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Safety of Polymyxin-B–based Hemoperfusion in Kidney and Liver Transplant Recipients

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ABSTRACT

Infection represents one of the primary barriers to successful organ transplantation. Our principal end point was to use a new assay, Entotoxin Activity Assay (EAA), which was developed to rapidly detect endotoxin activity (EA) for an early diagnosis of this complication. We also sought to prove the validity and safety of endotoxin removal using polymyxin-B–based hemoperfusion (PMX-DHP). The criterion for inclusion in the study was suspected infection when a patient experienced at least 2 of the 4 criteria of the systemic inflammatory response syndrome. EAA was performed on 71 patients: 29 liver transplantations and 42 kidney transplantations. Twenty-eight patients (39.5%) with EA >0.60 underwent PMX-DHP treatment to remove endotoxins. Each treatment was performed for 2 hours with a blood flow of 100 mL/min. All of the patients were treated with PMX-DHP until achieving an EA <0.4. Stabilization of hemodynamic and inflammatory frameworks was observed after the PMX-DHP. At 30 days follow-up, all of the patients were alive with good graft function and low levels of EA. We think it might be useful to determine EA routinely in transplant patients and look forward to large multicenter clinical trials to accurately assess the benefits of the EAA plus DHP-PMX to treat transplant patients with sepsis.

Bloodstream infection (BSI) remains an important cause of morbidity and mortality after solid-organ transplantation.[1] The risk of infection in transplant recipients is determined by their immunodeficiency compounded by chronic diseases at the time of transplantation, the intensity of exposure to potential pathogens (epidemiologic exposure), and the combined effect of all factors that contribute to a patient's susceptibility to infection (net state of immunosuppression).[2] For these reasons, recognition of infection is more difficult in transplant recipients than in individuals with normal immune functions. Our experience deals with kidney and liver transplant recipients. BSI affects 19%–35% of liver transplant patients and 15% of kidney recipients.[3,4] The immune system deficit of recipients with chronic disease at time of transplantation is different in kidney versus liver patients.

Chronic kidney disease patients show multiple biochemical, immune, and inflammatory alterations. The 2 major factors affecting these disorders are: 1) metabolic, biochemical, immune, or inflammatory alterations due to the uremic syndrome per se; and 2) alterations due to the therapeutic treatments of uremia, especially hemodialysis-induced stress, including activation of the proinflammatory transcription factor nuclear factor κB or endotoxin activity (EA),[5–6] conferring susceptibility to infection after kidney transplantation.

Chronic hepatic disease patients show deranged immune responses, as reflected by elevated serum levels of both proinflammatory and antiinflammatory cytokines. The mechanisms of the deficits include decreased hepatic production of complement (reduced C3 and C5 levels), impaired Kupffer cell function (phagocytosis), altered neutrophil chemotaxis, and impaired clearance of inflammatory cytokines.[7–11] Patients may also demonstrate decreased

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bactericidal activities of immunoglobulins against gram-negative pathogens.[12] Perhaps as a result of these deficits, BSI remains a significant concern in the early posttransplantation period; it is by far the most frequently occurring infectious complication after liver transplantation.[13–15] The timeline of posttransplantation infections has been used to establish a differential diagnosis: "early" imfections show a major predominance of bacteremia; "intermediate," viral pathogens and graft rejection as responsible for the majority of febrile episodes in this period; and "late", problems similar to those of the general community, primarily respiratory. Given that among both liver and kidney transplant patients, there is a high incidence of infections caused by gram-negative organisms and owing to fact that their inflammatory responses are profoundly altered, the clinical expressions of infection are often masked, and therefore any clinical signs or symptoms, even minor, require a comprehensive diagnostic approach. The presence of endotoxins, derived from gram-negative bacteria, confer greater resistance to standard antibiotic therapy and facilitate rapid evolution to septic shock and multi-organ failure (MOF). Therefore, it is important to determine an early diagnosis of EA and apply appropriate therapy.

Our principal end point was to use a new assay, Endotoxin Activity Assay (EAA; Spectral Diagnostics, Toronto, Canada), which was developed to rapidly detect EA in whole blood, to optimize targeted therapeutic interventions. We also sought to prove the validity and safety of endotoxin removal using polymyxin-B–based fiber hemoperfusion (PMX-DHP). Finally, survival, EA, and graft function were monitored for 30 days' follow-up. EAA measures the neutrophil-dependent respiratory burst activity in the presence of a specific lipopolysaccharide (LPS)–antibody reaction. Studies using this assay observed EA levels to significantly correlate with the severity of illness among intensive care unit patients, permitting the determination of risk to develop severe sepsis and septic shock. EAA has a predetermined high cut-off level of 0.60 EA units, which is associated with an increased risk for an adverse outcome.

METHODS

We studied all 191 adults with available follow-up data who underwent primary transplantation with a graft from a cadaveric donor from April 2008 to January 2011: 60 nonurgent liver and 131 kidney transplantations. Retransplantations and combined liver/kidney recipients were excluded. The criteria for inclusion were infection suspected by the presence of at least 2 of the 4 criteria of systemic inflammatory response syndrome (SIRS), ie, fever or hypothermia (temperature >38°C or <36°C, respectively), tachycardia (>90/min), tachypnea (>20/min), or $PaCO_2$ <32 mm Hg or mechanical ventilation, and a white blood cell count $>12.0 \times 10^4$/L or $<4.0 \times 10^4$/L or at least 10% immature neutrophils. Following these criteria, the Test EAA was performed on 71 patients: 29 liver and 42 kidney transplant recipients.

Twenty-eight enrolled patients (39.5%) with EA >0.60 received PMX-DHP treatment to remove endotoxins. The 11 female and 17 male subjects had a mean age of 54 ± 4.7 years. The Sequential Organ Failure Assessment (SOFA) was adopted to assess severity before performing DHP-PMX. For 48 hours postoperatively, all of the patients had received prophylactic antibiotics: ampicillin sodium and the beta-lactamase inhibitor sulbactam sodium (3 g/d). Blood and urine cultures were performed in all patients. When infection was suspected, meropenem (2 g/day) and vancomycin (2 g/d) were administered as initial empiric treatment, because in our clinical experience *Pseudomonas aeruginosa, Escherichia coli* and *Enterococcus* represented the most frequently occurring pathogens.

Transplant recipients generally received a calcineurin inhibitor, corticosteroids, and mycophenolate mofetil (MMF). The immunosuppressive regimens in the 28 patients included tacrolimus or cyclosporine MMF, and corticosteroids in 14 liver transplant patients (100%). Cyclosporine, MMF, and corticosteroids in 4 kidney transplant patients (28.5%), and tacrolimus, MMF, and corticosteroids in 10 kidney transplant patients (72.5%).

Standard harvest techniques used in situ cooling of the abdominal organs with University of Wisconsin preservation solution. Orthotopic liver transplantation was performed using the piggyback technique, leaving the inferior caval vein in situ without a temporary portocaval shunt in 13 patients. In the remaining live transplant patient, a venovenous bypass was performed using a centrifugal pump and heparinized tubing, connecting the femoral and portal veins to the axillary vein. Kidney transplantations were performed with a ureteroneocystostomy using the Lich-Gregoir extravesical technique positioning a double-J ureteral stent which remained for 6–8 weeks. The bladder catheter was routinely removed on day 6.

Endotoxin Activity Assay

EA in whole blood was measured as described by Romaschin et al[26] using the chemiluminescent EAA. Briefly, samples of EDTA anticoagulated whole blood were incubated with saturating concentrations of antibody, before stimulation with zymosan. The LPS/anti-LPS complex primes the patient's neutrophils for an augmented response. The resulting respiratory burst was detected as light release with the use of a chemiluminometer (SmartLine TL luminometer; Berthold). We measured basal (no antibody) and maximally stimulated (4,600 pg/mL LPS + antibody) responses in the same blood sample to calculate the EA of the test specimen (antibody only). EA was expressed in relative units derived from the integral of the basal and stimulated chemiluminescent responses. EA was classified as low (EA <0.4), intermediate ($0.4 \leq$ EA < 0.6), or high (EA ≥0.6) for a response set within 40 minutes. The EA levels were measured within 24 hours of the onset of signs of SIRS in association with suspicion of infection. If EA ≥0.60 was detected, patients were assigned to receive endotoxin removal therapy (PMX-DHP). EA levels were measured 1 hour after each hemoperfusion to evaluate the posttreatment effect, and again just before the start of the next PMX-DHP treatment.

Polymyxin-B–Based Hemoperfusion

Polymyxins have the unusual ability to bind to and neutralize endotoxin, an action in additions to their antibacterial effects that might be of clinical value. Polymyxins are a group of cyclic cationic polypeptide antibiotics derived from *Bacillus polymyxa.* Only polymyxin-B and colistin (polymyxin-E) have been used in clinical practice. Polymyxin-B differs from colistin by only 1 amino acid. The neutralization of the lethal effects of endotoxin in animal models by polymyxin-B has been known for more than 40 years, but their human clinical use has been limited owing to nephrotoxicity and neurotoxicity. This limitation has been overcome by a mechanism

which takes advantage of polymyxin without exposing patients to its systemic effects. PMX-DHP is the only extracorporeal device reported to be safe and effective in septic patients.[16–19] Polymyxin is covalently bound to polystyrene fibers by a reaction between one of the amino groups of the diaminobutyric acid residues, leaving at least 3–4 charged amino groups for LPS binding.

In the present study, we performed PMX-DHP (Toraymyxin; Toray Industries; Tokyo; Japan) with each treatment lasting 2 hours at a blood flow of 120 mL/min. All of the patients were treated with PMX-DHP daily until achieving an EA of <0.4. Vascular access was obtained by insertion of a double-lumen dialysis catheter (Arrow International, Reading, Pennsylvania) into the internal jugular vein (n = 10) or subclavian vein (n = 18). For anticoagulation, we injected a bolus of 2,500 units unfractionated heparin into the system for priming. None of the patients were on mechanical ventilatory support. The following clinical parameters were reported before and after each treatment cycle: kidney and liver parameters, urine output, mean arterial pressure (mAP), heart rate (HR), PaO_2/FiO_2 ratio, white blood cell count (WBC) with percentage of neutrophils, lactic acid, and inflammatory cytokine levels.

Cytokine Assays

Blood samples were drawn at the baseline and at the end of each PMX-DHP treatment. After 30 minutes samples were centrifuged to obtain serum aliquots that were stored frozen at −70°C for subsequent cytokine assay. Interleukin (IL) 6 was measured by a chemoluminescent assay (Immulite 2000; DPC Biermann, Bad Nauheim, Germany). Tumor necrosis factor (TNF) α was measured by enzyme-linked immunosorbent assay (Quantikine; R&D Systems, Minneapolis, Minnesota).

Statistical Analysis

All data were analyzed with SPSS for Windows version 16.0 (SPSS, Chicago, Illinois). Wilcoxon (Mann-Whitney) nonparametric tests compared unpaired data ($P < .05$) between baseline and end of PMX-DHP therapy. Hemodynamic data were analyzed with the use of 1-way analysis of variance (ANOVA) and ANOVA for repeated measures with a confidence interval of 95%. The analysis of mAP and HR was first performed by grouping data from both liver and kidney transplant patients to evaluate the effect of time. Treatment repetition on the same patient was indicated as T0 (basal) and T1, T2, T3, and T4 after the first, the second, the third, the fourth treatments, respectively. The 1-way ANOVA calculation was then performed separately 3 times for patients who had undergone 2 (HP2 group; n = 14), 3 (HP3 group; n = 8), or 4 (HP4 group; n = 6) PMX-DHP treatments. ANOVA for repeated measures was performed to compare differences in the efficacy of PMX-DHP therapy among various treatment schedules, ie, 2, 3, or 4 hemoperfusions. Pearson test was used to evaluate correlations among variables.

RESULTS

Fourteen liver (48.2%) and 14 kidney (38.7%) transplant patients showed positive EA levels and were treated with PMX-DHP: 11 liver transplant patients within 15 days and 3 within 48 days after transplantation and 6 kidney transplant patients within 30 and 8 within 74 days. There was no significant correlation between EA and immunosuppressive regimes. Before performing PMX-DHP treatments, the median EAs were 0.81 (range, 0.62–1.25) and 0.73 (range, 0.61–0.98) for liver and kidney transplant patients, respectively. A summary of the clinical parameters of these patients before and after PMX-DHP treatments is reported in Table 1. Two PMX-DHP treatments were performed on 16 patients, 3 on 8, and 4 on 4. No relevant adverse events were observed during the 72 treatments. Among liver transplant patients, 2 PMX-DHP treatments were performed on 7 patients [median EA 0.69 (0.62–0.76)], 3 treatments on 4 patients [median EA 0.84 (0.77–0.91)], and 4 treatment on 3 patients [median EA 1.11 (0.95–1.25)]. At the end of the endotoxin removal therapy, the median EA level was 0.33 [0.22–0.4]. After the last PMX-DHP treatment, when the EA levels were <0.4, we observed a significant improvement in the hemodynamic, liver parameters, and cytokine values. At the start, the evaluation of hepatic parameters showed an increase in bilirubin and transaminases. All parameters reached normal values within 5 ± 0.4 days after the end of therapy. Microbiologic findings showed the presence of gram-negative infections in 8 patients within 68.7 ± 5.2 hours after enrollment. The remaining 6 patients had negative blood or urine cultures.

Following the indications of EAA, PMX-DHP treatments were performed in kidney transplant patients: 2 on 9 patients [median EA 0.67 (0.61–0.72)], 3 on 4 [median EA 0.8 (0.78–0.82)], and 1 on 1 (EA 0.98) (Fig 2). Satisfactory values of daily urine output and kidney parameters were reached at the end of therapy. Significant improvements in hemodynamic parameters and cytokine values were observed, as in the liver transplant patients. Fig 1 shows a significant decrease ($P < .001$) in IL-6 and TNF-α at the end of extracorporeal therapy. Microbiologic findings showed the presence of gram-negative infections in 13 KT patients within 70 ± 2.4 hours after enrollment. Only 1 patient had negative blood and urine cultures. The KT patients revealed urinary tract infection to be the most common (69.2%).

Fig 3 shows the effects of each treatment on mAP and HR among all 28 patients. The overall effects of PMX-DHP therapy produced significant declines for both mAP (HP2: $P < .001$; HP3: $P < .001$; HP4: $P < .001$) and HR (HP2: $P < .001$; HP3: $P < .001$; HP4: $P < .001$). No significant difference was displayed for both mAP and HR after the first hemoperfusion (T1) compared with basal (T0) values. After the second treatment (T2), HP2 and HP3 groups showed a significant improvements in mAP (HP2: $P < .001$; HP3: $P < .001$) and HR (HP2: $P < .001$; HP3: $P = .001$) compared with basal (T0) values. HP4 group at T2 reached significance for HR (HP4: $P = .013$) but not for mAP (HP4: $P = .152$) compared with basal values. Significant differences were confirmed for mAP and HR in the HP3 group (mAP: $P < .001$; HR: $P < .001$), but mAP was significantly increased compared with the previous treatment of the same group ($P < .001$). The HP4 group reached significance for mAP after the third treatment ($P = .004$) compared with basal values as well as the previous treatment ($P = .004$). HR values of HP4 group did not significantly change after the second treatment. Analyses separately performed for liver versus kidney transplant patients

Table 1. Biochemical, Hemodynamic, and Clinical Changes of 28 Transplant Recipients Before and After PMX-DHP Treatments

	Liver Transplant (n = 14)		Kidney Transplant (n = 14)	
	Before	After	Before	After
T (C°)	37.4 ± 0.4	36.4 ± 0.5*	38.1 ± 0.9	36.1 ± 0.2*
mAP (mm Hg)	70 ± 2.88	82 ± 2.86*	72 ± 1.52	84 ± 0.21*
Heart rate (beats/min)	107 ± 3.55	78 ± 1.25*	110 ± 1.84	79 ± 2.67*
PaO_2/FiO_2 ratio	287 ± 14.25	316 ± 10.26*	300 ± 12	331 ± 9.56*
WBC ($10^3/mm^3$)	12,480 ± 1,680	5,875 ± 2,110*	11,250 ± 2,180	6,950 ± 1,430*
Neutrophilis (%)	82.22 ± 0.75	68.41 ± 4.22*	84 ± 1.50	70 ± 2.1*
Platelets ($10^3/\mu L$)	120 ± 46	119 ± 37	207 ± 34	205 ± 10
INR	1.42 ± 0.8	1.27 ± 0.4	0.8 ± 0.2	0.8 ± 0.1
Bilirubin (mg/dL)	8.4 ± 2.7	7.3 ± 1.8	0.9 ± 0.3	0.9 ± 0.3
ALT (IU/L)	782 ± 95	635 ± 84	44 ± 2.5	43.55 ± 1.8
AST (IU/L)	655 ± 64	495 ± 56*	37.5 ± 1.1	33 ± 1.6
γ-GT (IU/L)	225 ± 43	172 ± 40*	29 ± 0.4	29.2 ± 0.5
BUN (mg/dL)	49 ± 9.56	31 ± 6.22	184.6 ± 31	102 ± 15.20*
Creatinine (mg/dL)	1.05 ± 0.88	0.7 ± 0.16	5.9 ± 1.4	4.3 ± 0.7*
Diuresis (mL/d)	1,660 ± 345	2,100 ± 488*	940 ± 104	1,850 ± 225*
SOFA	9 ± 0.8	5 ± 0.3*	7 ± 0.4	4 ± 0.6*
Lactate (mmol/L)	3.5 ± 1.4	0.7 ± 0.8*	2.7 ± 0.4	0.6 ± 0.2*
IL-6 (ng/dL)	97.15 ± 21	48 ± 11.22*	84 ± 17.89	42 ± 0.4
TNF-α (ng/dL)	79.5 ± 16	32 ± 14.1*	61 ± 8.54	29 ± 6.25*
EA	0.81 [0.62–1.25]	0.33 [0.22–0.40]*	0.73 [0.61–0.98]	0.33 [0.29–0.39]*
30-day mortality	—	0/14	—	0/14

T = temperature; mAP = mean arterial pressure; BUN = blood urea nitrogen; ALT = alanine aminotransferase; AST = aspartate aminotransferase; SOFA = Sequential Organ Failure Assestment; INR = international normalized ratio; TNF = tumor necrosis factor; IL = interleukin; WBC = white blood cells; EA = endotoxin activity. [range].
*$P < .05$ (Wilcoxon signed rank test for paired data: basal vs end of therapy).

showed similar results to those obtained for the whole group. ANOVA comparing the 3 treatment schedules revealed significant differences for basal mAP (HP2 vs HP3: $P = .022$; HP2 vs HP4: $P = .034$; HP3 vs HP4: $P = .998$) and HR (HP2 vs HP3: $P = .025$; HP2 vs HP4: $P = .002$; HP3 vs HP4: $P = .463$) values among the HP2 group, whereas the other 2 groups were not significantly different from each other. This statistical framework did not change qualitatively after the first and the second treatments (T2). Taking into account the mAP and HR values at the end of all 3 schedules (EA <0.4), the significant differences disappeared. In fact, ANOVA showed a nonsignificant difference among groups for both mAP (HP2 vs HP3: $P = .646$; HP2 vs HP4: $P = .545$; HP3 vs HP4: $P = .973$) and HR (HP2 vs HP3: $P = .706$; HP2 vs HP4: $P = .977$; HP3 vs HP4: $P = .682$) values, ie, at the end of the therapies the 3 groups reached the same mAP and HR values.

Microorganisms isolated from blood cultures in 21 patients after 69.4 ± 1.5 hours of incubations were: *Pseudomonas aeruginosa* (n = 3), *Klebsiella pneumonae* (n = 2), *Proteus mirabilis* (n = 1), *Escherichia coli* (n = 12), and *Enterococcus faecium* (n = 3; Table 2). At 30 days' follow-up, all patients were alive with good graft function and low levels of endotoxin activity (Table 3).

DISCUSSION

Today, the prevention of infection has become a major goal of transplantation. In a retrospective observational study on 3,000 adult patients with suspected infection, Shapiro et al[20]

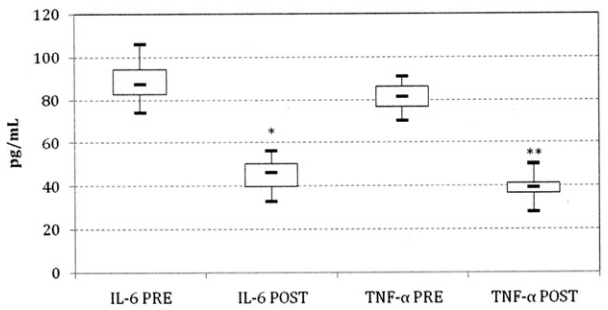

Fig 1. Cytokine values before and after PMX-DHP treatments in 28 transplant recipients. IL = interleukin; TNF = tumor necrosis factor. *$P < .001$.

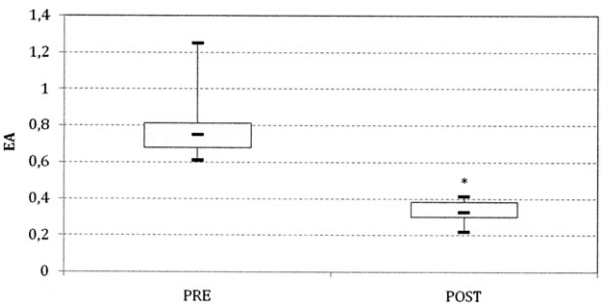

Fig 2. Modification of Endotoxin Activity (EA) with PMX-DHP treatments in 28 transplant recipients. *$P < .001$.

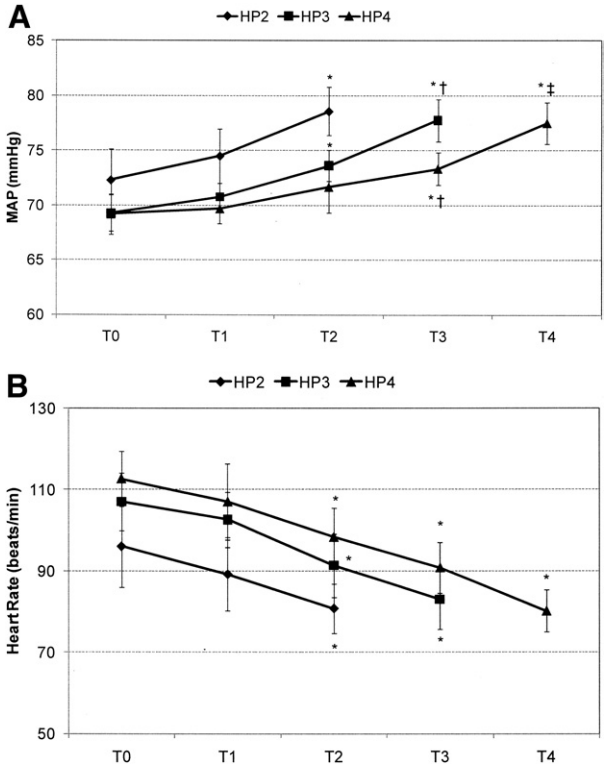

Fig 3. (A) Improvements of mean arterial pressure (mAP) during the PMX-DHP therapy observed for the 3 groups. *Statistical significance vs basal values; †statistical significance vs the second treatment; ‡statistical significance vs the third treatment. (B) Improvements of heart rate (HR) during the PMX-DHP therapy observed for the 3 groups *Statistical significance vs basal values.

reported the best outcomes (mortality 1.3%, odds ratio (OR) 0.8) among patients with SIRS compared with those with severe sepsis (mortality 9.2%, OR 4.0). Early recognition of EA may be the key to determine timing and results of treatment. EAA can help us to determine (after just 40 minutes), the presence of EA in the blood, leading us to PMX-DHP treatment. In 6 patients (4 LT and 2 KT), the first EAA test showed intermediate EA (median EA 0.56). The levels had changed to a median EA of 0.68 at a second

Table 2. Etiologies and Blood Culture Isolates in 21/28 Transplant Recipients*

	Liver Transplant (n = 14)	Kidney Transplant (n = 14)
Pathogen		
Escherichia coli	6	4
Pseudomonas aeruginosa	1	3
Enterococcus faecium	1	3
Klebsiella pneumoniae	—	2
Proteus mirabilis	—	1

*Seven patients showed negative blood cultures.

Table 3. Liver and Kidney Parameters and Endotoxin Activity at 30 Days' Follow-Up

	Liver Transplant (n = 14)	Kidney Transplant (n = 14)
Bilirubin (mg/dL)	>1.12 ± 0.25	—
ALT (IU/L)	69.5 ± 17.6	—
AST (IU/L)	82 ± 26.3	—
γ-GT (IU/L)	49 ± 34.2	—
BUN (mg/dL)	—	65.8 ± 24.3
Creatinine (mg/dL)	—	1.5 ± 0.8
Diuresis (mL/d)	—	2780 ± 450
EA	0.22 [0.17–0.36]	0.27 [0.19–0.32]

Abbreviations as in Table 1.

examination 24 hours later; these patients were shifted to the treatment group. This observation indicated that it is necessary to perform EAA even after a first intermediate result. The "dosing schedule" (number of PMX-DHP treatments) was determined by measuring EA levels before and after each treatment. EA levels were lowered after each treatment. As previously described,[21] we confirmed a higher percentage decrease (~40%) for treatments starting from EA ≤0.7 than from EA >0.9 (~20%), an observation that may be explained by the characteristics of the assay method and the PMX-DHP treatment. There is a nonlinear LPS dose-response curve for EAA. The amount of LPS removed by each PMX-DHP is fixed and determined by the number of available bonding sites of polymyxin-B within each cartridge. Seven patients (25%) showed high levels of EA in the absence of blood cultures positive for gram-negative infection. This probably occurred because kidney transplant recipients in particular show endotoxin persistence or possibly an increase in the circulation despite negative blood cultures owing to the actions of host defense or antibiotics. Furthermore, 6 liver transplant recipients showed translocation from the gastrointestinal tract as the source of endotoxin. Uncontrolled and excessive production of inflammatory cytokines *downstream of* microbial component stimulation appears to play a pivotal role in the development of MOF during sepsis. In this regard, it may be ideal to control the initial phases of the inflammatory cascade by treating sepsis with removal of microbial components (endotoxin) that trigger infectious inflammatory cascades. Our patients presented high cytokine levels with a hyperdynamic state. The antiendotoxin intervention significantly improved hemodynamics and significantly lowered the inflammatory state, as observed by decreased cytokines, WBC, and lactic acid. In this work, the detailed description of the hemodynamic framework of septic patients following an abdominal organ transplantation, ie, liver or kidney, confirmed the ability of PMX-DHP treatments added to standard therapy to improve hemodynamics in endotoxin-driven sepsis. mAP increased and HR decreased significantly after the therapy. EA progressively decreased after each PMX-DHP treatment, indicating deactivation of the immune system as a result of endotoxin removal and neutralization by means of polymyxin-B. Moreover, EA detection during therapy allowed

selection of a subgroup of patients that benefited from a larger number of hemoperfusions (3 or 4 instead of 2) to reach low EA values. Those patients were characterized by a worse hemodynamic framework than other patients at inclusion. The extension of the number of PMX-DHP treatments let them achieve mAP and HR values similar to the other patients at the end of the therapy (EA <0.4).

As reported by Tani et al,[22] cytokines themselves are not removed by PMX-DHP. The hypothesis is that removal of lipopolysaccharides inhibits cell activation and the release of proinflammatory cytokines, thereby controlling the inflammatory response and stabilizing the circulation. Positive changes among these parameters improved hepatic function in liver transplant patients within 5 ± 0.4 days and renal function in kidney transplant patients immediately after the end of the treatment. An improved SOFA score was observed at the end of the treatments with 100% survival of 28 patients at 30 days associated with good graft function. Marshall et al[23] in 2004 showed a significant correlation among EA levels and worsening of clinical and predictive parameters, such as SOFA score, PaO_2/FiO_2 ratio, and WBC. Liver transplant recipients (78.5%) showed higher EA than kidney transplant recipients (32%) within 15 days of transplantation. These data were consistent with those of Linares et al,[24] who performed a study on 67 transplant patients with sepsis, showing a major incidence of early bacteremia among liver (12%) versus kidney (4.8%) transplant recipients. Finally, in accordance with Bert et al,[25] we observed a major incidence of infections by *E coli* among liver (n = 6) versus kidney (n = 4) transplant patients. Because of the technical complexity of liver transplantation, many patients develop postoperative complications resulting from disruptions or defects of the gastrointestinal or biliary barriers. Such barrier defects may facilitate the passage of "low-virulence" *E coli* isolates into the peritoneal space and bloodstream which in a few hours can produce an early infection.

In conclusion, the immune system is partially compromised in transplant recipients owing to their pre-operative clinical status and immunosuppression therapy. Sometimes it is impossible to eradicate the pathogens. Infection represents one of the primary barriers to successful organ transplantation. We sought not to identify risks and variables associated with BSI, but rather early identification of EA or suspected infection among transplant patients. EAA is a diagnostic test to detect early infection or clarify the role of endotoxin translocation, helping to determine the correct timing for an intervention. This retrospective study with a small number of patients provided insights into the early management of endotoxemia. First, EAA showed the ability to identify patients eligible for a targeted therapy. Second, EAA efficiently measured the effects of PMX-DHP treatments, thus aiding in therapeutic dosing. And third, endotoxemia was also detected in cases without culture evidence of a gram-negative infection.

NOTE

Some data of this article were previously published in the following report: Novelli G, Ferretti G, Poli L, et al: Clinical results of treatment of postsurgical endotoxin-mediated sepsis with polymyxin-B direct hemoperfusion. Transplant Proc 42:1021, 2010. Fig 2.

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