SHOCK, Vol. 23, No. 5, pp. 400–405, 2005

A PILOT-CONTROLLED STUDY OF A POLYMYXIN B-IMMOBILIZED HEMOPERFUSION CARTRIDGE IN PATIENTS WITH SEVERE SEPSIS SECONDARY TO INTRA-ABDOMINAL INFECTION

Jean-Louis Vincent,[*] Pierre-François Laterre,[†] Jonathan Cohen,[‡] Hilmar Burchardi,[§] Hajo Bruining,[‖] Francisco Alvarez Lerma,[¶] Xavier Wittebole,[†] Daniel De Backer,[*] Stephen Brett,[**] Dolores Marzo,[¶] Haruji Nakamura,[††] and Stephanie John[‡‡]

[*]Department of Intensive Care, Erasme Hospital, Free University of Brussels, Brussels, Belgium; [†]Department of Intensive Care, Cliniques Universitaires Saint Luc, Brussels, Belgium; [‡]Brighton and Sussex Medical School, Falmer, United Kingdom; [§]Department of Intensive Care, Georg-August University, Göttingen, Germany; [‖]Department of Intensive Care, University Hospital, Rotterdam, The Netherlands; [¶]Intensive Care Unit, Hospital Universitorio del Mar, Barcelona, Spain; [**]Department of Intensive Care, Hammersmith Hospital, London W12 OHS, United Kingdom; [††]Toray Industries, Tokyo 103-8666, Japan; and [‡‡]Quintiles, Bracknell R942 1HX, United Kingdom

Received 29 Jun 2004; first review completed 27 Jul 2004; accepted in final form 8 Feb 2005

ABSTRACT—Endotoxin is an important pathogenic trigger for sepsis. The polymyxin B-immobilized endotoxin removal hemoperfusion cartridge, Toraymyxin (hereafter PMX), has been shown to remove endotoxin in preclinical and open-label clinical studies. In a multicenter, open-label, pilot, randomized, controlled study conducted in the intensive care unit in six academic medical centers in Europe, 36 postsurgical patients with severe sepsis or septic shock secondary to intra-abdominal infection were randomized to PMX treatment of 2 h (n = 17) or standard therapy (n = 19). PMX was well tolerated and showed no significant side effects. There were no statistically significant differences in the change in endotoxin levels from baseline to 6 to 8 h after treatment or to 24 h after treatment between the two groups. There was also no significant difference in the change in interleukin (IL)-6 levels from baseline to 6 to 8 h after treatment or to 24 h after treatment between the two groups. Patients treated with PMX demonstrated significant increases in cardiac index (CI; $P = 0.012$ and 0.032 at days 1 and 2, respectively), left ventricular stroke work index (LVSWI, $P = 0.015$ at day 2), and oxygen delivery index (DO_2I, $P = 0.007$ at day 2) compared with the controls. The need for continuous renal replacement therapy (CRRT) after study entry was reduced in the PMX group ($P = 0.043$). There was no significant difference between the groups in organ dysfunction as assessed by the Sequential Organ Failure Assessment (SOFA) scores from day 0 (baseline) to day 6. Treatment using the PMX cartridge is safe and may improve cardiac and renal dysfunction due to sepsis or septic shock. Further studies are needed to prove this effectiveness.

KEYWORDS—Sepsis, endotoxin, hemodynamics, organ dysfunction, extracorporeal renal support, Polymyxin B

INTRODUCTION

Sepsis and septic shock continue to be life-threatening complications and major causes of death in the medical and surgical intensive care unit (ICU). In the last decade, research into the pathogenesis and pathophysiology of sepsis, septic shock, and subsequent multiple organ failure (MOF) has clarified some of the mechanisms of these disease processes (1–4). Various mediators, including cytokines, nitric oxide (NO), reactive oxygen species, and lysosomal enzymes, are liberated by activation of leukocytes, macrophages, and endothelial cells, and are thought to play a pivotal role in the pathophysiology of septic shock and MOF. Hemodynamics in patients with sepsis or septic MOF may be improved with continuous hemofiltration (CHF) or continuous hemodiafiltration (CHDF), possibly due to the removal of some of these humoral mediators (5), and the addition of coupled plasma filtration adsorption to hemodialysis may be more effective at improving hemodynamic status in patients with sepsis than hemodialysis alone (6).

The PMX cartridge (Toraymyxin, Toray Industries, Tokyo, Japan) is an extracorporeal hemoperfusion device that uses polymyxin-B fixed to α-chloroacetamide-methyl polystyrene-derived fibers that are packed in the cartridge. Polymyxin-B is chemically immobilized in the polystyrene-derived fiber through covalent bonds and does not leach from the fiber. Polymyxin-B is known to bind to endotoxin, an outer membrane component of gram-negative bacteria that is thought to be an important pathogenic trigger for the production of inflammatory mediators. Several preclinical studies demonstrated that hemoperfusion or plasmapheresis over immobilized polymyxin-B could remove endotoxin from the blood (7–9). Subsequently, open-label clinical trials using PMX were conducted in Japan, demonstrating the safety of PMX in the treatment of septic shock and its capacity to decrease endotoxin levels (10). More recent studies have shown improved hemodynamic status (11) and improved survival (12) in patients with sepsis treated with PMX. Since 1994, PMX has been listed as a blood purification device in Japan and is reimbursed by Japanese national health insurance. The PMX device received its product certification (CE mark) for use in Europe, according to the directives of the European Community in 1998, and is classified as a Class IIb medical device.

In a recent randomized phase II trial (13) of another hemofiltration endotoxin-adsorbing system, based on immobilized

Address reprint requests to Dr. Jean-Louis Vincent, Department of Intensive Care, Erasme University Hospital, Route de Lennik 808, B-1070 Brussels, Belgium. E-mail: jlvincen@ulb.ac.be.

DOI: 10.1097/01.shk.0000159930.87737.8a

human serum albumin, no significant differences in endotoxin elimination or organ function (as assessed by the Sequential Organ Failure Assessment [SOFA]) were identified between treated patients and controls, although there was a trend to improved renal function and shorter length of ICU stay in the subgroup of treated patients with peritonitis.

The current study is the first prospective randomized controlled trial (RCT) of the PMX device in patients with sepsis. The hypothesis was that, in line with previous *in vitro* and clinical studies, treatment with PMX would reduce endotoxin levels and improve hemodynamic status. To increase homogeneity in the study population, we included only patients with sepsis of abdominal origin.

PATIENTS AND METHODS

Six European, academic medical centers were involved in the study. The ethics committees for each center approved the study protocol. Informed consent was obtained from each patient or the patient's relative or surrogate.

Study population

The study population consisted of surgical patients with severe sepsis (with or without shock) presumed to be caused by gram-negative infection, arising from the abdominal cavity, and still present after surgery. Patients who had initially undergone elective surgery were eligible if the PMX treatment could be started within 24 h of diagnosis of severe sepsis. Patients admitted to hospital for intra-abdominal infection requiring emergency surgery were also eligible provided the PMX treatment was started within 48 h of diagnosis of severe sepsis. Inclusion criteria were presumed gram-negative infection arising from the abdominal cavity within 2 weeks of abdominal surgery, at least two of the systemic inflammatory response syndrome (SIRS) criteria (14), and at least one organ dysfunction defined as follows: cardiovascular—systolic blood pressure of below 90 mmHg (11.7 kPa) or a decrease in systolic blood pressure of at least 40 mmHg from baseline, which was transient or required treatment with vasoactive drugs at any dose; respiratory—PaO_2 of less than 75 mmHg (9.75 kPa) or PaO_2/FiO_2 ratio of less than 250 mmHg; renal—creatinine of more than 2 mg/dL (171 μmol/L) or urine output of less than 500 mL/day or a urine output of less than 30 mL/h for 2 h during patient screening; coagulation—a 50% decrease in platelet count or a decrease in the platelet count to less than $100 \times 10^3/mm^3$; central nervous system (CNS)—Glasgow Coma Scale of less than 13 or decrease of greater than 1 from baseline; and hepatic—bilirubin of more than 10 mg/dL. Patients were excluded from the study for the following reasons: life expectancy less than 30 days (as assessed by the attending physician); "do not resuscitate" order; HIV infection; uncontrolled hemorrhage within 24 h before study entry; organ transplantation during the year before study entry; history of sensitivity to polymyxin-B or anticoagulant and/or extracorporeal circulation; severe thrombocytopenia (<30,000 cells/mm^3) and/or granulocytopenia (<500 cells/mm^3); an APACHE II score (15) >30, a SOFA score (16) >12 or >4 organ failures by Goris score (17); and end-stage organ failure defined as follows: respiratory—end-stage chronic obstructive airways disease and corpulmonale; cardiac—New York Heart Association score = IV; CNS—brain stem death (confirmed by the attending physician and an independent physician registered for more than 5 years, on two separate occasions) or persistent vegetative state; renal—requiring maintenance hemodialysis; hepatic—chronic liver disease complicated by hepatic encephalopathy, coagulopathy, fluid retention, and/or hepatocellular jaundice. Patients without evidence of gram-negative infection or endotoxemia at baseline were excluded from the analysis at the end of the study (patients with only gram-positive or fungal cultures were kept in the final study analysis as long as a significant endotoxin level had been measured).

Treatment

Patients who fulfilled the study criteria were randomized, using sealed envelopes, to receive standard therapy plus PMX (PMX group) or standard therapy only (control group). Patients of both groups received full intensive care management, including fluid resuscitation, vasopressors, antimicrobial therapy, ventilatory support, appropriate surgical management (including drainage of the nidus of infection), and continuous renal replacement therapy (CRRT) and hemodialysis, if required. PMX hemoperfusion was conducted in the ICU. All patients in the PMX group underwent PMX treatment only once, on day 0, after severe sepsis had been diagnosed. For venous access, a double lumen catheter was inserted into the femoral vein and blood was drawn from the vein and returned to it. The blood flow rate was permitted to be no less than 80 mL/min and no greater than 120 mL/min, according to the study protocol. Each individual session of hemoperfusion lasted for 2 h. Heparin was used as an anticoagulant. The activated clotting time was measured (0, 30, 90, and 120 min) and the heparin infusion was adjusted, if needed, to keep the activated clotting time within the range of 180 to 240 s. A hemofiltration machine, or a direct hemoperfusion machine with a heparin pump and monitors for inlet and outlet pressure, was used for PMX hemoperfusion. When the hemoperfusion was discontinued within the first 90 min of treatment due to clotting problems, a second PMX treatment was considered. Patients whose PMX treatments were discontinued within 90 min of the second treatment were withdrawn from the study. However, all data were included in the intention to treat analysis.

Study procedures

Patients were followed up for 28 days. All patients had a pulmonary artery catheter inserted as part of routine management. In each patient, the following hemodynamic and blood gas variables were assessed at baseline, day 1, and day 2: heart rate (HR), mean arterial pressure (MAP), right atrial pressure (RAP), pulmonary arterial pressure (PAP), pulmonary capillary wedge pressure (PCWP), cardiac index (CI), systemic vascular resistance (SVR), left ventricular stroke work index (LVSWI), oxygen delivery index (DO_2I), oxygen consumption index (VO_2I), and PaO_2/FiO_2 (P/F ratio). The following were also recorded during the study: routine observations (body temperature, HR, mean blood pressure, and respiratory rate), general blood chemistry/hematology for safety assessment, urine output, concomitant devices and medication, bacteriology, length of ICU stay, and survival. Organ dysfunction was assessed using the SOFA and Goris scores. Endotoxin and IL-6 levels were measured at baseline, 120 min (post-treatment and PMX group only), 6 to 8 h, and 24 h after the first blood sampling. All endotoxin and IL-6 assays were done at a central laboratory (Hammersmith Hospital, London, UK). IL-6 was measured using a commercial kit (Quantikine, R & D Systems, Minneapolis, MN), and endotoxin was measured with the modified limulus amebocyte lysate (LAL) assay (18) using a commercial kit (COATEST, Diapharma, West Chester, OH). The LAL assay is highly sensitive (0.005 EU/mL in saline; 0.05 EU/mL in serum or plasma) and is virtually specific for endotoxin.

Outcome measurements

The primary endpoint was improvement of organ dysfunction in patients and was assessed by the changes in the SOFA score and Goris score from baseline at day 1 to day 6. Secondary endpoints were reduction in plasma endotoxin and IL-6 concentrations (pretreatment to 120 min, 6-8 h and 24 h post-treatment), patient survival over 28 days after PMX treatment, and length of ICU stay. After data analysis, the outcome of the study was not as expected, and due to this, an additional analysis was performed on the hemodynamic data, comparing the change from baseline to day 1 and day 2; the pulmonary artery catheter enabling hemodynamic measurements was present in the majority of patients for up to 2 days post-treatment. Also, the need for renal replacement therapy was assessed for the first 5 days after study entry.

Data analysis

Efficacy and safety analyses were performed on a per protocol analysis. The change from baseline was compared between the two groups using an analysis of covariance (ANCOVA) or nonparametric test (Wilcoxon test). The normal distribution of the data was checked using the Shapiro-Wilk's test and a normal QQ-plot. When the data were normally distributed, the Student's *t* test was used. Wilcoxon test was used for the data that were not normally distributed. The endotoxin and IL-6 levels were compared using the Wilcoxon test. For the changes in hemodynamic and blood gas data, the data were compared using ANCOVA. For SOFA and Goris scores, the mean area under the curve from baseline to day 6 was calculated. The total scores were carried forward for patients who died before day 6. Shock-free days were defined as the number of days a subject was alive and not receiving vasopressor medication during the 28-day period. Mechanical ventilation (MV)-free days and ICU-free days were calculated likewise. The difference in the free days between the two groups was compared by a Student's *t* test. The two groups were also compared for frequency of initiation of CRRT using a Fisher's exact test. Patient survival over the 28 days was assessed by product-limit life table analyses using the log-rank test. Hypothesis tests were two sided and at the 5% level of statistical significance.

RESULTS

Baseline demographics

A total of 36 patients were randomly assigned into the study over 2 years, with 17 patients in the PMX group and 19 patients in the control group. One patient, in the control group, was excluded from analysis (as per protocol) for not showing

evidence of a gram-negative infection or endotoxemia at baseline. The demographics and baseline data of the 35 patients remaining in the analysis are summarized in Table 1. There was no significant difference between the two groups in baseline demographics, APACHE II score, or number of failed organs.

Gram-negative bacteria were detected between day −4 and day 0 (baseline) in 13 (72%) control and 14 (82%) PMX patients; seven of the control and 10 of the PMX patients were infected with mixed bacteria (Table 1). In the control group, four of 18 patients had gram-negative bacteremia. In the PMX group, three of 17 patients had gram-negative bacteremia. There were no significant differences in antibiotic administration at baseline (Table 1).

Endotoxin and IL-6

The median, 25% percentile, and 75% percentile of blood endotoxin and IL-6 levels are shown in Table 2. The baseline endotoxin and IL-6 levels were similar in both groups. Endotoxin levels in both groups did not change significantly between baseline and 120 min. There were no statistically significant differences in the endotoxin levels between the two groups at any time point. IL-6 levels in both groups showed a tendency to decrease after 24 h, but this was not statistically significant.

TABLE 1. **Summary of baseline characteristics**

	Control Group (n = 1)	PMX Group (n = 1)
Gender (%)		
Male:female	9 (50):9 (50)	13 (76):4 (24)
Age (year)		
Mean (range)	62.3 (29-79)	52.7 (28-76)
APACHE II score at study entry (mean ± SD)	18.7 ± 6.1	16.7 ± 5.9
Baseline score (mean ± SD)		
SOFA score	10.2 ± 3.2	10.0 ± 2.3
Goris score	7.1 ± 2.4	6.8 ± 1.9
Baseline organ dysfunction (%): 16 patients in each group had two or more failed organs		
Any	18 (100)	17 (100)
Shock	17 (94)	17 (100)
Respiratory	14 (78)	10 (59)
Renal	9 (50)	8 (47)
Central nervous system	1 (6)	2 (12)
Hepatic	0 (0)	1 (6)
Hematologic	8 (44)	5 (29)
Type of intra-abdominal infection (%)		
Abscess in abdominal cavity	3 (17)	3 (18)
Secondary to gut perforation	7 (38)	3 (18)
Secondary to gut resection	2 (11)	6 (34)
Secondary to pancreatitis	2 (11)	1 (6)
Cholecystitis	1 (6)	2 (12)
Other	3 (17)	2 (12)
Bacteriology between day −4 and day 0 (%)		
Mixed	7 (38)	10 (59)
Gram-negative only	6 (33)	4 (24)
Gram-positive only*	3 (17)	2 (12)
Yeast only*	1 (6)	1 (6)
None*	1 (6)	0
Use of vasopressor (%)		
Norepinephrine/epinephrine	11 (61)	15 (88)
Dopamine	6 (33)	12 (71)
Dobutamine	7 (39)	3 (18)
Use of antibiotic (%)		
Amikacin	8 (44)	5 (29)
Cefuroxime	6 (33)	6 (35)
Fluconazole	6 (33)	7 (41)
Metronidazole	11 (61)	8 (47)

*Endotoxin positive at baseline.

Hemodynamic and blood gas data

Changes in hemodynamic variables from baseline to days 1 and 2 were evaluated. There were no significant differences in hemodynamic variables at baseline between the groups (Table 3). In the PMX group, MAP increased significantly from baseline to day 2 ($P = 0.006$). In the control group, PAPs increased significantly from baseline to day 1 ($P = 0.034$) and day 2 ($P = 0.004$). CI was significantly greater in the PMX than in the control group at days 1 and 2 ($P = 0.012$ and 0.032). In the PMX group, the LVSWI increased from baseline to day 2 and was greater than in the control group on day 2 ($P = 0.015$). The PCWP remained unchanged (Fig. 1). DO_2I increased in the PMX group to become significantly greater than in the control group on day 2 ($P = 0.007$).

At baseline, 14 (78%) control and 17 (100%) PMX patients were receiving at least one inotropic agent (Table 1). There were no significant differences between groups in the doses of each inotropic or vasopressive agent on days 1 and 2. There was no significant difference in shock-free days (control: 15.2 ± 11.1, range 0-29 days; PMX: 14.4 ± 12.5 days, range 0-28 days).

Organ function

The mean serum creatinine value at baseline was similar in both groups (control, 2.03 ± 0.96 mg/dL; PMX, 1.94 ± 1.03 mg/dL). Nine (50%) of 18 control patients and 8 (47%) of 17 PMX patients had renal failure (oliguria or creatinine >2 mg/dL) at study entry, and a total of seven (39%) control patients and two (12%) PMX patients received CRRT (hemofiltration including CVVH and/or hemodialysis) for the first 5 days of the study. The frequency of CRRT initiation after study entry in the control group (seven of 18 cases with an average CRRT duration of 9.3 days) was statistically greater ($P = 0.043$) than in the PMX group (one of 16 cases [one patient was already receiving CRRT at baseline] with a duration of 1 day only).

At baseline, 16 (89%) control and 17 (100%) PMX patients were already receiving MV. The mean number of MV-free days was 13.9 ± 11.3 days in the PMX group and 11.7 ± 10.7 days in the control group (differences not significant). There were no significant differences in the SOFA and Goris Scores between the control and the PMX groups.

Survival over 28 days and time in ICU

Five of 18 (28%) control patients and five of 17 (29%) PMX patients died during the study period. The survival analysis of time (days) to death showed no statistical significance between the two groups ($P = 0.749$). There was no statistically significant difference in the mean duration of ICU stay between the two groups (PMX, 13.2 ± 9.4 days; control, 17.0 ± 9.4 days, ns). The number of ICU-free days was 6.8 ± 9.4 and 9.2 ± 10.4 days in the control and PMX groups, respectively (ns).

TABLE 2. **Actual endotoxin and IL-6 levels, pg/ml**

	Control Group					PMX Group				
Time Point	Minimum	25% Percentile	Median	75% Percentile	Maximum	Minimum	25% Percentile	Median	75% Percentile	Maximum
Endotoxin										
Baseline	0	12.0	72.5	165.0	277	0	3.8	28.0	96.8	647
120 min	N/A	N/A	N/A	N/A	N/A	0	20.0	26.0	53.8	845
6-8 h	0	20.0	41.0	123.0	187	0	7.0	29.0	81.5	1956
24 h	0	36.0	68.5	99.0	478	0	9.0	38.5	69.0	1330
IL-6										
Baseline	136	1066	1605	7242	26,520	82	350	1632	4316	47,481
120 min	N/A	N/A	N/A	N/A	N/A	73	304	1437	3473	22,740
6-8 h	111	486	1213	4484	11,900	70	253	1175	4964	22,511
24 h	69	172	638	954	15,168	34	187	512	1746	12,486

N/A, Not applicable.

Adverse events

During the study, all patients in both groups reported at least one treatment-emergent adverse events (AE). The control group patients had a greater number of AEs than the PMX patients (control, 127 events [7.1 per patient]; PMX, 80 events [4.7 per patient]). Only one PMX patient had a treatment-emergent AE considered to be possibly device related (fever). The most frequent AEs that occurred during the study were pleural effusions (five control and three PMX patients), anemia (three control and four PMX patients), and fluid overload (three patients in each group). None of the AEs occurred during PMX treatment, and the AEs reported during the study were not indicative of polymyxin-B toxicity (nephrotoxicity, including albuminuria and cellular casts; neurotoxicity, including irritability and progressive weakness).

There were no significant differences in the white blood cell or platelet counts between the two groups. Twelve (71%) PMX patients received PMX treatment within 48 h after abdominal surgery. Despite the administration of 2500 to 7000 U of unfractionated heparin (bolus plus continuous administration), no AEs due to bleeding were observed during or after PMX treatment.

Device failure

A total of 21 PMX cartridges were used for 17 patients in the PMX group. Four (24%) of 17 treatments were incomplete at the first attempt due to clotting. Two of these cases were successfully treated with an additional cartridge. In the other two patients, the second treatment was not completed because clotting occurred again after 51 and 34 min, respectively. At this point, these two patients were withdrawn from the study; however, their data were included in the intention to treat analysis.

DISCUSSION

This is the first multicenter RCT using PMX hemoadsorption in severe sepsis or septic shock. The results indicated that CI, LVSWI, and DO_2I were significantly improved in the PMX group. The need for CRRT was also decreased in the PMX group, and the device was proven to be safe.

PMX is a hemoperfusion cartridge designed to remove endotoxin from the patient's circulation. During this pilot study, there was no significant change in endotoxin levels in the PMX group (at 120 min), but the endotoxin levels showed significant variability within the two groups at each time point measured and across time. This degree of variability may confound the assessment of the PMX device, but is entirely consistent with other clinical studies using the LAL assay to measure plasma endotoxin levels in critically ill patients (19–22). The LAL assay is highly sensitive (0.005 EU/mL in saline; 0.05 EU/mL in serum or plasma) and is virtually specific for endotoxin. Although activated by β-glucan, a component of fungal cell walls, this has to be present in extremely high concentrations before it affects the assay and, therefore, has not been felt to be a confounding factor in the many clinical studies

TABLE 3. **Hemodynamic and blood gas data: changes from baseline (mean ± SD)**

	Control Group		PMX Group	
	Day 1	Day 2	Day 1	Day 2
MAP (mmHg)	4.3 ± 17.15	9.0 ± 18.87	4.2 ± 13.74	10.8 ± 13.65
RAP (mmHg)	0.5 ± 3.79	0.07 ± 3.10	−0.56 ± 5.67	−0.56 ± 4.77
PAP (mmHg)	3.71 ± 6.58	3.2 ± 3.67	2.22 ± 5.60	0.06 ± 4.48
PCWP (mmHg)	0.44 ± 5.40	−1.07 ± 3.24	−1.06 ± 5.18	−1.81 ± 4.96
CI (L/min/m^2)	−0.11 ± 1.06**	−0.21 ± 0.88**	0.49 ± 0.87**	0.32 ± 1.13**
SVR (dynes/cm^{-5})	38.5 ± 248.92	154.7 ± 259.78	−13.0 ± 156.39	95.4 ± 352.71
LVSWI (g/min/m2)	3.27 ± 13.15	4.20 ± 13.85**	9.05 ± 10.68	14.31 ± 14.58**
DO_2I (mL/min/m2)	−21.06 ± 138.78	−48.43 ± 120.91**	22.43 ± 97.94	29.96 ± 88.92**
VO_2I (mL/min/m2)	−8.14 ± 34.51	−19.48 ± 27.16	5.53 ± 40.11	9.90 ± 42.27
P/F ratio (mmHg)	22.08 ± 92.44	0.43 ± 93.36	−9.47 ± 60.88	29.54 ± 83.38

**Significant difference in change from baseline between groups.

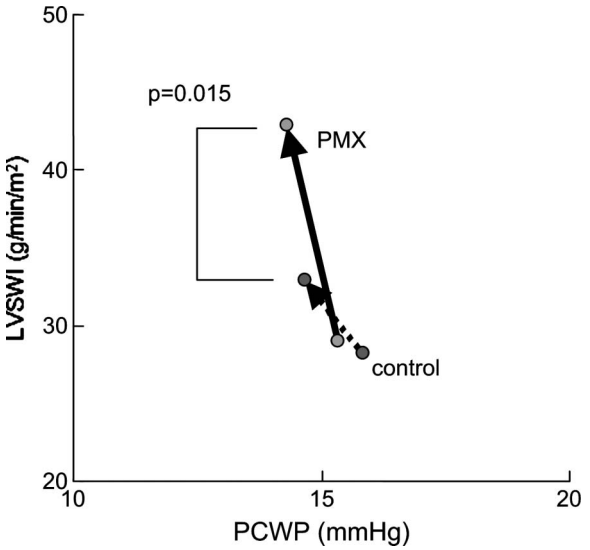

FIG. 1. **Effects of PMX on left ventricular function.** Arrows show the direction of the changes of LVSWI and PCWP from baseline to day 2.

that have evaluated the LAL test. The inability to show a significant decrease in endotoxin levels has been described previously in Japanese clinical studies using this device (23). Furthermore, patients in this study were treated with PMX up to 48 h after the diagnosis of severe sepsis and this also may have affected the endotoxin results. In addition, PMX was applied to patients only once in this study. In cases with overwhelming endotoxin invasion into the blood stream, one session of hemoperfusion with PMX might not be enough. More frequent treatments may have been more effective.

In this pilot study, there was no limitation to ICU patient management apart from the endotoxin adsorption therapy, and differences in infusion therapy among centers may have influenced the hemodynamic results. However, any such effects would be the same in the placebo and PMX groups, thus minimizing any influence on comparisons between groups. In addition, CRRT may have an affect on hemodynamics. Hirasawa et al. (5) reported that hemodynamics in patients with sepsis or septic MOF were improved by the removal of a variety of humoral mediators, including IL-6, with CHF or CHDF. In the present study, there was no significant difference in IL-6 levels between the groups.

Several experimental studies have reported beneficial hemodynamic effects of PMX. In a canine model of endotoxin-induced shock, Aoki et al. (24) investigated the cardiovascular improvement with hemoperfusion carried out using PMX, a sham cartridge (carrier fiber without polymyxin-B), charcoal, and an anion exchange resin; only the PMX-treated group demonstrated an increase in blood pressure. In a later canine study (25), PMX was compared with a sham treatment (blood tubing only) and an anion exchange resin; only the PMX group experienced a sustained increase in blood pressure. In a recent sheep model, PMX improved systemic hemodynamics and oxygenation compared with treatment with a sham column (26). In open-label, nonrandomized clinical studies in Japan, PMX treatment has been associated with earlier withdrawal from dopamine infusion, as a result of increased MAP and increased SVR (CI unchanged) (27, 28). In the present RCT, MAP in the PMX group also increased significantly from baseline to day 2, but there was no significant difference between the two groups. An improvement in cardiac function by PMX was indicated in this study by an increase in LVSWI without notable changes in PCWP.

The favorable effects of PMX on cardiac function may be secondary to the elimination of myocardial-depressant mediators. Various candidates have been suggested. Recently, Nakamura et al. (29) reported that cardiac troponin T levels, a potential marker of septic myocardial injury (30), were higher in hemodialysis patients with sepsis than in those without sepsis and healthy control subjects, and that PMX treatment reduced these levels (29). Other possible candidates that may be removed by PMX treatment with resulting beneficial hemodynamic effects include endogenous cannabinoids, such as macrophage-derived anandamide (31) and tetrahydrbiopterin, an essential cofactor for inducible nitric oxide synthase (32). Further clinical studies are needed to investigate the exact mechanisms involved in PMX treatment.

We acknowledge the study has some limitations, including potential confounding effects of the heparin used in the hemoperfusion device, the limitations of the assays used to measure endotoxin and IL-6 levels, and the inability to measure all mediators or constituents that may have been removed by the cartridge.

In conclusion, although this pilot study failed to demonstrate reduced blood endotoxin levels compared with the control group, the results do show that PMX treatment is safe, and is associated with an improved hemodynamic status and cardiac function. Larger, adequately powered, multicenter clinical trials now need to be conducted to confirm the effects of PMX treatment on hemodynamics and to determine its effects on outcome.

ACKNOWLEDGMENTS

The authors thank all the nursing staff and the physicians of the ICU and Nephrology Departments for their major contribution in the daily care of the study patients, and Peter Fenwick for technical assistance with the assays. They also thank Toray Industries, Inc. for the donation of the PMX cartridges and Hisataka Shoji (Toray Industries, Inc.) for his advice in the preparation of this manuscript.

REFERENCES

1. Oberholzer A, Oberholzer C, Moldawer LL: Sepsis syndromes: understanding the role of innate and acquired immunity. *Shock* 16:83–96, 2001.
2. Power C, Fanning N, Redmond HP: Cellular apoptosis and organ injury in sepsis: a review. *Shock* 18:197–211, 2002.
3. Opal SM, Huber CE: Bench-to-bedside review: Toll-like receptors and their role in septic shock. *Crit Care* 6:125–136, 2002.
4. Amaral A, Opal SM, Vincent JL: Coagulation in sepsis. *Intensive Care Med* 30:1032–1040, 2004.
5. Hirasawa H, Oda S, Shiga H, et al.: Endotoxin adsorption or haemodiafiltration in the treatment of multiple organ failure. *Curr Opin Crit Care* 6:421–425, 2000.
6. Ronco C, Brendolan A, Lonnemann G, Bellomo R, Piccinni P, Digito A, Dan M, Irone M, La Greca G, Inguaggiato P, Maggiore U, De Nitti C, Wratten ML, Ricci Z, Tetta C: A pilot study of coupled plasma filtration with adsorption in septic shock. *Crit Care Med* 30:1250–1255, 2002.
7. Cohen J, Aslam M, Pusey CD, et al.: Protection from endotoxemia: a rat model for plasmapheresis and specific adsorption with polymyxin B. *J Infect Dis* 155:690–695, 1987.

8. Aoki H, Kodama M, Tani T, Hanasawa K: Treatment of sepsis by extracorporeal elimination of endotoxin using polymyxin B-immobilized fiber. *Am J Surg* 167:412–417, 1994.
9. Tetta C, Gianotti L, Cavaillon JM, Wratten ML, Fini M, Braga M, Bisagni P, Giavaresi G, Bolzani R, Giardino R: Coupled plasma filtration-adsorption in a rabbit model of endotoxic shock. *Crit Care Med* 28:1526–1533, 2000.
10. Kodama M, Tani T, Hanasawa K, et al.: Treatment of sepsis by plasma endotoxin removal: Hemoperfusion using a polymyxin-B immobilized column. *J Endotoxin Res* 4:293–300, 1997.
11. Uriu K, Osajima A, Hiroshige K, Watanabe H, Aibara K, Inada Y, Segawa K, Anai H, Takagi I, Ito A, Kamochi M, Kaizu K: Endotoxin removal by direct hemoperfusion with an adsorbent column using polymyxin B-immobilized fiber ameliorates systemic circulatory disturbance in patients with septic shock. *Am J Kidney Dis* 39:937–947, 2002.
12. Nemoto H, Nakamoto H, Okada H, Sugahara S, Moriwaki K, Arai M, Kanno Y, Suzuki H: Newly developed immobilized polymyxin B fibers improve the survival of patients with sepsis. *Blood Purif* 19:361–368, 2001.
13. Reinhart K, Meier-Hellmann A, Beale R, Forst H, Boehm D, Willatts S, Rothe KF, Adolph M, Hoffmann JE, Boehme M, Bredle DL: Open randomized phase II trial of an extracorporeal endotoxin adsorber in suspected gram-negative sepsis. *Crit Care Med* 32:1662–1668, 2004.
14. ACCP-SCCM Consensus Conference: Definitions of sepsis and multiple organ failure and guidelines for the use of innovative therapies in sepsis. *Crit Care Med* 20:864–874, 1992.
15. Knaus WA, Draper EA, Wagner DP, Zimmerman JE: APACHE II: a severity of disease classification system. *Crit Care Med* 13:818–829, 1985.
16. Vincent JL, de Mendonça A, Cantraine F, Moreno R, Takala J, Suter P, Sprung C, Colardyn FC, Blecher S: Use of the SOFA score to assess the incidence of organ dysfunction/failure in intensive care units: results of a multicentric, prospective study. *Crit Care Med* 26: 1793–1800, 1998.
17. Goris RJ, te Boekhorst TP, Nuytinck JK, Gimbrere JS: Multiple-organ failure. Generalized autodestructive inflammation? *Arch Surg* 120:1109–1115, 1985.
18. Cohen J, McConnell JS: Observations on the measurement and evaluation of endotoxemia by a quantitative limulus lysate microassay. *J Infect Dis* 150: 916–924, 1984.
19. Bates DW, Parsonnet J, Ketchum PA, Miller EB, Novitsky TJ, Sands K, Hibberd PL, Graman PS, Lanken PN, Schwartz JS, Kahn K, Snydman DR, Moore R, Black E, Platt R: Limulus amebocyte lysate assay for detection of endotoxin in patients with sepsis syndrome. AMCC Sepsis Project Working Group. *Clin Infect Dis* 27:582–591, 1998.
20. Maury E, Barakett V, Blanchard H, Guitton C, Fitting C, Vassal T, Chauvin P, Guidet B, Offenstadt G: Circulating endotoxin during initial antibiotic treatment of severe gram-negative bacteremic infections. *J Infect Dis* 178:270–273, 1998.
21. Cohen J: The detection and interpretation of endotoxaemia. *Intensive Care Med* 26(suppl 1):S51–S56, 2000.
22. Venet C, Zeni F, Viallon A, Ross A, Pain P, Gery P, Page D, Vermesch R, Bertrand M, Rancon F, Bertrand JC: Endotoxaemia in patients with severe sepsis or septic shock. *Intensive Care Med* 26:538–544, 2000.
23. Tani T: Review of endotoxin-adsorbing direct hemoperfusion therapy using a column containing polymyxin B immobilized fiber. *Curr Opin Crit Care* 6:416–420, 2000.
24. Aoki H, Hanasawa K, Tani T, et al.: Comparative study between PMX-F and other adsorbents for treatment of septic shock. *Therapeutic Plasmapheresis* 312:345–350, 1988.
25. Sato T, Orlowski JP, Zborowski M: Experimental study of extracorporeal perfusion for septic shock. *ASAIO J* 39:M790–M793, 1993.
26. Yamamoto H, Koizumi T, Kaneki T, Fujimoto K, Kubo K, Honda T: Direct hemoperfusion with polymyxin B-immobilized fiber improves shock and hypoxemia during endotoxemia in anesthetized sheep. *J Endotoxin Res* 8:419–426, 2002.
27. Tani T, Hanasawa K, Kodama M, Imaizumi H, Yonekawa M, Saito M, Ikeda T, Yagi Y, Takayama K, Amano I, Shimaoka H, Ohta M, Okahisa T, Koga N, Fujita N, Yamasa H: Correlation between plasma endotoxin, plasma cytokines, and plasminogen activator inhibitor-1 activities in septic patients. *World J Surg* 25: 660–668, 2001.
28. Tani T, Hanasawa K, Endo Y, Yoshioka T, Kodama M, Kaneko M, Uchiyama Y, Akizawa T, Takahasi K, Sugai K: Therapeutic apheresis for septic patients with organ dysfunction: hemoperfusion using a polymyxin B immobilized column. *Artif Organs* 22:1038–1044, 1998.
29. Nakamura T, Ushiyama C, Shoji H, Koide H: Effects of hemoperfusion on serum cardiac troponin T concentrations using polymyxin B-immobilized fibers in septic patients undergoing hemodialysis. *ASAIO J* 48:41–44, 2002.
30. Fernandes CJ Jr, Akamine N, Knobel E: Cardiac troponin: a new serum marker of myocardial injury in sepsis. *Intensive Care Med* 25:1165–1168, 1999.
31. Wang Y, Liu Y, Ito Y, Hashiguchi T, Kitajima I, Yamakuchi M, Shimizu H, Matsuo S, Imaizumi H, Maruyama I: Simultaneous measurement of anandamide and 2-arachidonoylglycerol by polymyxin B-selective adsorption and subsequent high-performance liquid chromatography analysis: increase in endogenous cannabinoids in the sera of patients with endotoxic shock. *Anal Biochem* 294:73–82, 2001.
32. Hoshiai K, Hattan N, Fukuyama N, Tadaki F, Hida M, Saito A, Nakanishi N, Hattori Y, Nakazawa H: Increased plasma tetrahydrobiopterin in septic shock is a possible therapeutic target. *Pathophysiology* 7:275–281, 2001.