EXTHERA MEDICAL

CLINICAL PROTOCOL

Safety and Performance Evaluation of the Seraph® 100 Microbind® Affinity Blood Filter (Seraph 100) for Reducing Bacteremia in Patients on Hemodialysis

Document Number:	**CP001**	**Revision: F**
Number of Pages:	**53**	
Number of Associated Forms:	**N/A**	
Author:	**KATHLEEN WHITE**	
Supersedes:	**Rev E**	
Effective Date:	**September 19, 2017**	
Released By:	**KATHLEEN WHITE**	

Table of Contents

Investigator Signature Page

I have read the protocol and I agree to conduct the study as outlined and in accordance with all applicable laws and regulations, including Good Clinical Practice guidelines. In addition, I agree to provide all the information requested in the case report forms presented to me by the Sponsor in a manner to assure completeness, legibility and accuracy.

I agree to actively enroll patients into this study and confirm that I am not currently participating in any clinical investigations for similar types of medical devices.

I also agree that all information provided to me by the Sponsor, including pre-clinical data, protocols, case report forms, and any verbal and written information, will be kept strictly confidential and confined to the clinical personnel involved in conducting the study. It is recognized that this information may be relayed in confidence to the Ethics Committee.

In addition, no reports or information about the study or its progress will be provided to anyone not involved in the study other than the Sponsor or the Ethics Committee. Any such submission will indicate that the material is confidential.

_____ _____

Investigator Signature Date

_____

Investigator Printed Name

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Investigational Site Name

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Protocol Synopsis

Title	Safety and Performance Evaluation of the Seraph 100 Microbind Affinity Blood Filter for Reducing Bacteremia in Patients on Hemodialysis
Protocol Number	CP001
Device Under Investigation	Seraph 100 Microbind Affinity Blood Filter
Indication(s) for Use	The Seraph100 Microbind Affinity Blood Filter is indicated as an adjunctive treatment for blood stream infection (BSI) in patients on renal replacement therapy.
Subject Population	Patients who require renal replacement therapy and have bacteremia.
Objectives	• To confirm safety and performance of using the ExThera Medical Seraph 100 Microbind Affinity Blood Filter in a hemodialysis circuit. • To confirm as secondary endpoints the demonstrating of effectiveness for pathogen reduction.
Primary Endpoints	Safety: • SAEs occurring while connected to the device. • Clinically significant changes in hematology indices. • Clinically significant changes in chemistry indices. • Device complications.
Secondary Endpoints	Safety: • All clinical and laboratory adverse events post procedure 1-14 days. Performance: • Throughput flow effects • Compatibility with hemodialysis systems. • Changes in hemodynamics. Effectiveness: • A mean pathogen reduction of > 40% measured as colony forming units (CFU) or a mean increase in time to positivity (TTP) of > 22 minutes in blood passed through the Seraph 100 cartridge. The pre-cartridge blood sample must have a detectable bacterial count (as measured by CFU or TTP) • Duration of bacteremia • Bacteremia recurrence rate • Rate of cardiac and non-cardiac infectious complications • Changes in cytokine levels

Study Design	This trial is a prospective, non-randomized study in patients as an adjunctive treatment for blood stream infection (BSI) in patients on renal replacement therapy. The heparin surface being studied is currently marketed on extracorporeal circuits. It has been shown to absorb various types of Gram positive and Gram negative bacteria and to reduce toxins and cytokines in *in vitro* studies using blood. For this study, patients on renal replacement therapy who develop bacteremia will have the Seraph 100 Microbind Affinity Blood Filter included in the dialysis circuit for up to 4 hours, on one day, with scheduled monitoring. Patients will be followed for 14 days post treatment. Patients will be monitored by vital signs and laboratory indices on the day of treatment and on post procedure day 1, 2, 3, 4, 5, 6, 7 and 14.
Number of Subjects	15 patients will be enrolled.
Number of Sites	Five European sites will enroll patients.
Estimated Times	Enrollment Period: 9-12 months Follow-Up: 14 days

Inclusion Criteria

Patients must meet ALL the following inclusion criteria to potentially be included in the study:

1. Require renal replacement therapy.
2. Be ≥ 18 years old and ≤ 90 years old
3. Positive blood culture and one of the following:
    a. Clinical evidence of a catheter exit site to tunnel infection as evidenced by redness, tenderness or purulence.
    b. Bacteremia is proven with two separate blood cultures from independent vein punctures.
    c. A blood culture where the time to positivity is within 14 hours.
    d. Growth from a blood culture taken from the hemodialysis catheter 2 or more hours before the growth of a blood culture drawn peripherally at the same time.

    NOTE: Written results for bacteremia and pathogen identification do not necessarily need to come from the hospital lab and written results can be provided by an outside clinic. The test results from the external source should have been generated within one (1) week prior to hospital admission.
4. The patient or patient's legal representative is able to understand the requirements of the study and signs an approved informed consent form prior to enrollment which explains aggressive care.

Exclusion Criteria. Patients who meet ANY the following exclusion criteria will NOT be included in the study:

1. Have an arteriovenous polytetrafluoroethylene (PTFE) graft.
2. Lack of a commitment to full aggressive support.
3. Have inability to maintain a minimum mean arterial pressure of ≥ 65 mm Hg despite vasopressor therapy and fluid resuscitation.
4. Have had chest compressions as part of cardiopulmonary resuscitation (CPR)
5. Have had an acute myocardial infarction (MI) within the past 3 months.
6. Have had serious injury within 36 hours of screening.
7. Have uncontrolled hemorrhage.
8. Are not expected to live > 14 days.
9. Have malignancy and are not expected to live 42 days.
10. Have neutropenia (absolute neutrophil count <500 cells/µL).
11. Have Child-Pugh Class C cirrhosis.

12. Have New York Heart Association Class IV Heart Failure or an ejection fraction <30%.
13. Have known Antithrombin III deficiency.
14. Have platelet count <30,000/µL.
15. Cannot have intravenous (IV) supplemental iron halted during trial period.
16. Are currently involved in an investigational drug or device trial.
17. Have been previously enrolled in this clinical trial.
18. Next hemodialysis treatment will not take place for at least 24 hours after enrollment.
19. Serious bleedings and clotting disorders, determined by blood transfusion of > 2 units of packed red blood cells, **or**, An acute (48 h) hemoglobin decline of at least 2 g/dL, transfusion requirement of >4 units over 48h, objective evidence of bleed, documented by physician.
20. Breast feeding and pregnant women
21. Contraindications for heparin sodium for injection are:
    a. Have heparin sensitivity
    b. Severe thrombocytopenia.
    c. With an uncontrolled active bleeding state, except when this is due to disseminated intravascular coagulation
    d. In whom suitable blood coagulation tests, e.g. whole blood clotting time, partial thromboplastin time, etc cannot be performed at appropriate intervals (this contraindication refers to full-dose heparin; there is usually no need to monitor coagulation parameters in patients receiving low-dose heparin)
22. Serious injuries, which have occurred more than 36 hours, have to be excluded.

1. Introduction

1.1. Literature Review

Many microorganisms including bacteria, viruses and parasites attach to heparan sulfate receptors (glycosaminoglycans) on the surface of mammalian cells. Heparin is a similar glycosaminoglycan that has also been demonstrated to have binding sites for microorganisms. Mattsby-Baltzer (2011) reported that beads coated with end-point attached heparin and contained within a column were demonstrated to remove >65% of *Staphylococcus aureus* (SA) and Methicillin-Resistant SA (MRSA) in inoculated whole blood after a single pass through the miniature column. These findings suggest that the bio-active surface may offer a device-based treatment for patients with SA or MRSA induced sepsis.

Axelson (2010) previously studied blood from septic patients and demonstrated that the level of one pro-inflammatory cytokine, TNF-α, was restored to normal levels after passage over beads coated with the same biologically active heparin surface with no evidence of inflammatory system activation in the blood (no increase in cytokine RANTES). Use of untreated beads as controls had no effect on cytokine levels and caused *elevation* in cytokine RANTES. This is consistent with experiences reported during use of the bio-active surface on cardiopulmonary bypass circuits.

1.2. Background

End-point attached heparin has been commercially available on extracorporeal circuits and oxygenators since 1989. The Medtronic Maxima Oxygenator and Extracorporeal Circuits with Heparin Surface were cleared by FDA under 510(k) K891687 and are widely used today to provide thrombo-resistant blood-contacting surfaces for complex medical procedures. One trade name for end-point attached heparin is the Carmeda BioActive Surface (CBAS), which has proven durable, non-leaching and is supported by the largest body of peer-reviewed clinical and scientific evidence of any biocompatible surface used for cardiopulmonary bypass devices today, according the Medtronic website (http://www.medtronic.com/for-healthcare-professionals/productstherapies-/cardiovascular/cardiopulmonary-products/carmeda-bioactive-surface/index.htm) Medtronic promotes the benefits of end-point attached heparin covalently bonded to surfaces of extracorporeal circuits based on published clinical and scientific information as follows:

- Less blood product use
- Less perioperative blood loss
- Shorter ventilator time
- Shorter hospital length of stay
- Less postoperative body temperature rise
- Significantly greater urine output during CPB
- Lower costs, as related to improved clinical outcomes
- Less negative impact on the body's defense systems, including the:
    - contact system
    - coagulation system
    - fibrinolytic system
    - complement system
    - cytokine proteins

- Reduced impact on the blood's formed elements, including:
  - platelets
  - red blood cells
  - leukocytes

As of May, 2012, the Medtronic website reports the studies referenced include over 1,000 patients with no reports of adverse effects to the patient attributed to the Carmeda Bio-Active Surface.

The media in the Seraph 100 Microbind Affinity Blood Filter uses end point attached heparin to remove bacteria and cytokines from the blood, thus supplementing conventional antibiotic treatment, without affecting hepatic or renal function.

1.3. Device Description

The Seraph 100 Filter is a single use, disposable column packed with ultra-high molecular weight polyethylene beads which have been modified to contain end-point attached heparin on the surface. The amount of heparin as measured by the 3-methyl-2-benzothiazolinone hydrazone (MBTH) method is 2.0 ± 0.5 mg heparin/g bead. The cartridge contains nominally 160 grams of beads with a void volume/priming volume of 160-170 mL. The assembled unit weighs approximately 400 grams. The devices are sterilized using a standard ethylene oxide cycle, following ISO 11135-1:2007. Chemical Indicator labels are located near the product label. A green label indicates that the device has been exposed to ethylene oxide gas.

Material used to manufacture the column:

Component	***Material***	***Blood Contacting***
Column Body and End Caps	Copolyester, DuraStar™ Polymer, MN611	Yes
End Plate	Hydrophilic porous polyethylene, Porex, pore size 90-130 microns	Yes, only after heparinization of surface.
Adsorption media, beads	Ultra-high molecular weight polyethylene (UHMWPE), Inhance® UH-1045, particle size 300 microns	Yes, only after heparinization of surface.
O-Ring	Platinum cured silicone	No
End Point Attached Heparin	Heparin Sodium,	Yes
Adhesive	Two-part epoxy, Loctite® M-31CL	No

Manufacturing process:

Ultra High Molecular Weight Polyethylene (UHMWPE) particles and end plates with amine groups on their surface are further modified with 'end-point-attached' heparin. This resulting surface is referred to as the CARMEDA® BioActive Surface (CBAS® Heparin Surface) by others, and is used in commercially-available medical devices including oxygenators, dialyzers and vascular grafts.

During processing the end point attached heparin particles and endplates are exhaustively washed with purified water to remove unattached heparin. Quality control testing is performed on dry end-point attached heparin beads and end plates to determine the quantity of heparin attached, the heparin activity and the endotoxin content.

The Seraph 100 filters are assembled by placing and end plate in each end cap. An O-ring is placed in the recess at each end of the column. One end cap is glued, using the epoxy, to one end of the column. The column/end cap assembly is held vertically and filled with 160 ± 5 grams of end point attached heparin UHMWPE particles. A second end cap is glued to the open end of the column.

A label is attached to the column body. The assembled unit weights approximately 400 grams. The column is placed in an inner peel pouch and then placed in a larger, outer peel pouch. The column/peel pouches are placed in a 4 inch x 4.25 inch x 11 inch box.

The devices are sterilized using a standard ethylene oxide cycle. Chemical Indicator labels are included and positioned near the product label. A green label indicates that the device has been exposed to ethylene oxide gas.

Quality control includes a visual inspection of each assembled Seraph 100 cartridge, flow test and a pressure test using one cartridge selected at random from each lot of 30. After sterilization cartridges are selected at random for the Ethylene Oxide residual testing and sterility confirmation test.

1.4. Indication for Use

The device is intended to be used concurrent with hemodialysis at the onset of bacteremia to remove bacteria and cytokines from the blood, thus supplementing conventional antibiotic treatment, without affecting hepatic or renal function.

Seraph® 100 Microbind® Affinity Blood Filter

1.5. Device Properties

Biocompatibility testing was performed in accordance with current ISO 10993 standards.

Test	Result
MEM Elution	**PASS**: The test article extract did not cause any cellular destruction.
ASTM Hemolysis (Extract Method), human blood	**PASS**: The difference between the hemolytic indexes of the test article and the negative control equals 0.00 percent. This places the test article in the non-hemolytic range (0-2) according to the Hemolytic Index and Grade table.
ASTM Hemolysis (Direct Contact Method), human blood	**PASS**: The difference between the hemolytic indexes of the test article and the negative control equals 0.83 percent. This places the test article in the non-hemolytic range (0-2) according to the Hemolytic Index and Grade table.
Sensitization: Magnusson-Klingman Method, polar and nonpolar extractions	**PASS**: The test article Extracts demonstrated no sensitization reactions under the conditions of this assay.
Intracutaneous Reactivity Irritation Test in Rabbits, polar and nonpolar extractions	**PASS**: The test article extracts did not cause skin irritation under the conditions of this assay.
Acute Systemic Toxicity in Mice, polar and nonpolar extractions	**PASS**: The test article extracts did not cause adverse effects under the conditions of this assay.
Materials Mediated Pyrogen Test in Rabbits	**PASS**: The test article extracts did not cause a febrile reaction after IV administration under conditions of the assay.

1.6. Pre-clinical Investigations

Animal and *in vitro* studies involved investigation as to whether the heparin coated beads could remove bacterial toxins from solution. Protective Antigen (PA) is one component of the toxins generated by *Bacillus anthracis*, responsible for anthrax. Protection of macrophages from toxin-mediated death was demonstrated by removal of PA.

In Vitro Studies

Seraph's heparinized media has been shown to absorb various types of Gram positive and Gram negative bacteria and to reduce toxins and cytokines in *in vitro* studies using blood. An *in vitro* study was performed at the Department of Infectious Diseases at the University of Gothenburg under the direction of Tomas Bergstrom. In brief, a mixture of methicillin-resistant *staphylococcus aureus* (MRSA) and blood (4 x 10^5 CFU/mL) was passed through the Seraph adsorption media. The bound bacteria were eluted from the beads and quantified using r-t PCR. The MRSA mean percent reduction and standard deviation of six (6) samples was 66 % ± 9 %.

In vitro studies were performed by Microchem Laboratories (Round Rock, Texas) using inoculated blood and using a single pass through a miniature column containing the Seraph adsorption media. Test samples were conditioned with 2.0 mL of PBS, then, 2.0 mL of FBS, then, another 2.0 mL of PBS prior to inoculation. Test inoculum was prepared from overnight culture of test microorganism. Cultures were diluted in Defibrinated Horse Blood to ~2-3x10^5 CFU/mL prior to use. 2.0 mL of dilute test inoculum were repeatedly filtered through the test syringes, with enumerations of remaining bacteria on the 3rd filtrate. Standard dilution and plating techniques were used to enumerated remaining bacteria after filtration.

Bacteria	Single Pass Binding (%)
MRSA	92
S. aureus	66
K. pneumoniae (CRE)	99.9
K. pneumoniae	37
E. coli (CRE)	99.9
E. coli	99.7
S. pneumoniae	53
E. faecalis	99.0
E. faecalis(VRE)	91
E. faecium	56
A. baumannii	79
S. epidermidis	58
Methicilin resistant *S. epidermidis*	66
S. pyogenes	76
Serratia marcescens	73

GLP Animal Safety Study (Protocol 13A0385G-X01G)

The Seraph 100 Filter is attached to a dialysis machine in series with a dialysis cartridge. The Seraph 100 Filter contains media in which heparin is covalently bonded to small polymer beads using a proprietary end-point attachment process. In vitro studies have shown the heparinized media is effective for removal of bacteria from infected blood. The purpose of this chronic study was to evaluate the safety of the Seraph 100 Microbind Affinity Blood Filter during dialysis (test) in comparison to standard dialysis without the Seraph 100 cartridge (control) in the non-diseased swine model. The eight animals (3 control and 5 test) received 4 hours of dialysis and were then survived for 3 +/- 1 days.

The animals were considered healthy based on a veterinarian exam and blood work prior to the study procedure. There were no clinically significant clinical observations related to the test article during the study period.

Blood work collected during dialysis revealed a slight decrease in red blood cell count, hemoglobin, platelets and hematocrit at the 5 minute time point for all animals. The values returned to within normal limits or remained at or near baseline during the remainder of the study. There were no clinically significant abnormalities in the erythrogram.

The white blood cell count overall decreased for all animals throughout dialysis. Any slight increases throughout the procedure are likely attributed to stress as a result of the procedure and anesthesia and are not considered

unexpected. With the exception of one animal (173), the white blood cell counts remained within normal limits even though the values decreased over time. The overall percentage of white blood cell loss ranged from 7.9-40.2% by the end of dialysis. The white blood cell with the highest percent loss during dialysis was monocytes, eosinophils and lymphocytes. The loss was approximately 53.5-73.8% for monocytes, 0-100% for eosinophils and 1.9-64.1% for lymphocytes. Monocyte, eosinophil and lymphocyte percent reference ranges are 1-7%, 0-6% and 41-81%. Monocytes and eosinophil make up a relatively small percentage of the overall white blood cells.

This white cell loss was found in both the test and control animals. It appears that cell loss is due to the dialysis procedure with possible loss due to cell attachment to various blood-contacting surfaces of the dialysis circuit (e.g. tubing, catheter, dialyzer etc). There may be some cell loss in the test article from cells adhering to the beads, however, it does not appear that this is a significant contributor since there was very little difference in values between test and control animals.

There were no clinically significant abnormalities in the clinical chemistry that were considered due to the test article. The test article performed as expected. There were no cavitations, leaks or abnormal pressures. In addition, there were no leaks, obstructions or unusual flow patterns for these animals during dialysis. The test article performed as expected and evaluation was considered successful.

1.7. Current Standard of Care

Hemodialysis access-related bacteremia is a frequent complication in patients on renal replacement therapy. These patients are normally treated with catheter removal to achieve source control in conjunction with empirical antimicrobial therapy aimed at the most common offending pathogens. The antibiotic therapy is then focused after 24-48 hours when the identification and antibiotic sensitivity of the infecting organism is known.

1.8. Study Rationale

Infection is the second leading cause of death in the hemodialysis patient population. (Suzuki, et al. 2016) Hemodialysis access-related bacteremia is a frequent complication in patients on renal replacement therapy and these patients are at a 26-fold higher risk for infection. (Suzuki, et al. 2016) *Staphylococcus aureus* and methicillin-resistant *Staphylococcus aureus* account for 50% of the bacteremia. (D'Amato-Palumbo, et al. 2013) The incidence rate of *Staphylococcus aureus* bacteremia (SAB) in this population has been found to be 1.2 per 100 patient-months and ~8.5% of these patients have an SAB event during their hemodialysis career. (Marr, et al. 1998) (Li 2009) The development of infectious endocarditis occurs in 12-17% and non-cardiac infectious complication such as septic arthritis and osteomyelitis occur in 10-17%. Mortality estimates due to *Staphylococcus aureus* bacteremia in the hemodialysis population are 14-19%. (Marr, et al. 1998) (Li 2009) In a recent large trial of Daptomycin versus standard therapy for Bacteremia and Endocarditis due to *Staphylococcus aureus* the 42-day mortality was 15-16%. (Fowler 2006). While *Staphylococcus aureus* (including MRSA) account for the majority of infections in the hemodialysis patient, several other bacteria cause bacteremia. The causative pathogens and the percent of cases are summarized below. (D'Amato-Palumbo, et al. 2013)

Pathogen	Bacteremia Cases (%)
Staphylococcus aureus (including MRSA)	50.9
Other *Staphylococcus aureus*	10.7
Streptococcus	2.7
Enterococcus	8.9
E. coli	4.5
Klebsiella spp.	5.9
Pseudomonas spp.	8.4
Candida spp.	3.6

Bacteremia that is prolonged in duration (> 72 hours) is an independent predictor of patients who go on to suffer cardiac and non-cardiac infectious complications. (Fowler 2003). The working hypothesis for this trial is that hemoperfusion over the Seraph 100 Filter should remove circulating bacteria and, in conjunction with appropriate antibiotic therapy shorten the duration the bacteremia and thereby prevent infectious complications. Including the Seraph 100 Microbind Affinity Blood Filter in the hemodialysis circuit does not expose the patient to any additional circuitry during the study.

2. Intended Use and Study Objectives

2.1. Intended Use Statement

The Seraph 100 Microbind Affinity Blood Filter is indicated as an adjunctive treatment for blood stream infection (BSI) in patients on renal replacement therapy.

2.2. Study Objectives

This study is designed to confirm the safety and performance of using the Seraph 100 Microbind Affinity Blood Filter in the hemodialysis circuit for reducing the bacterial load and pro-inflammatory markers in the bloodstream of patients who have catheter-related bacteremia. The effectiveness endpoint is a mean pathogen reduction of > 40% in blood passed through the Seraph 100 Filter. The pre-cartridge blood sample must have a detectable bacterial count. Hematology indices will be measured and the incidence of adverse events will be recorded to study safety for up to 2 weeks post treatment.

3. Patient Population

3.1. Number of Patients

15 patients will be enrolled in the study. Enrollment will be sequential initially, with the first patient being treated and followed for 14 days. The Data Safety Monitoring Board (DSMB) will review results before a second patient can be treated. After two patients have been enrolled and followed under this scheme, enrollment will be allowed to proceed as patients who meet the inclusion/exclusion criteria become available.

3.2. Inclusion Criteria

Patients must meet ALL the following inclusion criteria to potentially be included in the study:

1. Require renal replacement therapy.
2. Be ≥ 18 years old and ≤ 90 years old
3. Positive blood culture and one of the following:
   a. Clinical evidence of a catheter exit site to tunnel infection as evidenced by redness, tenderness or purulence.
   b. Bacteremia is proven with two separate blood cultures from independent vein punctures.
   c. A blood culture where the time to positivity is within 14 hours.
   d. Growth from a blood culture taken from the hemodialysis catheter 2 or more hours before the growth of a blood culture drawn peripherally at the same time.
   NOTE: Written results for bacteremia and pathogen identification do not necessarily need to come from the hospital lab and written results can be provided by an outside clinic. The test results from the external source should have been generated within one (1) week prior to hospital admission.
4. The patient or patient's legal representative is able to understand the requirements of the study and signs an approved informed consent form prior to enrollment which explains aggressive care.

3.3. Exclusion Criteria

Patients who meet ANY the following exclusion criteria will NOT be included in the study:

1. Have an arteriovenous polytetrafluoroethylene (PTFE) graft.
2. Lack of a commitment to full aggressive support.
3. Have inability to maintain a minimum mean arterial pressure of ≥ 65 mm Hg despite vasopressor therapy and fluid resuscitation.
4. Have had chest compressions as part of cardiopulmonary resuscitation (CPR)
5. Have had an acute myocardial infarction (MI) within the past 3 months.
6. Have had serious injury within 36 hours of screening.
7. Have uncontrolled hemorrhage.
8. Are not expected to live > 14 days.
9. Have malignancy and are not expected to live 42 days.
10. Have neutropenia (absolute neutrophil count <500 cells/µL).
11. Have Child-Pugh Class C cirrhosis.
12. Have New York Heart Association Class IV Heart Failure or an ejection fraction <30%.
13. Have known Antithrombin III deficiency.
14. Have platelet count <30,000/µL.

15. Cannot have intravenous (IV) supplemental iron halted during trial period.
16. Are currently involved in an investigational drug or device trial.
17. Have been previously enrolled in this clinical trial.
18. Next hemodialysis treatment will not take place for at least 24 hours after enrollment.
19. Serious bleedings and clotting disorders, determined by blood transfusion of > 2 units of packed red blood cells, **or**, An acute (48 h) hemoglobin decline of at least 2 g/dL, transfusion requirement of >4 units over 48 h, objective evidence of bleed, documented by physician.
20. Breast feeding and pregnant women
21. Contraindications for heparin sodium for injection are:
    a. Have heparin sensitivity
    b. Severe thrombocytopenia.
    c. With an uncontrolled active bleeding state, except when this is due to disseminated intravascular coagulation.
    d. In whom suitable blood coagulation tests, e.g. whole blood clotting time, partial thromboplastin time, etc cannot be performed at appropriate intervals (this contraindication refers to full-dose heparin; there is usually no need to monitor coagulation parameters in patients receiving low-dose heparin)
22. Serious injuries, which have occurred more than 36 hours, have to be excluded.

3.4 Justification of Certain Inclusion and Exclusion Criteria

a. Inclusion criteria No. 2: Be ≥ 18 years old and ≤ 90 years old

According to the United States Renal Data System the incidence rate of dialysis requiring chronic kidney disease is highest in the age group > 75 years. This age group is also the fastest growing age group of dialysis patients. (http://www.usrds.org/2012/pdf/v2_ch1_12.pdf). In Europe 45 % of all new dialysis patients are aged >70 years (Nephrol Dial Transplant. 2014 Oct;29(10):1956-64. doi: 10.1093/ndt/gfu253. Epub 2014 Jul 24. Use of vascular access for haemodialysis in Europe: a report from the ERA-EDTA Registry).

Age statistics published in the *United States Renal Data System. 2016 USRDS annual data report: Epidemiology of kidney disease in the United States. National Institutes of Health, National Institute of Diabetes and Digestive and Kidney Diseases, Bethesda, MD, 2016,* indicates that the prevalence of ESRD in the age group greater than 75 years increasing.

In Germany the average age at the start of hemodialysis therapy is 68 years, hence an age limit of 90 years would exclude patients for which this therapy is targeted.

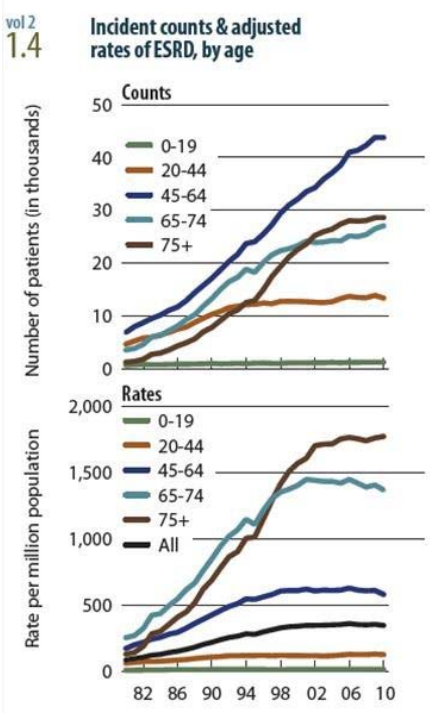
The age limit of < 90 years is requested due to

- There is an unmet medical need by patients in the greater than 80-year age group because the incidence of infections in tunneled catheters is significantly higher in the older patient population. The lower number of patients in the 80-90 age group is more than compensated by their increased incidence of catheter infections. This group could really benefit from a well tolerated treatment (see next bullet) administered during dialysis which is capable of removing a wide range of pathogens from the blood stream.
- Regarding safety: This group is already on dialysis thus demonstrating their tolerance of extracorporeal treatments. The 3 patients already treated in this study were treated in a standard dialysis unit and not in

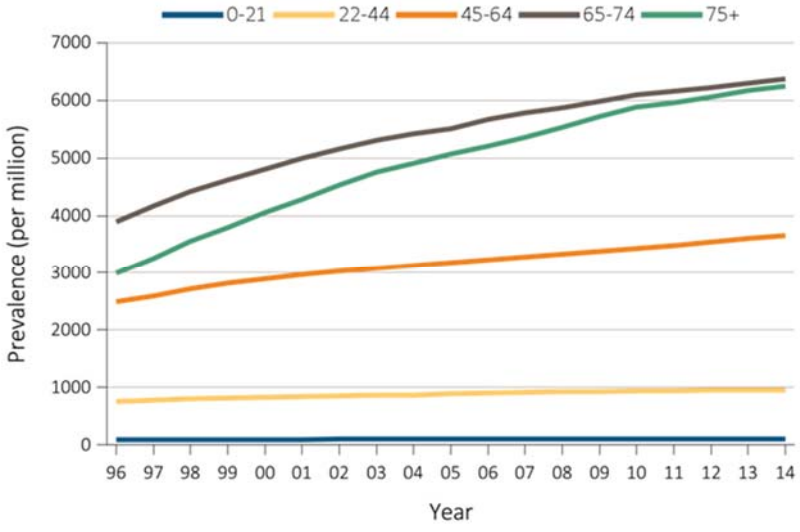
the ICU. In each case the Seraph treatment was well tolerated and essentially transparent to the patient. Compatibility with the Fresenius dialysis machines was excellent with no alarms sounding during the treatments. No negative effect on vital signs was seen in any of the three patients. On the contrary, their vital signs remained the same or improved during treatment.

Table 1. Baseline characteristics of patients new on HD

	Total[a]
n (%)	13 044
Age, mean (SD)[#]	65.1 (14.9)
Age categories (years, %)[$]	
20–44	11.5
45–59	19.9
60–69	22.9
70–80	31.1
> 80	14.5



Prevalence of Hemodialysis Patients by Age Group



b. Inclusion criteria No. 3: Definition bacteremia

"It is recommended to collect 2 - 4 blood cultures from different punctures sites, including intravascular catheters where appropriate." Hence in patients central venous catheters (in this case dialysis catheters) it is advised to draw blood from a peripheral puncture side as well from the central venous catheter at the same time (so called "paired blood cultures"). If the time to positivity differs >2 hours between the two sides, i.e. the blood culture from the catheter growth bacteria earlier than the blood culture from the peripheral vein, this is regarded as prove for a catheter related infection.

http://www.divi.de/images/Dokumente/Empfehlungen/Leitlinien/S2/Sepsis_Leitlinie_20104.pdf

Inclusion Criteria No3: Definition Positive Blood Culture

The criterion of 'Positive Blood Culture' by automated Time to Positivity (TTP) analysis is a TTP of < 36 hours. The bacteria to be included within this TTP range are MRSA, *S. aureus, K. pneumoniae (CRE), K. pneumoniae, E. coli (CRE), E. coli, S. pneumoniae, E. faecalis, E. faecalis (VRE), E. faecium, A. baumannii, S. epidermidis, Methicilin resistant S. epidermidis, S. pyogenes, Serratia marcescens*.

c. Inclusion criteria No 3.c: A blood culture where the time to positivity is within 14 hours.

Patients that have a positive blood culture within 14 hours after the initiation of incubation will be included in this study. Khatib *et. al.* demonstrated that a positive blood culture within 14 hours may identify patients with significant risk of metastatic infections. It is also a strong indicator of a catheter related infection.

Khatib, Riad, et al. "Time to positivity in Staphylococcus aureus bacteremia: possible correlation with the source and outcome of infection." *Clinical infectious diseases* 41.5 (2005): 594-598.

Patients with E. coli bacteremia – "Median TTP was significantly shorter for patients who died than for those who survived (9.7 h, inter-quartile range 7.85–11.05 h vs. 11.2 h, inter-quartile range 10.1–11.4 h; p <0.001). Patients with TTP in the lowest quartile were more likely to be female, to have a non-urinary tract or an unknown origin of bacteraemia, to have severe sepsis or shock, and to subsequently die."

Peralta, G., et al. "Time-to-positivity in patients with Escherichia coli bacteraemia." Clinical microbiology and infection 13.11 (2007): 1077-1082.

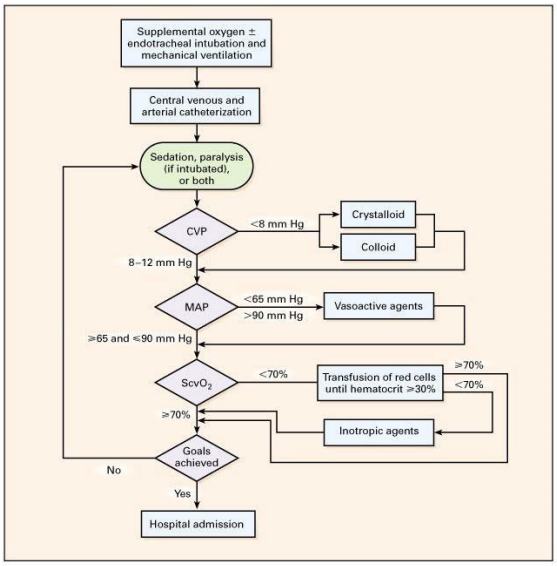
Patients with K. pneumoniae bacteremia – "TTP <7 h (46 patients, 19.9%) was associated with a higher Pittsburg bacteraemia score (6.2 ± 5.5 vs. 3.7 ± 4.3, p 0.002), fewer non-fatal diseases by the McCabe classification (39.1% vs. 64.9%, p 0.002), a higher percentage of patients with liver cirrhosis, active malignancy, and chemotherapy within 3 months (28.3% vs. 11.9%, p 0.007; 28.3% vs. 14.6%, p 0.031; 23.9% vs. 5.4%, p <0.001), more primary bacteraemia (45.7% vs. 22.2%, p 0.002), and a higher 30-day mortality rate (47.8% vs. 21.1%, p <0.001)."

Liao, C-H., et al. "Correlation between time to positivity of blood cultures with clinical presentation and outcomes in patients with Klebsiella pneumoniae bacteraemia: prospective cohort study." Clinical microbiology and infection 15.12 (2009): 1119-1125.

d. Exclusion criteria No.3: Have inability to maintain a minimum mean arterial pressure of ≥ 65 mm Hg despite vasopressor therapy and fluid resuscitation.

A MAP of >65 mm Hg is one of the three pillars of sepsis therapy and an established threshold to identify the unstable patient according to Early goal-directed therapy in the treatment of severe sepsis and septic shock.

Rivers E, Nguyen B, Havstad S, Ressler J, Muzzin A, Knoblich B, Peterson E, Tomlanovich M; Early Goal-Directed Therapy Collaborative Group. N Engl J Med. 2001 Nov 8;345(19):1368-77. A MAP < 65 mm Hg despite administration of volume and catecholamine therapy would identify a patient at increased risk for cardiovascular stability, i.e. a patient in whom this therapy could confer adverse effects.



e. Exclusion criteria No. 10

Have neutropenia (absolute neutrophil count <500 cells/µL).

Neutropenia increases the risk of infection and is also a sign of severe sepsis. According to national guidelines (http://www.awmf.org/leitlinien/aktuelle-leitlinien.html) it also requires a broad spectrum of medical measures including the administration of granulopoiesis stimulating hormone (Granulocyte-Colony Stimulating Factor, G-CSF). Further, as bacterial clearance is dependent on the number of leucocytes neutropenic patients It is unclear whether this treatment could be influenced by the procedure we aim to examine.

f. Exclusion Criteria No. 14.

Have platelet count <30,000/µL

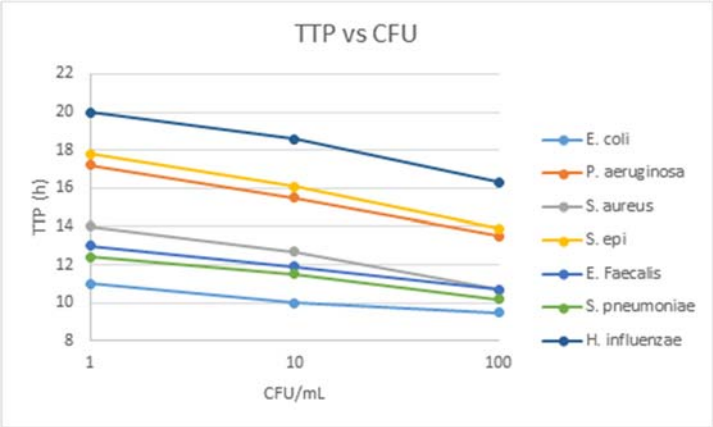
Extracorporeal adsorptive procedures may impose the risk of reducing the platelet count (Ther Apher. 2002 Jun;6(3):189-92. Hemoadsorption in critical care. Ikeda T), therefore we aim to exclude patients that do already have a low platelet count before the procedure.

g. Add Time to Positivity (TTP) Measurement

The measurement of CFU/mL requires manual counting of Colony Forming Units on inoculated culture plates in the microbiology laboratory. However, current clinical practice in all of our clinical centers is to use Automated

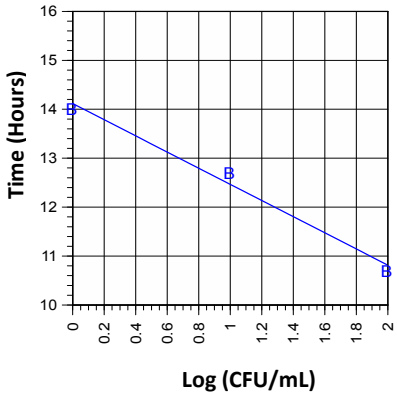
Blood Culture Instruments. It is therefore much more common to report the Time to Positivity (TTP) measured by an automated instrument and to use that time as an inverse measure of the intensity of the infection. That is, a shorter time to positivity is caused by a higher sample pathogen concentration, whereas a longer TTP is associated with a sample that contains less pathogen and therefore requires more time in culture for the pathogen to grow to the level that gives a measurable positive endpoint.

Idelevich et al, from our clinical center in Muenster, have published data that confirm a linear relationship between TTP and log[CFU/ml] for several pathogens.



Time to positivity vs. log[CFU/mL] for seven different pathogens plotted using data from Idelevich *et al* [1] showing a near linear relationship in the concentration range typical of human bacteremia, e.g., 1 to 100 CFU/mL.

As an example, replotting the *Staph. Aureus* data from Idelevich and fitting it to a linear regression line gives the results below. A 40% reduction in *Staph. aureus* per pass of blood through the Seraph 100 filter is estimated to increase TTP by only 22 minutes. The mean increase in TTP in our first patient was measured at 468 minutes or 7.8 hours, equivalent to >99% reduction per pass.



Linear Regression:

TTP = 14.12 - 1.65*Log[CFU/mL] **r^2 = 0.99**

40% Reduction in CFU/mL per pass = + 0.37 hr (22 min) change in TTP

Mean change in TTP during treatment = 7.8 hr (470 min)

Calculation of the change in TTP for *Staph. aureus* due to a 40% reduction in CFU/mL using data from Idelevich et al.

Based on the considerations presented here, and the assumption that the expected change in TTP per pass of blood through the Seraph 100 Filter will produce a measurable increase in TTP, we believe that using TTP from automated blood culture constitutes a necessary and reasonable change to our protocol.

4. Study Design

4.1. Informed Consent

The investigator must obtain written informed consent prior to patient's enrollment and participation in the study. The rationale, information of the study specifics, including the potential risks and benefits of the Seraph 100 Microbind Affinity Blood Filter will be explained to the patient prior to obtaining the written consent from the patient.

4.2. Screening Procedures/Baseline Visit

All patients who meet the inclusion criteria will be included in the screening log. If a patient has an exclusion criterion this will be noted in the screening log. Patients meeting all inclusion and no exclusion criteria will be asked to sign an informed consent prior to any protocol procedures being performed, then the patient is considered enrolled. The investigator will follow the Baseline tests as per the schedule in the table in Section 5.1 All patients are tested by the microbiology laboratory to have bacteremia.

4.3. Concomitant Medications

Patients can be prescribed any concomitant medication or other medical procedure during the study period as deemed appropriate by the Investigator. Concomitant medications/procedures prescribed at baseline should be listed on the Medication Log Case Report Form (CRF). If medications are added or changed, they should be indicated on the Concomitant Medication Log.

Standard of care medication regimens are provided below as guidelines:

- Empiric Therapy for Gram positive bacteremia (pending sensitivities): Vancomycin loading dose 25mg/kg IV + Cefazolin 2gm IV.
- Methicillin-sensitive *Staphylococcus aureus* (MSSA): MSSA: Cefazolin 2 gms, 3x weekly IV.
- Anaphylaxis or hives with penicillin: Daptomycin intradialytic dose of 7 mg/kg for low permeability dialyzers or 9 mg/kg for high-permeability dialyzers.
- Methicillin-resistant *Staphylococcus aureus* (MRSA):
    - Vancomycin Minimum Inhibitory Concentration ≤ 2: Vancomycin 25mg/kg IVloading dose, then 500mg at the end of each dialysis session.
    - Vancomycin Minimum Inhibitory Concentration ≥ 2: Daptomycin 6mg/kg IV every 48 hours.
- Empiric therapy for Gram negative bacteremia (pending sensitivities): Cefepime 1 gm q24IV or Meropenem 1 gm q24IV or Ceftazadime 1 gm load and 1 gm post HD
- Targeted therapy for Gram negative bacteremia should be based on the antibiotic susceptibility of the Gram negative organism

4.4. Treatment Procedure

If the patient is receiving supplemental IV iron during dialysis treatments, the delivery of IV iron must be stopped during the trial period (day of procedure + 14 days of follow-up).

Once the patient is enrolled, treatment with the Seraph 100 Filter will be concurrent with the next dialysis treatment, to be scheduled no later than 24 hours from enrollment. The dialysis cartridge may be a Fresenius Medical Care, FX 80 Capillary High-Flux Dialysers/Haemodiafilter or any legally marketed dialysis cartridge.

Systemic heparinization during hemodialysis with concomitant filtration with the Seraph 100 Filter is recommended with a 3-5 minute waiting period after the initial heparin bolus before beginning dialysis. The Seraph 100 Filter should be placed in the dialysis circuit before the dialysis cartridge. The dialysis circuit will be primed and de-air as recommended by the manufacturer's directions, with an additional 1 L of prime solution to accommodate the Seraph 100 Filter.

Dialysis should be run at 250-350 mL/min for 4 ± 1 hour. The concurrent treatment procedure will be in accordance with the Instructions for Use, except that during the trial dialysis, blood samples will be taken before and after the Seraph 100 Filter cartridge using Sponsor supplied tubing sets. The schedule for taking the pre- and post- cartridge blood samples is as follows: 5 minutes, 30 minutes, 60 minutes, 120 minutes, 180 minutes and 240 minutes during dialysis. The trial device and blood samples will be saved for further analysis.

4.4.1 Instructions for procedure

Refer to Instructions for Use, Document CP003 'Seraph® 100 Microbind® Affinity Blood Filter, Instructions for Use'

5. Follow-up Evaluations

Section 5.1 contains a schedule of events including testing that is performed on the blood samples that are taken during the patient enrollment in the study. The following are some highlights of the schedule of events during the procedure:

- Pre- and post- cartridge blood samples at: 5 minutes, 30 minutes, 60 minutes, 120 minutes, 180 minutes and 240 minutes during treatment to determine pathogen content by measuring colony forming units (CFU/mL) or time to positivity (TTP).
- Pre cartridge blood samples are taken before the Seraph cartridge.
- Post cartridge blood samples are taken after the Seraph cartridge but before the dialysis cartridge.
- Hematology, clinical chemistry, and coagulation tests are performed at start of procedure (t=0), end of procedure (t=4hours) and 30 ± 5 minutes post procedure.
- Complement levels are measured at start of procedure (t=0), end of procedure (t=4hours).
- Cytokine levels are measured at 5, 120 and 240 minutes.
- Kt/V urea is measured only as part of the standard dialysis treatment.
- Measurement of antibiotic level is optional and is measured pre- and post-cartridge during the procedure.
- Rotem analysis is optional but desirable. Samples may be taken pre- and post-Seraph at start of procedure and at 30 ± 5 minutes post procedure

5.1. Test Schedule

PPD = Post Procedure Day

Schedule of Events

Schedule of Tests:	**Baseline**	**Procedure (Day 0) 0-4 hrs**	**PPD Day 1**	**PPD Day 2,3,4,5,6**	**PPD Day 7 - 0, +1**	**PPD Day 14 ± 1**
Inclusion/exclusion	X					
Informed Consent	X					
Demographics	X					
Medical history	X					
Physical Exam	X	X	X	X	X	X
Vital Signs • Body weight • Temp • Respiratory Rate • Heart Rate • Main Arterial Pressure • Oxygen saturation	X	X	X	X	X	X

Schedule of Tests:	**Baseline**	**Procedure (Day 0) 0-4 hrs**	**PPD Day 1**	**PPD Day 2,3,4,5,6**	**PPD Day 7 - 0, +1**	**PPD Day 14 ± 1**
Quantitative Blood cultures 5, 30 ,60, 120, 180 and 240 minutes during procedure – pre and post filter using Sponsor supplied tubing sets • CFU or TTP Identify pathogen		X				
Post Procedure Quantitative Blood cultures • CFU or TTP Identify pathogen			X	X	X	X
Antimicrobial susceptibility on all bloodstream isolates		X	X			
Transthoracic Echo					X	
Laboratory - hematology • WBC with diff • Hgb • HCT • Platelets • Thrombocytes	X	X (start and end of procedure and 30 ± 5 minutes post procedure)	X	X	X	X
Antibiotic Levels (optional)		X (during procedure at 30 minutes)				

Schedule of Tests:	**Baseline**	**Procedure (Day 0) 0-4 hrs**	**PPD Day 1**	**PPD Day 2-6**	**PPD Day 7 - 0, +1**	**PPD Day 14 ± 1**
Laboratory - chemistry • Total protein • Albumin • AST • ALT • Alkaline phosphatase • Total bilirubin • Direct bilirubin • Indirect bilirubin • Urea • Creatinine • Glucose • Sodium • Potassium • Calcium • CPK • Immunoglobulins • Kt/V urea (only during dialysis treatment)	X	X (start and end of procedure and 30 ± 5 minutes post procedure)	X	X	X	X
Laboratory - coagulation • PT-INR • PTT • D-dimer • Fibrinogen • TATc • Antithrombin • Rotem Analysis (Optional)	X	X (start and end of procedure and 30 ± 5 minutes post procedure)	X	X	X	X
Laboratory – complement levels • CH50 • C3a • C5a		X (start and end of procedure)				
Laboratory – cytokines • TNF-α • IL-6 • IL-10		X (during procedure at 5, 120 and 240 minutes)				
Adverse Events (record all)		X	X	X	X	X

5.2. Post study care

Post study, the patient is following the standard care as part of the hemodialysis treatment.

6. Patient Safety and Adverse Event Reporting

6.1. Adverse Event

An Adverse Event (AE) is defined as any untoward medical occurrence, unintended disease or injury or any untoward clinical signs (including an abnormal laboratory finding) in subjects, users or other persons whether or not related to the investigational medical device.

A new condition or the worsening of a pre-existing condition will be considered an AE.

Stable chronic conditions present prior to study entry and do not worsen during the study will not be considered as an AE.

Any symptom, sign, illness, injuries or experience that develops or worsens in severity during the course of the study should be regarded as adverse events. Moreover, abnormal results of diagnostic procedures are considered to be adverse events if the abnormality:

- results in study withdrawal,
- is associated with a serious adverse device effect,
- is associated with clinical signs or symptoms,
- leads to additional treatment or to further diagnostic tests,
- is considered by the investigator to be of clinical significance.

6.2. Serious Adverse Event

Serious adverse event is every occurred unwanted event in a clinical study requiring approval or a performance evaluation requiring approval which has led, may have lead or may lead:

- Immediate or mediate to dead or
- A serious deterioration of the health status of a subject, user or another person without considering if the event has been caused by the medical device;

6.3. Serious Adverse Device Effects (SADEs)

Serious adverse device effects (SADEs) are adverse device effects that have resulted in any of the consequences characteristic of a serious device event or that might have led to any of these consequences if suitable action had not been taken or intervention had not been made or if circumstances had been less opportune.

A Serious Adverse Device Effect is a SAE attributed to the study device; an adverse device effect that has resulted in any of the consequences characteristic of a serious adverse event.

6.4. Unanticipated Serious Adverse Device Effect

An Unanticipated Serious Adverse Device Effect (USADE) is any Serious Adverse Event (effect, problem) on health or safety, or any life-threatening problem or death caused by or associated with the investigational device, if that event, effect, problem, or death was not previously identified in nature, severity, or degree of incidence in the investigational plan/protocol or application (including a supplementary plan or application), or any other unanticipated serious problem associated with the investigational device that relates to the rights, safety, or welfare of the subjects.

6.5. Severity Definition

- Mild: AE/SAE which is easily tolerated;
- Moderate: AE/SAE sufficiently discomforting to interfere with daily activity;
- Severe: AE/SAE that prevents normal daily activities.

The investigator will document his opinion of the relationship of the AE/SAE to the investigational medical device using the criteria summarized below.

- Unrelated: The AE/SAE has no temporal relationship to the investigational device or study procedure, and/or there is evidence of alternative cause such as concurrent medication or illness;
- Possibly related: A temporal relationship with the investigational device or study procedure is not clear, alternative causes are also possible;
- Probably related: A clear cut temporal relationship to the use of the investigational device or study procedure and potential alternative etiology is not apparent;
- Definitely related: A clear-cut temporal relationship to the use of the investigational device or study procedure and no other possible cause.

6.6. Potential Adverse Events

Potential risk associated with the standard hemodialysis are:

- Hypotension
- Hypoxemia
- Thrombus formation
- Bruising at the catheter entry site
- Arrhythmia
- Cramps
- Death
- Device leaks or obstructions
- Edema
- Hematoma
- Infection
- Hemodynamic instability

Anticipated adverse events under this study that are not expected to require reportable events are:

- Infection
- Bruising at the catheter entry site
- Hematoma
- Stable chronic conditions present prior to study entry and do not worsen during the study will not be considered as an AE.

Additional potential adverse events which may be associated with the use of the investigational device include, but are not limited to:

- Leaks, obstructions, or unusual flow patterns during dialysis
- Cavitations, or cartridge leaks
- Thrombus formation on the device

6.7. Management and Reporting of SAE and SADE

Any serious adverse events, device related or not, and irrespective of the potential causal relationship to the study, must be reported as soon as possible, preferably within 24 hours of the Investigator's knowledge of the event to the sponsor and to the monitor. Suspected serious adverse events should also be reported. As per relevant local regulatory requirements, the sponsor will communicate SAE information to the appropriate regulatory agencies.

Serious adverse device effects must be reported as soon as possible, by the investigator/Co-investigator to the sponsor and to the monitor, preferably within 24 hours of the Investigator's knowledge of the event. Suspected serious adverse events should also be reported. As per relevant local regulatory requirements, the Sponsor will communicate SADE information to the appropriate regulatory agencies.

Any adverse event or adverse device effect must be followed until it has resolved, has a stable level of sequelae or in the investigator's opinion is no longer clinically relevant. Supporting studies, treatment modalities and resolution status are recorded on the adverse event CRF.

As described above, in the case of an observed SAE, which may or may not be related to the use of the study device, the Sponsor, represented by Medpace, must be notified immediately (no later than two (2) working days after detection) by fax and/or by emailing a completed adverse event form and SAE notification form:

Medpace Germany GmbH
Tel: +49 (0)89 89 55 718 44
Fax: +49 (0)89 89 55 718 104
E-mail: medpace-safetynotification@medpace.com

If any further information is required, the Sponsor, represented by Medpace, will contact the investigator. All other adverse events must be reported with five (5) days of the investigator's knowledge of the event to:

Medpace Germany GmbH
Tel: +49 (0)89 89 55 718 44
Fax: +49 (0)89 89 55 718 104
E-mail: medpace-safetynotification@medpace.com

As described below, following Sponsor evaluation of each adverse event, health authorities and/or responsible Ethics Committees (EC) will be notified as appropriate.

6.8. Notification to the EC or Competent Authority

Notification to German authority (BfArM)

The BfArM is to be informed about the event as defined per Directive 93/42/EEC, the Medical Device Law (Medizinproduktegesetz - MPG) of 07Aug2002 (Federal Law Gazette I Page 3146) and the Medical Device Safety Plan Ordinance (Medizinprodukte-Sicherheitsplanverordnung - MPSV) of 24Jun2002 as amended from time to time.

Device- or procedure related Serious Adverse Events, occurring in Germany, will be reported by the Sponsor to the Competent Authority immediately by using the Serious Adverse Event Report Form available on the BfArM website. Device- or procedure related Serious Adverse Events occurring abroad in a study also performed in Germany will be reported by the Sponsor to the Competent Authority immediately using the MEDDEV 2.7/3 SAE Reporting Form. Serious Adverse Events deemed unrelated to the device or the procedure, occurring both in Germany and abroad, will be reported by the sponsor to the Competent Authority on request or at least quarterly, by using the MEDDEV 2.7/3 SAE Reporting Form.

Contact point for the authorities:

Krisztina Nagy
Senior Clinical Safety Manager
Clinical Safety

Medpace UK
26-28 Hammersmith Grove
London W6 7HA
United Kingdom
Tel: +44 (0) 208 563 5900 Ext: 25712
E-mail: medpace-safetynotification@medpace.com

6.9. Device Deficiencies and Malfunctions

All device deficiencies related to the identity, quality, durability, reliability, safety or performance of the medical devices shall be documented. This includes unexpected outcomes or device deficiencies that might have led to a serious adverse event if a) suitable action had not been taken or b) intervention had not been made or c) if circumstances had been less fortunate. The adverse event is reported on the Adverse Event Case Report Form. Definitions for device deficiencies are included in the Appendices.

The Product Experience Form is specific for reporting all device deficiencies, or malfunctions that occur during the course of the trial, whether to not they were associated with an adverse event. Product Experience Forms should be submitted to the sponsor within 24 hours of the occurrence defining the device deficiency. This includes unexpected outcomes or device deficiencies that might have led to a serious adverse event if a) suitable action had not been taken or b) intervention had not been made or c) if circumstances had been less fortunate.

6.10. Safety Related Stopping Rules/Data Safety Monitoring Board

A Data Safety Monitoring Board (DSMB) comprised minimally of a statistician, and two independent physicians, one of whom is a nephrologist and one of whom specializes in infectious diseases, all experienced with clinical trials, will monitor the progress of the trial and the safety of participants. Members of the DSMB will be selected based on their experience, reputation for objectivity, absence of conflicts of interest (and the appearance of same), and knowledge of clinical trial methodology. Responsibilities of the DSMB will include the following:

- Be familiar with the clinical protocol and plans for data and safety monitoring.
- Recommend any updates to the informed consent documents if warranted by new safety information.
- Review reports of related studies to determine whether the monitored study needs to be changed or terminated.
- Review all adverse events with supporting documentation as appropriate and recommend halting the trial according to pre-established stopping rules. The DSMB will be informed of SAEs in real time. This requirement, in addition to formal stopping rules which are provided below, will be incorporated by the DSMB in their charter.
- Meet regularly and following each DSMB meeting, provide the Sponsor with written findings for the trial as a whole and any relevant recommendations related to continuing, changing, or terminating the trial. The DSMB will also be contacted for a review if unanticipated adverse events occur.

The Safety Data Monitoring Board will review the first two patients sequentially before additional patients can be subjected to the use of the Seraph Microbind Affinity Blood Filter concurrent with their hemodialysis. After two patients have been enrolled and followed for 14 days (consecutively), enrollment will be allowed to proceed as patients who meet the inclusion/exclusion criteria become available.

The following stopping rules will be incorporated into the DSMB charter. The trial will be halted if any of the following situations occur. Restart may not occur until the DSMB has reviewed the information and recommended a restart.

- Any death within 7 days of the procedure or during the period of hospitalization, whichever is longer, that is considered to be possibly related to the use of the device.
- Any significant bleeding event requiring 3 or more units of packed cells for 2 consecutive days.
- Any significant clotting event
- The DSMB will examine the data from all patients at the completion of the study, and prepare a final report in which their opinion of the relative safety of the Seraph Microbind Affinity Blood Filter is described.

7. Study Administration

7.1. Investigator Training

Prior to enrolling subjects in the trial, Investigators will be provided specific training on the procedural steps required to use the Seraph 100 Microbind Affinity Blood Filter.

7.2. Monitoring

For this study, the sponsor will be responsible for ensuring that appropriate monitoring of the study is performed. ExThera Medical can assign a CRO to perform the clinical monitoring. The CRO written procedures will be audited first to assure the monitoring will adequately assess the quality of the study and to assure that each person involved in the monitoring process carries out his defined duties. Standardized written procedures, sufficiently detailed to cover the general aspects of clinical investigations, will be used as a basic monitoring plan and will be supplemented by more specific or additional procedures, as required by the clinical investigation.

A pre-investigation "Qualification Visit" will be conducted to ensure that the Investigator clearly understands and accepts the obligations incurred in undertaking the clinical investigation and that the facilities are acceptable throughout the clinical investigation. Only qualified investigational sites will participate in the study. Periodic monitoring visits will be conducted with adequate frequency to ensure that the Investigator's obligations are being fulfilled and that the facilities continue to be acceptable. Direct contact between the Clinical Monitor and the Investigator will be maintained throughout the investigation, and the Clinical Monitor will visit the Investigator at the site of the investigation.

Monitoring activities will be in compliance with sponsor SOPs, monitoring plan good clinical practice (GCP) principles and ISO 14155:2011.

A study close-out monitoring visit will be conducted at the completion of the study to ensure that all clinical study materials and patient data are properly documented and returned to ExThera Medical.

7.3. Case Report Forms

CRFs for individual subjects will be provided by ExThera Medical. The investigator must keep a separate log of patient names and current addresses.

CRFs are used to record study data and are an integral part of the study and subsequent reports. Therefore, the reports must be legible and complete. All forms should be filled out using a black ball point pen. Errors should be lined out but not obliterated and the correction inserted, initialed and dated by the investigator.

7.4. Source Documentation Requirements

The Study Coordinator delegated by the Investigator will perform primary data collection drawn from participating subject's hospital source documentation review. Data to be collected for study purposes must not be transcribed directly into the applicable CRF before being recorded first in the hospital medical record. The data must be recorded from original source documents and available for review by the study monitor.

7.5. Data Management

Data items from the CRF will be entered centrally into a study database. Discrepancies will be checked against the original source documentation and any required corrections will be made to the database. The data will then be fully validated, using study-specific range and consistency checks and database listings. Obvious error is corrected and a query will be sent to the site/clinical designee for confirmation of the correction by the investigator. Other errors or omissions will also be sent to the clinical designee for resolution by the investigator. Resolved queries are signed by Investigator and corrected data entered into the database.

Original completed and monitored CRFs will be stored at the Sponsor offices, while copies will be maintained at the investigational site for a period according to the site's procedures.

Once the study data is clean, it will be statistically analyzed as described on the section 8 of this protocol.

7.6. Protocol Amendments

Amendments will originate from the Sponsor and will be implemented after regulatory approval from the competent authorities or ethics committee. It should be noted that when an amendment to a protocol substantially alters the study design or increases potential risk to the study subjects, the informed consent should be revised and, if applicable, a subject's consent to continue participation should again be obtained.

No deviations from the protocol should be made except where alternative treatment is necessary to protect the life or physical well-being of subjects in an emergency. In situations requiring a departure from the protocol, the Investigator or other physician in attendance will contact the Sponsor's clinical representative by fax or telephone within 5 days of the emergency. In all cases, contact with the Sponsor's clinical representative must be made as soon as possible in order to discuss the situation and agree on an appropriate course of action. Except in an emergency, prior approval from the Sponsor is required for changes in the protocol and, if these changes affect the soundness of the study, the rights, safety, or welfare of the subjects, the Sponsor must notify the Competent Authority and/or the EC. The case report form and source document will describe any departure from the protocol and the circumstances requiring it.

7.7. Protocol Deviations

The investigator should not implement any deviation from or changes to the protocol without prior approval by ExThera Medical. Reports of any deviation from the protocol conducted in an emergency situation, to protect the life or physical well-being of a patient, will be reported to the Sponsor or representative and to the Ethics Committee as soon as possible after detection, but no later than twenty four (24) hours from the time of the deviation. Deviations should be documented on the appropriate case report form. Some examples of deviations are post procedure testing/visits out of time window, medication regime not followed, patient consent not obtained. This list is not exhaustive and there may be other deviations.

Any report of withdrawal of Ethics Committee approval will be submitted to the Sponsor within five (5) working days.

7.8. Withdrawal of Patients

Subjects have the right to withdraw from the study at any time, for any reason, without jeopardizing their medical care. The Investigator, the EC and Exthera Medical also have the right to withdraw subjects from the study for the following reasons:

- when continuation may jeopardize the health of the subject,
- protocol violations,
- being lost to follow-up,
- adverse events or concurrent conditions,
- administrative,
- death,
- or other reasons.

Subjects can leave the study at any time for any reason without prejudice or any impact on subsequent medical treatment from the physician or site staff. The reason for withdrawal will be investigated and carefully documented in the subject's medical file and the appropriate section of the CRF. When a subject withdraws or is withdrawn from the study. All subjects withdrawn from the study will undergo a final study visit consisting of a safety evaluation; as such subjects will continue to receive normal, standard of care follow-up by the study physician outside the scope of the approval clinical study protocol.

The investigator can decide to withdraw a subject from the study for urgent medical reasons, appropriate as a safety measure and/or if the subject's medical condition contraindicates further study participation. When a subject is withdrawn from the study, the final evaluation and the follow-up will be performed as completely as possible (to the extent to which the subject agrees to). In addition, any comments (spontaneous or elicited) or complaints made by the subject of any physician not related to the investigation but taking care of the subject subsequently will be carefully recorded in the subject medical file and related section of the CRF. After the study exit the subject would be followed up as per standard care.

Data for study subjects who withdraw their consent prior to planned study completion, are lost to follow up, or expire, are to be recorded on the Study Completion Form.

In case of "lost to follow up", every attempt must be made by the site to contact the subject, their relatives and their General Practitioner before categorizing them as "lost to follow up". The Sponsor must be notified of the reason for subject discontinuation and, as indicated above, the site records this information into the corresponding CRF and subject's hospital source documents.

Withdrawn subjects will be replaced to complete enrollment.

7.9. Maintaining Records

Source documents may include a patient's medical record, hospital charts, clinic charts, the investigator's study files, questionnaires, and the results of diagnostic tests such as laboratory tests, and echocardiograms. The investigator's copy of the CRFs serves as part of the investigator's record of a patient's study-related data.

The following information should be included in the patient's medical record:

- Patient's name and contact information;
- The study title, number/name, and sponsor name (ExThera);
- The date the patient was enrolled into the study and the patient ID number;
- A statement that written informed consent was obtained;
- Date of procedure, procedural type, and investigational device lot number;
- Hospitalization course;
- Visit dates;
- All medications;
- Occurrence of any adverse events;
- Date patient exited the study, and a notation as to whether the patient completed the study or discontinued, with the corresponding reason.

The investigator is responsible for ensuring that data are properly recorded on each patient's CRFs and related documents. The investigator who signs the protocol signature page should personally sign the final CRF to ensure that the observations and findings are recorded correctly and completely. The original CRFs are to be submitted to the data management group in a timely manner at the intervals specified by ExThera Medical.

All forms must be filled out completely. An explanation must be provided for any missing data points. Correction of data on the CRFs should be made by crossing out the incorrect data with a single line and writing the correct values next to those crossed out. Never obliterate incorrect entries. Each correction must be initialed and dated by the person making the correction. All CRFs will be reviewed for completeness, accuracy and clarity. Queries for missing or unclear data will be made as necessary.

7.10. Submitting Reports

An (interim) report and final report is submitted to the competent authorities and/or ethics committee per local requirements.

7.11. Site Record Retention Policy

All study related documentation, including CRFs, informed consents, regulatory documents, study manuals, training materials, device accountability records, and source documents (including electronic echocardiograms), must be maintained until ExThera Medical notifies the investigator in writing that they may be discarded or as long as the local law required (which ever comes later). ExThera Medical requires that it be notified in writing if the investigator wishes to relinquish ownership of the data so that mutually agreed upon arrangements can be made for transfer of ownership to a qualified entity.

8. Statistical Methods

8.1. Study Endpoints

Primary Endpoint:

- Safety:
  - SAEs occurring while connected to the device.
  - Clinically significant changes in hematology indices.
  - Clinically significant changes in chemistry indices.
  - Device complications.

Secondary Endpoints:

- Safety:
  - All clinical and laboratory adverse events post procedure 1-14 days.
- Performance:
  - Throughput flow effects
  - Compatibility with hemodialysis systems.
  - Changes in hemodynamics.
- Effectiveness:
  - A mean pathogen reduction of > 40% measured as colony forming units (CFU) or a mean increase in time to positivity (TTP) of > 22 minutes in blood passed through the Seraph 100 filter. The pre-cartridge blood sample must have a detectable bacterial count (as measured by CFU or TTP)
  - Duration of bacteremia
  - Bacteremia recurrence rate
  - Rate of cardiac and noncardiac infectious complications
  - Changes in cytokine levels

8.2. Sample Size Calculations

The primary endpoint of the study is to confirm the safety of the ExThera Medical Seraph 100 Microbind Affinity Blood Filter in a hemodialysis circuit. From discussions with DEKRA, the notified body used by ExThera Medical Corporation, DEKRA suggested that 15 patients would be sufficient to demonstrate safety.

An *in vitro* study was performed at the Department of Infectious Diseases at the University of Gothenburg under the direction of Tomas Bergstrom. In brief, a mixture of methicillin-resistant *staphylococcus aureus* (MRSA) and blood (4×10^5 CFU/mL) was passed through the Seraph adsorption media. The bound bacteria were eluted from the beads and quantified using real-time PCR. The MRSA mean percent reduction and standard deviation of six (6) samples was 66 % ± 9 %.

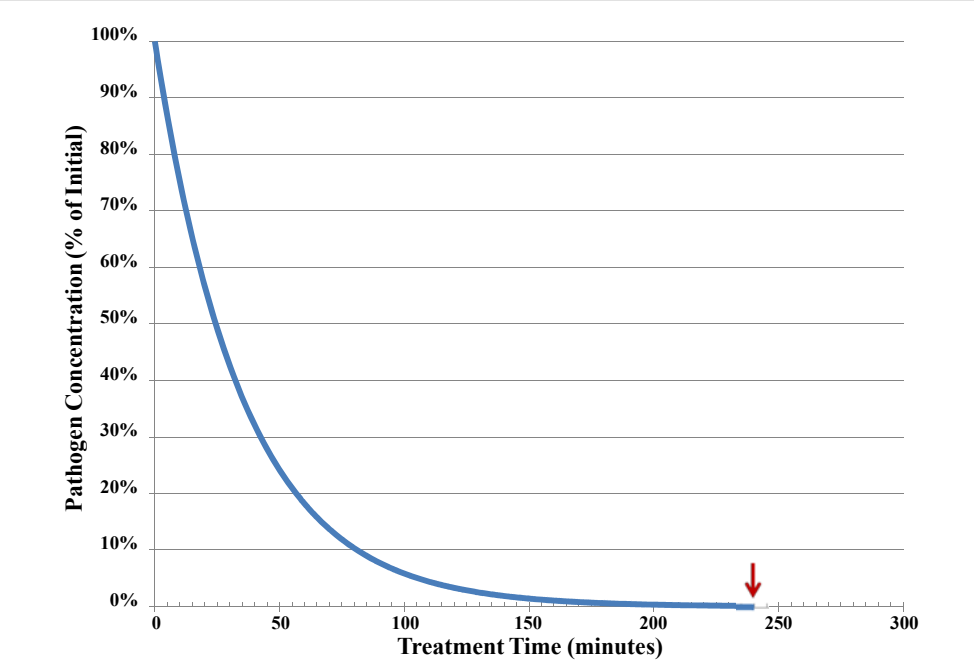


Figure 1. Expected reduction in bloodstream concentration of S.A or MRSA by Seraph® 100 Microbind® Affinity Blood Filter at 40% reduction per pass and 350 mL/min. At 240 minutes of treatment a 3 log (99.9%) reduction in bacteria concentration is expected.

Bacteremia patients with bloodstream infections typically have 100 to 1000 CFU/mL of blood. Blood from the patient is passed through the Seraph 100 filter during a dialysis treatment lasting approximately 4 hours. The MRSA or SA is removed from the blood and the 'clean' blood is returned to the patient. Whereas our model (Figure 1) assumes complete mixing of the returning clean blood with 5 liters of the patient's blood, this assumption is unlikely to accurately represent the clinical situation using veno-venous blood access.

For this reason we have chosen a more conservative goal of: a mean of 40% reduction as the secondary endpoint. Using the data from the in vitro study, mean reduction of 66% with 9% standard deviation only 3 patients would provide 80% power to confirm the hypothesis of >40% reduction. A 40% reduction in the MRSA or SA for a 4 hour treatment time (typical dialysis treatment) should result in a 3 log (99.9%) reduction by the end of a four-hour treatment. This sample size calculation was based on a one-sample t-test with a 0.05 one-sided level of significance.

The following populations will be considered for analysis of the data:

- Population treated with the investigated device.
- Population completed with follow-up through the primary and secondary assessment endpoints.
- Population who completed the study with no significant protocol deviations.

8.3. Statistical Analysis

In general, categorical variables will be summarized using frequencies and percentages of patients in each category. Ninety-five percent confidence intervals for percentages will be computed. Continuous variables will be summarized using descriptive statistics and 95% confidence intervals for means will be computed where appropriate.

Adverse events will be coded using MedDRA dictionary. SAE and treatment emergent adverse events will be summarized by system organ class and preferred term. Statistic modeling will be used to identify potential predictors of treatment success as well as safety events and performance.

8.4. Methods to Eliminate Bias

The site coordinator will maintain a log indicating the subjects eligible for the study at their centers and the resulting participation or non-participation with exclusion rationale. These data will be utilized in the design of future studies. The reported complications will be reviewed for accuracy at the time of reporting and verification of event to determine device relatedness.

8.5. Missing data

Attempts will be made to complete any missing data but missing data will not be imputed. Subjects with a missing outcome will not be included in the analyses. The number of subjects with missing outcomes will be identified for each of the analyses. A sensitivity analysis will be conducting to assess the potential impact of missing data on the study conclusions.

9. Clinical Risk/ Benefit Analysis

9.1. Risk Assessment

A risk analysis according to ISO 14971 Application of risk management to medical devices has been conducted. The risks associated with this investigational device, the Seraph® Microbind® Affinity Blood Filter, have been identified by performing Failure Mode and Effect Analysis (FMEA) / Risk Analysis, biocompatibility testing according to the ISO 14971 as well as a risk/benefit assessment.

Identified additional potential adverse events which may be associated with the use of the investigational device include, but are not limited to:

Adverse Event	Mitigation	Likelihood	Severity	Device Related	Risk Factor
Incorrect placement of device in dialysis circuit	Instruction for Use and Label, user training.	1	4	5	20
Blood loss by disconnection of tubing or tubing rupture	Instruction for Use, user training.	1	4	5	20
Air embolism	Instruction for Use, user training.	1	4	5	20
Hemolysis	Device design to accommodate blood flows up to 400 mL/min.	2	3	3	18
Particle embolism	Endplates hold particles in the column, as verified in performance testing.	1	3	5	15
Infection	Sterilization validation.	2	3	3	18
Damage to environment or property	Instructions for Use describe proper biohazard disposal is required.	1	1	5	5
Negative tissue interaction	Biocompatible materials, as verified in performance testing.	2	3	5	30
Thrombocytopenia	Heparin is permanently attached to adsorption media and does not leach off, as verified in performance testing.	2	3	3	18
Pyrogenic reaction	Device is non-pyrogenic, as verified in performance testing.	1	3	5	15
Toxic response	Biocompatibility testing and sterilization validation verified that device is biologically safe.	1	4	4	16
Device is ineffective and does not remove bacteria	Verification of material specification and lot release testing.	1	4	5	20
Total risk: 215					

Risk Event Severity Classification

1	No Clinical Effect – Physician/Patient expectation not met
2	Marginal – Reduction in device performance or minor patient injury not requiring intervention
3	Serious – Patient injury which may require intervention to preclude death or permanent disability
4	Critical – Patient injury which requires intervention to preclude death or permanent disability
5	Catastrophic – Product failure which will directly result in patient death or permanent disability

Adverse Event and Likelihood Classification

1	Improbable: <0.001%
2	Remote: 0.001 to 0.01%
3	Occasional: 0.01 to 1 %
4	Probable: 1 to 10%
5	Frequent [>10%]

Adverse Event Related to Device Classification

1	Unrelated to device
2	Unlikely related to device
3	Possibly related to device
4	Probably related to device
5	Definitively related to device

Risks have been proven, minimized or eliminated through appropriate design control, confirmed by pre-clinical bench, laboratory and animal testing. A summary of preclinical results is disclosed in the Clinical Investigator's Brochure.

Risks associated with the Seraph 100 Filter are similar to those associated with other filters used during hemodialysis. In addition, there are standard risks associated with standard hemodialysis procedure. Known and unexpected risks are relatively mitigated by working with an Investigator who is experienced and skilled in hemodialysis procedure with patients who show signs of sepsis. Additionally, the Investigator will be thoroughly trained on proper device properties and operation. Risks will also be minimized under this study protocol through adherence to the inclusion/exclusion criteria. Complications that may occur may include the following, but are not limited to:

Potential Adverse Event	Likelihood Rating
Hypotension	4
Cramps	4
Febrile reactions	4
Arrhythmia	4
Hemolysis	4
Hypoxemia	4
Thrombosis	3
Hematoma	3
Device failure/malfunction	1

9.2. Benefit Analysis

Benefits of the Seraph 100 Microbind Affinity Blood Filter during the hemodialysis procedure may include potential reduction in the duration of bacteremia and the incidence of metastatic infections including endocarditis, septic arthritis and osteomyelitis. As part of early treatment of sepsis, Seraph 100 could also theoretically prevent the development of multi-system organ dysfunction and lower mortality.

9.3. Risk/Benefit Analysis

Based on bench and pre-clinical testing with the Seraph® Microbind® Affinity Blood Filter it is expected that subjects enrolled in this study will benefit from a shortened duration of bacteremia with no significant additional risk. By monitoring the blood characteristics and hemodynamics the potential risk is controlled and monitored.

The literature shows that the main clinical challenges with standard of care for these hemodialysis patients are:

- A broad-spectrum antibiotic treatment has to be initiated prior to determining the exact bacteria causing the bacteremia.
- Despite standard of care, catheter-related bloodstream infections still cause unacceptably high morbidity and mortality.

9.4. Risk mitigation

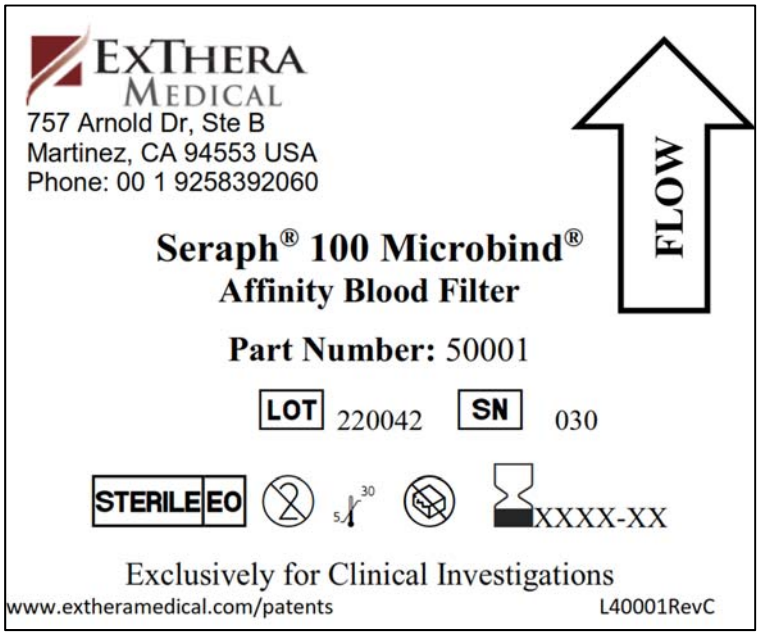
The study will be monitored to ensure the identification, documentation and analysis of adverse events, compliance with the protocol, and ethical requirements are in place for conducting research to protect the safety and well-being of all subjects.

Only operators with substantial experience in using hemodialysis/dialysis machine will be allowed to participate in this clinical study.

10. Device Management

10.1. Packaging and Labeling

The packaging for each cartridge consists of double peel pouches which are heat sealed. The peel pouch package is then placed in an 11 inch x 4.25 inch x 4.25 inch box. The cartridge and box are labeled with identical labels. Each label contains the product name, part number, lot number, serial number, sterilized by EO, one use only, and expiration date. The label also states "Exclusively for Clinical Investigations". An example of a label is:



10.2. Storage at Study Center

The devices for use in this study must be stored in a secure area. The secure area will have restricted access and the study devices will be kept separate from other medical devices. The study devices will only be handled by trained personnel and will not be supplied to any individual not involved in the investigation.

10.3. Device Accountability

The study devices will be inventoried at regular intervals during the study, and all unused or expired devices will be returned to the Sponsor when study enrollment is closed.

A form will be provided to the site that will log the model, lot number, and date received by the site. As the devices are used, the site will record the subject study identification number and date of procedure. A space will be provided for recording returned product and the reason for the return.

11. Ethics and Responsibility

11.1. Compliance to the Laws and Guidelines

The Investigator agrees that the study will be conducted according to the medical device directive 93/42/EEC as amended by Directive 2007/47/EC; COUNCIL DIRECTIVE 93/42/EEC of 14 June 1993 concerning medical devices as implemented into the local laws, GCP principles, ISO 14155:2011, and the principles of the World Medical Association Declaration of Helsinki 1964 (including all amendments and Notes of Clarification, up to and including the Scotland 2000 amendment and Tokyo 2004 Note of Clarification). The Investigator will conduct all aspects of this study in accordance with all national, state, and local laws or regulations.

Prior to starting the study at the site, the regulatory requirements have to be completed at the Competent Authority and the Ethics Committee and written documentation of the regulatory approvals of both the protocol and the informed consent form, which must comply with all requirements outlined by the Sponsor, must be provided to ExThera Medical. This approval must refer to the informed consent form and the study by title and the protocol number as given by ExThera Medical.

If an Investigator is a member of the Ethics Committee, he may not participate in the study approval decision. This non-participation must be noted in the approval letter.

No device supplies will be shipped to the Investigator for clinical research use until Ethics Committee and Competent Authority approval for the country in which the study is being conducted has been provided in writing to ExThera Medical.

11.2. Role of ExThera Medical

The sponsor's responsibilities include the following: (A) selection of the principal investigators/study sites and any other consultants who participate in the study, (B) provision to the principal investigator of a signed Clinical Investigation Agreement, Investigator's Brochure and study protocol with all applicable case report forms, (C) provision of the Investigational Product, (D) instruction of the investigators and their staff in the proper use of the Investigational Product, (E) provision of financial support to each investigational site, along with product liability and no-fault clinical trial insurance, (F) establishment of regulatory standards per federal regulations for all investigational sites, (G) performance of site monitoring at the investigational sites, and (H) maintenance of a Clinical Trial Master File. The sponsor is also responsible for the site initiation meeting and study monitoring. The site initiation meeting will be held at each center prior to the start of the study, the purpose of which is to insure that the investigator and staff thoroughly understand the protocol and their responsibilities as participants in the study.

Shipment of the investigational device to a site will not occur until the following documents are received by the study director: (A) written EC approval for conducting the study, (B) the text of the site's Patient Consent Form if different from the sample provided in the protocol, (C) a signed Clinical Investigation Agreement (Investigator Signature Page), (D) a signed Study Contract, and (E) curricula vitae for the principal investigator, sub-investigators, and study coordinator.

Each site will be visited at regular intervals to ensure that the study is conducted in full compliance with the protocol and all applicable regulations. The study monitor assigned will maintain personal contact with the investigator and

staff throughout the study by phone, mail, and on-site visits. At the close of the study, the clinical monitor will make a final site visit to collect all outstanding case report forms, assure that the investigator's files are accurate and complete, review record retention requirements with the investigator, account for all study supplies shipped to the investigator, provide for disposition of remaining supplies, and assure that all applicable study requirements have been met. All observations made or actions taken at this visit will be documented in a final report that will be given to the Sponsor.

11.3. Investigator's Responsibilities

The principal investigator is responsible for obtaining Ethics Committee approval for the study at each respective study site. The investigator is required to comply with the national requirements and the Declaration of Helsinki and agrees to do so by signing the Investigator Agreement provided in the attachments.

This clinical study shall be conducted in accordance with ISO 14155, with Good Clinical Practices and all other regulatory requirements. The study site will perform all of the requirements specified in this protocol. The Sponsor will provide the study site with the investigational plan, report of prior investigations, case report forms and all other necessary study related materials to conduct the trial. The Sponsor will perform or oversee, all aspects of the data quality assurance (data collection, data auditing and monitoring of the investigational sites).

If a Clinical Monitor becomes aware that an Investigator is not complying with the signed Investigator's Agreement, the Investigational Plan, the requirements of ISO 14155 or other applicable regulations, or any conditions of approval imposed by the reviewing Ethics Committee, ExThera Medical will immediately either secure compliance or discontinue shipments of the device to the Investigator and terminate the Investigator's participation in the investigation. The Investigator will be required to return all investigational products to ExThera Medical.

The investigator is responsible for submitting a study summary to ExThera Medical within a short time after the completion of the study, and for supplying this also to the governing Ethics Committee.

11.4. Informed Consent

The patient must provide a written informed consent, as approved by the EC, prior to participating in the trial. The patient must be adequately informed of the risks and benefits of study participation and treatment with the Seraph Microbind Affinity Blood Filter, the protocol requirements and of their expected participation in the trial.

The Investigator shall provide a copy of the signed informed consent form to the subject and/or legal guardian. The original form shall be maintained in the subject's medical records at the site.

11.5. Publications

ExThera Medical, as the Sponsor, has a proprietary interest in this study. Information obtained from the Sponsor by the investigator over the course of this study or generated under the protocol is the property of the Sponsor, and should therefore not be disclosed to anyone without prior consent of the Sponsor. The Sponsor will allow the investigator to publish results from this study with Sponsor approval. Scientific manuscripts, presentations or other communications relating to this study should be sent to the Sponsor for review at least 2 months prior to submission for publication or presentation. The Sponsor will complete its review within 1 month, and then forward its comments back to the

investigator. If there is disagreement, the opinions of both the Sponsor and investigator will be fairly and adequately represented.

12. Confidentiality

12.1. Patient Identification

Patients will be identified on the study Case Report Forms by the site number and a sequential number. No patient's initials or full date of birth will be collected. Case Report Forms are confidential documents and will only be available to the Investigator, Sponsor, trial statistician, and if requested, the EC and regulatory authorities.

All subject related medical data in the study will be handled confidential and not be released without the written permission of the subject (or the subject's guardian). The data will be handled in an anonymously.

13. Revision History

Rev.	DCO #	Description of Changes	Originator
A	00006	Initial document release	Kathleen White
B	00011	Revised for better specify protocol details. See DCO for specifics.	Kathleen White
C	00017	Add inclusion and exclusion criteria to Protocol Synopsis to meet German requirements.	Kathleen White
D	00044	Revise protocol to respond to BfArM questions and comments. See DCO for specific changes.	Kathleen White
E	00072	Revise protocol to increase age group and add additional in vitro pathogen data. See DCO for specific changes.	Kathleen White
F	00129	Change of EU Sponsor Representative Update of legal basis for safety notifications Change of contact point for the authorities regarding clinical safety Correction of example label	Kathleen White

APPENDIX I: Bibliography

Axelsson, J., M. Ferreira, L. Adolfsson, K. McCrea, R. Ward, and O. Larm. 2010. Cytokines in blood from septic patients interact with surface-immobilized heparin. ASAIO J. **56**: 48-51.

Mattsby-Baltzer, I., T Bergstrom, K, McCrea, R. Ward, L Adolfsson, O. Larm. 2011. Affinity apheresis for treatment of bacteremia caused by *Staphylococcus aureus* and/or Methicillin-Resistant *S. aureus* (MRSA). J. Microbiol. Biotechnol. **21**(6): 659-664.

TTP and CFU correlation

Idelevich, Evgeny A., et al. "Direct blood culturing on solid medium outperforms an automated continuously monitored broth-based blood culture system in terms of time to identification and susceptibility testing." New microbes and new infections 10 (2016): 19-24.

Haimi-Cohen, Yishai, Ernestine M. Vellozzi, and Lorry G. Rubin. "Initial concentration of Staphylococcus epidermidis in simulated pediatric blood cultures correlates with time to positive results with the automated, continuously monitored BACTEC blood culture system." Journal of clinical microbiology 40.3 (2002): 898-901.

Blot, François, et al. "Earlier positivity of central-venous-versus peripheral-blood cultures is highly predictive of catheter-related sepsis." Journal of clinical microbiology 36.1 (1998): 105-109.

Bacterial load (TTP and CFU) correlation to outcome.

Yagupsky, P. A. B. L. O., and F. S. Nolte. "Quantitative aspects of septicemia." Clinical microbiology reviews 3.3 (1990): 269-279.

Whimbey, Estella, et al. "Clinical significance of colony counts in immunocompromised patients with Staphylococcus aureus bacteremia." The Journal of infectious diseases (1987): 1328-1330.

Kluge, Ronica M., and H. L. Du Pont. "Factors affecting mortality of patients with bacteremia." Critical Care Medicine 1.5 (1973): 291.

Hsu, M-S., et al. "Sequential time to positivity of blood cultures can be a predictor of prognosis of patients with persistent Staphylococcus aureus bacteraemia." Clinical Microbiology and Infection 20.9 (2014): 892-898.

Khatib, Riad, et al. "Time to positivity in Staphylococcus aureus bacteremia: possible correlation with the source and outcome of infection." Clinical infectious diseases 41.5 (2005): 594-598.

Marra, Alexandre R., et al. "Time to blood culture positivity as a predictor of clinical outcome of Staphylococcus aureus bloodstream infection." Journal of clinical microbiology 44.4 (2006): 1342-1346.

D'Amato-Palumbo, S, et al. "Retrospective study of microoganisms associated with vascular access infections in hemodialysis patients." Oral Surg Oral Med Oral Pathol Oral Radiol 2013 January; 115(1)

Suzuki, M, et al. "Bacteremia in hemodialysis patients." World J Nephrol 2016 November 6; 5(6) 489-496

Fowler. 2003. "Clinical Identifiers of Complicated Staphylococcus aureus Bacteremia." Arch Intern Med 2066-2072.

Li, Y., Friedman, J.Y., O'Neal, B.F., Hohenboken, M.J., Griffiths, R.I., Stryjewski, M.E., Middleton, J.P., Schulman, K.A., Inrig, J.K., Fowler, V.G. and Reed, S.D. 2009. "Outcomes of Staphylococcus aureus infection in hemodialysis-dependent patients." Clinical Journal of the American Society of Nephrology 4 (2): 428-434.

Marr, K.A., L Kong, V. Fowler, A. Gopal, D.J. Sexton, P.J. Conlon, and G.R. Corey. 1998. "Incidence and outcome of Staphylococcus aureus bacteremia in hemodialysis patients." Kidney International 54: 1684-1689.

Fowler Jr, Vance G., Helen W. Boucher, G. Ralph Corey, Elias Abrutyn, Adolf W. Karchmer, Mark E. Rupp, Donald P. Levine et al. "Daptomycin versus standard therapy for bacteremia and endocarditis caused by Staphylococcus aureus." New England Journal of Medicine 355, no. 7 (2006): 653-665.