Cytokine Profile and Correlation to the APACHE III and MPM II Scores in Patients with Sepsis

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In 35 patients fulfilling the criteria of systemic inflammatory response syndrome (SIRS) of infectious origin, tumor necrosis factor-α (TNF-α), interleukin-6 (IL-6), tumor necrosis factor-soluble receptor (TNF-sR), and interleukin-12 (IL-12), C-reactive protein (CRP) levels and the Acute Physiology, And Chronic Health Evaluation III score (APACHE III) were determined on days 1 to 7, 14, 21, and 28. The Mortality Probability Models (MPM) II sepsis score was assessed at the time of admission into the study. The MPM II sepsis score correlated with IL-6 plasma levels on day 1. The APACHE III scores correlated with plasma levels of TNF-sR on days 2–7, with IL-6 levels on days 3–7, and with CRP levels on days 4–7 ($p < 0.01$). The MPM II sepsis score, the APACHE III score, and the IL-6, TNF-sR, and CRP levels were significantly different between survivors and nonsurvivors and between patients with and without shock ($p < 0.05$). A significant decrease of the APACHE III scores, IL-6, and CRP levels was observed over the study period in the survivor group only ($p < 0.05$), while neither the dynamics of TNF-α nor IL-12 plasma levels contributed to the risk estimation of mortality. Presterl E, Staudinger T, Pettermann M, Lassnigg A, Burgmann H, Winkler S, Frass M, Graninger W. Cytokine profile and correlation to the APACHE III and MPM II scores in patients with sepsis.

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Sepsis is a major problem in the care of critically ill patients (1, 2). Despite the availability of potent antibiotics and intensive supportive care, mortality of septic patients approximates 30% and ranges from 20 to 60% depending upon the population being evaluated (3). Several prognostic scoring systems, including the Acute Physiology and Chronic Health Evaluation (APACHE) III or the Mortality Probability Models (MPM) II (4), have been used so far to assess the acute clinical condition of seriously ill patients with sepsis (5–7), to predict prognosis and to assist physicians' clinical decisions. Endogenous mediators which are released following to the interaction of microbial products with macrophages, lymphocytes, platelets, and other cells, include tumor necrosis factor-alpha (TNF-α), various interleukins, platelet activating factor, prostaglandins, leukotrienes, and many others (8). TNF-α is the principal mediator in sepsis, particularly in septic shock and lethal sepsis (9). TNF-receptors occur in a 75- and a 55-kD form. The circulating forms of both receptors have been found in the serum of septic patients and may modify the response to endogenous TNF-α during sepsis. These molecules appear to act as both TNF-α carriers and TNF-α antagonists (10, 11). Interleukin-6 (IL-6) can be induced by TNF-α and interleukin-1 (IL-1). IL-6 induces the synthesis of acute-phase proteins and stimulates growth of activated T-cells, and, together with IL-10 and IL-1, is a potent inhibitor of TNF-α production by peripheral blood mononuclear cells (12). IL-12 is produced by macrophages and B-lymphocytes, and may contribute to the inflammatory process via the induction of gamma-interferon.

The aim of this study was to define the role of the TNF-α, TNF-sR, IL-6, IL-12, and the C-reactive protein (CRP) during the course of sepsis in patients with proven infections. The cytokine levels were correlated with the clinical condition expressed by the initially assessed MPM II score customized for sepsis and by daily assessed APACHE III scores, to prove a possible correlation between the scoring systems and blood cytokine levels, and to evaluate whether repeated measurement of blood cytokine levels may contribute to mortality risk estimation of clinical scoring systems.

METHODS

The study was performed at the University Hospital of Vienna in 1995. The protocol was reviewed and approved by the Institution's Ethics Committee. Thirty-five patients were consecutively included after informed consent had been obtained. The demographic data of the patients (age, underlying diseases, date of admission, date of death) were documented. The study period was 28 d. In all patients, at least two blood cultures, together with any other culture as clinically indicated, were drawn before study entry.

Sepsis was defined as a systemic inflammatory response syndrome (SIRS) associated with infection along the criteria of the American College of Chest Physicians and the Society of Critical Care Medicine Consensus Conference (13): The signs of sepsis were (1) body temperature $< 35.6°$ or $> 38.3°$ C, tachycardia (> 90 beats per min), a respiratory rate > 20 breaths per minute or a $Pa_{CO_2} < 32$ mm Hg (unless the patient was mechanically ventilated), a white cell count ≥ 12 G/L or

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TABLE 1

PATIENTS' DISEASES, PATHOGENS, AND ANTIBIOTIC TREATMENT (EMPIRIC OR ADJUSTED TO THE ISOLATED PATHOGEN)

Patient No.	Focus	Bacteremia	Underlying Disease(s)	Sample Site	Pathogen Isolated	Antibiotic Treatment	Died
1	Pneumonia	No	Chronic obstructive pulmonary disease	Blood Bronchial fluid	— —	Ceftazidime	No
2	Pneumonia	No	Brain injury, ventricular shunt	Blood Bronchial fluid	— —	Piperacillin + Tazobactam	Yes
3	Pneumonia	No	Renal failure, aspiration	Blood Bronchial fluid	— —	Cefotaxime, Netilmicin	Yes
4	Unknown	Yes	—	Blood	*Salmonella typhi*	Ciprofloxacin	No
5	Unknown	Yes	Chronic obstructive pulmonary disease	Blood Urine	*Escherichia coli* *E. coli*	Ceftriaxone	No
6	Pneumonia	No	Chronic cardiac output failure	Blood Bronchial fluid Urine	— — —	Imipenem, Teicoplanin, Gentamicin	Yes
7	Unknown	Yes	—	Blood Urine	*Enterobacter cloacae*	Pefloxacin	No
8	Pyelonephritis	No	Kidney anomaly	Blood Urine	*E. coli* —	Cefodizime	No
9	Osteomyelitis	Yes	Diabetes mellitus, renal failure	Blood Wound pus	*Staphylococcus aureus* *S. aureus*	Vancomycin, Rifampicin	No
10	Unknown	Yes	Cardiac failure, emphysema	Blood Urine	*Morganella morgagnii* —	Cefodizime Netilmicin	Yes
11	Pneumonia	No	Chronic cardiac failure	Blood CVC Bronchial fluid	*S. epidermidis* *Enterococcus faecalis* —	Vancomycin	Yes
12	Unknown	Yes	Coronary heart disease, diabetes mellitus	Blood	*S. aureus*	Vancomycin	Yes
13	Unknown	Yes	—	Blood	*Salmonella paratyphi*	Fleroxacin	No
14	Pneumonia	No	—	Blood Bronchial fluid	— —	Ceftriaxone	No
15	Osteomyelitis	Yes	—	Blood Bone biopsy	*S. aureus* *S. aureus*	Teicoplanin	No
16	Mediastinitis	Yes	Coronary heart disease pacemaker	Blood Aspirate	*S. epidermidis* *S. epidermidis*	Vancomycin	No
17	Pneumonia	No	—	Blood Bronchial fluid	— —	Ceftriaxone Netilmicin	No
18	Pyelonephritis	Yes	Renal calculi	Blood Urine	*E. coli* *E. coli*	Cefodizime	No
19	Pyelonephritis	Yes	Renal calculi, renal failure	Blood Urine	*E. coli* *E. coli*	Ciprofloxacin	Yes
20	Pneumonia	No	—	Bronchial fluid	*Klebsiella oxytoca*	Imipenem	Yes
21	Pneumonia	Yes	Diabetes mellitus, renal fialure	Blood	*S. aureus*	Vancomycin	Yes
22	Endocarditis	Yes	Aortal valve replacement	Blood	*Streptococcus mitis*	Penicillin G Netilmicin	Yes
23	Meningitis	Yes		Blood CSF	*Neisseria meningitidis* *N. meningitidis*	Ceftriaxone	No
24	CVC-sepsis	Yes	Aortocoronary bypass coronary heart disease	Blood CVC	*S. epidermidis* *S. epidermidis*	Teicoplanin Netilmicin	No
25	Pneumonia	No	Coronary heart disease; myocardial infarction	Blood Bronchilal fluid	— *S. marcescens*	Piperacillin/ Tazobactam	No
26	Osteomyelitis	Yes	—	Blood Aspirate	*Bacillus fragilis* *B. fragilis*	Metronidazole	No
27	Unknown	Yes	Coronary heart disease Diabetes mellitus	Blood	*P. aeruginosa*	Ceftazidime Tobramycin	No
28	Pyelonephritis	Yes	Diabetes mellitus	Blood Urine	*E. cloacae* *E. cloacae*	Ofloxacin	Yes
29	Wound infection	Yes	Psoariasis	Blood Wound	*E. cloacae* *E. cloacae*	Imipenem	Yes
30	Pyelonephritis	Yes	Diabetes mellitus	Blood Urine	*S. aureus* *S. aureus*	Flucloxacillin	No
31	Pneumonia	No	Diabetes mellitus	Blood Bronchial fluid	— *K. pneumoniae*	Imipenem	Yes

(*Continued*)

TABLE 1

CONTINUED

Patient No.	Focus	Bacteremia	Underlying Disease(s)	Sample Site	Pathogen Isolated	Antibiotic Treatment	Died
32	Meningitis	Yes	Diabetes mellitus	Blood CSF	*Streptococcus pneumoniae*	Penicillin G	No
33	Pneumonia	No	Diabetes mellitus	Blood Bronchial fluid	*E. coli*	Ciprofloaxcin Amikacin	Yes
34	Pneumonia	Yes	Glioblastoma	Blood Bronchial fluid	*Legionella pneumophila* *L. pneumophila*	Erythromycin Ciprofloxacin	Yes
35	Pneumonia	No	Myelodysplastic syndrome	Blood Bronchial fluid	— *P. aeruginosa*	Imipenem Vancomycin	Yes

$\leq$ 4 G/L or $>$ 10% immature neutrophils (bands). Sepsis syndrome was defined as the evidence of organ dysfunction and hypoperfusion abnormality: Acute alteration of the mental status; elevated plasma lactate, unexplained metabolic acidosis with arterial pH $<$ 7.3; hypoxemia ($P_{A_{O_2}} <$ 70 mm Hg breathing room air, or an acute drop in Pa_{O_2} of $>$ 15 mm Hg below baseline with breathing room air or hypoxemia requiring mechanical ventilation); prolonged prothrombin time, or a decrease of the platelet count of more than 50% or $\leq$ 100 G/L; oliguria; and hypotension defined as systolic blood pressure of $<$ 90 mm Hg or a decrease of $>$ 40 mm Hg from baseline. Septic shock was defined as hypotension additional to sepsis syndrome persisting despite adequate fluid resuscitation and requiring vasopressor treatment.

Patients were not eligible for the study if they were under 18 yr of age; if they had a rapidly progressive underlying disease with a general life expectation of at most 3 mo (including AIDS); liver cirrhosis; burns involving $>$ 20% of the body surface area; uncontrolled hemorrhage; cardiogenic shock; granulocytopenia ($<$ 1 G/L); antineoplastic chemotherapy within 7 d before inclusion into the study; preceding elective surgery; or treatment with any anticytokine drug. All patients included were not enrolled in any other clinical study or sepsis trial.

Standard supportive care, surgical procedures (drainage of abscesses, removal of an infected foreign body, etc.), as well as broad spectrum antibiotics were provided to all patients. Pneumonia was diagnosed if infiltrations were present on the chest X-ray, and, if possible, a positive culture from purulent sputum or bronchial fluid could be obtained. Abscesses were diagnosed by sonography or by CT-scan, together with growth of a pathogenic bacteria from aspirated pus. Urinary tract infections were diagnosed by the evidence of leucocyturia, hematuria and growth of a pathogen in midstream urine culture. Meningitis was diagnosed by the evidence of high numbers of leucocytes and the growth of bacteria in the cerebrospinal fluid. Endocarditis was diagnosed when the echocardiography revealed valvular vegetations, and blood cultures were repeatedly positive. Osteomyelitis was diagnosed by the radiological evidence of osteomyelitic destruction of the bone, a positive bone scintigraphy and the growth of bacteria from pus or a bone biopsy.

Cytokine Measurement

Blood for cytokine measurement was drawn on days 1–7, 14, 21, and 28 after inclusion to the study. All plasma samples were collected in Vacutainer® tubes (Becton Dickinson, Rutherford, NJ) containing EDTA. Samples were centrifuged at 3,000 g at 4° C for 10 min. Plasma samples were then stored at −70° C.

Plasma concentrations of TNF-α were determined by using an enzyme-linked immunosorbent assay (ELISA) with a sensitivity of 5 pg/mL (Quantikine; R&D Systems, Minneapolis, MN). Briefly, a monoclonal antibody specific for TNF-α is coated onto a microtiter plate. Standards (0.2 mL) and samples (0.2 mL) were pipetted into the wells, and any TNF-α present was bound by the immobilized antibody. After washing, an enzyme-linked polyclonal antibody specific for TNF-α was added to the wells to "sandwich" the TNF-α immobilized. A substrate solution was added. The color development is stopped and the intensity of the color is measured with an automatic reader at 450 nm wave length. Standards of recombinant TNF-α were used in a concentration of 15.6 to 1,000 pg/mL. Plasma concentrations of IL-6 and IL-12 were determined by solid phase ELISA (Quantikine, R&D Systems) similar to the TNF-α assay. The sensitivity of the test was 0.7 pg/mL for IL-6 and 0.5 pg/mL for IL-12.

Plasma concentrations of soluble TNF-receptor (TNF-sR, 60 kDA) was determined by an enzyme-linked immunosorbent assay (Bender MedSystems, Vienna, Austria). Similar to the other tests a monoclonal antibody specific for the 55 kDa protein of the s-TNF-R was used. The sensitivity was 0.08 ng/ml.

APACHE III and MPM II Sepsis Score

The Acute Physiology and Chronic Health Evaluation APACHE III score was assessed daily by using the worst clinical parameters (7). The APACHE III score was calculated by a computer using the Statistical Analysis Software (Cary, NC).

The Mortality Probability Model (MPM) II customized for sepsis (14) was retrospectively assessed for the time of inclusion into the study. The MPM II includes the length of stay at the ICU, and has been developed so far for the assessment at 0, 24, 48, and 72 h after admission to the ICU. Thus, the MPM II_{24} was assessed in 16 patients, the MPM II_{48} in seven and the MPM II_{72} in four patients. The other patients had been admitted to the ICU longer than 72 h before developing sepsis.

Statistical Analysis

All statistical analysis was done with the Statistical Analysis Software (Cary, NC). Wilcoxon's rank-sum test was used for comparing two groups (survivors–nonsurvivors; bacteremic–nonbacteremic) on day 1–28. The repeated measures ANOVA (General Linear Models Procedure by SAS) was used to compare the change of plasma cytokine levels and the APACHE III from day 1 to day 28. The Spearman's rank correlation was used to define a correlation between APACHE III and the plasma cytokine levels, and among the cytokine levels. The levels of significance were set at $p < 0.05$ for the ANOVA and the comparisons between groups, and at $p < 0.01$ for the correlation coefficient.

RESULTS

Demography

Thirty-five patients (22 men, 13 women) were included into the study. The mean age was 52 years (range 19–83 yr), the mean body weight 82 kg (range 52–130 kg), and the mean height 1.72 m (range 1.55–1.86 m). The mean duration of the hospitalization prior to study entry was 2.5 days (range 0–11 days). Chronic underlying diseases were found in 24 patients; heart failure (n = 7), coronary heart disease (n = 4), chronic obstructive pulmonary disease (n = 3), diabetes mellitus (n = 8) for mean 4 yr, and neoplastic malignancies (n = 2). All patients fulfilled the inclusion criteria of sepsis. Twenty patients were in septic shock and showed signs of organ dysfunction: Respiratory failure requiring mechanical ventilation (n = 9), acute cerebral coma (n = 3), acute renal failure requiring dialysis or hemofiltration (n = 4), hepatic failure (n = 3), and dis-

TABLE 2

CORRELATION MATRIX (p VALUES) FOR CYTOKINES AND APACHE III AND MPM II SEPSIS SCORES

	APACHE III			MPM II Sepsis		
Day	TNF-sR	IL-6	CRP	TNF-sR	IL-6	CRP
1	0.0200	0.0390	0.8513	0.049	0.004	0.400
2	0.0004	0.0230	0.5504			
3	0.0008	0.0001	0.3290			
4	0.0001	0.0001	0.0038			
5	0.0005	0.0001	0.0001			
6	0.0009	0.0001	0.0001			
7	0.0001	0.0015	0.0001			

seminated intravasal coagulation (n = 1). In 24 patients a defined focus of infection could be found: pneumonia (n = 12), pyelonephritis (n = 5), osteomyelitis (n = 3), meningitis (n = 2), endocarditis (n = 1), mediastinitis (n = 1), and wound infection (n = 1). Eight patients had bacteremia without evident focus. Pathogens and infection sites are listed in Table 1. Patients in whom the causative pathogens could be identified (n = 33), received antibiotic treatment appropriate to the susceptibility pattern of the organisms. The daily dose was adjusted to renal function, if necessary. Sixteen patients died within the first 14 d.

Correlation of TNF-sR, CRP, TNF-α, IL-6, and the APACHE III Score (n = 35)

The initially assessed MPM II sepsis score correlated with IL-6 plasma levels ($p < 0.01$) and TNF-sR ($p < 0.03$) on day 1. APACHE III scores did not correlate with TNF-sR, IL-6, and CRP on day 1. The correlation between daily assessed cytokines and corresponding APACHE III scores started on day 2, and was persistent for several days, as shown in Table 2.

Patients with Bacteremia (n = 20) Versus Patients without Bacteremia (n = 15)

APACHE III and MPM II sepsis scores, CRP- and cytokine plasma levels did not differ significantly between both groups, nor did they change from day 1 to day 28 (Table 3). IL-12 levels were negative in all patients except for one in whom *Staphylococcus aureus* was isolated from multiple blood cultures, and who died on day 6.

Survivors (n = 19) Versus Nonsurvivors (n = 16)

MPM II sepsis score, APACHE III score, CRP-, TNF-α, IL-6, and TNF-sR plasma levels of both groups are summarized in Table 4. The MPM II sepsis scores and the APACHE III scores were significantly higher in nonsurvivors at the time. The area under the curve of the APACHE III scores were also significantly greater in nonsurvivors ($p < 0.01$). Cytokines and CRP levels were higher in nonsurvivors in a time-dependent fashion: IL-6 and TNF-sR levels differed significantly from day 2 to day 7, the CRP levels from day 4 to day 7, and all three were different on day 14.

The APACHE III scores and plasma concentrations of IL-6 and CRP decreased steadily from day 1 to day 7, 14, 21, and 28 in the survivor group. There was no significant change in the TNF-α and TNF-sR levels. In the nonsurvivor group, cytokine levels, plasma CRP, or APACHE III scores did not change throughout the study period. Figure 1a–e shows the cytokine levels and the APACHE III scores (median and range) on days 1, 4, and 7.

Patients with Septic Shock (n = 20) Versus Patients without Septic Shock (n = 15)

Six of nine patients without bacteremia and five of eleven patients with bacteremia had septic shock from the beginning of the study. Septic shock occurred more frequently in the nonsurvivor group (16 of 16 patients) than in the survivor group (4 of 19 patients). The differences of APACHE III score, CRP-, TNF-α, IL-6, and TNF-sR plasma levels between both groups are presented in Table 5. APACHE III scores of the patients with septic shock were significantly higher at any time compared with those of patients without shock. The area under the curve of the APACHE III scores was also significantly higher ($p < 0.01$). In patients with shock, IL-6 levels were sig-

TABLE 3

CYTOKINE PLASMA CONCENTRATION AND APACHE III SCORE IN THE PATIENTS WITH BACTEREMIA AND WITHOUT BACTEREMIA

Day	TNF-α (*pg/ml*)	IL-6 (*pg/ml*)	TNF-sR (*ng/ml*)	CRP (*mg/dl*)	APACHE III Score	n	MPMII Sepsis Score	n (MPMII)
1	9.3 (0–203)	171.9 (7.1–7,637)	17.2 (0.35–49.4)	17.8 (9.3–57.5)	51 (23–115)	20	0.389 (0.044–0.844)	13
	7 (0–16.6)	199.2 (22.5–1,888)	13.5 (6.1–66)	15.2 (2.9–36.8)	66 (28–136)	15	0.376 (0.05–0.705)	14
2	10.2 (0–41.9)	226.1 (5.4–9,349)	19.2 (5.5–83)	20.6 (8.2–49.8)	46 (13–115)	20	—	
	6.8 (0–509.9)	235.9 (4.6–1,420)	13.3 (3.4–64.1)	17.2 (9.2–31.1)	61 (7–145)	15		
3	10 (0–41)	152.4 (2.1–9,999)	16.6 (4–64)	16.35 (8.3–42.3)	44 (9–130)	20	—	
	5.6 (0–18.8)	191 (0–500)	8 (3.9–48.1)	14.3 (10.1–27.1)	71 (4–118)	15		
4	5 (0–24.5)	130.6 (0–9,999)	15.7 (5–82.9)	11.8 (4.4–32)	44 (0–120)	19	—	
	4.7 (0–17.2)	103.4 (0–748)	9.2 (4.5–46.4)	14.2 (5.2–28.2)	57.5 (3–112)	14		
5	11.4 (1–30.7)	21.5 (0–9,999)	11.8 (0.2–63.5)	77.8 (3.2–35)	29.5 (0–129)	19	—	
	5 (0–25.1)	16.5 (0–600)	6.8 (1.9–136.7)	10.8 (2.5–26.5)	41 (0–129)	14		
6	2.7 (0–13.8)	1.6 (0–213.5)	9 (3.3–15.5)	4.8 (1.5–27.8)	28.5 (0–125)	15	—	
	3.8 (0–10.2)	5.8 (0–225.4)	7.2 (3–32.9)	6.6 (1.3–24.5)	31.5 (7–91)	12		
7	1.45 (0–29.7)	62.3 (0–9,999)	10 (3–81.1)	4.4 (1–39)	31 (0–127)	15	—	
	7.1 (0–78.4)	30.3 (0–433.6)	9.4 (2.7–75.1)	6.3 (0.2–24.3)	32.5 (0–111)	12		
14	11.6 (0–12.8)	9.1 (0–205.1)	7.2 (2–47.2)	2.8 (0.2–18)	26 (0–67)	14	—	
	5.8 (0–40.9)	27.4 (0–412.4)	6.8 (0–59.5)	6.7 (0–46.6)	58 (5–108)	8		
21	0 (0–13.8)	3 (0–38.1)	6.4 (1–40.1)	5.4 (0.2–17.8)	10 (0–57)	11	—	
	1.9 (0–10.1)	6.2 (0–76.3)	6.4 (2–13.2)	3.2 (0.2–12.2)	21.5 (0–50)	8		
28	0.2 (0–12.5)	0.5 (0–85.4)	3.25 (0–9.2)	1.2 (0–12.5)	8 (0–49)	11	—	
	0.5 (0–9.7)	0 (0–9.7)	4.3 (1.8–11.7)	1.5 (0–8.1)	13 (0–46)	8		

First line: median (range) of patients with bacteremia; second line: median (range) of patients without bacteremia.

TABLE 4

CYTOKINE LEVELS AND APACHE III SCORE OF NONSURVIVORS AND SURVIVORS FROM DAYS 1–28, MEDIAN (RANGE)

Day	TNF-α (*pg/ml*)	IL-6 (*pg/ml*)	TNF-sR (*ng/ml*)	CRP (*mg/dl*)	APACHE III SCORE	n	MPMII Sepsis SCORE	n (MPMII)
1	3.9 (0–203)	158 (7.1–877)	6.8 (0.5–49.4)	17.5 (9.3–36.8)	46 (24–76)†	19	0.237 (0.444–0.652)†	15/12
	12 (0–124.5)	370.7 (18.7–7,637)	27.2 (0.35–66)	16.1 (2.9–57.5)	75 (23–136)	16	0.544 (0.237–0.844)	
2	5.5 (0–41.9)	51.2 (4.6–457)*	9.1 (84.3–50.5)†	21.3 (8.2–37.8)	39 (7–70)†	19	—	
	10.9 (0–509.9)	412.6 (41.4–9,340)	27.2 (13.3–83)	17.7 (9.2–49.8)	79.5 (22–145)	16		
3	4.5 (0–23.8)*	22.1 (0–448)†	8.6 (3.9–47.8)†	14.9 (8.3–39.8)	34 (4–71)†	19	—	
	14.2 (0–41)	301 (54.1–9,999)	20.3 (7.6–64)	14.9 (10.3–42.3)	85 (17–130)	16		
4	2.2 (0–24.4)	14.4 (236.5)†	8.9 (4.5–51.8)†	9.8 (4.4–32)†	22 (0–82)†	19	—	
	9.1 (0–19.9)	382 (100.8–9,999)	23.8 (9.4–52.9)	18.4 (9.3–32)	86.5 (27–120)	14		
5	4 (0–16.5)†	2.8 (0–292.1)†	6.8 (0.2–60.3)†	6.1 (2.5–26.5)†	26 (0–59)†	19	—	
	12 (6.1–30.7)	243 (2.3–9,999)	32.6 (1.9–136.7)	13.2 (8.6–35)	78 (20–129)	14		
6	2 (0–13.8)	1.6 (0–225.4)*	7.5 (1.3–27.8)*	4.8 (1.3–27.8)	23 (0–63)†	19	—	
	7.6 (6.7–9.2)	52 (1.5–98.8)	13.8 (8.7–32.9)	11.5 (7.5–12.8)	72 (51–125)	8		
7	1.5 (0–13.9)†	13.9 (80–512)*	7.4 (2.7–60.8)*	3.6 (0.2–23)†	17 (0–68)†	19	—	
	12.7 (0–78.4)	195.7 (0–9,999)	17 (9.4–81.1)	21.6 (8.1–39)	96 (40–127)	8		
14	2.2 (0–12.8)†	6.1 (0–237.9)*	5.4 (0–47.2)*	2.7 (0–49.6)*	26 (0–70)†	19	—	
	13.2 (10.7–40.9)	155.2 (65.7–412.4)	29.4 (16.1–59.5)	12.7 (10.3–16.2)	91 (91–108)	4		
21	1.2 (0–13.8)	3 (0–76.3)	6.4 (1–40.1)	5.4 (0.2–17.8)	10 (0–57)	19	—	
	—	—	—	—	—	0		
28	0.2 (0–9.7)	0 (0–85.4)	3.7 (0–11.7)	1.2 (0–12.5)	12 (0–49)	19	—	
	—	—	—	—	—	0		

First line: median (range) of nonsurvivors; second line: median (range) of survivors.
* Significant difference ($p < 0.05$).
† Significant difference ($p < 0.01$).

nificantly increased from day 1 to day 7, TNF-sR from day 1 to day 4, and the CRP levels from day 3 to day 7.

DISCUSSION

In this study, two clinical scoring systems, the MPM II sepsis score and the APACHE III score, correlated with plasma levels of IL-6, TNF-sR, and the C-reactive protein at different times over the course of sepsis. The MPM II sepsis score correlated with IL-6 levels on day 1, but the APACHE III score did not. The APACHE III scores correlated with TNF-sR levels on days 2–7, with IL-6 levels on days 3–7, and with CRP levels on days 4–7 ($p < 0.01$). Nonsurvivors had significantly higher MPM II and APACHE III scores at any time. Compared with survivors, nonsurvivors had significantly higher IL-6 and TNF-sR concentrations from day 2 to day 7, and CRP levels from day 3 on. Compared with patients without septic shock, the IL-6 and TNF-sR levels of patients with septic shock were always significantly higher beginning on day 1. A difference in plasma CRP levels was only observed from day 3 on. The MPM II and APACHE III scores were significantly different between the patients with septic shock and those without. Between patients with bacteremia and patients without bacteremia, no significant difference of cytokine and CRP levels, or any scoring system was observed.

TNF-α, IL-6, and interferon-γ play a pivotal role in sepsis and septic shock (15). Serum levels of TNF-α and IL-6 were elevated, although variable, in patients with Gram-negative bacteremia or septic shock (16, 17). An association of serum TNF-α levels and mortality was observed in patients with septic shock of meningococcal origin (18). TNF-α levels were increased in patients with fatal outcome, and correlated inversely with survival (19). In another study, IL-6 levels carried a similar relation to mortality (20). Persistent elevation of TNF-α levels after 12 h in patients with multiple organ failure suggested a relationship between TNF-α levels and organ dysfunction, although TNF-α plasma levels were not considered a good predictor of mortality (21). Neither TNF-α nor IL-6 were regarded specific for infection, but increased serum concentrations of TNF-α and IL-6 were found in patients with septic shock compared with patients with non-septic shock (22, 23). Persistently increased IL-6 plasma levels rather than peak values were considered predictive for poor outcome in patients with septic shock (23). Increased IL-6 levels in patients with sepsis and septic shock correlated with severity of shock (24). The authors suggested that monitoring IL-6 levels could be a predictor of mortality because IL-6 levels correlated best with outcome. A significant correlation of initial serum levels of TNF-α, IL-6, and other cytokines and the APACHE II scores in 13 patients with meningococcal and pneumococcal septic shock was reported by Gardlund and colleagues (25). In the present study, TNF-α did not correlate with any scoring system. This may be due to the fact that TNF-α is produced locally, and plasma concentrations do not reflect the paracrine activity of this cytokine (26). IL-6, however, correlated with the MPM II sepsis score on day 1, and with the APACHE III score beginning on day 2. A possible explanation is that the MPM II was adjusted to patients with sepsis and the length of ICU admission. A persistent correlation between IL-6 and the MPM II sepsis score during the course of sepsis would be of interest, but so far, the MPM II sepsis score is only applicable for evaluation during the first 4 d after admission to the ICU.

Increased TNF-sR levels were found to indicate poor outcome in patients with meningococcemia and in cancer patients with febrile neutropenia (27, 28). TNF-sR correlated with the APACHE II score in 12 critically ill patients, and a persistent evaluation was considered to be predictive for an adverse outcome in few patients (29). TNF-sR had also been found to be increased in patients with cytomegalovirus-pneumonitis after lung transplantation when compared with transplant patients experiencing rejection or to healthy transplant patients (30).

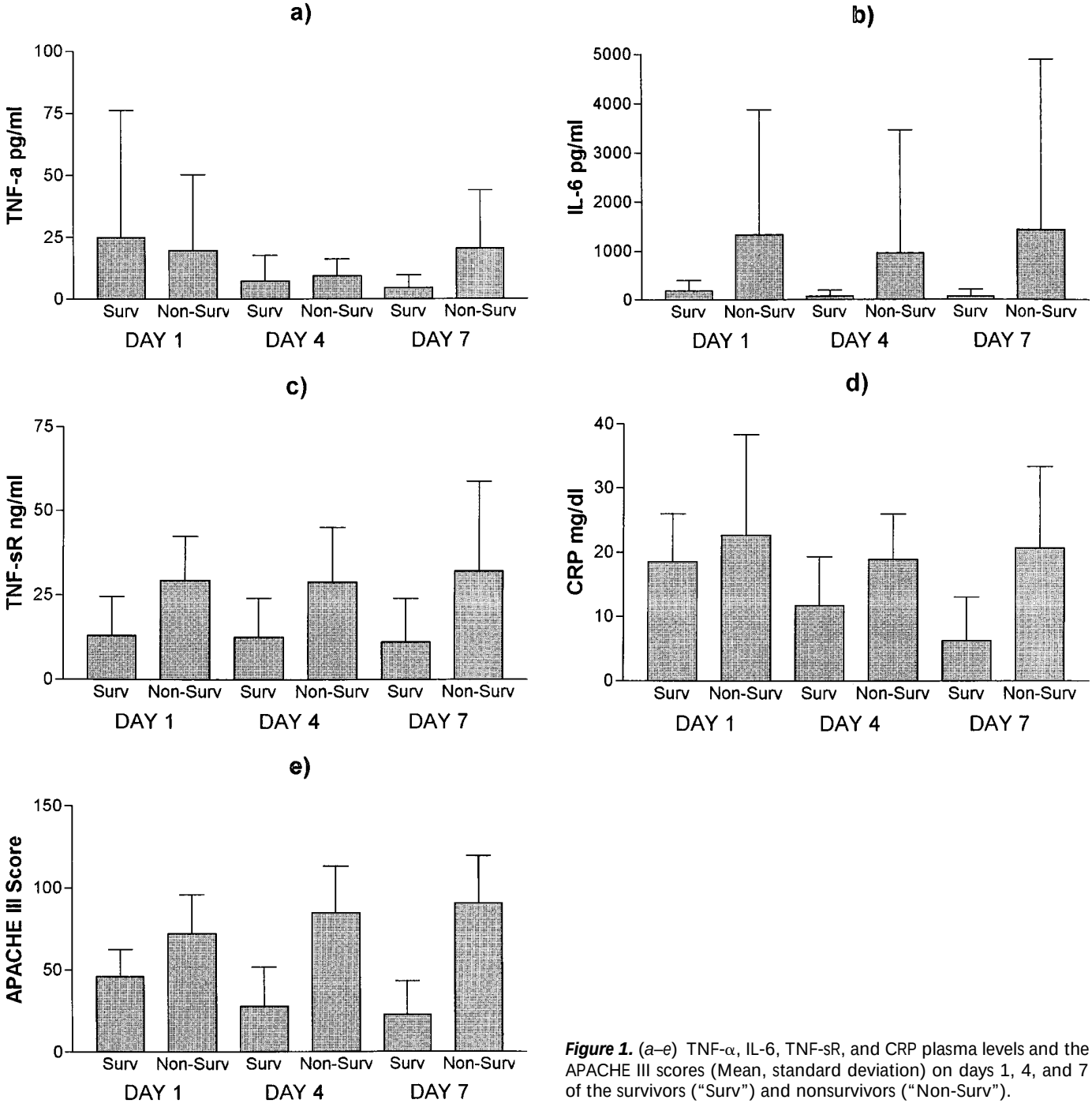

Figure 1. (*a–e*) TNF-α, IL-6, TNF-sR, and CRP plasma levels and the APACHE III scores (Mean, standard deviation) on days 1, 4, and 7 of the survivors ("Surv") and nonsurvivors ("Non-Surv").

In the present study, TNF-sR was significantly higher in patients with septic shock compared with those without. TNF-sR levels were significantly higher in nonsurvivors than in survivors, but only starting on day 2, which could be explained as the response to the high TNF-α concentrations locally produced. Plasma levels of TNF-α, however, were not increased. TNF-sR plasma levels are more easily measurable because of the longer half-life of TNF-sR compared with TNF-α. TNF-sR might therefore be used as an indicator of the TNF-α production, and hence as a parameter to quantify inflammatory response. A significant correlation between the APACHE II score and TNF-sR plasma levels was found by Kern and coworkers (28). In our study, TNF-sR levels correlated with APACHE III scores starting on Day 2. In contrast to IL-6, TNF-sR levels did not correlate to the MPM II sepsis scores on Day 1. Thus, IL-6 seems to be the better parameter for assessment of severity of sepsis, as it is produced as early as 2–4 h after initiation of the inflammatory response (31).

IL-12 stimulates interferon-γ production, which is a mediator of endotoxin and TNF-α effects in experimental sepsis (32, 33). Heinzel and coworkers detected peak levels of IL-12 within 2 to 4 h after endotoxin injection (34), thus IL-12 plasma levels should be increased in patients with septicemia. However, in the present study, IL-12 could not be measured in most of the patients.

The APACHE III score was reported to be more predic-

TABLE 5

CYTOKINE LEVELS AND APACHE III SCORE OF PATIENTS WITH SEPTIC SHOCK AND PATIENTS WITHOUT SEPTIC SHOCK FROM DAYS 1–28, MEDIAN (RANGE)

Day	TNF-α (*pg/ml*)	IL-6 (*pg/ml*)	TNF-sR (*ng/ml*)	CRP (*mg/dl*)	APACHE III Score	n	MPMII Sepsis Score	n (MPMII)
1	8.6 (0–203)	370.7 (18.7–7,637)†	24.5 (0.4–66.5)*	16.3 (2.9–57.5)	73 (23–136)†	20	0.504 (0.237–0.884)†	16
	3.9 (0–120)	56.1 (7.1–312.1)	6.9 (0.5–49.4)	15.5 (9.3–36.8)	42 (24–76)	15	0.084 (0.044–0.512)	11
2	10 (0–510)	350 (41.4–9,340)†	24.5 (3.4–83)*	17.9 (9.2–49.8)	67 (22–145)†	20		
	7.1 (0–41.9)	41.8 (4.6–398.1)	10 (4.7–50.5)	18.3 (8.2–37.8)	32 (7–70)	15		
3	10.2 (0–41)	245.6 (54.1–9,999)†	19 (6.6–64)*	16.4 (10.3–42.3)*	77 (17–130)†	20		
	5.2 (0–23.8)	15.7 (0–208)	8.3 (3.9–47.8)	11.3 (8.3–39.8)	15 (4–71)	15		
4	5 (0–19.9)	235 (1.4–9,999)†	19 (7–52.9)*	18.7 (9.3–32)†	77 (22–120)†	18		
	2.5 (0–24.4)	13 (0–236.5)	8.6 (4.5–51.8)	8.5 (4.4–15.9)	15 (0–82)	15		
5	12 (0–30.7)*	144.6 (0–9,999)*	32.5 (1.9–136.7)*	13.2 (8.6–35)†	50 (20–129)†	18		
	4 (0–16.5)	2.8 (0–292)	6.1 (0.2–60.3)	4.6 (2.5–23.6)	19 (0–59)	15		
6	6.7 (0–9.2)	97 (1.5–225.4)*	10.6 (8.7–32.9)	12.8 (6.6–25)*	51 (31–125)†	12		
	2.4 (0–13.8)	0.5 (0–213.5)	5.5 (83–51.5)	3.3 (1.3–27.8)	15 (0–63)	15		
7	9.7 (0–78.4)	195.7 (4.5–9,999)†	11.6 (9.4–81.1)	19.7 (4–39)†	70 (31–127)†	12		
	1.5 (0–13.9)	0.2 (0–248.8)	5.6 (2.7–60.8)	2 (0.2–18.3)	12 (0–68)	15		
14	6.6 (0–40.9)	155.2 (27.4–412.4)*	16.1 (0.2–59.5)	10.3 (6.7–49.6)*	70 (24–108)†	7		
	2.4 (0–12.8)	0.9 (0–205.1)	3.6 (0–47.2)	2.2 (0–18)	18.5 (0–67)	15		
21	2.5 (0–10.1)	23 (12.3–76.3)	9.7 (7–13.2)	6.2 (6–12.2)	43 (14–45)	4		
	0.6 (0–13.8)	0.7 (0–38.1)	4.5 (1–40.1)	1.8 (0.2–17.8)	4.5 (0–57)	15		
28	1 (0–9.7)	0 (0–9.9)	11.1 (0–11.7)	5 (1.4–8.1)	20 (7–46)	4		
	0 (0–6.1)	0 (0–85.4)	3.5 (1–9.2)	0.4 (0–12.5)	11 (0–49)	15		

First line: median (range) of patients with septic shock; second line: median (range) of patients without septic shock.
* Significant difference ($p < 0.05$).
† Significant difference ($p < 0.01$).

tive for mortality than plasma levels of TNF-α, interleukin-1β, IL-6, and IL-8 so far, a persistent correlation between the APACHE III and cytokine concentration was not observed in a nonhomogeneous ICU patient population (35). In our study, correlations between the APACHE III score and MPM II sepsis score, and plasma levels of IL-6, TNF-sR and CRP were observed: IL-6 levels correlated with the MPM II customized for sepsis on day 1. Beginning on day 2, the APACHE III score correlated with TNF-sR, and then with IL-6 and CRP plasma levels beginning on days 3 and 4, respectively. Both scoring systems as well as the plasma IL-6 and CRP levels were significantly higher in the nonsurvivors compared with the survivors. Now that reliable laboratory tests for cytokines become more and more available and less costly, IL-6 and CRP should be included into the assessment models of outcome prediction in patents with sepsis, either associated to or included into a clinical scoring system. Further, this new scoring system should be applicable repeatedly like the APACHE III score, and adapted for sepsis and the length of ICU admission like the MPM II score.

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