Serum CRP and IL-6 levels after trauma

Not predictive of septic complications in 31 patients

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We studied 31 blunt trauma victims, Injury Severity Score (ISS) mean 14 (9–57), for the pattern of release of C-reactive protein (CRP) and cytokine interleukin-6 (IL-6). Blood samples were taken on admission (within 6 hours of injury), as well as at 24 hours, and 3, 5 and 7 days. Serum CRP and IL-6 were measured by ELISA. Subsequent surgical events and sepsis were noted.

Serum IL-6 levels on admission were considerably higher (median 135 pg mL^{-1}) than our laboratory reference range (< 5 pg mL^{-1}), slowly returning towards reference values during the study. Serum CRP levels were similar to laboratory normal values on admission (median 8.5 mg L^{-1} vs 7.5 mg L^{-1}), reaching peak values (median 110 mg L^{-1}) after 3 days. There was a correlation between IL-6 release and ISS but not between CRP and ISS. Patients undergoing surgery showed further increases in IL-6 and CRP levels postoperatively. Of 24 surgical patients, 9 developed postoperative sepsis.

In blunt trauma patients, early assessment of the markers CRP or IL-6 were not useful for the diagnosis of sepsis. Levels of CRP following accidental or surgical trauma should be assessed with caution.

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C-reactive protein (CRP) can rise dramatically after the onset of inflammation, being synthesized in the liver, following release of interleukin-6 by activated macrophages (Whicher and Dieppe 1985) and increasing as much as 1000-fold (Pepys 1984). After surgery, CRP rises rapidly from 6 hours after skin incision, reaching a peak at 48 hours (Whicher and Evans 1990). The magnitude of the CRP and acute phase protein response is said to be related to the mass of inflamed tissue (Shakespeare et al. 1989).

An additional well recognized element in the acute phase reaction is interleukin-6 (IL-6), a multifunctional protein that plays important roles in host defense, immune response, acute phase reaction and hematopoiesis (Akira et al. 1993). It is responsible for switching on many acute protein genes such as C-reactive protein, complement components C3 and factor B, fibrinogen and 1-antitrypsin (Gauldie et al. 1988, Gauldie et al. 1990). Elevated IL-6 following surgical and accidental trauma has been reported by several authors (Cruickshank et al. 1990, Hoch et al. 1993, Giannoudis et al. 1996).

A prerequisite for the use of acute phase protein levels such as CRP and IL-6 as diagnostic aids in the early detection of postoperative infections in clinical work is a knowledge of the baseline CRP and IL-6 responses induced by accidental trauma. We studied the pattern of release of serum CRP and serum IL-6 concentrations in patients following blunt orthopedic trauma, in order to determine whether surgical procedures such as secondary inflammatory insults or sepsis affect the pattern of release.

Patients and methods

Between May 1995 and July 1996 we studied 31 adults, following accidental blunt trauma. Research Ethics Committee approval was obtained and all patients or their representatives prior to entry into the study gave verbal or written consent (Table 1). All patients presented within 6 hours of injury, had an injury severity score (ISS) ≥ 9 and were inpatients for at least 7 days. Venous blood samples were collected on arrival in the emergency department (within 6 hours of injury) and after 24 hrs, 3 days, 5 days and 7 days following injury. Blood samples were also collected from 20 (11 women) healthy, uninjured individuals with a median age of 54 (23–85) years who formed the control group for IL-6 assay. Standard laboratory normals were used for CRP.

Blood samples were collected into tubes containing clot activator. Samples were centrifuged at 1000 rpm for a period of 10 minutes and serum was stored

Table 1. Patients' details

Case	ISS	Age	Sex	Injuries, fracture
1	57	34	M	pelvis,humerus,T8, bilateral pleural contusion, rupture spleen
2	33	21	M	scapula, lacerations external, right tension hemothorax, head injury
3	27	29	M	scapula, right hemothorax, ruptured subclavian artery
4	29	40	F	pelvis opened, ribs, perineal floor tear, lacerations external
5	30	26	M	pelvis, humerus, forearm, dislocated knee, head injury, lacerations
6	25	27	M	bilateral tibias, pelvis, head injury
7	22	27	M	femur, ribs, splenic laceration, minor head injury
8	17	33	F	acetabulum, femur, ribs, forearm, lacerations
9	22	26	M	femur, right tibia, right pubic rami, right pneumothorax, head injury
10	17	26	M	femur, right forearm, degloving injury right leg, head injury
11	13	55	M	ribs, acetabulum, lacerations external
12	13	50	F	pelvis, lacerations external, minor head injury
13	13	86	F	pelvis, head injury
14	13	31	M	pelvis, head injury
15	14	19	M	pelvis, lumbar vertebrae 4-5, head injury
16	11	18	M	forearm, ruptured saphenous vein, head injury
17	9	45	M	right tibia
18	9	36	M	left tibia
19	9	30	M	femur, pubic rami
20	9	46	M	femur, Bennet's
21	9	85	F	femur
22	9	50	M	femur
23	9	27	M	bilateral femurs
24	9	20	F	femur
25	10	24	M	femur, laceration forehead
26	10	25	M	femur, lacerations
27	9	37	M	femur,
28	9	45	M	femur, humerus
29	14	79	M	femur, humerus, forearm, head injury, lacerations
30	13	55	M	scapula, left lung contusion, lacerations
31	10	46	M	tibia, lacerations external

at –70 °C.

Serum IL-6 was measured using enzyme-linked immunoabsorbant assay kits (ELISA) (R & D Systems Inc, Minneapolis, USA). All measurements were performed in duplicate. Serum CRP levels were determined using a CRP kit and a synchrom CX-7-Delta analyzer (Beckman).

Sepsis was defined as clinical evidence of an infection site or a positive bacterial culture with a systemic response, characterized by 2 or more of the following: peripheral white blood count of less than $2 \times 10\ L^{-1}$ or more than $15 \times 10\ L^{-1}$; temperature higher than 38 °C; twofold increase in serum alkaline phosphatase levels; bilirubin levels higher than 20 mol/L^{-1}; hypotension, systolic blood pressure less than 100 mmHg for 2 hours; tachycardia greater than 100 beats per minute for 2 hours (Bone et al. 1989).

Statistics

Assumption of normality was tested with a one-sample Kolmogorov-Smirnov test. Data are expressed as mean (SEM) or median (range), as appropriate. Parametric and non-parametric data were compared, using the unpaired Student's t-test and Mann-Whitney U-test, respectively. Comparisons with laboratory reference ranges were made with the one-sample Student's t-test. Trends with time were assessed by repeated measures analysis of variance. Correlations were explored by backward forced-entry multiple regression and logistic regression.

Results

Of the 31 patients studied, 25 were operated on. 1 patient from the operated group died 32 hours following surgery (bilateral intramedullary nailing procedures for bilateral femoral shaft fractures) from ARDS/fat embolism syndrome.

Serum IL-6 levels were above control levels ($p < 0.0001$) on admission, slowly returning towards control values over the study period. Serum CRP levels were normal on presentation, subsequently increased, significantly peaking between 24 and 72 hours after injury. CRP subsequently decreased, but was still significantly elevated above controls at day 7, the end of the study period (Figure 1).

Serum IL-6 levels correlated closely with ISS ($r = 0.61$, $p = 0.0002$). A severe injury is considered to be associated with an ISS ≥ 16. Patients with ISS ≥ 16

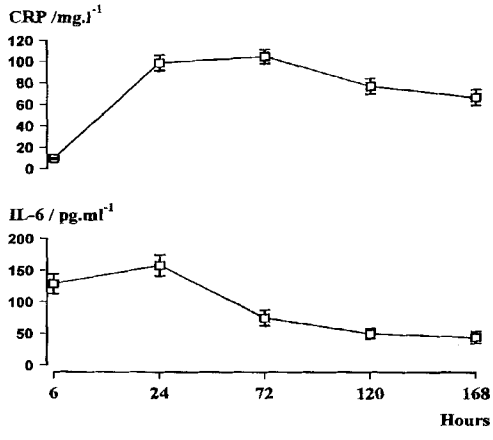

Figure 1. Mean (standard error) levels of IL-6 and C-reactive protein vs time after injury. Pattern of release following blunt orthopedic trauma. The 95% cut-off in the control group is 7.5 mg L^{-1} for CRP and 5 pg mL^{-1} for IL-6. Trend with time $p < 0.001$ (ANOVA)

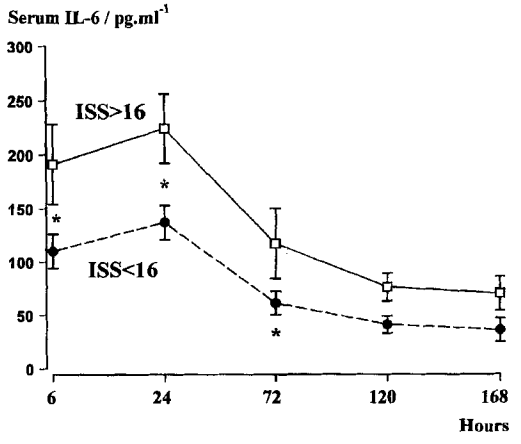

Figure 2. IL-6 high vs low injury severity score. ISS > 16 (n 9) and ISS < 16 (n 22). *$p < 0.05$

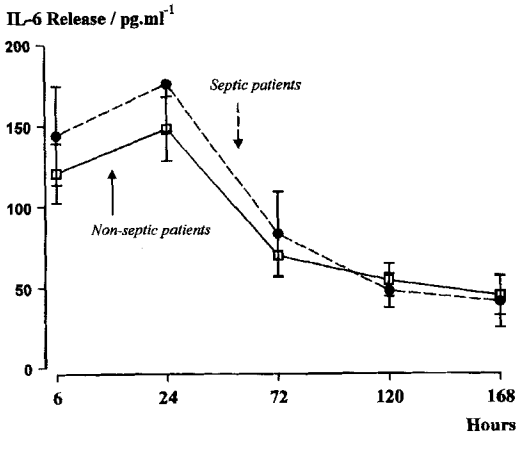

Figure 3. IL-6 concentration vs time after injury. Septic patients (n 9) and non-septic patients (n 15).

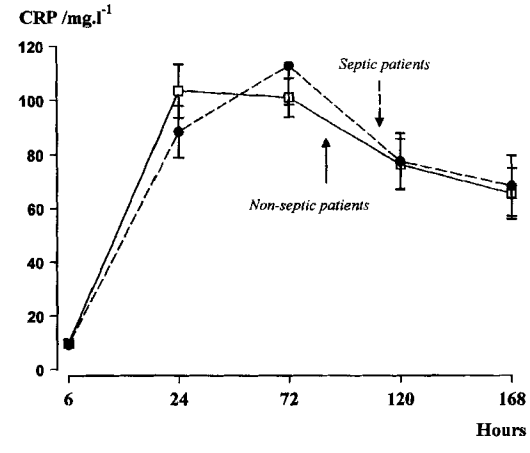

Figure 4. CRP concentration vs time after injury. Septic patients (n 9) and non-septic patients (n 15).

had higher serum IL-6 than those with moderate injury ($p = 0.01$) (Figure 2). No correlation was found between CRP and ISS at any time-point.

Table 2. Types of surgical procedures

Procedure	Uncomplicated	Septic
Subclavian artery repair	0	1
IM nailing femur	12	4
IM nailing tibia	3	1
ORIF forearm	0	1
ORIF humerus, splenectomy	0	1
ORIF pelvis	0	1
Total	15	9

The surgical procedures were all performed within 24 hours of injury. The serum CRP levels in patients who required surgery were higher than those in the non-surgery group 120 hours after the trauma ($p = 0.04$). Peak serum CRP was higher in the operated group. The peak IL-6 levels were also higher in the operated group ($p = 0.03$). 2 subgroups of patients were identified: group A, operated within 6 hours of injury; and group B, operated between 6 and 24 hours following injury. Serum IL-6 and CRP levels at all time-points after 6 hours were similar between group A and group B patients.

9 patients developed sepsis, all made a good recovery. These 9 patients formed the septic group (Table 2). The early levels of serum IL-6 in the septic group

were higher (until day 3), but did not reach statistical significance. Similarly, the CRP in the septic group was not significantly different from the non-septic group (Figures 3 and 4).

Discussion

We describe the normal pattern of IL-6 and CRP release after injury and subsequent surgical treatment in blunt trauma patients: the levels of IL-6 rapidly rose and those of the subsequently synthesized acute phase protein CRP increased later. In our patients shortly after trauma, serum CRP was similar to our reference range, rising within 24 hours of injury and reaching maximum levels on day 3 of the study. There was no biochemical evidence of liver damage in consequence of the trauma. This pattern of release reflects the cytokine-dependent induction of CRP synthesis, unlike other acute phase proteins (Whicher and Evans 1990). Although the degree of CRP response was greater in the operative group than in the nonoperative group, there was no relationship between CRP and the ISS. This finding agrees with those of Gosling and Dickson (1992) and Yoon et al. (1993).

Moody et al. (1985), studying burn patients, reported the highest serum CRP concentrations were in patients with the most severe injuries and that prolonged CRP elevations were associated with sepsis.

However, Colley et al. (1983) reported a poor relationship between serum concentration of CRP and the extent of tissue damage, the response being considered an "all-or-nothing" phenomenon.

In our study, IL-6 was released shortly after trauma and its levels were significantly higher than in controls. Prior to the presentation in the trauma room, where the first blood sample was taken, none of the patients studied had blood transfusions. A correlation between initial IL-6 levels and ISS was observed. These findings confirm those of Cruickshank et al. (1990), Pullicino et al. (1990) and Giannoudis et al. (1997).

Waydhas et al. (1996) have reported that surgery may act as a second insult and inflammatory stimulus. In our study, the levels of both IL-6 and CRP were higher in the group requiring surgical treatment. Patients operated within 6 hours of injury and those operated after 6 hours, but within 24 hours, showed similar patterns of CRP or IL-6 release. No later procedures were performed in these patients.

A raised IL-6 level has previously been associated with unexpected postoperative complications, poor outcome in septic patients and the development of posttraumatic infections or multiple organ failure syndrome after polytrauma (Waage et al. 1989, Burgie et al. 1992, Bank et al. 1995). In our study, serum IL-6 in the septic group of patients was higher up to day 4 than in the nonseptic group, but this trend did not reach significance.

Elevated CRP has been shown to reflect infection after major joint replacement (Niskanen et al. 1996) while serial measurements are recommended to assess posttraumatic or postoperative deep wound infections (Kallio et al. 1990). In our study, serum CRP levels in the septic group were higher on day 3 and at the end of the study period but did not reach statistical significance.

Our findings show that, following blunt trauma, the serum IL-6 increases immediately and correlates with ISS. CRP increase in within 24 hours, but no correlation with ISS was noted. The increase in these markers is similar to that after elective surgery and represents the normal inflammatory response to injury. The IL-6 and CRP are insensitive markers of additional sepsis at this stage.

Acknowledgments

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