program Official or Project Officer in an expedited manner (within 24 hours). In accordance with central IRB requirements, the site investigator may be required to report such unanticipated problems to the central IRB in addition to notifying the Data Coordinating Center. In the event that the medical monitor believes that such an event warrants emergent suspension of enrollment in the trial, and National Institute for Child Health and Human Development (NICHD) staff cannot be reached expeditiously, the Data Coordinating Center will notify the study investigator (Dr.Hall) and all site investigators to cease enrollment in the trial. Resumption of enrollment will not occur without consent of the National Institute for Child Health and Human Development (NICHD) staff after discussion with the DSMB.

11.2.4 Monitoring Serious Adverse Events

The Principal Investigator of the Data Coordinating Center (Dr. Dean) will act as the medical monitor for this study. If Dr. Dean is unavailable, a qualified physician will be designated to fulfill this function. Site investigators and/or research coordinators will report serious adverse events to the Data Coordinating Center within 24 hours. A detailed completed report will be required to be sent to the Data Coordinating Center within 3 working days of the event, and the medical monitor will assess all serious adverse events reported from site investigators.

For each of these serious adverse events, the site investigator will provide sufficient medical history and clinical details for a safety assessment to be made with regard to continuation of the trial. The medical monitor will sign each SAE report after review. All SAE reports will be retained at the Data Coordinating Center, and all SAE reports will be available for review by DSMB members and National Institute for Child Health

and Human Development (NICHD) staff.

In the unlikely event that the medical monitor believes an unexpected and study-related SAE warrants emergent cessation of enrollment in the trial, National Institute for Child Health and Human Development (NICHD) staff and the DSMB chairperson will be immediately consulted. If these individuals concur with the judgment of the medical monitor, or if the National Institute for Child Health and Human Development (NICHD) staff and the DSMB chairperson cannot be reached expeditiously, the Data Coordinating Center will notify the study investigator (Dr.Hall) and all site investigators to cease enrollment in the trial. Resumption of enrollment will not occur without consent of the National Institute for Child Health and Human Development (NICHD) staff after discussion with the DSMB.

In accordance with central IRB requirements, the site investigator may be required to report such events to the central IRB in addition to notifying the Data Coordinating Center.

After notification of the National Institute for Child Health and Human Development (NICHD) Program Official or Project Officer, and the DSMB chairperson, of *serious, unexpected, and study-related* adverse events or unanticipated problems, decisions will be made whether to continue the study without change, and whether to convene the entire DSMB for an emergent meeting. If a decision is made to suspend enrollment in the trial, this will be reported to the study investigator (Dr. Hall) and all clinical investigators, who will be instructed to report this to their central IRB.

The DSMB will review all adverse events (not necessarily serious, unexpected, and study-related) during scheduled DSMB meetings. The Data Coordinating Center will prepare a summary report of adverse events for the DSMB meetings, classified with the MedDRA coding system.

12 Study Training

A formal training program for investigators and research staff will be held prior to the start of enrollment. The training program will cover regulatory topics and Good Clinical Practice. The training will also provide in depth explanations regarding study procedures, clinical care, adverse event reporting, data entry procedures, quality assurance, site monitoring, and the informed consent process. A manual of operations will be provided to each investigator prior to the start of enrollment. The manual will detail specific information about the study procedures, regulatory information, safety reporting, and

other necessary information. Updates and revisions to the manual will be made available electronically. The Data Coordinating Center, in collaboration with the study investigator (Dr. Hall), will be the main contact for study questions.

Each participating clinical site that has not previously participated in the GIFT study (which also uses whole blood ex vivo LPS-induced TNFα production capacity testing) will undergo one on-site visit prior to enrollment start to insure familiarity with clinical and laboratory procedures. The remote monitoring process for the GRACE study will then validate that appropriate and attributable source data are utilized for specific data points.

13 Study Monitoring

13.1 Site Monitoring Plan

A study-specific risk-based monitoring plan, separate from the protocol will be completed which outlines specific criteria for monitoring. This plan will include frequency of monitoring, the data elements to monitor, and a follow up plan for non-compliant sites. Remote site monitoring schedules will be determined by the Data Coordinating Center in coordination with the study principal investigator.

13.2 Remote Monitoring

The Data Coordinating Center remotely monitor study activities. Remote monitoring involves detailed review of the data entered by the Clinical Center and consultations with the Clinical Center investigator and/or research coordinator to review safety and data quality. This may require uploading de-identified copies of specific parts of the medical record, patient study file, regulatory documentation, or other source documents to the Data Coordinating Center staff, who review those materials against the data recorded in the electronic data capture system. This helps assure protocol compliance and accurate data collection. The Data Coordinating Center may conduct more remote monitoring activities early in the trial to assure protocol compliance and identify any training issues that may exist. Remote monitoring documents will be retained in accordance with federal requirements. Safety of subjects will be monitored and ensured in accordance with the Data and Safety Monitoring Board (DSMB) plan.

13.3 Pharmacy Monitoring

The Clinical Center pharmacy must maintain adequate records of all dispensed study drug. Each pharmacy will be monitored and may be requested to send copies of these documents to the Data Coordinating Center.

14 Regulatory Issues

14.1 Food and Drug Administration

This trial is being conducted under an Investigational New Drug application approved by the Food and Drug Administration. The clinical investigator at each participating site will complete a Form FDA 1572, "Statement of Investigator." Copies of signed forms will be kept at the Data Coordinating Center, and original signature versions will be kept at the sponsor's institution (Dr. Mark Hall, Nationwide Children's Hospital).

14.2 Health Insurance Portability and Accountability Act

Data elements collected include the date of birth and date of admission. Prior to statistical analyses, dates will be used to calculate patient age at the time of the study events.

Data elements for race, ethnicity, and gender are also being collected. These demographic data are required for Federal reporting purposes to delineate subject accrual by race, ethnicity, and gender.

For purposes of the DCC handling potential protected health information (PHI) and producing the de–identified research data sets that will be used for analyses, all study sites have been offered a Business Associate Agreement with the University of Utah. Copies of executed Business Associate Agreements are maintained at the DCC.

14.3 Inclusion of Women and Minorities

There will be no exclusion of patients based on gender, race, or ethnicity.

14.4 ClinicalTrials.gov Requirements

This trial will be registered at ClinicalTrials.gov in accordance with Federal regulations.

14.5 Retention of Records

For federally funded studies subject to the Common Rule, records relating to the research conducted shall be retained for at least 3 years after completion of the research. Completion of the research for this protocol should be anticipated to include planned primary and secondary analyses, as well as subsequent derivative analyses. Completion of the research also entails completion of all publications relating to the research. All records shall be accessible for inspection and copying by authorized representatives of the regulatory authorities at reasonable times and in a reasonable manner [45 CFR §46.115(b)].

15 Record Access

The medical record and study files (including informed consent, permission, and assent documents) must be made available to authorized representatives of the Data Coordinating Center, upon request, for source verification of study documentation. In addition, medical information and data generated by this study must be available for inspection upon request by representatives (when applicable) of the Food and Drug Administration (FDA), NIH, other Federal funders or study sponsors, and the Institutional Review Board (IRB) for each study site.

15.1 Public Use Data Set

After subject enrollment and follow up have been completed, the DCC will prepare a final study database for analysis. A releasable database will be produced and completely de-identified in accordance with the definitions provided in the Health insurance Portability and Accountability Act (HIPAA). Namely, all identifiers specified in HIPAA will be recoded in a manner that will make it impossible to deduce or impute the specific identity of any patient. The database will not contain any institutional identifiers.

The DCC will also prepare a data dictionary that provides a concise definition of every data element included in the database. If specific data elements have idiosyncrasies that might affect interpretation or analysis, this will be discussed in the dictionary document. In accordance with policies determined by the investigators and funding sponsors, the releasable database will be provided to users in electronic form.

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