Message

From: Marc Thibonnier [/O=THERANOS ORGANIZATION/OU=FIRST ADMINISTRATIVE GROUP/CN=RECIPIENTS/CN=MTHIBONNIER]
Sent: 12/10/2009 5:05:02 PM
To: Carolyn Balkenhol [/O=THERANOS ORGANIZATION/OU=FIRST ADMINISTRATIVE GROUP/CN=RECIPIENTS/CN=Cbalkenhol]; Danise Yam [/O=THERANOS ORGANIZATION/OU=FIRST ADMINISTRATIVE GROUP/CN=RECIPIENTS/CN=Dyam]; Elizabeth Holmes [/O=THERANOS ORGANIZATION/OU=FIRST ADMINISTRATIVE GROUP/CN=RECIPIENTS/CN=Eholmes]
Subject: FW: ABA RESCUE Trial (UNCLASSIFIED)

Carolyn and Danise:

This is the trail of emails related to the services that Theranos will be providing for the ABA Rescue Trial headed by Major Kevin Chung, MD (see copy attached).

The budget for point of care analysis by Theranos is $72,000 per year for 4 years for a total of $288,000 (pages 23-26 of the grant application).

We should submit an invoice for:

- $250,000 paid upfront to cover the immediate cost of production of the readers and cartridges
- the remaining $38,000 should be paid when recruitment of the patients starts in 2010.

Because of their accounting rules, that second payment of $38,000 may have to be spread over the next 3 years, but let's summit an invoice with two payments only and see with Dr. Chung can do.

Thank you.

Marc

Marc Thibonnier, M.D., M.Sc., F.A.H.A.
Chief Medical Officer
Theranos
Office: 650-470-6192
Cell: 239-682-3791

------ Forwarded Message
From: "Chung, Kevin K MAJ MIL USA MEDCOM AISR" <KEVIN.CHUNG@US.ARMY.MIL>
Date: Mon, 30 Nov 2009 19:04:38 -0600
To: <mthibonnier@theranos.com>
Subject: Re: ABA RESCUE Trial (UNCLASSIFIED)

I am not back in my office until the 15th...but in the meantime, if you could send me an invoice with a reasonable quote for years 1-4, I think we can get things rolling.

K
Kevin Chung, MD
Medical Intensivist

Sent from BlackBerry

From: Marc Thibonnier
To: Chung, Kevin K MAJ MIL USA MEDCOM AISR
Sent: Mon Nov 30 18:58:36 2009
Subject: Re: ABA RESCUE Trial (UNCLASSIFIED)
Hi Kevin:

Thanks for the good news. As you know, the total contract with Theranos is for $288,000, most of it will be spent upfront for production of the readers and the cartridges. So indeed, "a big chunk upfront" will certainly be welcome.

Let me know when we can talk about it this week.

Take care,

Marc

On 11/24/09 8:39 AM, "Chung, Kevin K MAJ MIL USA MEDCOM AISR" <KEVIN.CHUNG@US.ARMY.MIL> wrote:

Classification: **UNCLASSIFIED**

Caveats: NONE

Marc,

The money is in for the ABA RESCUE trial. I need to know what your upfront costs will be to preposition analyzers in at the 11 participating sites.

Can't give you an advance payment for all 4 years but I can try to get you a big chunk upfront for set-up costs and such.

Let me know.

KC

Classification: **UNCLASSIFIED**

Caveats: NONE

Marc Thibonnier, M.D., M.Sc., F.A.H.A.
Chief Medical Officer
Theranos
Office: 650-470-6192
Cell: 239-682-3791

------ End of Forwarded Message

TITLE OF PROJECT: The ABA RESCUE Trial: The **A**merican **B**urn **A**ssociation **R**andomized controlled **E**valuation of high-volume hemofiltration in adult burn patients with **S**eptic sho**C**k and mild ac**U**te kidn**E**y injury

Principle Investigator: Kevin K. Chung, MD

Co-Investigator: Amy M. Sprague, MD, CTI, Augusta Georgia

Mailing Address:
US Army Burn Center
US Army Institute of Surgical Research
3400 Rawley E. Chambers Avenue
Fort Sam Houston, TX 78234-6315

Telephone: 210-916-3301 **Facsimile:** 210-271-0830

E-mail: kevin.chung@us.army.mil

This proposal is for: Multicenter randomized clinical trial

Anticipated Length of Project to completion: 4 years

Anticipated Budget (including indirect costs – 15% overhead):
Year One: **$820,175**
Year Two: **$699,806**
Year Three: **$711,767**
Year Four: **$725,231**
Total: **$2,956,980**

Brief Synopsis of Project:
This is a multicenter non-blinded randomized controlled trial comparing intervention with early high-volume hemofiltration (HVHF) plus 'contemporary' care versus 'contemporary' care alone in critically ill adult burn patients who develop septic shock with mild acute kidney injury (AKI) to be performed in 10 US burn centers. Simply, HVHF is Continuous Venovenous Hemofiltration (CVVH) applied at 2-3 times the normal 'renal' dose. This technique has been widely reported as having positive hemodynamic, pulmonary, and immuno-protective effects in sepsis mediated shock in both animal and human studies.[1] To date, no randomized prospective trials of similar design and patient population have been performed. To maintain clinical equipoise, our plan is to recruit burn centers that do not utilize HVHF as part of their routine clinical care. We will enroll patients who exhibit signs and symptoms of septic shock (requiring vasopressor support despite adequate volume expansion) with early and mild AKI and randomize them to treatment with early HVHF in addition to 'contemporary' care verses 'contemporary' care alone which will reflect sepsis and AKI management based on the most recent clinical practice guidelines. As part of 'contemporary' care for sepsis, a burn specific 24-hour sepsis bundle based on the Surviving Sepsis Campaign will be applied.[2] The primary endpoint for this study is vasopressor-free days in the first 14 days, as defined as the number of days (24 hours) the study patients are alive and free on any vasopressor support. Major secondary

endpoints will be 14 day mortality, 28 day mortality, and in-hospital mortality. Sixty patients in each arm are required to detect a difference of 2 vasopressor-free days with adequate power (>80%).

The ABA RESCUE Trial

The **A**merican **B**urn **A**ssociation **R**andomized controlled **E**valuation of high-volume hemofiltration in adult burn patients with **S**eptic sho**C**k and mild ac**U**te kidn**E**y injury

I. Statement of Work

This is a multicenter non-blinded randomized controlled trial comparing intervention with high-volume hemofiltration (HVHF) plus 'contemporary' care versus 'contemporary' care alone in critically ill burn patients who develop septic shock with mild acute kidney injury (AKI). We intend to recruit 10 burn centers that do not prescribe HVHF as part of their standard care. We will enroll patients who exhibit signs and symptoms of septic shock (American Burn Association Consensus definition[2]) with early and mild AKI (AKIN Stage I[2]). Both groups will receive 'contemporary' care for septic shock as defined by adherence to a burn specific 24-hour sepsis bundle based in the Surviving Sepsis Campaign recommendations. In addition to 'contemporary' care, those assigned to the treatment group will be initiated on HVHF at an ultrafiltration dose of at least 70 ml/kg/hr for a total duration of 72 hours. Management of AKI and the application of renal replacement therapy for renal indications will be based on institution-specific customary practice and will be applied equally to both groups. This study is specifically designed to assess the efficacy of early HVHF as an adjunctive therapy in septic shock in a critically ill burn population managed in accordance with the Surviving Sepsis Campaign guidelines. The primary endpoint for this study is vasopressor-free days in the first 14 days, as defined as the number of days (24 hours) the study patients are alive and free on any vasopressor support. Major secondary endpoints will be 14 day mortality, 28 day mortality, and in-hospital mortality.

This trial will be conducted in accordance with the policies and procedures of the ABA-MultiCenter Trials Group (ABA-MCTG) which can be found via the following link (www.ameriburn.org).

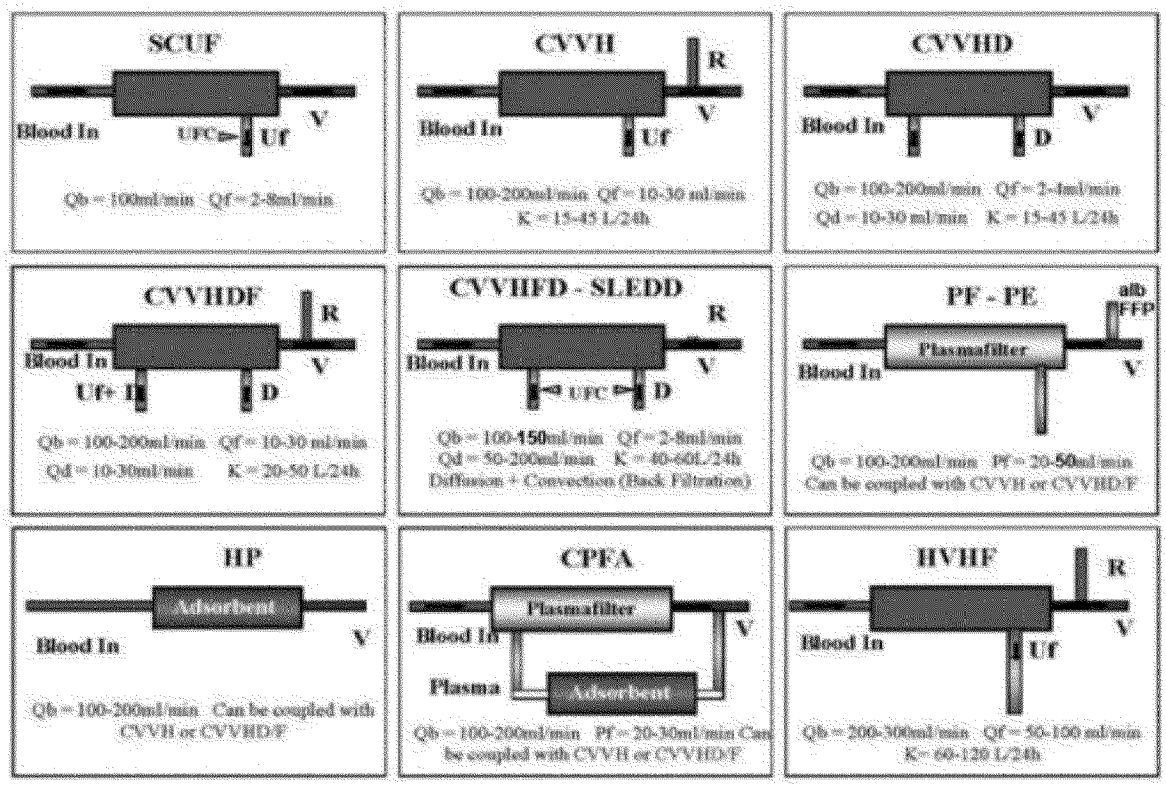
II. Background and Significance

The importance of sepsis and infection in patients with severe burns was highlighted by the recent American Burn Association Consensus Conference to standardize definitions for future studies.[3] Sepsis is common, with a reported incidence of 2.26 cases per 100 hospital discharges, expensive, costing $22,100 per case, and deadly with an average mortality of 28.6%.[5] Septic shock, the most severe form of sepsis, is defined as persistent hypotension despite adequate fluid resuscitation and/or Lactate >4mmol (36 mg/dl).[3, 4] Development of septic shock often leads to organ dysfunction and death.[6-8] The epidemiology of severe sepsis and septic shock in burn patients is not abundantly described in the literature. Recently, authors reported findings of a one year prospective observational study conducted at Parkland Memorial Hospital. Of the 85 patients admitted with 20% Total Body Surface Area (TBSA) burns, 12 (14%) developed septic shock.[5] The same group reported that mortality associated with septic shock to be 63%.[5]

Acute kidney injury is also a common and devastating complication in critically ill burn patients with mortality reported to be between 80 and 100%.[6-10] Despite recent advances in burn care, the unacceptably high mortality rate in this subgroup has not changed over time. The pathogenesis of

AKI in burns, similar to other critically ill populations, is often multi-factorial with one major component being sepsis induced ischemic tubular necrosis. Thus, AKI secondary to septic shock is a common and devastating condition in the burn ICU.

Extracorporeal blood purification, originally developed for the treatment of renal failure, has recently evolved to encompass various techniques applied to the treatment many extra-renal problems in the critically ill.[1, 11-14] Continuous Renal Replacement Therapy (CRRT) is a method to achieve extracorporeal blood purification and is an umbrella term use to describe a variety of continuous modes of renal support, to include Slow Continuous Ultrafiltration (SCUF), CVVH, CVVHD, CVVHDF, and C-SLED. CRRT generally describes modalities applied to support patients who develop renal failure. Other extracorporeal blood purification techniques exist to treat a variety of conditions, to include plasmapheresis, plasma-exchange, hemoperfusion, and HVHF. These techniques along with all the CRRT modalities are illustrated on the next page.[15]



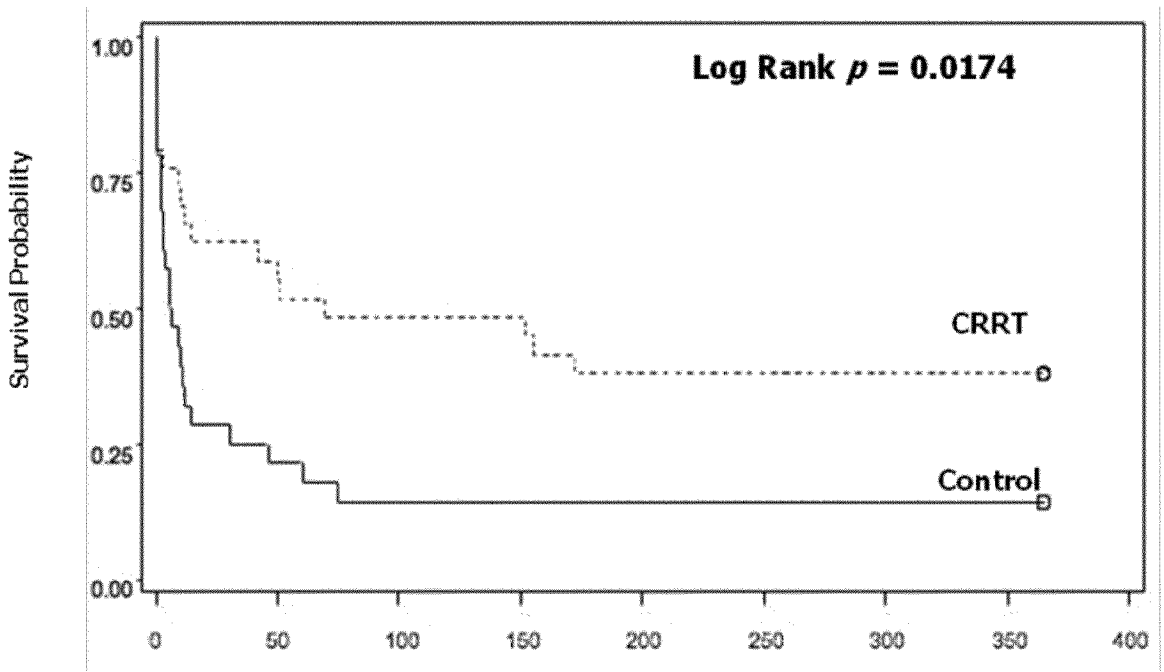
Different extracorporeal therapy techniques
Blood In: blood inlet; SCUF: slow continuous ultrafiltration; CVVH: continuous venovenous hemofiltration; CVVHD: continuous venovenous hemodialysis; CVVHDF: continuous venovenous hemodiafiltration; CVVHFD-SLEDD: continuous venovenous high-flux dialysis-sustained low efficiency daily dialysis (6-10 h duration); PF-PE: continuous plasmapheresis-plasma exchange; HP: continuous hemoperfusion; CPFA: coupled plasmafiltration adsorption; HVHF: high-volume hemofiltration (applied continuously or as short pulses); Qb: blood flow rate; Qd: dialysate flow rate; Qf: ultrafiltration rate; K: urea clearance; R: replacement solution; Uf: ultrafiltrate; D: dialysate; UFC: ultrafiltration control; V: venous return; alb: albumin; FFP: fresh frozen plasma

Of the various techniques available to potentially treat septic shock, the most well described, safe, practical and immediately available is HVHF.[1, 15, 17-19] On the surface, hemofiltration shares many similarities to hemodialysis. Both techniques utilize an extracorporeal circuit that directs blood through

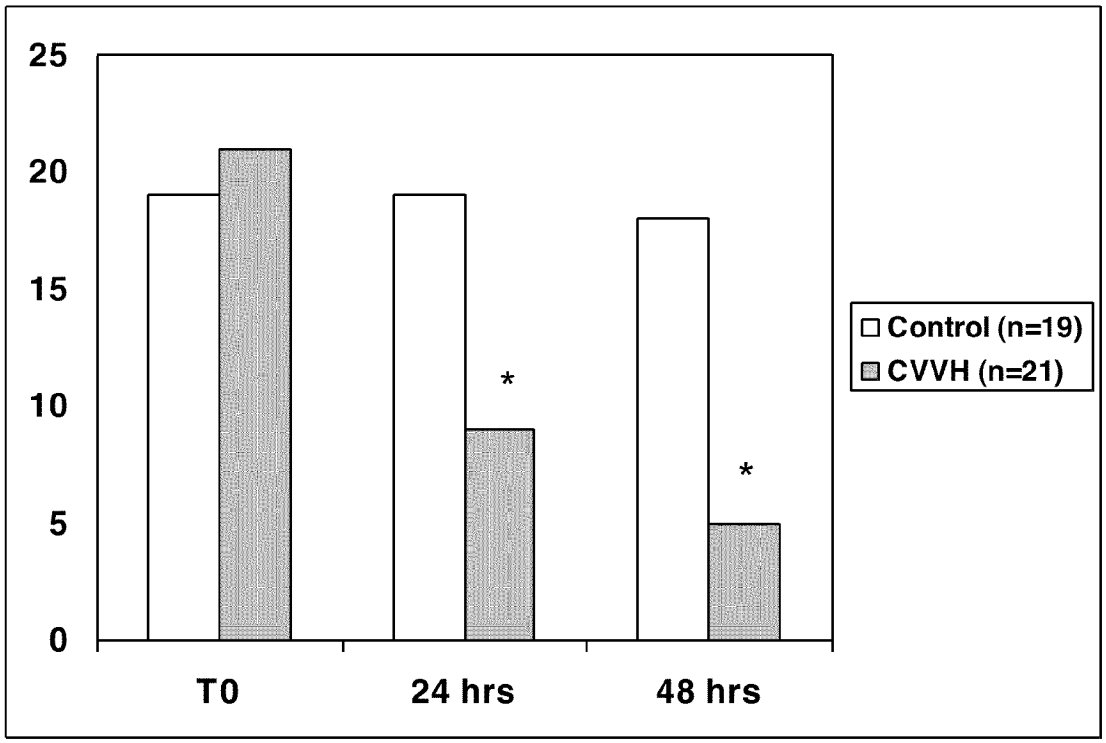
a hemofilter. Hemofiltration is a convective clearance of solutes across a membrane, by a continuous negative pressure that pulls ultrafiltrate from the serum across a semi-permeable membrane, thus 'dragging' and essentially clearing unwanted solutes from the blood. In contrast, dialysis accomplishes this through use of a countercurrent of an 'ideal' fluid (dialysate) which creates an osmotic gradient that facilitates the movement of molecules down the concentration gradient. The complex pathogenesis of sepsis is a systems level interplay between the immune, neurological, endocrine and coagulation systems all triggered by the recognitions of a pathogen by the innate immune system.[22, 23] The complexity of this system, perhaps explains the lack of success demonstrated by the specific cytokine targeting approach devised and clinically tested over the last two decades.[16] Several experimental and clinical studies have suggested that hemofiltration at sufficient doses can 'clear' inflammatory mediators in septic shock, and that this non-specific 'cytokine removal' is associated with positive physiologic effects including improvement of hemodynamic instability and other important clinical outcomes.[1, 15, 17-19, 21]

Our group recently demonstrated an absolute reduction of 55% and 32% in 28-day mortality and hospital mortality in critically ill burned military casualties with acute kidney injury (AKI) (n=18) aggressively treated with high volume continuous venovenous hemofiltration (CVVH) when compared to a closely matched historical cohort (n=16).[22]

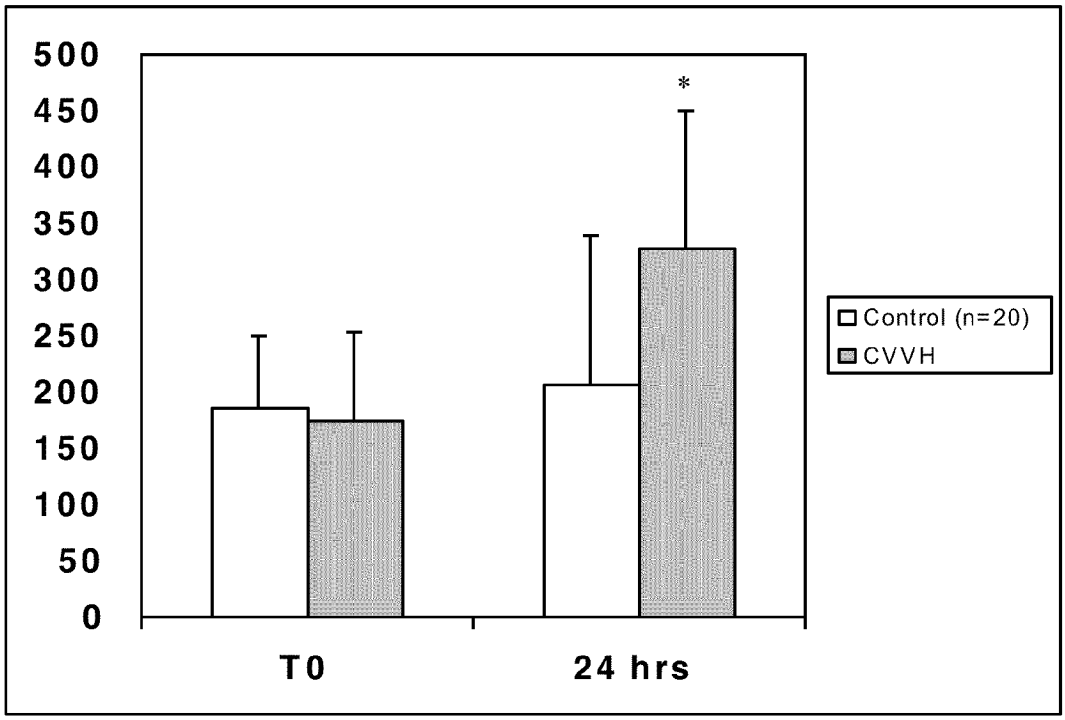
We have recently performed a repeat analysis, this time including all civilian patients, that demonstrates similar results with a 33% and 23% decrease in 28-day and hospital mortality (total n=57).[Chung et al, submitted to Crit Care] A Kaplan-Meier Estimates of Survival between the two groups demonstrates that CVVH was associated with a significantly higher rate of survival out to beyond a year, with the majority of the benefit realized in the first month.



In a subgroup of patients with shock (n=21), a majority of them with septic shock, we observed a dramatic reduction in the vasopressor requirement at 24 and 48 hours that did not exist in the historical cohort. (*$p < 0.05$ both to baseline and between groups.)



In those with Acute Lung Injury/Acute Respiratory Distress Syndrome (ALI/ARDS), there was a significant increase from baseline in the partial-pressure-of-arterial-oxygen to fraction-of-inspired-oxygen ratio (PFR) at 24 hours in the CVVH group (n=16, 174+/-78 to 327+/-122, p=0.003) but not the control group (n=20, 186+/-64 to 207+/-131, p=0.98).



Thus, there is a strong indication that the survival benefit realized was a result of the treatment effect demonstrated in the specific subgroup of burn patients with shock and/or ALI/ARDS as confounders to their AKI.

We are not alone in this observation. Piccini, et al. described improvement in hemodynamics, gas exchange, and 28 day survival compared to historical controls after the institution of early, isovolemic hemofiltration for the treatment of oliguric patients with septic shock. [17] The 28 day survival of 55% was significantly higher than in the historical control arm (27%, $p<0.05$) The authors hypothesized that early, high volume hemofiltration may non-specifically affect mediators (both pro-inflammatory and anti-inflammatory) and improve outcomes by modulating both early, multiple organ dysfunction due to systemic inflammation and allowing for increased immunocompetence later in the course of sepsis.

Ronco et al reported in a randomized controlled study that increasing the dose of hemofiltration from 20 ml/kg/hr to 35 ml/kg/hr improved survival in ICU patients with acute renal failure.[18] It is interesting to note that in a third group of patients who received a higher dose of hemofiltration (45 ml/kg/hr) there was a significantly increased survival in the subgroup of patients with sepsis.

A few others have reported similar results using a 'pulse' high dose hemofiltration technique in sepsis.[19, 20]

In a multicenter prospective randomized trial involving 61 patients with shock after out-of-hospital cardiac arrest, high-volume hemofiltr ation (200 ml/kg/hr) was associated with improved 6-month survival (OR, 4.4; 95% CI, 1.1-16.6) when compared to controls.[21] This study demonstrates the efficacy and long term safety of this therapy in a non-specific shock state at a very high dose of therapy.

Together, there is a strong signal that this mode of therapy is efficacious in various shock states and safely administered even at the highest doses. Based on these findings a large multi-center prospective randomized trial is already underway in Europe. The IVOIRE (hIgh VOlume in Intensive caRE) study will compare early high volume hemofiltration (70 ml/kg/hr) versus a standard volume hemofiltration (35 ml/kg/hr) in patients with septic shock and mild AKI (NCT00241228, clinicaltrials.gov) in a mixed ICU population. As of March 2009, they have enrolled 109 patients and plan to do an interim analysis when they reach 120 patients. Target enrollment remains 240 patients. [Honore, P. Personal communications 2009].

It is important to note that 'contemporary' care of severe sepsis and septic shock has evolved over the past 5 years. Recently, critical care and infectious disease experts representing 11 international organizations developed and published a consensus management document with various recommendations in the management of severe sepsis and septic shock.[4] From these guidelines, two sepsis 'bundles' were created, a sepsis resuscitation bundle and a sepsis management bundle, that consolidated various evidenced based elements important to improving outcomes in these patients.[22] The sepsis resuscitation bundle (also called the 6-hour bundle) combines 5 performance tasks to be accomplished in the initial management period, to include the measurement of lactate, blood cultures prior to antibiotic administration, early antibiotic administration, management of hypotension with volume and vasopressors, and application of CVP (8 mmHg) and ScvO2 (>70%) goals as endpoints. The sepsis management bundle (also called the 24-hour bundle) combines 4 elements to include consideration for low-dose steroids as indicated, consideration for recombinant human activated protein C as indicated, tight glycemic control, and application of lung protective ventilation. Evidence suggests that adaptation of and compliance with an institution-specific 'bundle' based on these guidelines in the management of patients with severe sepsis and septic shock is associated with a significant improvement in in-hospital mortality.[28, 29]

We believe critically ill burn patients who develop septic shock along with AKI have a high risk of death. Our data suggests a number needed to treat of 4 patients for one life saved to hospital discharge. It is against this backdrop that we propose a multicenter trial in this high risk population to compare pulse high-volume hemofiltration along with 'contemporary' care against 'contemporary' care alone.

III. Objectives/Hypothesis

Our overall hypothesis is that **High-Volume Hemofiltration in addition for 'contemporary' care will result in an <u>improvement of all measured clinical outcomes</u> when compared to 'contemporary' care alone in the treatment of critically ill patients with septic shock with mild AKI**. The specific aims are:

<u>Specific Aim 1:</u> To evaluate pertinent physiologic outcomes with HVHF.

Hypothesis 1: The HVHF group will have a least two more vasopressor-free days in the first 14 days when compared to contemporary care alone.

Hypothesis 2: The HVHF group will have a significantly better pulmonary function (as evidenced by the Oxygenation Index) at 24, 48, 72 hours and 7 and 14 days when compared to contemporary care alone.

Hypothesis 3: The HVHF group will have significantly lower MODS scores at 24, 48, 72 hours and 7 and 14 days when compared to contemporary care alone.[3]

Specific Aim 2: To evaluate pertinent clinical outcomes.

Hypothesis 1: The HVHF group will have a significantly lower 14-day mortality rate when compared to contemporary care alone.

Hypothesis 2: The HVHF group will have a significantly lower 28-day mortality rate when compared to contemporary care alone.

Hypothesis 3: The HVHF group will have a significantly lower in-hospital mortality rate when compared to contemporary care alone.

Specific Aim 3: To assess the molecular pathophysiologic effects of HVHF.

Hypothesis 1: HVHF will result in significantly altered serum levels of various pro and anti inflammatory mediators when comparing various time points to baseline or to the control group (TNF-α, IL-6, IL-8, IL-10, IL-12, and IFN-γ).

Hypothesis 2: Analysis of the ultrafiltrate fluid will reveal significant levels of various pro and anti inflammatory mediators.

IV. Technical Objectives

Objective 1: Ensure at least 70% compliance to the burn specific sepsis bundle in both groups.
Objective 2: Ensure at least a 90% compliance with the treatment dose of 70 ml/kg/hr (delivered) for 72 hours in the HVHF group.

V. Project Milestones

June 2009 – Selection of sites who meet criteria for participation in this study. On-site PI will also be identified.
July 2009 – Begin Institutional Review Board approval process at each site.
July-October 2009 – Study site visits by co-Investigators for site investigator and coordinator training.
October 2009 – Begin enrolling patients at sites with previous CRRT capability.
October 2011 – Perform interim analysis.
October 2013 – Study completion.

VI. Military Significance

Severely burned soldiers often develop septic shock and AKI as complications of their injury and prolonged hospitalization. In both burn and general multi-trauma populations, the development of septic shock and AKI is associated with a high likelihood of death. Since November, 2005 the US Army Burn Center began to treat patients with CVVH for AKI and have observed, like others, an extra-renal benefit. Although it is now standard of care for many trauma centers and our ICU, CVVH has not been widely adopted throughout the military. Application of this mode of therapy as an adjunctive treatment for septic shock is considered novel and not performed on a routine basis. Findings from this study and future studies in this field have the potential to change practice in all medical facilities in the Department of Defense and impact the care of the combat wounded at the highest risk of death.

VII. Public Purpose

Severe sepsis and septic shock remains one of the most important causes of mortality in the ICU. As already stated, sepsis is common, with a reported incidence of 2.26 cases per 100 hospital discharges, expensive, costing $22,100 per case, and deadly with an average mortality of 28.6%.[23] Due to a progressively aging population and advances in other areas of medicine continuing to

impact life expectancy, we can expect these numbers to rise in the future. Despite the many advances in this field outlined in the current Survival Sepsis Campaign guidelines, there is significant room for improvement. Multiple clinical trials have evaluated pharmacologic and other therapeutic approaches that have targeted specific key mediators in sepsis without success. A non-specific 'systems' approach to modulate the inflammatory response appears to be the logical next step. A recent review highlighted hemofiltration as a potential novel therapy that needs further investigation.[11] The burn population is an ideal population to study this type of therapy due to the relatively homogenous nature of the population as well as the relatively high mortality rate when septic shock ensues. It is easier to demonstrate benefit when mortality is high. Thus for a therapy such as high-volume hemofiltration, the most ideal population would be one that has the highest mortality rate. The severe burn patient who subsequently develops septic shock fits that bill. Furthermore, the burn model is widely considered the universal model for trauma. This type of intervention, if proven to be beneficial in the burn population has potential applicability in vast number of other populations at high risk of death. The significance of this type of study in the field of critical care and extracorporeal blood therapy cannot be overstated.

VIII. Methods

A. Study design and treatment protocols

Design: This is an interventional multicenter randomized controlled study to be conducted at approximately 10 ten burn centers that currently do not prescribe HVHF as part of their standard practice. Centers will begin patient enrollment immediately upon approval of the protocol by the IRB and protocol education of all the staff. Once patients are deemed eligible and agree to participate (via surrogate consent in most cases) they will be randomized to therapy (with HVHF) in addition to 'contemporary' care or 'contemporary' care only.

Septic shock: Patients will be screened on a daily basis to determine if they meet criteria for septic shock as recently defined by the ABA specific for the burn patient.[3] Sepsis is defined as a change in the burn patient that triggers the concern for infection. The trigger includes at least three of the following.

- Temperature >39 degrees or 36.5 degrees C
- Progressive tachycardia > 110 bpm
- Progressive tachypnea >25 bpm
- Thrombocytopenia <100,000/mcl
- Hyperglycemia (in the absence of pre-existing diabetes mellitus)
    - Untreated plasma glucose>200 mg/dl
    - Insulin resistance
- Inability to continue enteral feedings >24 hours
    - Abdominal distension
    - Enteral Feeding intolerance (residuals 2X the feeding rate in adults)
    - Uncontrolled diarrhea (>2500 ml/day)

In addition, the following documentation are required.

- Culture positive infection
- Pathologic tissue source
- Clinical response to antimicrobials

Septic shock is defined as 'sepsis' as above in addition to persistent hypotension despite adequate fluid resuscitation and/or a serum Lactate of >4 mmol. For the purposes of this study, anyone with circulatory shock secondary to 'presumed sepsis' will be included.

Acute Kidney Injury: (See section XIII for Acute Kidney Injury Network staging system)[24] Once patients are identified as being in septic shock, their renal function will be assessed and classified. In order to be enrolled into the study, patients must be at least an AKIN stage I. This corresponds to a serum creatinine increase of at least 0.3 ml/dl or a urine output less than 0.5 ml/kg/hr for greater than 6 hours.

Contemporary care: Both groups will receive 'contemporary' care by applying the Burn-specific Sepsis Bundle adapted from the most recent Surviving Sepsis Campaign (SSC) recommendations and specifically modified to our patient population.[4] This will ensure **benefit to all participants** regardless of randomization group. The following section describes in detail what this involves.

Burn-Specific Sepsis Bundle (six elements)
This bundle consists of six elements extracted from the 6 and 24 hour bundles advocated for the optimal treatment of patients in septic shock and slightly modified for the purposes of this study. The following elements will be followed and tracked. Lactate will be drawn at the time of randomization. CVP (if a central venous catheter is in place - preferred) will be measured at the time of randomization. A central line in the subclavian or internal jugular vein is preferred but not required. If the CVP is less than 8, the patient will receive a fluid challenge (crystalloid or colloid) with the intent of raising the CVP above a specific goal *as determined by the bedside clinician*. A central venous saturation (ScvO2) will be measured (via continuous measurement or a spot venous blood gas). If the ScvO2 is <70% the patient will be given up to 2 units of PRBCs if the hemoglobin is less than 7 mg/dl. If the ScvO2 is still less than 70%, the patients will be initiated on dobutamine at a low dose at the discretion of the bedside provider. The administration of low-dose steroids will also be at the discretion of the bedside provider. For the purpose of the study, as long as low-dose steroids has been considered, then the requirement per the Burn-Specific Sepsis Bundle will be met. Patients can receive 100 mg of hydrocortisone every 8 hours for a minimum of three doses and then stopped. There will be no cosyntropin stimulation test required for the study. Continuation or weaning of steroids will be at the discretion of the bedside provider. Every attempt will be made to achieve reasonable glucose control utilizing the already existing institution insulin/glucose protocols. If the patient is on mechanical ventilation, every effort will be made to maintain plateau pressures less than 30 mmHg. Three elements on the original 6 and 24 hour bundles were left out. It is assumed that 'standard' care of all patients at the participating burn centers involves timely initiation of antibiotics and the acquisition of blood cultures prior to the initiation of antibiotics. Assuming patients enrolled in this study will be outside the 6 hour window at the time of randomization, it is reasonable to exclude it from this protocol. The prescription of Activated Protein C, due to the bleeding risk, should be considered on a case by case basis at the discretion of the provider. Compliance with this bundle will be recorded using a simple collecting tool similar to the one below at T+1. Our stated objective is compliance with this bundle for both groups of >70%.

Y/N	Lactate measured?
Y/N	CVP>8 mmHg?
Y/N	ScvO2>70%?
Y/N	Low-dose steroids considered?
Y/N	Serum glucose < 150 within 24 hours?
Y/N	Plateau Pressure < 30 within 24 hours?

It is recommended that nephrology consultation be requested for patients considered to be candidates for renal replacement (for renal indications) in either group. Initiation of renal replacement outside the 'intervention' in the HVHF group and in the 'contemporary' care only arm will be determined in accordance with nephrology recommendations or ICU-specific normal practices.

Intervention group: After consent has been obtained, those randomized to the treatment group will undergo placement of a central venous access. A double lumen dialysis catheter, minimum 13.5 french or larger, will be placed in the internal jugular or femoral vein. This will ensure optimum performance of the therapy. High-volume hemofiltration using the NxStage System One (Lawrence, MA), via 1.5m2 Polyethelsulfone filter, (or comparable institution-specific machine) at a prescribed dose 70 ml/kg/hr will be infused pre-filter for a duration of **72 hours**. No regional anticoagulation will be needed. The filter will be changed every 24 hours to optimize the adsorptive effects of the filter. Interruption of therapy due to clogging of the filter, patient transfers, or operative procedure will be recorded daily. For simplicity, no attempt should be made to 'catch-up' with a higher dose when the patient is placed back on therapy. Patients will be continued on therapy until the 3th day. After **72 hours** of therapy, regardless of patient status, treatment at the *study dose* (70 ml/kg/hr) will be discontinued. From that point on, the bedside clinicians, with or without consultation with nephrology, will decide the dose, mode (CVVH, CVVHD, IHD, or SLED), and duration of therapy based on institution-specific standard practice based on the patient's condition. The control arm may receive any mode of renal replacement therapy at any time at the standard doses usually prescribed for severe AKI under the direction of nephrology and/or critical care staff. The maximum dose used for renal replacement should be 35 ml/kg/hr in most centers. This standard practice will be one of the criteria used for site selection. (See next section)

Site selection criteria:
It is vital that we select the appropriate sites for participation in this study. At a minimum, participating center must have the following characteristics.

- Burn center with at least 350 patient admissions per year.
- Identification of study advocates who will act as the on-site investigator.
- Existing CRRT capability with volume of at least 2 treatments a month.
- Existing research support infrastructure (research nurse support with availability for patient enrollment and data collection)
- Capability to perform HVHF (70 ml/kg/hr) with existing machines.
- No prescription of HVHF (as defined as use of a dose greater than 35 ml/kg/hr) as part of standard practice for extra-renal indications.

All interested sites will be screened in order to determine eligibility as one of our 10 participating site. The US Army Burn Center and Joseph Still Burn Center will be excluded from patient enrollment due to heavy institutional bias in favor of HVHF in septic shock.

Cytokine Assay:
There are more than 100 cytokines that function as short-range mediators of many biological processes like inflammation, infection and immune responses.[25] Pro- and anti-inflammatory cytokines are produced during sepsis and the study of their production and trends has both diagnostic and predictive values. In this proposal, we will measure panels of cytokines reflecting inflammation (TNF-α, IL-6, IL-8), type 1 helper T cells (IFN-γ, IL-12), type 2 helper T cells (IL-10), and chemokines (IL-8) at the bedside of the patients with the integrated TheranOS system (Palo Alto, CA) described in section XI. Each analyte costs $25, totaling $72,000 a year in analytical costs for 30 patients sampled 16 times (10 serum and 6 ultrafiltrate fluid) for 6 analytes. This system is a point of care device that

requires less than 3 ml of blood per cartridge, containing all 6 analytes. Results are available **within the hour** and saved in the internal hard drive of the TheranOs system.

B. Study population

Adult patients admitted to a burn center after sustaining a burn injury of any size.

C. Subject recruitment

Subjects eligible for this study are unlikely to be able to provide their own consent. Surrogate consent will be obtained as necessary. Potential subjects will be screened for participation in this study by an onsite research coordinator on a daily basis. The research coordinator or on-site PI will approach the patient or surrogate and obtain informed consent. Study participation will begin when a subject or legally authorized representative consents to participate in the study. Participation ends if the patient is withdrawn from the study, transfers out of the hospital, or dies.

Randomization: Patients meeting inclusion and exclusion criteria will be randomized to either the HVHF arm or the Control arm by paired groupings to ensure equal distribution of burn size and age. Computerized randomization will occur at the Data Coordinating Center (DCC) and performed by the Director of Research Operations (DROP). After identification within one of the defined groupings below, the computer will randomly identify the study arm. The subsequent patient enrolled within that same group will be in the opposite study arm (i.e. – Control vs HVHF). After each complete pair, a new random identification will be performed.

Group 1: age 18-65 and <40%TBSA
Group 2: age 18-65 and >40%TBSA
Group 3: age >65 and <40%TBSA
Group 4: age >65 and >40%TBSA

D. Inclusion criteria

- All adult patients admitted to the burn ICU with burns of any size who develop septic shock with concurrent AKI (at least AKIN stage 1) >48 hours after admission. AKIN staging is described in Section XIII reproduced by Mehta et al.[24]
- Patients 18 or older

E. Exclusion criteria

- Age <18
- Non-thermal injury (exfoliating skin disorders or necrotizing fasciitis)
- Pre-admission diagnosis of end stage renal failure
- Diagnosis of acute kidney injury (AKIN stage 3) with anticipation for renal replacement therapy

F. Duration

4 years

G. Baseline data collection and monitoring

Baseline admission data as well as baseline data at the time of randomization (T0) will be collected and recorded at that time on data sheets that will be faxed or scanned and e-mailed to the ABA Director of Research Operations (DROP) within 48 hours of randomization. Subsequent data sheets on days T+1, T+2, T+3, T+7, T+14 and T+28 will also be sent to the database manager for

processing all within 48 hours of collection.

1. The following ADMISSION characteristics will be collected within 24 hours of randomization.
   - Register/hospital number, Age, Gender
   - Admission weight
   - Percent burn on admission, Percent full thickness burn on admission
   - Smoke inhalation injury (Y/N)
   - Injury Severity Score (on admission)
   - Urine output (24 hours prior to randomization)
   - Total Fluids in (24 hours prior to randomization)

2. The following data will be collected at the time of randomization (T0).
   - Modified Marshall Score (on day of randomization)
   - PaO2, FiO2, Ventilator mode, Tidal Volume, Mean Airway Pressure, PEEP
   - Heart Rate, Systolic Blood Pressure, Diastolic Blood Pressure, Mean Arterial Pressure
   - Type of central venous access
   - CVP, ScvO2
   - Type of vasopressor and dose
     - Norepinephrine (mcg/min)
     - Vasopressin (Units/min)
     - Dobutamine (mcg/kg/min)
     - Neosynephrine (mcg/min)
     - Epinephrine (mcg/min)
     - Dopamine (mcg/kg/min)
     - other
   - Total bilirubin, Platelet count, Blood urea nitrogen, Creatinine, Lactate, serum glucose
   - Serum Cytokine Panel (TNF-α, IL-6, IL-8, IL-10, IL-12, and IFN-γ)
   - For HVHF Group
     - Hemofiltration Access (Femoral, IJ)
     - Prescribed Blood Flow Rate
     - Prescribed Replacement Fluid Rate
     - Prescribed fluid removal
     - Ultrafiltrate Cytokine Panel
3. The following data will be collected at **24 hours** from time of randomization (T+1).
   - Burn Sepsis Bundle Compliance (Y/N)
     - Antibiotics initiated <6 hours? (Y/N)
     - Blood cultures taken before antibiotics? (Y/N)
     - Serum lactate measured? (Y/N)
     - CVP>8 mmHg? (Y/N)
     - ScvO2>70%? (Y/N)
     - Low-dose steroids? (Y/N)
     - Serum glucose < 150 within 24 hours? (Y/N)
     - Plateau Pressure < 30 within 24 hours? (Y/N)
   - Modified Marshall Score
   - PaO2, FiO2, Ventilator mode, Tidal Volume, Mean Airway Pressure, PEEP

- Heart Rate, Systolic Blood Pressure, Diastolic Blood Pressure, Mean Arterial Pressure
- Type of central venous access
- CVP, ScvO2
- Total urine output in last 24 hours
- Total fluids in last 24 hours
- Type of vasopressor and dose
  - Norepinephrine (mcg/min)
  - Vasopressin (Units/min)
  - Dobutamine (mcg/kg/min)
  - Neosynephrine (mcg/min)
  - Epinephrine (mcg/min)
  - Dopamine (mcg/kg/min)
  - other
- Total bilirubin, Platelet count, Blood urea nitrogen, Creatinine, Lactate, serum glucose
- Rh-APC initiated? (Y/N)
- Serum Cytokine Panel (at hours12 and 24)
- For HVHF Group
  - Mean Blood Flow Rate
  - Total Replacement Fluid Infused in last 24 hours
  - Total volume removed in last 24 hours
  - Number of filter changes in last 24 hours
  - Total hours OFF therapy
  - Ultrafiltrate Cytokine Panel (at hours 12 and 24)

4. The following data will be collected at **48 hours** from time of randomization (T+2).
   - PaO2, FiO2, Ventilator mode, Tidal Volume, Mean Airway Pressure, PEEP
   - Heart Rate, Systolic Blood Pressure, Diastolic Blood Pressure, Mean Arterial Pressure
   - Type of central venous access
   - CVP, ScvO2
   - Total urine output in last 24 hours
   - Total fluids in last 24 hours
   - Type of vasopressor and dose
     - Norepinephrine (mcg/min)
     - Vasopressin (Units/min)
     - Dobutamine (mcg/kg/min)
     - Neosynephrine (mcg/min)
     - Epinephrine (mcg/min)
     - Dopamine (mcg/kg/min)
     - Other
   - Modified Marshall Score
   - Total bilirubin, Platelet count, Blood urea nitrogen, Creatinine, Lactate, serum glucose
   - Serum Cytokine Panel (at hours 36 and 48)
   - For HVHF Group
     - Mean Blood Flow Rate

  - Total Replacement Fluid Infused in last 24 hours
  - Total volume removed in last 24 hours
  - Number of filter changes in last 24 hours
  - Total hours OFF therapy
  - Ultrafiltrate Cytokine Panel (at hours 36 and 48)

5. The following data will be collected at **72 hours** from time of randomization (T+3)
   - PaO2, FiO2, Ventilator mode, Tidal Volume, Mean Airway Pressure, PEEP
   - Heart Rate, Systolic Blood Pressure, Diastolic Blood Pressure, Mean Arterial Pressure
   - Type of central venous access
   - CVP, ScvO2
   - Total urine output in last 24 hours
   - Total fluids in last 24 hours
   - Type of vasopressor and dose
     - Norepinephrine (mcg/min)
     - Vasopressin (Units/min)
     - Dobutamine (mcg/kg/min)
     - Neosynephrine (mcg/min)
     - Epinephrine (mcg/min)
     - Dopamine (mcg/kg/min)
     - other
   - Modified Marshall Score
   - Total bilirubin, Platelet count, Blood urea nitrogen, Creatinine, Lactate, serum glucose
   - Serum Cytokine Panel (at hours 60 and 72)
   - For HVHF Group
     - Mean Blood Flow Rate
     - Total Replacement Fluid Infused in last 24 hours
     - Total volume removed in last 24 hours
     - Number of filter changes in last 24 hours
     - Total hours OFF therapy
     - Ultrafiltrate Cytokine Panel (at hours 60 and 72)
6. The following data will be collected at **7 days** from time of randomization (T+7).
   - PaO2, FiO2, Ventilator mode, Tidal Volume, Mean Airway Pressure, PEEP
   - Heart Rate, Systolic Blood Pressure, Diastolic Blood Pressure, Mean Arterial Pressure
   - Type of central venous access
   - CVP, ScvO2
   - Type of vasopressor and dose
     - Norepinephrine (mcg/min)
     - Vasopressin (Units/min)
     - Dobutamine (mcg/kg/min)
     - Neosynephrine (mcg/min)
     - Epinephrine (mcg/min)
     - Dopamine (mcg/kg/min)
     - other
   - Modified Marshall Score

- Total bilirubin, Platelet count, Blood urea nitrogen, Creatinine, Lactate, serum glucose
- Serum Cytokine Panel

7. The following data will be collected at **14 days** from time of randomization (T+7).
   - PaO2, FiO2, Ventilator mode, Tidal Volume, Mean Airway Pressure, PEEP
   - Heart Rate, Systolic Blood Pressure, Diastolic Blood Pressure, Mean Arterial Pressure
   - Type of central venous access
   - CVP, ScvO2
   - Number of vasopressor-free days in the first 14 days from randomization. One vasopressor-free day is defined as a 24 hour period that the patient is alive and free of any continuous vasopressor infusion for any period of time.
   - Modified Marshall Score
   - Total bilirubin, Platelet count, Blood urea nitrogen, Creatinine, Lactate, serum glucose
   - Serum Cytokine Panel
8. The following data will be collected at **28 days** from time of randomization (T+28)
   - Ventilator-free days in first 28-days
   - Pneumonia (Y/N)
     - Organism
   - Venous Thromboembolism (VTE) (Y/N)
     - Site
   - Blood Stream Infection (Y/N)
     - Organism
   - ARDS (Y/N)
   - Modified Marshal Score
   - Serum Cytokine Panel
   - Died (Y/N)
9. The following data will be collected on day of death or discharge from the hospital (T+D).
   - ICU days, Hospital days
   - Day of death (from time of randomization)
   - Day of discharge (from time of randomization)
   - Renal Loss (Y/N) – Defined as requiring long term intermittent hemodialysis.

H. Outcome measures

Primary Endpoints: Number of vasopressor-free days in the first 14 days.

Secondary Endpoints: Survival at 14 days, 28 days and survival to discharge, ICU days, hospital days, ventilator-free days in the first 28 days after enrollment, renal loss (need for long term RRT – greater than 28 days), cytokine analysis (serum and ultrafiltrate), and dose of vasopressors (before, during, and after radomization).

I. Sample size analysis

To achieve an 80% power to detect a difference in vasopressor-free days of 1.85 days between the two groups using a two-sided t-test (level of significance 0.05) a sample size of 60 per group is required. **(120 Patients TOTAL)**

J. Risks

Burn patients who develop septic shock are at a high risk of death. The most immediate risk will be related to central venous access for the therapy. Double lumen dialysis catheters will be placed via seldinger technique. We will limit the placement of the catheter to the internal jugular or femoral site to minimize risk and optimize therapy.

Another risk will be related to the therapy itself. The literature is abundant with regards to the safety and possible efficacy of this therapy in this type of patient. Still, we will be enrolling subjects at the highest risk of death with already existing hemodynamic instability. Any unanticipated blood loss due to machine malfunction or operator error may result in an adverse event or even death. Every effort will be made to ensure adequate staff competencies (with extensive upfront training for naïve sites) and minimization of this risk. The type of therapy selected for this study (hemofiltration), compared to other modes of extracorporeal blood purification techniques, is the most simple, and as a result least prone to significant error such as massive volume or electrolyte shifts.

Also, extra blood samples will be taken on days 1-3, 7, 14 and 28 days from time of admission for cytokine analysis. Neither blood sample nor ultrafiltrate collection pose a significant risk to the patient. No other laboratory tests will be performed outside of what is clinically indicated for the patient.

An additional risk of this study is compromise of subject confidentiality. However, the investigators will make every effort to protect subject confidentiality through secure data management and de-identification.

K. Safety

Burn patients experience a number of adverse events as a result of their burn injuries. All unanticipated problems involving risk to subjects or serious adverse events **related to participation in the study** and deaths **related to participation in the study** will be promptly reported to the appropriate IRB**.** A written report will follow the initial notification.

L. Patient withdrawal from the study

Active participation will end if and when a patient withdraws from the study. However, with permission, data collected prior patient withdrawal may still be subject to analysis.

M. Reporting adverse events

Given the interventional nature of this trial, a **Data Safety Monitoring Board (DSMB)** will be created to provide independent oversight and review. The DSMB will consist of three members (statistician, medical provider, and an attorney or clergy member) who will serve as an independent monitoring body to help in assuring that all study subjects receive safe and ethical treatment, that unforeseen complications are detected and evaluated, that methodology is appropriate and adhered to, and that the results of the study are analyzed in a rigorous and objective manner.

Adverse events will be reported to each the Institutional Review Board at the respective site according to institution specific guidelines for reporting adverse events from human use participants. Unanticipated problems involving risk to subjects or others, serious adverse events related to participation in the study and all subject deaths will be promptly reported to the ABA's Director of Research Operations (DROP) who will in turn submit it to the DSMB and along with a report to the

Army Surgeon General's Human Subjects Research Review Board (HSRRB). A complete written report should follow the initial telephone call. In addition to the methods above, the complete report can be sent to the U.S. Army Medical Research and Materiel Command, ATTN: MCMR-ZB-QH, 504 Scott Street, Fort Detrick, Maryland 21702-5012.

The potential study related adverse events that will be reported are treatment related blood loss, venous thromboembolic events, and cardiac arrest and/or death while undergoing therapy.

N. Data management

All study related datasheets, identified only by subject and site number, will be turned in to the ABA's Director of Research Operations (DROP) at the Data Coordinating Center (DCC). Once data is entered onto a password protected secure database, the datasheet will be stored in study binder kept for each site at a secure central location. Data generated during the performance of this trial will become the property of the ABA. Use of study data in publications, presentations, grant proposals, and other uses will require the permission of the ABA Multicenter Trials Group.

When the results of the research are published or discussed in conferences, no information will be included that would reveal the subject's identity. Records of the subject's participation in this study may only be disclosed in accordance with federal law, including the Federal Privacy Act, 5 U.S.C. 552a, and its implementing regulations. DD Form 2005, Privacy Act Statement-Health Care Records, contains the Privacy Act Statement for the records. By signing the consent form document, the subject gives permission for information gained from participation in this study to be published in medical literature, discussed for educational purposes, and used generally to further medical science. The subject will not be personally identified; all information will be presented as anonymous data.
If photographs, videos, or audio tape recordings of the subject will be used for educational purposes, the subject's identity will be protected or disguised.

Authorized representatives of the US Army Institute of Surgical Research, the Brooke Army Medical Center Institutional Review Board, ABA's DROP, the U.S. Army Medical Research and Materiel Command, the Human Subject Research Review Board (HSRRB), the Clinical Investigation Regulatory Office (CIRO), or other federal agencies may need to review records of individual volunteers.

IX. References Cited

1. Honore PM, Joannes-Boyau O, Merson L, et al. The big bang of hemofiltration: the beginning of a new era in the third millennium for extra-corporeal blood purification! Int J Artif Organs 2006;29(7):649-59.
2. Mehta R, Kellum J, Shah S, et al. Acute Kidney Injury Network: report of an initiative to improve outcomes in acute kidney injury. Critical Care 2007;11(2):R31.
3. Greenhalgh DG, Saffle JR, Holmes JHt, et al. American Burn Association consensus conference to define sepsis and infection in burns. J Burn Care Res 2007;28(6):776-90.
4. Dellinger RP, Levy MM, Carlet JM, et al. Surviving Sepsis Campaign: international guidelines for management of severe sepsis and septic shock: 2008. Crit Care Med 2008;36(1):296-327.

5. Fitzwater J, Purdue GF, Hunt JL, O'Keefe GE. The risk factors and time course of sepsis and organ dysfunction after burn trauma. J Trauma 2003;54(5):959-66.
6. Chrysopoulo MT, Jeschke MG, Dziewulski P, Barrow RE, Herndon DN. Acute renal dysfunction in severely burned adults. Journal of Trauma-Injury Infection & Critical Care 1999;46(1):141-4.
7. Coca SG, Bauling P, Schifftner T, Howard CS, Teitelbaum I, Parikh CR. Contribution of acute kidney injury toward morbidity and mortality in burns: a contemporary analysis. Am J Kidney Dis 2007;49(4):517-23.
8. Kim G-H, Oh KH, Yoon JW, et al. Impact of burn size and initial serum albumin level on acute renal failure occurring in major burn. Am J Nephrol 2003;23(1):55-60.
9. Leblanc M, Thibeault Y, Querin S. Continuous haemofiltration and haemodiafiltration for acute renal failure in severely burned patients. Burns 1997;23(2):160-5.
10. Schiavon M, Di Landro D, Baldo M, De Silvestro G, Chiarelli A. A study of renal damage in seriously burned patients. Burns Incl Therm Inj 1988;14(2):107-12.
11. Azevedo LCP, Park M, Schettino GPP. Novel potential therapies for septic shock. Shock 2008;30 Suppl 1:60-6.
12. Nakada T-A, Oda S, Matsuda K-I, et al. Continuous hemodiafiltration with PMMA Hemofilter in the treatment of patients with septic shock. Mol Med 2008;14(5-6):257-63.
13. Kellum JA, Bellomo R, Mehta R, Ronco C. Blood purification in non-renal critical illness. Blood Purif 2003;21(1):6-13.
14. Venkataraman R, Subramanian S, Kellum JA. Clinical review: extracorporeal blood purification in severe sepsis. Critical Care 2003;7(2):139-45.
15. Ratanarat R, Permpikul C. Roles of extracorporeal blood purification in sepsis. J Med Assoc Thai 2007;90(5):1021-31.
16. Cohen J. The immunopathogenesis of sepsis. Nature 2002;420(6917):885-91.
17. Piccinni P, Dan M, Barbacini S, et al. Early isovolaemic haemofiltration in oliguric patients with septic shock.[see comment]. Intensive Care Med 2006;32(1):80-6.
18. Ronco C, Bellomo R, Homel P, et al. Effects of different doses in continuous veno-venous haemofiltration on outcomes of acute renal failure: a prospective randomised trial.[see comment]. Lancet 2000;356(9223):26-30.
19. Honore PM, Jamez J, Wauthier M, et al. Prospective evaluation of short-term, high-volume isovolemic hemofiltration on the hemodynamic course and outcome in patients with intractable circulatory failure resulting from septic shock.[see comment]. Crit Care Med 2000;28(11):3581-7.
20. Ratanarat R, Brendolan A, Ricci Z, et al. Pulse high-volume hemofiltration in critically ill patients: a new approach for patients with septic shock. Semin Dial 2006;19(1):69-74.
21. Laurent I, Adrie C, Vinsonneau C, et al. High-volume hemofiltration after out-of-hospital cardiac arrest: a randomized study. J Am Coll Cardiol 2005;46(3):432-7.
22. Levy MM, Pronovost PJ, Dellinger RP, et al. Sepsis change bundles: converting guidelines into meaningful change in behavior and clinical outcome. Crit Care Med 2004;32(11 Suppl):S595-7.
23. Angus DC, Linde-Zwirble WT, Lidicker J, Clermont G, Carcillo J, Pinsky MR. Epidemiology of severe sepsis in the United States: analysis of incidence, outcome, and associated costs of care. Crit Care Med 2001;29(7):1303-10.
24. Mehta RL, Kellum JA, Shah SV, et al. Acute Kidney Injury Network: report of an initiative to improve outcomes in acute kidney injury. Crit Care 2007;11(2):R31.
25. Feldmann M. Many cytokines are very useful therapeutic targets in disease. J Clin Invest 2008;118(11):3533-6.

26. Ventilation with lower tidal volumes as compared with traditional tidal volumes for acute lung injury and the acute respiratory distress syndrome. The Acute Respiratory Distress Syndrome Network. N Engl J Med 2000;342(18):1301-8.

X. Facilities and Other Resources

TheranOS (Palo Alto, CA) - Point of Care Testing (POCT) is going to play an increasing role in the more efficient delivery of individualized medical care to patients, especially in intensive care units. POCT systems which can efficiently close the loop **in real time** between a unique patient and the health care team (HCT) are needed. The TheranOS system allows the HCT to monitor drugs, their metabolites, and relevant biomarkers from whole blood samples in real time at any testing frequency in a clinic, hospital setting or any POC. Its infrastructure allows for actionable information to be extracted by the HCT through the three major interconnected units described in the next section.

XI. Equipment

The TheranOS system (Palo Alto, CA) - Each participating center will be equipped with the capability to perform POCT for the cytokines. This includes:

a. A reagent component which includes:
   - The use of reagents directed against drugs, their metabolites and any circulating biomarkers.
   - Disposable cartridges pre-loaded with chemiluminescent immunoassays able to test simultaneously up to six different analytes with a total system CV of <15%. The cartridges can be customized to measure any combination of drug and biomarkers together to map indicators/trends of subject status.

b. A remote portable patient care device which currently performs three functions:
   - On site, immediate and rapid automatic measurement using the disposable cartridges, without any human intervention, of various analytes from blood samples.
   - Graphical user interface allowing the patient to enter a variety of relevant information such as caloric consumption, energy expenditure and current treatments, and to initiate the assays.
   - Two-way communication portal between patients, instruments, mainframe computer, HCT and back.

c. An information integration and exploitation infrastructure which permits:
   - Data acquisition and storage in real-time of point-of-care results.
   - The integration of blood parameters and patient diary data with all other physiologically relevant information into in the electronic health record.
   - A central mathematical software program to graphically visualize, help to interpret, and analyze all data in one place, and link any new information into a disease management system mapping the information into a probability space of clinical outcomes
   - The graphical display of clinically relevant and actionable information back to the HCT and/or the patient.

XII. Other Attachments

N/A

XIII. Acronyms and Symbol Definition

Modified Marshal Scoring System[3]

Organ System	0	1	2	3	4
Cardiovascular (heart rate, inotropes, lactate)	≤120	120–140	>140	Inotropes	Lactate >5
Respiratory, PO_2/FIO_2	>300	226–300	151–225	76–150	≤75
Renal (creatinine, μmol/L)	≤100	101–200	201–350	351–500	>500
Central nervous system (Glascow Coma Scale)	15	13–14	10–12	7–9	≤6
Hepatic (total bilirubin, μmol/L)	≤20	21–60	61–120	121–240	>240
Hematologic (platelet count $\times 10^3$)	>120	81–120	51–80	21–50	≤20

Six domains of the MODS. The scores can range between 0 and 24. The heart rate is defined as beats per minute (bpm). "Inotropes" indicates the need for inotropes more than dopamine >3μg · kg^{-1} · min^{-1}. Lactate is measured in mmol/L.

Pneumonia - As defined by the ABA consensus conference[3]

Acute Respiratory Distress Syndrome

- American-European Consensus definition[26]
  - P/F Ratio < 300 (ALI), < 200 (ARDS)
  - Bilateral pulmonary infiltrates on chest radiograph.
  - No clinical evidence of left atrial hypertension or (if measured) a pulmonary-capillary wedge pressure of 18 mm Hg or less.

Blood Stream Infection - As defined by the ABA consensus conference[3]

Acute Kidney Injury Network staging criteria for AKI.

Classification/staging system for acute kidney injury[a]

Stage	Serum creatinine criteria	Urine output criteria
1	Increase in serum creatinine of more than or equal to 0.3 mg/dl (≥ 26.4 μmol/l) or increase to more than or equal to 150% to 200% (1.5- to 2-fold) from baseline	Less than 0.5 ml/kg per hour for more than 6 hours
2[b]	Increase in serum creatinine to more than 200% to 300% (> 2- to 3-fold) from baseline	Less than 0.5 ml/kg per hour for more than 12 hours
3[c]	Increase in serum creatinine to more than 300% (> 3-fold) from baseline (or serum creatinine of more than or equal to 4.0 mg/dl [≥ 354 μmol/l] with an acute increase of at least 0.5 mg/dl [44 μmol/l])	Less than 0.3 ml/kg per hour for 24 hours or anuria for 12 hours

[a]Modified from RIFLE (Risk, Injury, Failure, Loss, and End-stage kidney disease) criteria [26]. The staging system proposed is a highly sensitive interim staging system and is based on recent data indicating that a small change in serum creatinine influences outcome. Only one criterion (creatinine or urine output) has to be fulfilled to qualify for a stage. [b]200% to 300% increase = 2- to 3-fold increase. [c]Given wide variation in indications and timing of initiation of renal replacement therapy (RRT), individuals who receive RRT are considered to have met the criteria for stage 3 irrespective of the stage they are in at the time of RRT.

XIV. Brief Description of Animal and Human Use

This study involves human subjects. Enrollment of a total of 120 patients is anticipated for the study over a 4 year period. There is intent to benefit all participants of this study regardless of randomization group. Both groups will be subject to 'contemporary' care to ensure that state of the art, consensus guideline driven care is delivered in the care of patients who develop septic shock. It is likely even those not enrolled in the study may benefit from the institution of the burn-specific

sepsis bundle at each site. Study participants randomized to the intervention arm will undergo placement of a CRRT catheter and will be initiated on CVVH for 72 hours. Other than cytokine analysis, no additional blood draws outside what is already necessary for routine ICU care will be performed. These additional blood draws pose minimal additional risk as the amount of blood required for each cytokine panel is 1 cc. It is anticipated that each patient will undergo approximately 7-10 of these blood draws for the duration of the study. In addition, data will be collected prospectively from the participant's medical records for the duration of the hospital stay. Basic demographic information as well as specific laboratory values, hemodynamic data, and specific diagnoses will be recorded for the purpose of data tabulation and analysis. All gathered data will be coded and stored on a secure server behind a firewall and password protected drive accessible only to the study coordinators and the co-investigators.

XV. Budget

PRINCIPLE INVESTIGATOR: Chung, Kevin, K								
DETAILED BUDGET FOR YEAR:		1st	2nd	3rd	4th	Total	From 2009	Through 2010
PERSONNEL		TYPE APPT. (Mo.S)	ANNUAL BASE SALARY		EFFORT ON PROJECT	DOLLAR AMOUNT REQUESTED		
NAME	ROLE ON PROJECT					SALARY REQUESTED	FRINGE BENEFITS	TOTALS
Kevin K Chung, MD	PI	3	140K		25%	0	0	0
Amy M. Sprague, MD	Co-I	1.2	190K		10%	22,800	6,612	29,412
ISR Research Coordinator	Coordination, training	6	110K		50%	37,243	18,070	55,313
Site Research Nurse (estimated)	Enrollment, data collect.	24			N/A	296,471	(N/A)	296,471
SUBTOTALS								381,196
CONSULTANT COSTS								
MAJOR EQUIPMENT								
MATERIALS, SUPPLIES AND CONSUMABLES (ITEMIZE BY CATEGORY) 1)Replacement Fluids/Filters set/IV tubing ($4000/ptX30=120,000) 2)Point of Care Cytokine Analysis ($2400/pt X 30 = $72,000)								192,000
RESEARCH-RELATED PATIENT COSTS								0
MEDICAL CARE FOR RESEARCH-RELATED INJURY COSTS								0
OTHER DIRECT COSTS 1)IRB FEES ($20,000) 2)INFORMED CONSENT TRANSLATION ($20,000)								40,000
PUBLICATION/REPORTING COSTS								0
TRAVEL COSTS								40,000
SUBTOTAL OF DIRECT COSTS FOR THIS BUDGET PERIOD								272,000
CONSORTIUM/ SUBAWARD COSTS	DIRECT COST							0
	INDIRECT COST ($2000 per patient enrolled X 30 patients)							60,000
TOTAL DIRECT COST FOR THIS BUDGET PERIOD								713,196
TOTAL INDIRECT COSTS FOR THIS BUDGET PERIOD (15% overhead)								106,979
TOTAL COST FOR THIS BUDGET PERIOD								820,175

YEAR 2								
PRINCIPLE INVESTIGATOR: Chung, Kevin, K								
DETAILED BUDGET FOR YEAR:		1st	2nd	3rd	4th	Total	From 2010	Through 2011
PERSONNEL		TYPE APPT. (Mo.S)	ANNUAL BASE SALARY	EFFORT ON PROJECT	DOLLAR AMOUNT REQUESTED			
NAME	ROLE ON PROJECT				SALARY REQUESTED	FRINGE BENEFITS	TOTALS	
Kevin K Chung, MD	PI	3	145K	25%	0	0	0	
Amy M. Sprague, MD	Co-I	0.6	196K	5%	9,785	2,838	12,623	
ISR Research Coordinator	Coordination, training	3	114K	25%	19,233	9,306	28,539	
Site Research Nurse (estimated)	Enrollment, data collect.	24		N/A	305,365	(N/A)	305,365	
SUBTOTALS							346,527	
CONSULTANT COSTS (ICON)							0	
MAJOR EQUIPMENT (CRRT Machines X 10)							0	
MATERIALS, SUPPLIES AND CONSUMABLES (ITEMIZE BY CATEGORY) 1)Replacement Fluids/Filters set/IV tubing ($4000/ptX30=120,000) 2)Point of Care Cytokine Analysis ($72,000)							192,000	
RESEARCH-RELATED PATIENT COSTS							0	
MEDICAL CARE FOR RESEARCH-RELATED INJURY COSTS							0	
OTHER DIRECT COSTS 1)IRB FEES 2)INFORMED CONSENT TRANSLATION							0	
PUBLICATION/REPORTING COSTS							0	
TRAVEL COSTS							10,000	
SUBTOTAL OF DIRECT COSTS FOR THIS BUDGET PERIOD							202,000	
CONSORTIUM/ SUBAWARD COSTS	DIRECT COST						0	
	INDIRECT COST ($2000 per patient enrolled X 30 patients)						60,000	
TOTAL DIRECT COST FOR THIS BUDGET PERIOD							608,527	
TOTAL INDIRECT COSTS FOR THIS BUDGET PERIOD (15% overhead)							91,279	
TOTAL COST FOR THIS BUDGET PERIOD							699,806	

YEAR 3								
PRINCIPLE INVESTIGATOR: Chung, Kevin, K								
DETAILED BUDGET FOR YEAR:		1st	2nd	3rd	4th	Total	From 2011	Through 2012
PERSONNEL NAME	ROLE ON PROJECT	TYPE APPT. (Mo.S)	ANNUAL BASE SALARY	EFFORT ON PROJECT	DOLLAR AMOUNT REQUESTED: SALARY REQUESTED	FRINGE BENEFITS	TOTALS	
Kevin K Chung, MD	PI	3	149K	25%	0	0	0	
Amy M. Sprague, MD	Co-I	0.6	202K	5%	10,083	2,924	13,007	
ISR Research Coordinator	Coordination, training	3	119K	25%	19,810	9,585	29,395	
Site Research Nurse (estimated)	Enrollment, data collect.	24		N/A	314,526	(N/A)	314,526	
SUBTOTALS								356,928
CONSULTANT COSTS (ICON)								0
MAJOR EQUIPMENT (CRRT Machines X 10)								0
MATERIALS, SUPPLIES AND CONSUMABLES (ITEMIZE BY CATEGORY) 1)Replacement Fluids/Filters set/IV tubing /other($4000/ptX30=120,000) 2)Point of Care Cytokine Analysis ($72,000)								192,000
RESEARCH-RELATED PATIENT COSTS								0
MEDICAL CARE FOR RESEARCH-RELATED INJURY COSTS								0
OTHER DIRECT COSTS 1)IRB FEES 2)INFORMED CONSENT TRANSLATION								0
PUBLICATION/REPORTING COSTS								0
TRAVEL COSTS								10,000
SUBTOTAL OF DIRECT COSTS FOR THIS BUDGET PERIOD								202,000
CONSORTIUM/ SUBAWARD COSTS	DIRECT COST							0
	INDIRECT COST ($2000 per patient enrolled X 30 patients)							60,000
TOTAL DIRECT COST FOR THIS BUDGET PERIOD								618,928
TOTAL INDIRECT COSTS FOR THIS BUDGET PERIOD (15% overhead)								92,839
TOTAL COST FOR THIS BUDGET PERIOD								711,767

YEAR 4								
PRINCIPLE INVESTIGATOR: Chung, Kevin, K								
DETAILED BUDGET FOR YEAR:		1st	2nd	3rd	4th	Total	From 2012	Through 2013
PERSONNEL NAME	ROLE ON PROJECT	TYPE APPT. (Mo.S)	ANNUAL BASE SALARY	EFFORT ON PROJECT	DOLLAR AMOUNT REQUESTED: SALARY REQUESTED	FRINGE BENEFITS	TOTALS	
Kevin K Chung, MD	PI	3	154K	25%	0	0	0	
Amy M. Sprague, MD	Co-I	0.6	208K	5%	10,385	3,012	13,397	
ISR Research Coordinator	Coordination, training	3	125K	25%	20,404	9,873	30,277	
Site Research Nurse (estimated)	Enrollment, data collect.	24		N/A	323,962	(N/A)	323,962	
SUBTOTALS								367,636
CONSULTANT COSTS (ICON)								0
MAJOR EQUIPMENT (CRRT Machines X 10)								0
MATERIALS, SUPPLIES AND CONSUMABLES (ITEMIZE BY CATEGORY) 1)Replacement Fluids/Filters set/IV tubing /other($4000/ptX30=120,000) 2)Point of Care Cytokine Analysis ($72,000)								192,000
RESEARCH-RELATED PATIENT COSTS								0
MEDICAL CARE FOR RESEARCH-RELATED INJURY COSTS								0
OTHER DIRECT COSTS 1)Publication Costs								1,000
PUBLICATION/REPORTING COSTS								0
TRAVEL COSTS								10,000
SUBTOTAL OF DIRECT COSTS FOR THIS BUDGET PERIOD								203,000
CONSORTIUM/ SUBAWARD COSTS	DIRECT COST							0
	INDIRECT COST ($2000 per patient enrolled X 30 patients)							60,000
TOTAL DIRECT COST FOR THIS BUDGET PERIOD								630,636
TOTAL INDIRECT COSTS FOR THIS BUDGET PERIOD (15% overhead)								94,595
TOTAL COST FOR THIS BUDGET PERIOD								725,231

YEAR 1-4								
PRINCIPLE INVESTIGATOR: Chung, Kevin, K								
DETAILED BUDGET FOR YEAR:		1st	2nd	3rd	4th	Total	From 2009	Through 2013
PERSONNEL		TYPE APPT. (Mo.S)	ANNUAL BASE SALARY		EFFORT ON PROJECT	DOLLAR AMOUNT REQUESTED		
NAME	ROLE ON PROJECT					SALARY REQUESTED	FRINGE BENEFITS	TOTALS
Kevin K Chung, MD	PI							
Amy M. Sprague, MD	Co-I							
ISR Research Coordinator	Coordination, training							
Site Research Nurse (estimated)	Enrollment, data collect.							
SUBTOTALS								1,452,287
CONSULTANT COSTS								
MAJOR EQUIPMENT								
MATERIALS, SUPPLIES AND CONSUMABLES (ITEMIZE BY CATEGORY) 1)Replacement Fluids/Filters set/IV tubing 2)Point of Care Cytokine Analysis								
RESEARCH-RELATED PATIENT COSTS								
MEDICAL CARE FOR RESEARCH-RELATED INJURY COSTS								
OTHER DIRECT COSTS 1)IRB FEES 2)INFORMED CONSENT TRANSLATION								
PUBLICATION/REPORTING COSTS								
TRAVEL COSTS								
SUBTOTAL OF DIRECT COSTS FOR THIS BUDGET PERIOD								879,000
CONSORTIUM/ SUBAWARD COSTS	DIRECT COST							0
	INDIRECT COST							240,000
TOTAL DIRECT COST FOR THIS BUDGET PERIOD								2,571,287
TOTAL INDIRECT COSTS FOR THIS BUDGET PERIOD (15% overhead)								385,693
TOTAL COST FOR THIS BUDGET PERIOD								2,956,980

XVI. Budget Justification

I. Salaries:

A. PI - Due to his current status as a government employee, cannot request any grant related salary.

B. Co-I - This salary is based on the NIH maximum allowable annual salary for investigators plus 29% benefits. We have requested a 10% salary for the first year and 5% for each subsequent year based on the level of anticipated involvement of the co-investigator. This includes a 3% raise per year adjustment.

C. ISR Research Nurse – We estimate a 50% full time equivalent research nurse will be required for the purpose of coordination, data collection, and administrative duties in the first year of the study. This will drop to a 25% full time equivalent for each subsequent year. This includes a 3% raise per year adjustment.

D. Site-specific Nurse Time – We estimate 132 nurse hours will be required per patient. This accounts for the bedside administration of the 'intervention' for 72 hours along with 50 hours subsequent nurse hours needed for data collection and administrative duties. Assuming an enrollment rate of 30 patients per year, this totals 3960 nurse hours required per year across all the study sites. Given 1980 nurse hours/year, we have determined 2 full time equivalents will be needed across all the sites each year. The salaries are also adjusted for 3% inflation for each subsequent year.

II. Consultant Costs: N/A

III. Compensation per subject enrolled - $18,300 per subject: In order to cover the cost of the study for each site, compensation will be made per each subject enrolled into the study. This cost includes research/clinical nurse costs (as above), Materials/Supplies/Consumables (described below) and indirect costs. This does not include IRB costs or Informed Consent translation costs.

IV. Major Equipment: Centers with existing CRRT capability will be involved. Thus no major equipment purchase or lease is necessary.

V. Materials/Supplies/Consumables: We estimated an average disposable cost of $4000 per patient, for the 3 days of therapy (for the treatment arm) and subsequent blood draws. This figure includes replacement fluid, filter sets, catheters, IV tubing, and other disposables associated with therapy, as well as blood draws. For cytokine analysis, we estimate up to 96 analytes will be measured per person for the duration of the enrollment period. Each analyte measurement costs approximately $25. Thus, $2400 per patient will be dedicated for cytokine analysis.

VI. Research Related Patient Costs: Patients will not be compensated for participation in the study.

VII. Medical Care For Research-Related Injury Costs: The federal government is self insured. Participating sites will be responsible for the medical cost of any research-related injury.

VIII. Other Direct Costs: IRB fees and Informed consent translation fees are included in the budget.

IX. Publication/Reporting Costs: This cost was only included in the last year of the study.

X. ***Travel Costs:*** Assuming a max cost of $2000 per travel we have budgeted for 20 trips the first year to account for travel planned to each study site by the PI and/or the co-I. We anticipate no more than 4 trips per year for each subsequent year for follow-up visits as needed.

XI. ***Indirect Costs:*** We assume $2000 per patient enrolled for overhead costs for each participating center. The budget also includes a 15% overhead cost for the entire study for the ABA MCTG.