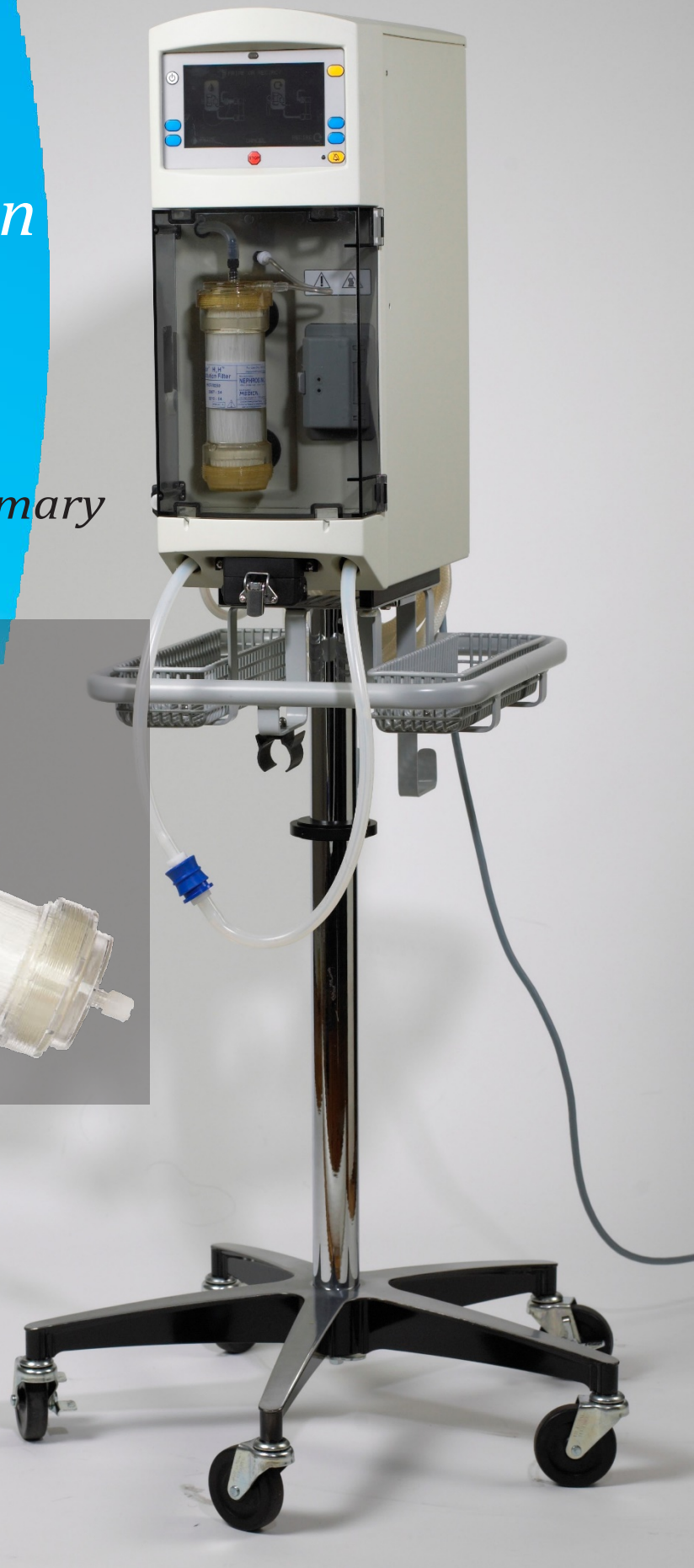
_PO

Nephros On-line Mid-Dilution Hemodiafiltration System

Clinicians' Overview with Safety and Efficacy Summary



Note: Federal (USA) law restricts these devices to sale by or on the order of a physician.

For further information contact:

Nephros, Inc.
380 Lackawanna Place
South Orange, NJ 07079 USA
Telephone: 201-343-5202
Fax: 201-343-5207
Web: www.nephros.com

Purpose

This document provides a potential clinician, user or reviewer with a brief summary of the Nephros On-line Mid-Dilution Hemodiafiltration (HDF) System and its components with salient comparisons between hemodialysis and hemodiafiltration. Also included are clinic requirements needed to use the Nephros On-line Mid-Dilution HDF System and a summary of safety and efficacy results obtained as part of a clinical investigational study with the Nephros system.

Introduction to HDF

Hemodiafiltration (HDF) is a renal replacement therapy that combines hemodialysis (solutes cleared primarily by diffusion) with hemofiltration (solutes primarily cleared by filtration or convection). During HDF, toxin rich plasma water is ultrafiltered across the dialyzer membrane and into the spent dialysate at rates of 100 to 300 ml/min (or 6 to 18 liters/hour). The increased ultrafiltration that occurs with HDF generally increases the clearance of uremic toxins, especially larger, middle molecular weight toxins, as these toxins are carried or "dragged" across the membrane as part of the %ow of ultrafiltered plasma water. To keep the patient in proper %uid balance, a "substitution %uid" must be continuously infused into the extracorporeal circuit to replace the plasma water that is being filtered out of the patient's blood as it passes through the hemodiafilter device.

"On-line HDF" refers to the method which produces the substitution %uid used during hemodiafiltration. In particular, in on-line HDF, substitution %uid is generated in real time using a portion of the dialysate produced by the dialysis machine. This is accomplished by redirecting some of the fresh dialysate and passing it through a series of redundant ultrafilters. These ultrafilters remove microbiologic contaminants (bacteria and endotoxin) from the dialysate before it is infused into the blood circuit as a substitution %uid.

On-line HDF using redundant ultrafilters to purify the dialysate for infusion is actively used in Europe. Nephros has developed the OLpūr® Hemodiafiltration H_2H Module) that works using the same principles as the European on-line HDF machines; however, it has been designed as a separate (standalone) device that works in conjunction with a standard ultrafiltration-controlled hemodialysis machine to generate on-line substitution %uid.

Nephros has also developed a unique hemodiafilter (OLpūr MD 220 Hemodiafilter) that overcomes the limitations observed with pre- and post-dilution HDF using a standard high-%ux dialyzer. The device is similar to a standard dialyzer but uses modified blood header caps to create a simple two-stage design whereby the substitution %uid is infused in a mid-dilution configuration as blood moves between the two stages of the device.

Further details of the Nephros OLpūr H_2H Module and the OLpūr MD 220 Hemodiafilter are provided in the device description section of this document.

What is needed to perform HDF with the Nephros On-line Mid-Dilution Hemodiafiltration System?

- Dialysis quality water meeting the current ANSI/AAMI/ISO standard
- Ultrapure dialysate meeting the current ANSI/AAMI/ISO standard
- The Nephros OLpūr MD 220 Hemodiafilter (MD 220 filter)
- The Nephros OLpūr H_2H Hemodiafiltration Module (H_2H Module)
- An H_2H compatible host hemodialysis machine (ultrafiltration-controlled)
- All accessory components for the Nephros H_2H Module

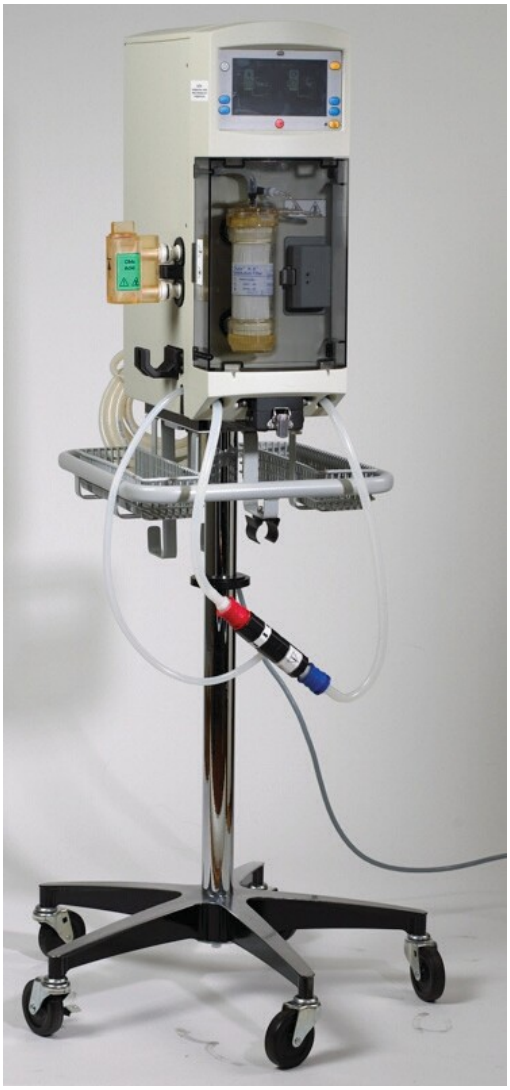
Indications for Use

The OLpūr H_2H Hemodiafiltration Module is indicated for use, with a UF-controlled hemodialysis machine that provides ultrapure dialysate in accordance with the current ANSI/AAMI/ISO standards, for treatment of patients with chronic renal failure as prescribed by a physician. The OLpūr MD 220 Hemodiafilter is indicated for use for hemodiafiltration of patients with chronic renal failure.

Contraindications

There are no absolute contraindications to HDF; however, HDF should be avoided or applied with extreme caution in patients with the following conditions or circumstances:

- Unable to receive or contraindications to adequate anticoagulation
- Hypercoagulation syndromes
- Hyperviscosity syndromes
- Pre-treatment Hematocrit ≥ 40%
- Single-Needle blood access
- Blood access with obvious recirculation



Description of the Nephros On-line Mid-Dilution HDF System

The Nephros OLpūr® H_2H Module

The H_2H Module is a software controlled electro-mechanical medical device designed to work in conjunction with an approved hemodialysis machine. The H_2H Module offers a simple way to enhance the functionality of a clinic's existing hemodialysis machine by enabling approved machines to be used to perform on-line HDF. The H_2H Module is a freestanding device on a

movable stand that is placed next to the host dialysis machine. The H_2H Module must be connected to the clinic's water supply, drain and electricity.

The H_2H Module serves two basic functions. The primary function is to generate on-line substitution %uid for HDF. During the course of an HDF treatment, the H_2H Module is continuously monitoring the dialysate %ow, blood %ow, and for any evidence of blood contamination back into its onboard substitution %uid filter. The H_2H Module's secondary function is to serve as a substitution %uid and water filter reprocessing system that automates management of the H_2H Module's dialysate purification system.

The H_2H Module may be used only with approved Nephros accessory components (i.e. H_2H Substitution filter, H_2H Water filter, and H_2H Infusion line). Only compatible host dialysis machine models can be used as indicated in the Nephros OLpūr H_2H Module User's Manual.

Accessory Components for the H_2H Module

The H_2H Substitution Fluid Filter

The H_2H Substitution filter is a hollow fiber, ultrafilter device that consists of two sequential (redundant) ultrafiltration stages in a single housing. During on-line HDF with the H_2H Module, fresh dialysate is redirected by the H_2H Module's hydraulic (substitution) pump and passed through this dual-stage ultrafilter before being infused as substitution %uid into the extracorporeal circuit. The H_2H Substitution filter remains on the H_2H Module for 14 days and is removed from the H_2H Module when its useful life has expired or if it has failed a filter integrity test performed by the H_2H Module. If the H_2H Substitution filter is not properly disinfected at least once every 72 hours by running a complete Filter Care routine, the Module's HDF treatment functions will be disabled, thereby prohibiting the user to perform online HDF with the H_2H Module until a successful Filter Care routine has been completed.

The H_2H Infusion Line

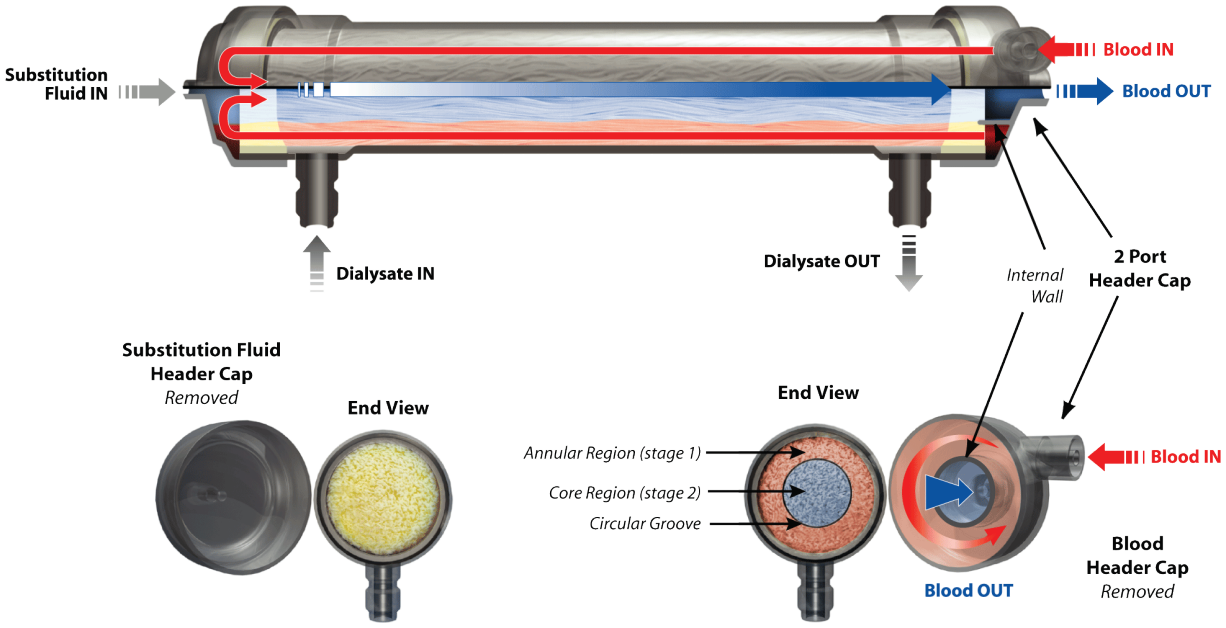
The H_2H Infusion line is a sterile, single-use, disposable tubing set that is replaced a(er each patient treatment. During on-line HDF, the H_2H Infusion line connects the outlet port of the H_2H Substitution filter to the substitution port on the Nephros MD 220 hemodiafilter. The H_2H Infusion line contains a detachable tubing segment (the rinse line) that contains a one-way check valve. During reprocessing of the H_2H Substitution filter, the distal portion of the rinse line is disconnected from the rest of the infusion set and is re-connected to a dedicated rinse port on the front of the H_2H Module. The one-way check valve, in addition to a so(ware controlled pinch valve on the H_2H Module, ensures that %uid (patient's blood) cannot %ow backward to contaminate the H_2H Substitution filter.

The H_2H Water Filter

The H_2H Water filter is a single stage ultrafilter. All water the H_2H Module uses for rinsing, priming and disinfection of the %uid path, including the H_2H Substitution filter, must %ow through this filter before entering the H_2H Module. The H_2H Water filter is a hollow fiber, membrane-type ultrafilter that is designed for continuous use over a 2 week (14 day) period when subjected to daily functional and structural integrity testing as part of H_2H Module's normal operation.

The OLpūr® MD 220 Hemodiafilter

Nephros has developed a unique hemodiafilter, the OLpūr MD 220 Hemodiafilter, to overcome limitations caused by using a standard high-%ux dialyzer for hemodiafiltration. The MD 220 is very similar to a typical hollow fiber dialyzer assembled with a single hollow fiber bundle made with a high-%ux (or high-permeability) membrane. At one end of the device, however, is a blood header cap having both a blood inlet port and a blood outlet port. The two-port cap has an internal dividing wall that inserts into a ring-shaped groove in the potting compound. A mechanical seal is formed that separates the incoming blood from the exiting blood at this end of the device. As a result, the fiber bundle is separated into two discrete but serially connected blood paths: an outer or annular path (Stage 1), and an inner or core path (Stage 2). At the opposite end of the device is the substitution header cap, which allows for the infusion of substitution %uid and subsequent mixing with the blood as it moves between the two stages (i.e. in a mid-dilution hemodiafiltration mode).



Theory of Operation of the Nephros On-line Mid-Dilution Hemodiafiltration System

The Volumetrically Static Dialysate Path

The dialysate path of a dialysis machine that is capable of functioning as host machine for the H_2H Module must behave as if it were a closed system relative to the expansion or contraction of its dialysate %uid volume. The dialysis machines that accomplish this are based on mechanical %uid balancing chambers or %ow measuring devices in feed-back loops with %ow control pumps. These ultrafiltration (UF) control systems are isovolumetric and assure that the volume of dialysate produced by the machine (fresh dialysate) is essentially equivalent to the volume of dialysate returned to the machine (spent dialysate). It is noted that a small difference between inlet and outlet dialysate %ows is necessary when a "net" amount of %uid is removed from the patient to reach their dry weight. This, however, is a normal function of a UF-controlled dialysis machine whereby the user enters a net volume of %uid to be removed from the patient (see section below).

The Volumetrically Variable Patient

The patient, in contrast to the above, does not behave as a closed %uid compartment. Fluid status of a patient can change in several ways which include the insensible net %uid loss from breathing and perspiration versus the net gain through drinking, saline infusion, and eating. During treatment of patients, it is generally necessary to remove %uid from the patient to offset the net gain of %uid and to return the patient to their "dry weight". This removal of %uid during treatment is accomplished as a net loss through ultrafiltration of plasma water across the dialyzer membrane.

Introducing the H_2H Module into the Dialysate Path

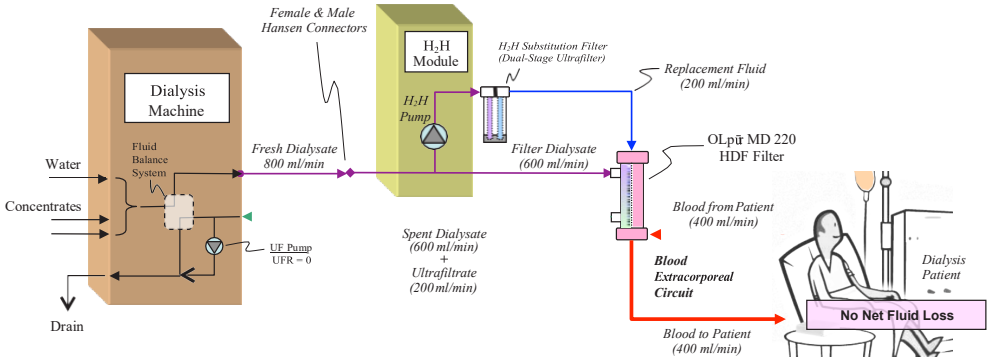
To accomplish on-line HDF with the H_2H Module, the H_2H Module is introduced as part of the closed dialysate path. As dialysate is "removed" from the "closed" dialysate path by the H_2H Module as illustrated below, the UF-control system of the host dialysis machine responds automatically by demanding an identical volume of plasma water (containing uremic toxins) to be forward filtered out of the blood and into the dialysate compartment of the hemodiafilter. This to assure the volume of spent dialysate returning to the host dialysis machine is the same as the volume of fresh dialysate it delivered to the H_2H Module and subsequently to the dialysate compartment of the hemodiafilter. The dialysate %uid that was removed from the "closed" dialysate path is purified by the H_2H Module by redundant ultrafiltration through its onboard substitution filter, and infused directly into the extracorporeal circuit as on-line replacement (substitution) %uid. The net effect is that plasma water containing the uremic toxins is transferred from the blood compartment to the dialysate compartment where it combines with the spent dialysate %uid being returned to the host dialysis machine in exchange for clean purified dialysate passing through the H_2H Substitution filter of the H_2H Module and into the extracorporeal circuit.

The host dialysis machine, controlled by user input, has complete authority over the dialysate production and dialysate %ow rate. The H_2H Module User's Manual requires a minimum dialysate %ow rate of 500 ml/min on procedures prior to starting the HDF treatment and recommends a minimum dialysate %ow rate of 600 ml/min during the

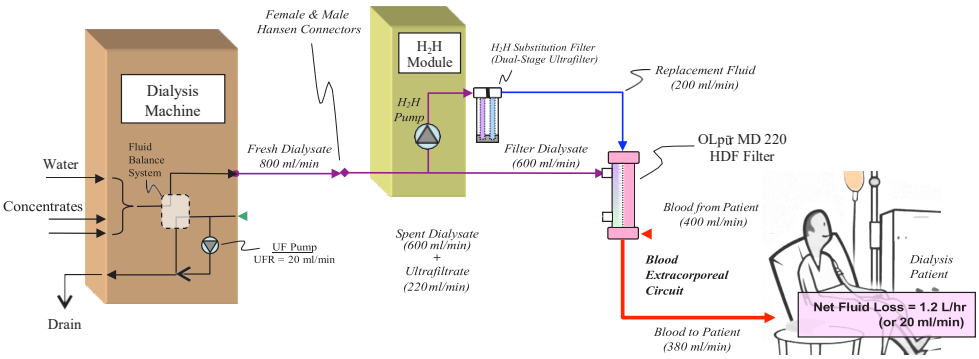
HDF treatment when using the H_2H Module in conjunction with the host dialysis machine. These %ow rates are within the range of dialysate %ow used in clinical practice and therefore do not require a shi(in normal management of the host's dialysate concentrate needs. The current recommended clinical practice with regard to dialysate concentrate is for the user to provide sufficient concentrate to complete the treatment at the user commanded dialysate %ow rate before starting the treatment.

Note that all references to "dialysate %ow rate" or "Q_d" refer to the value for dialysate %ow set on the host dialysis machine. This value (e.g. 800 ml/min) is also the dialysate %ow rate that exits the MD 220 filter and is returned to the host machine as spent dialysate. In conventional dialysis this is also the dialysate %ow rate entering the dialyzer. However, in the Nephros HDF system, the H_2H Module takes a portion of the fresh dialysate from the host machine (e.g. 200 ml/min) and purifies it for infusion into the blood side of the MD 220 filter. As a result, the dialysate %ow entering the MD 220 filter ("filter dialysate") is reduced by the volume of infusion %uid (200 ml/min). Therefore, in this example, the "filter dialysate" would be 600 ml/min. As a result of this lower filter inlet dialysate %ow rate (600 ml/min), the diffusive clearance of the MD 220 filter may be less than the diffusive clearance of a conventional dialyzer that would receive the full 800 ml/min dialysate inlet %ow. However, the MD 220 filter has a convective clearance component due to the 200 ml/min infusion %ow rate. As a result, the overall clearance of the MD 220 filter is equal to or greater than that of a conventional dialyzer at the same host machine Q_d setting.

On-line Mid-Dilution HDF with UF Rate Set = 0



On-line Mid-Dilution HDF with UF Rate = 1.2 L/hr



The Clinical Study

A multi-center, non-randomized clinical study was performed under an investigational device exemption (IDE) to evaluate safety and efficacy of the Nephros On-line Mid-Dilution Hemodiafiltration System. 28 patients who had been receiving maintenance high-%ux hemodialysis for a minimum of 3 months entered the test phase of the study using the Nephros OLpūr H_2H Hemodiafiltration Module, its accessory components, and the Nephros OLpūr MD 220 Hemodiafilter. A four (4) week period of routine high-%ux hemodialysis (without reuse) with dialysate %ow rate (Q_d) ranging between 600 to 800 ml/min with an average treatment time (95% CI) of approximately 230 min (219; 241) served as a control (baseline) period for comparison purposes. Patients then received 12 weeks of on-line HDF therapy on the Nephros Hemodiafiltration System devices at similar dialysate %ow rates (Q_d) and treatment times with an average substitution %uid %ow rate (95% CI) of approximately 185 ml/min (177;193) whereby similar data was collected during this period as the control period. Conventional dialysate was used as input to the H_2H Module and MD 220 filter during this clinical study, this representing a worse case condition relative to its intended use with ultrapure dialysate as input to the H_2H Module and MD 220 filter.

The primary objective of this study was to verify safety and clinical tolerance associated with performing on-line HDF with the Nephros MD 220 Hemodiafilter and the Nephros H_2H Module in conjunction with an approved (standard UF-controlled) host hemodialysis machine. This objective was measured by comparing the total number of inter- and intra-dialytic adverse events (AEs) and intra-dialytic adverse symptoms occurring over each study period.

The secondary objective of this study was to monitor subject blood chemistries, including beta-2-microglobulin and C-reactive protein (as an in%ammatory marker), and traditional performance parameters for determining adequacy of dialysis. In addition, Quality of Life assessments (Kidney Disease and Quality of Life™ (KDQOL™-36) were obtained during the fourth week of the control period (between visits 10 and 12) and during the final week of the test period (between visits 46 and 48).

The Study Results

The study demonstrated non-inferiority of treatment with the test device, Nephros OLpūr MD 220 Hemodiafilter and Nephros OLpūr H_2H Module in conjunction with a standard UF-controlled hemodialysis machine, to the standard control device (see table below). Reduction of beta-2-microglobulin with the test device was found to be significantly greater than that with the control device. Overall, use of the test device was safe and well-tolerated.

Adverse Events and Adverse Symptoms

The ITT (intent-to-treat) population included all subjects who underwent treatment at least once, which was used for all analyses. For the primary parameters Adverse Event Rate (AER) and Adverse Symptom Rate (ASR), a PP (per-protocol) analysis in addition to the ITT analysis was performed. No subjects were excluded, but intra-dialytic AEs and adverse symptoms were excluded only if they occurred following dialysis performed without the test device during the test period.

All AEs were analyzed according to the treatment-emergent principle in which an event was reported in the phase where it emerged (i.e., started a(er baseline or worsened versus baseline in the attributes severity, seriousness, relation to device, or action taken regarding the device), and not in the subsequent phases, even if the event continued to be present. Unless indicated otherwise, AEs and abnormalities discussed are treatment-emergent.

	Control Period Rate	Test Period Rate	Observed difference between test and control periods	95%CI on difference	Result[a]
AER[b] inclusion of hypo/ hyper volemic events					
ITT analysis	0.54	0.51	–0.025	–0.070; 0.019	non-inferior
PP analysis	0.54	0.51	–0.030	–0.074; 0.015	non-inferior
AER[b] exclusion of hypo/hyper volemic events					
ITT analysis	0.07	0.06	–0.017	–0.019; –0.015	non-inferior
PP analysis	0.07	0.05	–0.020	–0.022; –0.018	non-inferior
ASR[b]					
ITT analysis	0.15	0.20	0.051	0.042; 0.060	non-inferior
PP analysis	0.15	0.19	0.046	0.037; 0.054	non-inferior

[a] Non-inferiority of the test device to the control device was concluded if the upper confidence bound of the 95% CI interval was ≤ 0.02 for the difference in AER and ≤ 0.09 for the difference in ASR.

[b] The AER and ASR are the total number of AEs or adverse symptoms, respectively, in each study period divided by the total number of treatments in each study period.

Overall, all subjects were reported with at least one AE during both the control and the test period. Most of the AEs were intra-dialytic; only 9 (27%) subjects had an inter-dialytic AE during the 4-week control period and 8 (29%) subjects during the 12-week test period. Most of the intra-dialytic AEs concerned procedural complications (all subjects in the control and test period), which were mainly hemodialysis-induced symptoms, (i.e., hypovolemia and hypervolemia) which was based on a narrow tolerance around a measured subject %uid balance UF Error. Other reported intra-dialytic AEs were device leakage and procedural complication. Inter-dialytic AEs were mostly infections and gastrointestinal disorders.

Nineteen (57.6%) and 16 (57.1%) subjects were reported with at least one adverse symptom in the control and test period, respectively. Cramping and hypotensive episodes were the most frequently reported adverse symptoms in both treatment periods. Hypotensive episodes occurred at a 12% incident rate during the test period compared to an 8% incident rate during the control period.

Most AEs (intra- and inter-dialytic) were mild in severity. Moderate intra- and inter-dialytic AEs were reported for 1 subject each during the control period; none were reported in the test period. During the control period, severe inter-dialytic AEs were reported for 2 subjects (supraventricular tachycardia, upper gastrointestinal hemorrhage, chest pain, pain, staphylococcal bacteremia, and %ank pain), and no severe intra-dialytic AEs were reported. During the test period, severe intra-dialytic AEs were reported for 1 subject (catheter sepsis) and severe inter-dialytic AEs for 2 subjects (cellulitis and arteriovenous fistula aneurysm).

None of the subjects who received treatment with the test device expired. Only

one subject expired during the control period. Five other subjects experienced other SAEs: 2 during the control period (inter-dialytic supraventricular tachycardia and lobar pneumonia) and 3 subjects during the test period (1 intra-dialytic catheter sepsis and 2 inter-dialytic cellulitis and arteriovenous fistula aneurysm). Study treatment was permanently stopped due to an inter-dialytic SAE for 3 subjects during the control period and for 1 subject during the test period due to the hospitalization criteria outlined in the study protocol.

Lab Results

There were no consistent changes over time in mean laboratory values, except for a decrease in mean Alkaline Phosphatase (pre-treatment), beta-2-microglobulin (pre-treatment), and beta-2-microglobulin (post-treatment) levels. Between group comparison of the results obtained at Week 4 during the control period with those at Week 8, 12, and 16 during the test period, showed that for URR, Kt/Vdp, and PCR_n the difference was not statistically significant ($p > 0.05$). For β_2m–RR, the between group difference was statistically significantly different (< 0.001) in favor of the use of the Nephros test device at all 3 laboratory assessments.

Kt/Vdp values in both the test and control periods were approximately 1.5 on average. While these values are above the NKF KDOQI guidelines, they are not a necessary target for use of the Nephros HDF. They simply re%ect the standard practices of the clinics involved in the study.

Lab results appear below.

Lab Test	Mean Week 4 Dialysis	Mean Week 8 HDF	Mean Week 12 HDF	Mean Week 16 HDF
Albumin (g/dl)	3.8	3.8	3.9	3.9
Alkaline Phosphatase (u/l)	142.6	134.2	138	121.5
Beta 2 Microglobulin (mg/l) PRE	36.4	32.4	30.8	30.3
Beta 2 Microglobulin (mg/l) POST	17	7.9	7.1	7
Bicarbonate (mmol/l)	23	24.2	23.4	23.3
Blood Urea Nitrogen (mg/dl) PRE	59.3	63.7	63	62
Blood Urea Nitrogen (mg/dl) POST	14	16.2	15.3	15.6
C-Reactive Protein (mg/dl)	1.5	1.1	0.9	1
Calicium (mg/dl)	8.8	9	9	9
Chloride (mmol/l)	99	98.7	99.3	99.6
Creatinine (mg/dl)	9.1	9.4	9.4	9.6
Ferritin (ng/ml)	670.5	631.2	600.2	612.6
Hematocrit (%)	37.2	35	35.7	36.3
Hemaglobin (g/dl)	12.3	11.7	11.8	12.1
Iron (ug/dl)	70.4	69.1	69.9	66.6
Lactate Dehyrogenase (u/l)	164.5	173.8	168.6	169.2
Magnesium (mg/dl)	1.9	2	2	2
Phosphorus (mg/dl)	4.6	5.3	5.2	5.2
Platelet Count (X103)	187.8	189.3	183.8	182.8
Potassium (mmol/l)	4.7	5	4.8	5
Sodium (mmol/l)	138.9	138.8	139.3	139
Total Protein (g/dl) PRE	6.9	6.8	6.9	6.9
Total Protein (g/dl) POST	7.6	7.7	7.8	7.9
White Blood Count (X103)	6.9	6.7	6.7	6.9

Treatment Efficacy	Mean Week 4 Dialysis	Mean Week 8 HDF	Mean Week 12 HDF	Mean Week 16 HDF
Urea Reduction Ratio	0.761	0.751	0.757	0.755
KT/Vdp	1.557	1.43	1.439	1.45
Protein Catabolic Rate	1.154	1.216	1.193	1.168
Beta-2-m Reduction Ratio	0.574	0.785	0.798	0.797

Quality of Life- *KDQOL™-36*

The burden of kidney disease was assessed as low in comparison with the symptoms, problems, and effects of the disease. No statistical significant difference was observed between the control and test period for burden, symptoms and problems, and effects of kidney disease.

Risk versus Benefit Statement

Summarization of the clinic trial data in the section above and compliance of the Nephros devices with pertinent design safety standards indicates the Nephros On-line Mid-Dilution Hemodiafiltration System adds no additional risk for patients or users when compared to standard high-%ux hemodialysis; however, the clinical trial data showed that hypotensive episodes occurred at a 12% incident rate during the hemodiafiltration test period compared to an 8% rate during the standard high-%ux hemodialysis control period. The clinical trial data also indicated that with hemodiafiltration there is a significant improvement in the removal of the middle-molecular weight toxin beta-2-microglobulin when compared to standard high-%ux hemodialysis. As such, we believe the benefits of on-line HDF with the Nephros On-line Mid-Dilution Hemodiafiltration System outweigh the risks associated with its use.

First treatment performed with the Nephros On-line Mid-Dilution Hemodiafiltration System. Shown are Dr. Leonard Stern (left) and Edward Spence (right).

Nephros

Nephros, Inc. • 380 Lackawanna Place • South Orange, New Jersey 07079 USA

Telephone: 201-343-5202 • Fax: 201-343-5207 • www.nephros.com