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Interpreting the Mechanisms of Continuous Renal Replacement Therapy in Sepsis: The Peak Concentration Hypothesis

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Abstract: Severe sepsis and septic shock are the primary causes of multiple organ dysfunction syndrome (MODS), which is the most frequent cause of death in intensive care unit patients. Many water-soluble mediators with pro- and anti-inflammatory action such as TNF, IL-6, IL-8, and IL-10 play a strategic role in septic syndrome. In intensive care medicine, blocking any one mediator has not led to a measurable outcome improvement in patients with sepsis. CRRT is a continuously acting therapy, which removes in a nonselective way pro- and anti-inflammatory mediators; "the peak concentration hypothesis" is the concept of cutting peaks of soluble mediators through continuous hemofiltration. Furthermore, there is evidence of increased efficacy of high-volume hemofiltration compared to conventional CVVH, and other blood purification techniques that utilize large-pore membranes or sorbent plasmafiltration are conceptually interesting. **Key Words:** Acute renal failure—Multiple organ dysfunction syndrome—Sepsis—Continuous renal replacement therapy—Homeostasis—Peak concentration.

Acute renal failure (ARF) is increasingly seen as part of multiple organ dysfunction syndrome (MODS) in critically ill patients (1,2). MODS is the most frequent cause of death in patients admitted to intensive care units (3). Severe sepsis and septic shock are the primary causes of MODS (4,5) and develop as a result of the host response to infection of Gram-negative and Gram-positive bacteria (6). Infectious sepsis and noninfectious systemic inflammatory response syndrome (SIRS) encompass a complex mosaic of interconnected events. Molecules such as bacterial lipopolysaccharides (LPS), microbial lipopeptides, microbial DNA, peptidoglycan, and lipoteichoic acid trigger interactions with the Toll-like receptors and related molecules (MD-2, MyD88), the principal sensors of the innate immune response (7–9). Stimulus-receptor coupling activates different signal transduction pathways leading to exacerbated generation of cytokines, phospholipase A2-dependent, arachidonic acid-derived platelet-activating factor, leukotrienes, and thromboxanes. At the plasma level, activation of the complement (C3a, C5a, and their desarginated products) and coagulation pathways interacts with the process as products generated in the fluid phase may in turn trigger and sustain cell activation. Other agents play a role in the pathophysiology of sepsis, such as surface-expressed and soluble adhesion molecules, kinins, thrombin, myocardial depressant substance(s), endorphins, and heat shock proteins.

Continuous renal replacement therapies (CRRT) allow extracorporeal treatment in critically ill patients with hypercatabolism and fluid overload (10–12). CRRT have commonly used three types of depurative mechanisms: convection, diffusion, and adsorption by the filtering membrane. In addition to removing excess fluid and waste products in septic patients, the possibility that CRRT may remove bacterial LPS and other mediators introduced the concept of blood purification (13–22).

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In the present article, we will review some recent advances in the pathogenesis of sepsis in view of the possible effects of CRRT and related innovative techniques in restoring normal hemodynamic and immunologic homeostasis. Understanding how the mechanisms of sepsis may be affected by CRRT may open the concept of them being not only supportive but also preventive therapies, that is, preventing tissue and organ damage.

The pathogenesis of sepsis

Under physiological conditions, the biological activity of sepsis-associated mediators is under the control of specific inhibitors that may act at different levels. In sepsis, the homeostatic balance is altered and a profound disturbance of relative production of different mediators may be observed (as reviewed in [23]). On the one hand, the spillover into the circulation of mediators intended to have autocrine or paracrine effects generates systemic effects including endothelial damage (24), procoagulant, fibrinolytic, complement activities, hemodynamic shock, and vasoparalysis (25–31). On the other hand, monocytes present a profound inability to produce cytokines when they are challenged with different stimuli ex vivo (32,33).

The pathogenesis of sepsis was initially described as an overproduction of pro-inflammatory factors in the host. The concept was established on the basis of several studies. The injection of LPS into the experimental animal and healthy human subjects reproduces the initial phase of bacterial infection (34). In human subjects, LPS alters capillary integrity and affects the cardiovascular system (34), causes production of cytokines (26,27,35,36), and activates the coagulation-fibrinolytic pathways (37). Peak concentrations of IL-1, TNF, IL-6, and IL-8 occur within 2–3 hr of LPS infusion (26,27). Recent studies on knockout mice have shown that ICAM-1 mutant mice are resistant to the lethal outcome of endotoxin-induced pneumonitis (38).

What is the relevance of circulating cytokines? The presence or absence of detectable levels of cytokines within biological fluids reflects a rather complex balance between enhancing and inhibitory signals acting on producer cells, between production and catabolism, and between their binding to the target cells and the modulation of their receptors on the cell surface (23). Furthermore, their presence does not necessarily parallel their activity and a possible interplay between a given cytokine and its relative inhibitor (if known) should be considered (23). Cavaillon et al. coined the expression of circulating cytokines as being "the tip of the iceberg," implying that neither their presence nor their absence can reflect the complex interplay at the tissue level (23). Despite the fact that their peak concentrations may reflect an exacerbated production, these levels do not necessarily stand for enhanced bioactivity.

The concept of sepsis as a simply pro-inflammatory event has been subsequently challenged (32,33, 39,40).

In sepsis and SIRS, cell-associated cytokines in peripheral blood mononuclear cells (PBMC) are decreased, as is the capacity of these cells to produce several cytokines such as TNFα, IL-1α, IL-1β, IL-6, IL-10, and IL-12 (41,43), but not IL-1 receptor antagonist (44). Hyporesponsiveness is not only present in monocytes but occurs in whole blood (45), and is associated with increased plasma levels of IL-10 and prostaglandin E2, which are potent inhibitors of the production of pro-inflammatory cytokines (42,43,46). To describe the excessive anti-inflammatory counterpart of SIRS, Bone (47) coined the acronym CARS for "compensated anti-inflammatory response syndrome." Terms such as monocyte deactivation, immunoparalysis, or more simply, cell hyporesponsiveness, all indicate the inability of cells to respond ex vivo to LPS stimuli due to overproduction of anti-inflammatory cytokines. Adib-Conquy et al. (48) demonstrated that, upon LPS activation, PBMC of patients with SIRS show patterns of NF-κB expression that resemble those reported during LPS tolerance: global downregulation of NF-κB in survivors of sepsis and trauma patients and the presence of large amounts of the inactive homodimer in the nonsurvivors of sepsis. In the clinical setting of severe sepsis, Bone proposed that at a given time, SIRS or CARS predominates in patients, inducing shock or immune depression. However, a large amount of evidence now suggests that in patients with both infectious and noninfectious sepsis, SIRS and CARS may coexist, but in different compartments (Fig. 1). Based on these concepts, Cavaillon et al. (40) recently proposed that SIRS predominates within the inflamed tissues, whereas blood leukocytes show hyporeactivity. A restrained inflammatory response within the blood stream should avoid the endothelial activation leading to overexpression of adhesion molecules, and adherence and degranulation of leukocytes. It would avoid fatal clotting and organ failure. Nevertheless, peak concentrations of either LPS or different cytokines have been variably reported, and in some reports their levels, alone or in combination, have been even considered as useful markers in severity scores (49). Following the intravenous injection of LPS, a lag phase occurs and is followed after 1 hr by a steep increase in TNF, reaching a maximum at

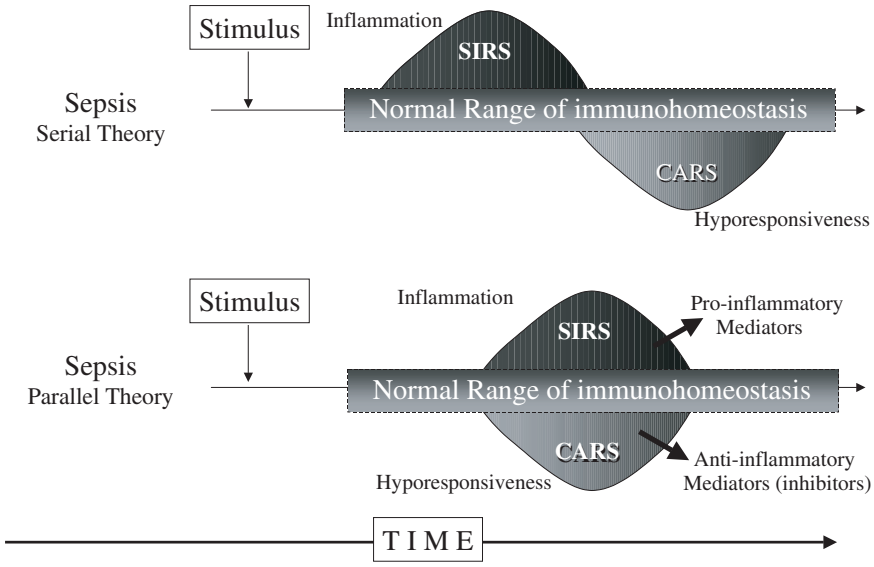

FIG. 1. The serial or sequential theory of sepsis and the parallel theory is shown. At the start, the sequence of events begins temporally, with a stimulus such as endotoxin dissemination; a systemic inflammatory response syndrome follows, with a spillover into the circulation of several pro-inflammatory mediators. Subsequently, a potent inhibition of the inflammatory process and a consequent cell hyporesponsiveness occurs. In the parallel theory, the processes occur simultaneously, and a parallel synthesis of pro- and anti-inflammatory mediators coexists in different districts of the body.

1.5 hr. TNF plasma concentrations are sharply reduced at 3 hr when both IL-6 and IL-8 sharply augment. Cytokine production is blunted by cyclooxygenase inhibition. Studies by Suffredini (34), Parrillo et al. (50), and van Deventer et al. (27) on human subjects infused with LPS characterized the initial inflammatory (TNF, IL-6, IL-8) and hemostatic (TAT, PAP, tissue plasminogen activator) responses out to 6–8 hr. Taylor et al. (30) studied the receptor and oxidative enzymatic responses of phagocytes in the human model of endotoxemia and correlated them with the response of molecular markers of hemostatic and inflammatory system activation and endothelial injury. These authors established that the compensated response to LPS consists of two stages: an immediate symptomatic inflammatory stage followed by an asymptomatic stage that is characterized by a recurrence of hemostatic activity, appearance of complement activation products complexed to C-reactive proteins, and evidence of endothelial injury (30). More in particular, these authors showed the occurrence of a two-step response: an early response characterized by the degranulation of neutrophils (as detected by elevation of elastase-α1/anti-trypsin complexes) coinciding with peak concentrations of TNF, IL-6, and regulatory responses such as IL-10 and activated protein C; and a late response characterized by hemostatic activity as reflected by the appearance of a second large peak of soluble fibrin and a second and greater decrease in circulating factor VIIa concentration coupled with the appearance of increased concentrations of plasma tissue factor antigen. This second phase was also characterized by sustained, markedly elevated levels of C-reactive protein (CRP) and CRP-bound activated complement components (30). A crucial aspect of these studies is that peak concentrations of LPS and of several cytokines appear at different time intervals. The intravenous injection of pro-inflammatory cytokines (TNF, IL-1) reproduces very closely the pattern of hemodynamic and intravascular changes induced by the injection of LPS (30).

In the human setting of sepsis, the finding of elevated levels of different pro- and anti-inflammatory cytokines has been variable, suggesting the possibility that their production may occur at a different time from sampling, their production in a given patient is compartmentalized, or both these two possibilities. We suggest that the events associated with sepsis/SIRS may happen in sequence (the sequential or serial sepsis theory) whereby pro- and anti-inflammatory mediators are alternately produced in high- or low-generation periods, thus ensuing in SIRS and CARS. On the other hand, the events associated with sepsis/SIRS may occur simultaneously (the parallel sepsis theory), in that SIRS and CARS may coexist in different districts or systems (Fig. 2). In the sequential sepsis theory, temporary prevalence of SIRS should probably be treated with high-dose steroids, assuming that a timely intervention is possible thanks to an early and accurate biological monitoring. Otherwise, the therapy may overlap with the next coming period of CARS and may even favor bacterial colonization and infection dissemination. Indeed, in this period, a protective antimicrobial therapy or even an immunostimulatory therapy should be administered. The time of intervention becomes crucial in order to prescribe the right therapy for the right disorder. Alternatively,

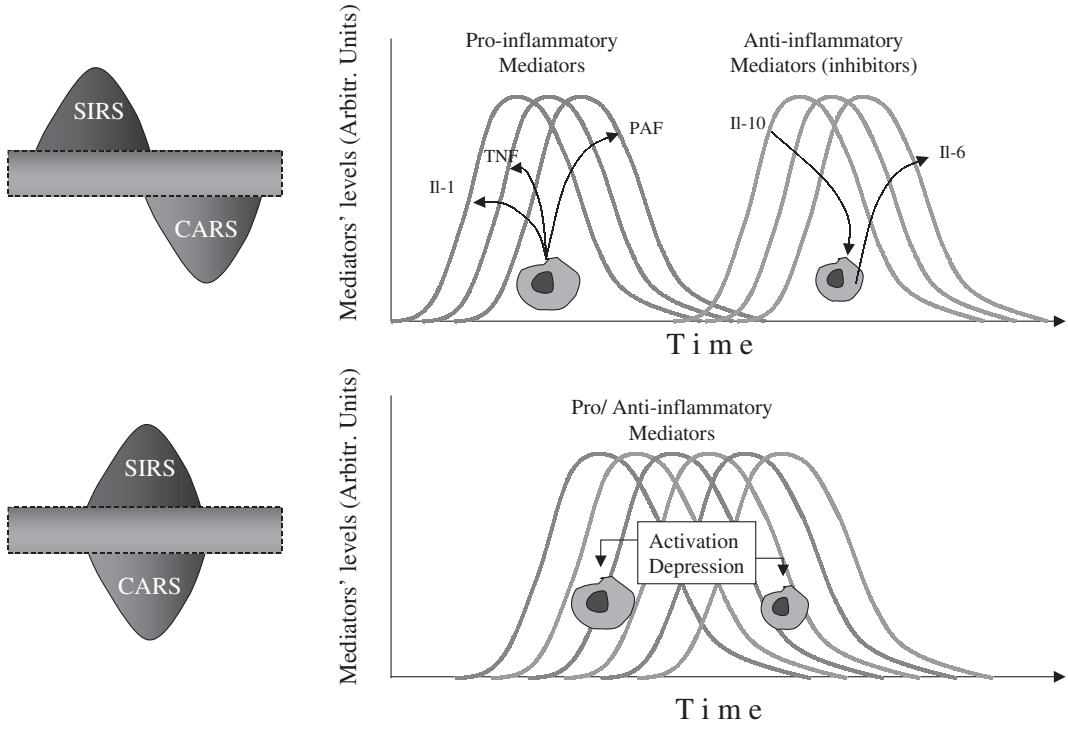

FIG. 2. In the sequential theory, peaks of pro-inflammatory mediators are followed by peaks of anti-inflammatory mediators. In the parallel theory, a mixture of pro- and anti-inflammatory mediators coexists. The sequential theory leads to the conclusion that, if pro- and anti-inflammatory activities could be monitored, specific therapies could be targeted for selected actions in different moment of the time course of the syndrome. In the parallel theory, one therapy or another may be effective on one side but deleterious on the other side.

if the parallel sepsis theory is considered, none of the two therapies may result, and a question remains as to the soundest therapeutic option.

In intensive care medicine, blocking one mediator has not led to measurable outcome improvement in patients with sepsis (51). Possibly more rigidly defined subgroups would gain by TNF-antagonizing treatments (52). On the other hand, it has been shown that antagonizing a cytokine may lead to deleterious consequences leading to substantially higher mortality (53). A low-level TNF response seems to be necessary for the host defense to infection (54,55), and high levels seemingly need to be modulated by an anti-inflammatory feedback. In sepsis, however, impaired regulation may cause an excessive anti-inflammatory response, which generates monocyte "immunoparalysis" and exposes the host to further infections. Both processes (inflammation and anti-inflammation) are designed to act in response to specific stimuli in a well-balanced fashion defined as immunohomeostasis.

Furthermore, the time point in the septic process of therapeutic intervention seems to be crucial. As the network acts like a cascade, early intervention would seem most beneficial. However, sepsis does not fit a one-hit model but shows complex and multiple rises in mediator levels that change over time. Neither single-mediator-directed nor one-time interventions therefore seem appropriate. One of the major criticisms attributed to continuous blood purification treatments in sepsis—its lack of specificity—could turn out to be a major strength. Unspecific removal of soluble mediators, be they pro- or anti-inflammatory, without completely eliminating their effect, may be the most logical and adequate approach to a complex and long-running process such as sepsis (Fig. 3). The concept of cutting peaks of soluble mediators, for example through continuous hemofiltration, is a paradigm we call "the peak concentration hypothesis" (56).

Continuous Renal Replacement Therapy

For several years, the issue of the capability of hemofiltration to remove inflammatory mediators has remained controversial. Numerous ex vivo as well as animal and human studies (reviewed in [70]) have shown that synthetic filters in common use in hemofiltration can extract nearly every substance involved in sepsis to a certain degree. Prominent examples are complement factors (57,58), TNF, IL-1,

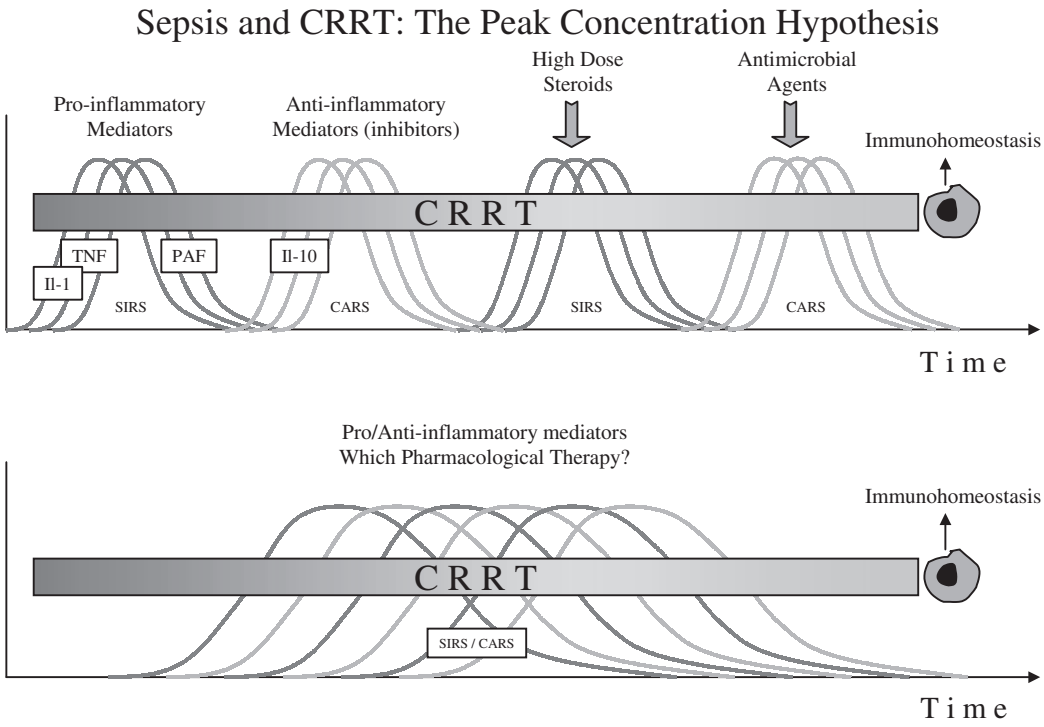

FIG. 3. In both theories (sequential and parallel), the concept introduced by the peak concentration hypothesis suggests that a nonselective control of the peaks of inflammation and immunoparalysis may contribute to bring the patient to a lesser degree of imbalance and close to the self-defenses induced by a nearly normal immunohomeostasis.

IL-6 (58–61), IL-8 (62), and PAF (63,64). Regarding plasma cytokine levels, their decrease appeared, nevertheless, to a minor degree. Other studies could not show any influence on cytokine plasma levels by CRRT (65–67). On the other hand, significant clinical benefits in terms of hemodynamic improvement have been achieved even without measurable decreases in cytokine plasma levels (68).

Obviously, the removal of substances different from the measured cytokines was responsible for the achieved effect. Bioactive substances including some of the measured cytokines were removed, causing the observed beneficial effect. In a recent study, Mariano et al. (62) evaluated the priming activity of sera from septic patients on polymorphonuclear neutrophils. This activity was related to ultrafiltrable mediators among which IL-8 seemed to be one of the most relevant ones. The results of the latter study further implicate the fact that several mediators may act in concert in altering the functional responses of the circulating leukocytes. When the response to sepsis is viewed in a network perspective, absolute values would be less relevant than relative ones within an array of interdependent mediators, as even small decreases could induce major balance changes. This makes measurement of cytokine plasma levels debatable while more local or tissue levels should be measured. These issues are extremely controversial and do not permit a definitive solution in favor or against the use of CRRT as a therapy for sepsis. In this context, a further step in clarifying the immunologic impact of CRRT has been taken by measuring a more downstream event integrating several cytokine influences: monocyte responsiveness (68,69).

In spite of some encouraging results as mentioned, the extent of achievable clinical benefit with conventional CRRT (using conventional filters and flow rates) in sepsis has generally been disappointing (70). Consequently, it was sought to improve the efficiency of the methodology regarding removal of soluble mediators of sepsis by increasing the amount of plasma water exchange, that is, increasing ultrafiltration rates.

Animal studies provided great support to this concept. Starting in the early 1990s, several studies using different septic animal models examined the effect of high ultrafiltration rates (up to 300 ml/kg/hr) on physiological parameters and outcome.

In a landmark study, a porcine model of septic shock induced by endotoxin infusion was investigated (71). The animals developed profound arterial hypotension and a decrease in cardiac output, stroke

volume, and right ventricular stroke work index. By high-volume hemofiltration (HVHF, 6 L/hr), right ventricular function, blood pressure, and cardiac output showed a remarkable improvement compared to control and sham-filtered animals (71,72). The same group extended their findings in the same model by i.v. adminstration of ultrafiltrate from LPS-infused animals into healthy animals. The latter ones developed hemodynamic features similar to septic shock while animals infused with ultrafiltrate from healthy animals showed a moderate blood pressure rise (73). In a further study by the same group, a bowel ischemia-reperfusion model in pigs was investigated. HVHF started before clamping of the superior mesenteric artery significantly diminished bowel damage and prevented hemodynamic deterioration (74).

These studies established that a convection-based treatment can remove substances with hemodynamic effects resembling septic shock when sufficiently high ultrafiltration rates are applied.

Several studies confirmed and refined these results. In three of them (75–77), the correlation of survival with ultrafiltration rate was specifically examined. A direct correlation could be demonstrated. Significant improvements in cardiac function, systemic and pulmonary vascular resistance, and hepatic perfusion (75) were found. Another study in lambs showed significant improvements in lung function (78). Only a minority of studies identified reduced mediator plasma levels (77,79).

A very recent study in pigs made septic by induced pancreatitis compared low-volume CVVH with HVHF of 100 ml/kg/hr. In the same study, the influence of frequent filter changes on survival and changes in TNF levels as well as monocyte and polymorphonuclear neutrophil function was analyzed (77). Early filter change allowed delineating the effect of cytokine removal by adsorption on the filter, as membrane capacity was saturated after a few hours. By changing filters, adsorption was continued to a certain extent. In this model, a hyperdynamic septic state is induced through an intervention that approximates underlying conditions encountered in human sepsis. Additionally, the intervention was started late to simulate real clinical conditions. Hemofiltration was commenced when the animals developed the clinical picture of hyperdynamic septic shock. HVHF was superior in all mentioned endpoints. Of relevance, increasing ultrafiltration had more effect than frequency of filter change (77).

Closer to human sepsis studies has been the finding that the ultrafiltration dose is correlated to outcome in critically ill patients with ARF. In a large randomized, controlled study including 425 patients, an ultrafiltration dose of 35 ml/kg/hr increased survival rate from 41% to 57% compared to a dose of 20 ml/kg/hr (80). Eleven percent to 14% (per randomization group) of the patients had sepsis. In these subgroups, there was a trend of a direct correlation between treatment dose and survival even above 35 ml/kg/hr, in contrast to the whole group where a survival plateau was reached.

This lends support to the concept of a "sepsis dose" of hemofiltration in septic patients contrasting to a "renal dose" in critically ill patients without systemic inflammation, the former being probably distinctly higher (without proven upper limit). Of note, there was no increase in adverse effects even with the highest ultrafiltration dose.

Over the last years, several human studies examined the clinical effects of HVHF. In 20 children undergoing cardiac surgery, zero-balanced HVHF was administered with UF-rates equivalent to 7–9 l/hr for a 70 kg adult (81). Endpoints correlating to the cardiopulmonary bypass-associated delayed inflammatory response were examined. There was a significant reduction in postoperative blood loss, time to extubation, and improvement in the arterial-alveolar oxygen gradient.

In a prospective cohort analysis in 306 critically ill patients with varying underlying diseases, a mean ultrafiltration rate of 3.8 l/hr was applied (82). Observed survival rates were significantly higher in the treated population compared to predicted survival by three well-validated scores.

In another trial in 11 septic patients with shock and MODS, a randomized crossover design of 6 L/hr versus 1 L/hr ultrafiltration was applied (83). The HVHF group displayed significantly greater reduction in vasopressor requirements. Both treatment groups showed a decrease in C3a and C5a plasma levels, which was significantly greater in the HVHF group.

Impressive clinical results were obtained in an evaluation of short-term HVHF in 20 points in catecholamine-refractory septic shock (84) comprising a patient cohort with very poor expected survival. A control group was not defined. Only one 4 hr session of HVHF removing 35 L of ultrafiltrate replaced by bicarbonate-containing fluid was applied as soon as mean blood pressure could not be stabilized above 70 mm Hg with dopamine, norepinephrine, and epinephrine after appropriate volume resuscitation. HVHF was followed by conventional CVVH. Endpoints were the increase in cardiac index, mixed venous oxygen saturation, and arterial pH, and decrease in norepinephrine requirements. Eleven patients reached all predefined endpoints and

showed impressively good survival (9 of 11) at 28 days. Nine patients did not reach all endpoints and had a 100% mortality rate. Apart from responding to HVHF, only time from ICU admission to the start of HVHF and body weight were survival-associated factors in the analysis. Patients with higher body weight did worse, possibly because they received a smaller ultrafiltration dose per body weight as speculated by the authors.

These trials still need cautious interpretation with respect to their limited design, but they certainly deliver sound evidence of feasibility and efficacy to set the stage for a large-scale trial on HVHF in sepsis.

Other approaches to achieving higher mediator clearance in sepsis have been sought. Apart from increasing ultrafiltration rates, higher removal rates of middle molecular weight molecules could be achieved by enlarging pore size of membranes. Animal data (85,86) as well as preliminary clinical data (87) demonstrate feasibility and probable superior removal rates of select cytokines using more open membranes. A study of 30 patients with severe sepsis using continuous plasmafiltration for 34 hr (88) found attenuation of the acute phase response and a trend toward clinical benefit although not significant (fewer failing organs).

A further step to increase mediator removal has been achieved with plasma filtration coupled with adsorption and followed by dialysis or filtration (CPFA) (89).

Using an experimental model of acute endotoxemia in the rabbit, Tetta et al. (90) showed that nonselective adsorption of cytokines and other pro-inflammatory mediators known to be produced in excess during sepsis could improve survival. The simultaneous removal of peak concentrations of TNF and PAF could also possibly prevent the formation of other biologically active substances, such as prostaglandins/leukotrienes, other cytokines, molecules up- or down-regulating membrane receptors, selectins, and adhesion molecules, thus amplifying and perpetuating the immunological disorders. In severe sepsis and septic shock, many new therapeutic strategies have targeted single molecules, but so far they have failed to reduce mortality rate (51). The results that stemmed from these studies strongly supported the concept that a relationship would link the simultaneous removal of different mediators to the improved survival. As stated above, the unselective, simultaneous removal of different mediators may not necessarily implicate that the goal of blood purification be achieved only on the basis of a significant reduction of the circulating cytokines. Much more effective would be the impact on the functional responses of cells implicated in the pathogenesis of sepsis (Fig. 3). In a very recent study, we tested the hypothesis that nonselective removal of mediators could improve hemodynamics and restore leukocyte responsiveness in patients with septic shock. The aim of this study was in response to the open question as to how CRRT may have a beneficial effect on the hemodynamic and immune response associated with severe sepsis (69). Immunomodulating substances (with molecular weight in the range of 5–50 kilodaltons) may be eliminated by diffusion, adsorption or convection depending on the rather variable cut-off of highly permeable membranes (range from 30 to 40 kilodaltons) (68). Adsorption is only a transient phenomenon. Thus, the effect of CRRT could be limited because of the low convective clearance of many soluble mediators. The use of a plasmafiltration membrane coupled with an adsorption device in CPFA could enhance unselective removal and improve hemodynamic stability compared to CRRT. CPFA was associated with the restoration of stable hemodynamics, particularly due to an increase in MAP. This change in blood pressure led to a significant reduction in norepinephrine requirement. CPFA also restored in vitro leukocyte responsiveness to LPS. The magnitude of this effect was significantly greater with CPFA than with CVVHDF. Plasma levels of TNFα and IL-10, however, were not significantly changed during CPFA or CVVHDF, but they did not display concentration peaks (69).

Patients' leukocyte responsiveness was evaluated by measuring spontaneous and LPS-stimulated production of TNFα. We used the whole-blood cytokine assay, as it measures cytokine production in the presence of a wide range of modulating factors (e.g., soluble receptors, natural inhibitors, proteases, etc.).

The spontaneous ex vivo production of TNFα by patients' whole blood was normal in all patients at the start but was increased at the end of treatment with CPFA. The spontaneous ex vivo production of TNFα was also further increased by passage through the hemodiafilter. These changes in ex vivo cell responsiveness could be due to a less biocompatible circuit, exposure to cytokine-inducing substances, removal of a dialyzable suppressive "uremic toxin," or removal of other inhibitors of cell responsiveness. Diffusion of suppressive uremic toxins may be important in acutely uremic patients. In agreement with this contention, a study of 12 critically ill patients with acute renal failure comparing low-volume CVVH (1,500 ml/hr) with a diffusive technique was performed in a nonrandomized, comparative fashion (91). High-flux bicarbonate dialysis amounting to 4,200 ml/hr was used and the

effect on monocyte responsiveness (ex vivo endotoxin-stimulated TNF production) was studied (91). Both techniques resulted in early improvement but only the diffusive technique displayed persistent effects. Ultrafiltrate contained monocyte-suppressive activity only with high-flux dialysis (91).

In their study, Ronco et al. (69) evaluated LPS-stimulated TNFα production in vitro. The latter was markedly inhibited at the start of treatment in all patients. An 8- to 10-fold increase in LPS-stimulated TNFα production was observed after 10 hr of CPFA at both sites 2 and 3, suggesting that this effect was not affected by diffusion/convection. Only a 5-fold increase could be seen in LPS-stimulated TNFα production after CVVHDF. Finally, the in vitro assessment of the effect of septic plasma on TNFα production by normal inflammatory cells showed that, at the start of treatment, LPS-stimulated TNFα production was suppressed by plasma obtained before and after the resin. Although such suppression was decreased by passage through the resin, it was not fully eliminated by it. We speculated that the resin was not able to completely adsorb suppressive soluble factors in a biologically significant amount in a single pass. However, after 10 hr of CPFA, the inhibitory effect of septic plasma was markedly attenuated. The ability of CPFA to restore immune cell responsiveness may be clinically beneficial.

Conclusion

Vast arrays of mostly water-soluble mediators play a strategic role in the septic syndrome. At variance with targeting single mediators, therapeutic intervention finalized at the nonselective removal of pro- and anti-inflammatory mediators seems a rational concept. A further advantage may be constituted by a continuously acting therapy, as in the case of CRRT. Therefore, sequentially appearing peaks of systemic mediator overflow could be curbed, as well as persistently high plasma levels reduced. This process is proposed as the underlying biological rationale for a series of innovative therapies in sepsis. The whole story of antagonizing pro- and anti-inflammatory processes by reducing the relative excess of active substances goes under the term of peak concentration hypothesis.

Recent animal and human trials have delivered much support to this concept. It has been conclusively shown that treatment dose in CRRT is a major factor concerning survival in ARF in the critically ill patient. There is accumulating evidence of increased efficacy of high-volume hemofiltration compared to conventional CVVH in terms of laboratory and clinical improvement, including survival. Machines to perform HVHF safely are available on the market.

Yet the evidence still is not strong enough to recommend HVHF outside clinical studies, taking into account possible adverse effects of the technique. A large-scale clinical trial is urgently needed to resolve the issue.

Other blood purification techniques using large-pore membranes or plasma filtration with adsorbent perfusion are in the early stages of clinical testing. They are conceptually promising and possibly constitute an important refinement.

REFERENCES

1. Cosentino F, Chaff C, Piedmonte M. Risk factors influencing survival in ICU acute renal failure. *Nephrol Dial Transplant* 1994;9(suppl 4):179–82.
2. Liano G, Pascual J. Acute renal failure. Madrid Acute Renal Failure Study Group. *Lancet* 1996;347:479.
3. Bellomo R, Ronco C. Indications and criteria for initiating renal replacement therapy in the intensive care unit. *Kidney Int* 1998;53(suppl 66):S106–9.
4. Bone RC, Balk RA, Cerra FB, et al. Definitions for sepsis and organ failure and guidelines for the use of innovative therapies in sepsis. *Chest* 1992;101:1644–55.
5. Camussi G, Montrucchio G, Diminioni L, Dionigi R. Septic shock: the unravelling of molecular mechanism. *Nephrol Dial Transplant* 1995;10:1808–13.
6. Glauser MP, Zanetti G, Baumgartner JD, Cohen J. Septic shock: pathogenesis. *Lancet* 1991;338:732–6.
7. Medzhitov R, Preston-Hurlburt P, Janeway CA Jr. A human homologue of the *Drosophila* Toll protein signals activation of adaptive immunity. *Nature* 1997;388:394–7.
8. Shimazu R, Akashi S, Ogata H, et al. MD-2, a molecule that confers lipopolysaccharide responsiveness on Toll-like receptor 4. *J Exp Med* 1999;189:1777–82.
9. Medzhitov R, Preston-Hurlburt P, Kopp E, et al. My88 is an adaptor protein in the hToll/IL-1 receptor family signaling pathways. *Mol Cell* 1998;2:253–8.
10. Von Bommel EFH, Bouvy ND, So KL, et al. Acute dialytic support for the critically ill: intermittent hemodialysis versus continuous arteriovenous hemodiafiltration. *Am J Nephrol* 1995;15:192–200.
11. Bellomo R, Mehta R. Acute renal replacement in the intensive care unit: now and tomorrow. *New Horiz* 1995;3:760–7.
12. Canaud B, Mion C. Extracorporeal treatment of acute renal failure: methods, indications, quantified and personalized therapeutic approach. *Adv Nephrol* 1995;24:271–81.
13. Silvester W, Bellomo R, Ronco C. Continuous versus intermittent renal replacement therapy in the critically ill. In: C. Ronco, R. Bellomo, eds. *Critical Care Nephrology*. Dordrecht: Kluwer Academic Publishers, 1998;1225–38.
14. Canaud B, Mion C. Extracorporeal treatment of acute renal failure: methods, indications, quantified and personalized therapeutic approach. *Adv Nephrol* 1995;24:271–81.
15. Bellomo R, Tipping P, Boyce N. Continuous veno-venous hemofiltration with dialysis cytokines from the circulation of septic patients. *Crit Care Med* 1993;21:522–6.
16. Schetz M, Ferdinande P, Van der Berghe G, et al. Removal of pro-inflammatory cytokines with renal replacement therapy: sense or nonsense? *Intensive Care Med* 1995;21:169–76.
17. van Bommel EF, Hesse CJ, Jutte NHPM, et al. Cytokine kinetics (TNF-alpha, IL-1 beta, IL-6) during continuous hemofiltration: a laboratory and clinical study. *Contrib Nephrol* 1995;116:62–75.

18. Bellomo R, Tipping P, Boyce N. Interleukin-6 and interleukin-8 extraction during continuous venovenous-hemodiafiltration in septic acute renal failure. *Ren Fail* 1995;17:457–66.
19. Millar AB, Armstrong L, van der Linden J, et al. Cytokine production and hemofiltration in children undergoing cardiopulmonary bypass. *Ann Thor Surg* 1993;56:1499–502.
20. Journois D, Pouard P, Greely WJ, et al. Hemofiltration during cardiopulmonary bypass in pediatric cardiac surgery. *Anesthesiology* 1994;81:1181–9.
21. Goldfarb S, Golper TA. Proinflammatory cytokines and hemofiltration membranes. *J Am Soc Nephrol* 1994;5:228–32.
22. Ronco C, Tetta C, Lupi A, et al. Removal for platelet-activating factor in experimental continuous arteriovenous hemofiltration. *Crit Care Med* 1995;23:99–107.
23. Cavaillon JM, Munoz C, Fitting C, Misset B, Carlet J. Circulating cytokines: the tip of the iceberg? *Circ Shock* 1992;38:145–52.
24. Reidy MA, Bowyer DE. Scanning electron microscopy: morphology of aortic endothelium following injury by endotoxin and during subsequent repair. *Atherosclerosis* 1997;26: 319–28.
25. Tellez Gil LY, Rosello AM, Torres AC, Moreno RL, Ferròn-Orihuela JA. Modulation of soluble phases of endothelial/leukocyte adhesion molecule 1, intercellular adhesion molecule 1, and vascular cell adhesion molecule 1 with interleukin 1β after experimental endotoxic challenge. *Crit Care Med* 2001;29:776–81.
26. Dinarello CA, Cannon JC, Wolff SM, et al. Tumor necrosis factor (cachectin) is an endogenous pyrogen and induces production of interleukin 1. *J Exp Med* 1986;163:1433–40.
27. van Deventer SJH, Buller HR, ten Cate JW, Aarden LA, Hack CE, Sturk A. Experimental endotoxemia in humans: analysis of cytokine release and coagulation, fibrinolytic and complement pathways. *Blood* 1990;76:2520–6.
28. Rosenberg RD, Aird WC. Vascular-bed specific hemostasis and hypercoagulable states. *N Engl J Med* 1999;340:1555–64.
29. Esmon CT. The protein C pathway. *Crit Care Med* 2000; 28:S44–8.
30. Taylor FB, Haddad PA, Hack E, et al. Two-stage response to endotoxin infusion into normal human subjects: correlation of blood phagocyte luminescence with clinical and laboratory markers of the inflammatory, hemostatic response. *Crit Care Med* 2001;29:326–34.
31. Faust SN, Levin M, Harrison OB, et al. Dysfunction of endothelial protein C activation in severe meningococcal sepsis. *N Engl J Med* 2001;345:408–16.
32. Pinsky MR. Sepsis: a pro- and anti-inflammatory disequilibrium syndrome. *Contrib Nephrol* 2001;132:354–66.
33. Cavaillon JM, Adib-Conquy M, Cloez-Tayarani I, Fitting C. Immunodepression in sepsis and SIRS assessed by ex vivo cytokine production is not a generalized phenomenon: a review. *J Endotoxin Res* 2001;7:85–93.
34. Suffredini AF, Fromm RE, Parker MM, et al. The cardiovascular response of normal humans to the administration of endotoxin. *N Engl J Med* 1989;321:280–7.
35. Matrich GD, Danner RL, Ceska M, et al. Detection of interleukin 8 and tumor necrosis factor in normal humans after intravenous endotoxin: the effect of antiinflammatory agents. *J Exp Med* 1991;173:1021–4.
36. Michie HR, Manogue KR, Spriggs DR, et al. Detection of circulating tumor necrosis factor after endotoxin administration. *N Engl J Med* 1988;318:1481–6.
37. Suffredini AF, Harpel PC, Parrillo JE. Promotion and subsequent inhibition of plasminogen activator after administration of intravenous endotoxin to normal subjects. *N Engl J Med* 1989;320:1165–72.
38. Kumasaka T, Quinlan W, Doyle N, et al. Role of the intercellular adhesion molecule (ICAM-1) in endotoxin-induced pneumonitis using ICAM-1 anti-sense oligonucleotides, anti-ICAM-1 monoclonal antibodies and ICAM-1 mutant mice. *J Clin Invest* 1996;97:2362–9.
39. Volk HD, Reinke P, Krausch D, et al. Monocyte deactivation: rationale for a new therapeutic strategy in sepsis. *Intensive Care Med* 1996;22(suppl 4):S474–81.
40. Cavaillon JM, Adib-Conquy M, Cloez-Tayarani I, Fitting C. Immunosuppression in sepsis and SIRS assessed by ex-vivo cytokine production is not a generalized phenomenon: a review. *J Endotoxin Res* 2001;7:85–93.
41. Munoz C, Carlet J, Fitting C, Misset B, Bleriot JP, Cavaillon JM. Dysregulation of in vitro cytokine production by monocytes during sepsis. *J Clin Invest* 1991;88:1747–54.
42. Randow F, Syrbe V, Meisel C, et al. Mechanism of endotoxin desensitization: involvement of interleukin-10 and transforming growth factor. *J Exp Med* 1995;181:1887–92.
43. Brandtzaeg P, Osnes L, Ostebo R, et al. Net inflammatory capacity of human septic shock plasma evaluated by a monocyte-based target cell assay: identification of interleukin-10 as a major functional deactivator of human monocytes. *J Exp Med* 1996;184:51–60.
44. Marie C, Muret J, Fitting C, et al. Interleukin-1 receptor antagonist production during infectious and noninfectious systemic inflammatory response syndrome. *Crit Care Med* 2000;28: 2277–82.
45. Granowitz EV, Porat R, Mier JW, et al. Intravenous endotoxin suppresses the cytokine response of peripheral blood mononuclear cells in healthy humans. *J Immunol* 1993;151: 1637–45.
46. Shapiro L, Panayotatos N, Meydani SN, Wu D, Dinarello CA. Ciliary neurotrophic factor combined with solubile receptor inhibits synthesis of proinflammatory cytokines and prostaglandin-E2 in vitro. *Exp Cell Res* 1994;215:51–6.
47. Bone RC, Grodzin CJ, Balk RA. Sepsis: a new hypothesis for pathogenesis of the disease process. *Chest* 1997;112:235–43.
48. Adib-Conquy M, Asehnoune K, Moine P, Cavaillon JM. Long-term-impaired expression of nuclear factor-kappa B and I kappa B alpha in peripheral blood mononuclear cells of trauma patients. *J Leukoc Biol* 2001;70:30–8.
49. Riche FC, Cholley BP, Laisne MJ, et al. Inflammatory cytokines, C reactive protein, and procalcitonin as early predictors of necrosis infections in acute necrotizing pancreatitis. *Surgery* 2003;133:257–62.
50. Parrillo JE, Burch C, Stelhamer JH, et al. A circulating myocardial depressant substance in humans with septic shock: septic shock patients with a reduced ejection fraction have a circulating factor that depresses in vitro myocardial cell performance. *J Clin Invest* 1985;76:1539–53.
51. Marshall JC. Such stuff as dreams are made on: mediator-directed therapy sepsis. *Nat Rev Drug Discov* 2003;2:391–405.
52. Abraham E, Glauser MP, Butler T, et al. p55 tumor necrosis factor receptor fusion protein in the treatment of patients with severe sepsis and septic shock. A randomized controlled multicenter trial. Ro 45-2081 study group. *JAMA* 1997;277: 1531–8.
53. Fisher CJ, Agosti JM, Opal SM. Treatment of septic shock with the tumor necrosis factor receptor: Fc fusion protein. *New Engl J Med* 1996;334:1697–702.
54. Echtenacher B, Falk W, Mannel D, Krammer PH. Requirement of endogenous tumour necrosis factor/cachectin for recovery from experimental peritonitis. *J Immunol* 1990;145: 3762–6.
55. van der Meer JWM. The effects of recombinant interleukin-1 and recombinant tumor necrosis factor on non-specific resistance to infection. *Biotherapy* 1988;1:19–25.
56. Ronco C, Ricci Z, Bellomo R. Importance of increased ultrafiltration volume and impact on mortality: sepsis and cytokine story and the role of continuous veno-venous hemofiltration. *Curr Opin Nephrol Hypertens* 2001;10:755–61.
57. Hoffmann JN, Hartl WH, Deppisch R, et al. Hemofiltration in human sepsis: evidence for elimination of immunomodulatory substances. *Kidney Int* 1995;48:1563–70.
58. Gasche Y, Pascual M, Suter PM, Favre H, Chevrolet JC, Schifferli JA. Complement depletion during haemofiltration

with polyacilonitrile membranes. *Nephrol Dial Transplant* 1996;11:117–9.
59. Kellum JA, Johnson JP, Kramer D, Palevsky P, Brady JJ, Pinsky MR. Diffusive vs. convective therapy: effects on mediators of inflammation in patients with severe systemic inflammatory response syndrome. *Crit Care Med* 1998;26:1995–2000.
60. Braun N, Giolai M, Rosenfeld S, et al. Clearance of interleukin-6 during continuous veno-venous hemofiltration in patients with septic shock. A prospective, controlled clinical study. *J Am Soc Nephrol* 1993;4:336.
61. Kellum JA, Bellomo R, Mehta R, Ronco C. Blood purification in non-renal critical illness. *Blood Purif* 2003;21:6–13.
62. Mariano F, Tetta C, Guida GE, Triolo G, Camussi G. Hemofiltration reduces the priming activity on neutrophil chemiluminescence in septic patients. *Kidney Int* 2001;60: 1598–605.
63. Ronco C, Tetta C, Lupi A, et al. Removal of platelet-activating factor in experimental continuous arteriovenous hemofiltration. *Crit Care Med* 1995;23:99–107.
64. Mariano F, Bussolati B, Migliori M, Russo S, Triolo G, Camussi G. Platelet-activating factor synthesis by neutrophils, monocytes, and endothelial cells is modulated by nitric oxide production. *Shock* 2003;19:339–44.
65. Sander A, Armbruster W, Sander B, et al. Haemofiltration increases IL-6 clearance in early systemic inflammatory response syndrome but does not alter IL-6 and TNF alpha plasma concentrations. *Intensive Care Med* 1997;23:878–84.
66. De Vriese AS, Colardyn FA, Philippe JJ, Vanholder RC, De Sutter JH, Lameire NH. Cytokine removal during continuous hemofiltration in septic patients. *J Am Soc Nephrol* 1999; 10:846–53.
67. Cole L, Bellomo R, Hart G, et al. A phase II randomized controlled trial of continuous hemofiltration in sepsis. *Crit Care Med* 2002;30:100–6.
68. Heering P, Morgera S, Schmitz FJ, et al. Cytokine removal and cardiovascular hemodynamics in septic patients with continuous venovenous hemofiltration. *Intensive Care Med* 1997; 23:288–96.
69. Ronco C, Brendolan A, Lonnemann G, et al. A pilot study on coupled plasma filtration with adsorption in septic shock. *Crit Care Med* 2002;30:1250–5.
70. De Vriese AS, Vanholder RC, Pascual M, Lameire NH, Colardyn FA. Can inflammatory cytokines be removed efficiently by continous renal replacement therapies? *Intensive Care Med* 1999;25:903–10.
71. Grootendorst AF, van Bommel EFH, van der Hoven B, van Leengoed LAM, van Osta ALM. High volume hemofiltration improves hemodynamics of endotoxin-induced shock in the pig. *J Crit Care* 1992;7:67–75.
72. Grootendorst AF, van Bommel EFH, van der Hoven B, van Leengoed LAM, van Osta ALM. High volume hemofiltration improves right ventricular function of endotoxin-induced shock in the pig. *Intensive Care Med* 1992;18:235–240.
73. Grootendorst AF, van Bommel EF, van Leengoed LA, van Zanten AR, Huipen HJ, Groeneveld AB. Infusion of ultrafiltrate from endotoxemic pigs depresses myocardial performance in normal pigs. *J Crit Care* 1993;8:161–9.
74. Grootendorst AF, van Bommel EFH, van Leengoed LAM, Naburus M, Bouman CSC, Groeneveld ABJ. High volume hemofiltration improves hemodynamics and survival of pigs exposed to gut ischemia and reperfusion. *Shock* 1994;2:72–8.
75. Lee PA, Matson JR, Pryor RW, Hinshaw LB. Continuous arteriovenous hemofiltration therapy for *Staphylococcus aureus*-induced septicemia in immature swine. *Crit Care Med* 1993; 21:914–24.
76. Rogiers P, Zhang H, Smail N, Pauwels D, Vincent JL. Continuous venovenous hemofiltration improves cardiac performance by mechanisms other than tumor necrosis factor-alpha attenuation during endotoxic shock. *Crit Care Med* 1999; 27:1848–55.
77. Yekebas EF, Eisenberger CF, Ohnesorge H, et al. Attenuation of sepsis-related immunoparalysis by continuous veno-venous hemofiltration in experimental porcine pancreatitis. *Crit Care Med* 2001;29:1423–30.
78. Nagashima M, Shin'oka T, Nollert G, Shum-Tim D, Rader CM, Mayer JE Jr. High-volume continuous hemofiltration during cardiopulmonary bypass attenuates pulmonary dysfunction in neonatal lambs after deep hypothermic circulatory arrest. *Circulation* 1998;98(suppl 19):II378–84.
79. Bellomo R, Kellum JA, Gandhi CR, Pinsky MR. The effect of intensive plasma water exchange by hemofiltration on hemodynamics and soluble mediators in canine endotoxemia. *Am J Respir Crit Care Med* 2000;161:1429–36.
80. Ronco C, Bellomo R, Homel P, et al. Effects of different doses in continuous veno-venous haemofiltration on outcomes of acute renal failure: a prospective randomised trial. *Lancet* 2000;356:26–30.
81. Journois D, Israel Biet D, Pouard P, et al. High-volume, zero-balanced hemofiltration to reduce delayed inflammatory response to cardiopulmonary bypass in children. *Anesthesiology* 1996;85:965–76.
82. Oudemans-van Straaten HM, Bosman RJ, van der Spoe JI, Zandstra DF. Outcome of critically ill patients treated with intermittent high-volume haemofiltration: a prospective cohort analysis. *Intensive Care Med* 1999;25:814–21.
83. Cole L, Bellomo R, Journois D, Davenport P, Baldwin I, Tipping P. High-volume hemofiltration in human septic shock. *Intensive Care Med* 2001;27:978–86.
84. Honore PM, Jamez J, Wauthier M, et al. Prospective evaluation of short-term, high-volume isovolemic hemofiltration on the hemodynamic course and outcome in patients with intractable circulatory failure resulting from septic shock. *Crit Care Med* 2000;28:3581–7.
85. Lee PA, Weger G, Pryor RW, Matson JR. Effects of filter pore size on efficacy of continuous arteriovenous hemofiltration therapy for *Staphylococcus aureus*-induced septicemia in immature swine. *Crit Care Med* 1998;26:730–7.
86. Kline JA, Gordon BE, Williams C, Blumenthal S, Watts JA, Diaz-Buxo J. Large-pore hemodialysis in acute endotoxin shock. *Crit Care Med* 1999;27:588–96.
87. Morgera S, Buder W, Lehmann C, et al. High cut off membrane haemofiltration in septic patients with multiorgan failure. A preliminary report. *Blood Purif* 2000;18:61.
88. Reeves JH, Butt WW, Shann F, et al., and the plasmafiltration in sepsis study group. Continuous plasmafiltration in sepsis syndrome. *Crit Care Med* 1999;27:2096–104.
89. Tetta C, Cavaillon JM, Schulze M, et al. Removal of cytokines and activated complement components in an experimental model of continuous plasma filtration coupled with sorbent adsorption. *Nephrol Dial Transplant* 1998;13:1458–64.
90. Tetta C, Gianotti L, Cavaillon JM, et al. Coupled plasma filtration-adsorption in a rabbit model of endotoxic shock. *Crit Care Med* 2000;28:1526–33.
91. Lonnemann G, Bechstein M, Linnenweber S, Burg M, Koch KM. Tumor necrosis factor-alpha during continuous high-flux hemodialysis in sepsis with acute renal failure. *Kidney Int* 1999;56(suppl 72):S84–7.