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Interleukin-6 and its soluble receptor during acute meningococcal infections: Effect of plasma or whole blood exchange

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Objectives: To determine the pattern of the soluble interleukin (IL)-6 receptor during acute meningococcal infections and recovery phases, and to measure the effect of plasma or whole blood exchange on the plasma concentrations of these mediators.

Design: Prospective, descriptive patient study.

Setting: University hospital intensive care unit.

Patients: Patients with bacteriologically proven meningococcal infections were entered in the study. Three groups were formed: a) patients with meningitis without shock (group A); b) patients with meningitis and shock (group B); and c) patients with shock only (group C).

Interventions: Part (n = 9) of the patients with shock underwent plasma or whole blood exchange.

Measurements and Main Results: Serum concentrations of interleukin-6 and soluble IL-6 receptors were determined sequentially during the acute and recovery phases. Peak concentrations of IL-6 were highest in group C, followed by group B and group A. Soluble IL-6 receptor concentrations showed an opposite pattern and were all below normal. Soluble IL-6 receptor concentrations were negatively correlated with the IL-6 concentrations. During recovery, IL-6 rapidly decreased and soluble IL-6 receptors increased to supranormal concentrations, after which concentrations returned to normal. Plasma or whole blood exchange did not significantly influence IL-6 concentrations but did increase the soluble IL-6 receptor concentration directly after an exchange session followed by a rapid decrease.

Conclusions: Soluble IL-6 receptor concentrations are low in acute meningococcal infections. Plasma or whole blood exchange temporarily increases these concentrations. It needs to be determined whether the effect of this therapy is beneficial to the patient.

Key Words: interleukin-6; Neisseria meningitidis; plasma exchange; interleukin-6 receptor; bacterial infection; mediators; cytokines; infection; critical illness

Among the acute bacterial infections, meningococcal infections are one of the most life-threatening diseases. The Gram-negative bacterium, Neisseria meningitidis, can induce a clinical disease that varies from meningitis to a rapidly deteriorating septic syndrome with severe shock and a mortality rate of 30% to 70% (1). High concentrations of the proinflammatory cytokines, interleukin (IL)-6 and tumor necrosis factor (TNF), are found in these patients, and these cytokine concentrations correlate with disease severity and outcome (2–4). The role of IL-6 in the pathogenesis of septic shock has not been elucidated. An important function of IL-6 is presumably to act as a mediator between damaged tissues and the liver, where it is a potent inducer of acute-phase proteins.

Soluble receptors for various cytokines have been shown to modulate the biological effects of their ligand (5). TNF activity, for example, is decreased in the presence of soluble TNF receptor by competitive inhibition (6); in contrast, low concentrations of soluble TNF receptor can increase the effects of TNF by prolonging its half-life (7).

Soluble IL-6 receptor, however, seems to play a different modulating effect on IL-6 activity. The ligand binding part of the cellular IL-6 receptor (gp80) is either shed from the surface or secreted by the cell (8, 9). A complex of the soluble IL-6 receptor and IL-6 can interact with the signal transducing part of the cellular IL-6 receptor (gp130) and generate a signal into the cell. Through this mechanism the presence of soluble IL-6 receptor enhances the biological activity of IL-6 (10, 11). Studies on the patterns and effects of this soluble receptor are necessary to understand the biological significance of this phenomenon.

We studied the pattern of IL-6 and its soluble receptor in the acute and recovery phases of meningococcal infections. A number of the study patients underwent plasma or whole blood exchange, which possibly improves clinical signs of sepsis and survival (12–15). Because the mechanism behind the beneficial effect of this procedure has yet to be elucidated, the effects of plasma or whole blood exchange on the patterns of the plasma concentrations of IL-6 and soluble IL-6 receptor were also measured in this study.

MATERIALS AND METHODS

Patients with bacteriologically proven meningococcal infections admitted to the intensive care unit.
were included in the study. Cultures for \textit{Neisseria meningitidis} had to be positive from at least one of the following sites: blood, cerebrospinal fluid (CSF), and/or skin lesion. According to Halstensen et al. (16), three groups were discriminated. Group A (n = 13) showed a meningococcal infection of the central nervous system without shock; group B (n = 5) showed a meningococcal infection of the central nervous system plus shock; and group C (n = 5) showed no sign of inflammation of the central nervous system but only shock. Infection of the central nervous system was defined as a leukocyte count of >100 cells x 10^9/L in CSF. During the first hours after admission, shock was defined prospectively as a systolic blood pressure of <100 mm Hg in patients >14 yrs of age, <85 mm Hg in children <14 yrs of age, and <75 mm Hg in patients <4 yrs of age. Demographics of the patients are given in Table 1. Four patients in group B and all patients in group C underwent plasma or whole blood exchange (12). In patients with a body weight of <25 kg, 50 to 60 mL of whole blood/kg were exchanged each session. In patients with body weight of ≥25 kg 30 to 40 mL plasma/kg were exchanged with fresh frozen plasma each session. The exchange was started as soon as possible after admission and was repeated after 12 hrs. When the patients' condition remained critical, the exchange was repeated 24 and 48 hrs later.

After admission, serum samples were collected serially during the first day and each following day. Blood was allowed to clot before centrifugation, and serum was stored at −20°C until tested. Samples were also taken directly before and after the exchange of plasma or blood.

Soluble IL-6 receptor and IL-6 concentrations were determined by enzyme-linked immunosorbent assays (ELISAs) (17). These ELISAs are not influenced by the presence of soluble IL-6 receptor in the IL-6 ELISA or IL-6 in the soluble IL-6 receptor ELISA (17). Detection limits for the IL-6 ELISA and the soluble IL-6 receptor ELISA were 20 pg/mL, and 0.4 ng/mL, respectively. Concentrations in normal volunteers for IL-6 and soluble IL-6 receptor are <20 pg/mL and 76.6 ± 19.3 (sb) ng/mL, respectively (17).

**Statistical Analysis.** Differences between groups were calculated with the Wilcoxon rank-sum test. Differences were considered significant with a two-tailed \( p \) value of <.05. Relationships between IL-6 and soluble IL-6 receptor were calculated using the Spearman correlation coefficient.

**RESULTS**

Figure 1 shows the peak concentrations of IL-6 and concomitant soluble IL-6 receptor concentration for each individual patient subdivided into the three groups. Median IL-6 peak concentrations were 703 pg/mL, 6942 pg/mL, and 120,800 pg/mL in group A (meningococcal infection of the central nervous system without shock), group B (meningococcal infection of the central nervous system with shock), and group C (meningococcal infection of the central nervous system with shock but no signs of inflammation of the central nervous system), respectively. For soluble IL-6 receptor, the concentrations were 56 ng/mL in group A, 12 ng/mL in group B, and 7 ng/mL in group C. The peak values for IL-6 differed significantly between groups A and B (\( p = .0009 \)), and groups A and C (\( p = .0002 \)), but not between groups B and C (\( p = .31 \)). For soluble

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**Table 1. Patient demographics**

<table>
<thead>
<tr>
<th>Group</th>
<th>Male/female</th>
<th>Age (yr)</th>
<th>Prehospital disease period (hr)</th>
<th>Mortality (n)</th>
<th>PE/BE (n/n)</th>
<th>Leukocytes in CSF (10^9/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group A</td>
<td>5/8</td>
<td>15 (2-64)</td>
<td>23.5 (12.5-84)</td>
<td>1</td>
<td>0/0</td>
<td>21674 (3900-70000)</td>
</tr>
<tr>
<td>Group B</td>
<td>3/2</td>
<td>15 (0-17)</td>
<td>14 (5-30)</td>
<td>1</td>
<td>3/1</td>
<td>417 (107-6000)</td>
</tr>
<tr>
<td>Group C</td>
<td>4/1</td>
<td>8 (2-18)</td>
<td>14 (7-16.5)</td>
<td>2</td>
<td>3/2</td>
<td>14 (1-45)</td>
</tr>
</tbody>
</table>

*Male/female n/n* = number of males/number of females of each individual patient; medians are indicated (A vs. B, and A vs. C; \( p < .001 \)).

Group A: meningococcal infection of the central nervous system without shock; Group B: meningococcal infection of the central nervous system with shock; Group C: meningococcal infection of the central nervous system with shock but no signs of inflammation of the central nervous system.
IL-6 receptor concentrations, the differences between groups A and B, and groups A and C were significant \( (p = .0009 \text{ and } .0005, \text{ respectively}) \). There was no statistically significant difference between groups B and C \( (p = .42) \).

All patients in each group had detectable IL-6 concentrations on entry in the study which gradually decreased during recovery. In contrast, soluble IL-6 receptor concentrations were low on admission and increased to supranormal concentrations before leveling off to normal concentrations. IL-6 concentrations returned to normal after \(-48\) hrs. Soluble IL-6 receptor concentrations, however, returned to normal ranges only after 120 to 160 hrs.

There was a strong negative correlation between IL-6 and soluble IL-6 receptor concentrations. For the peak concentration of IL-6 and its relationship with soluble IL-6 receptor concentration, the Spearman correlation coefficient was \(-0.82\).

Figure 2 shows the patterns of IL-6 and Figure 3 illustrates the patterns of soluble IL-6 receptor values over time in all patients of each group. In patients who underwent plasma or whole blood exchange, the sessions are indicated by thick lines. Soluble IL-6 receptor concentration is temporarily increased after a plasma or whole blood exchange session. At the onset of the disease, when IL-6 concentrations are high, this increase of soluble IL-6 receptor concentration is quickly nullified. At low IL-6 concentrations, the effect of plasma exchange is followed by an increase in the soluble IL-6 receptor concentration. This phenomenon can be visualized by the calculation of the change of soluble IL-6 receptor concentration per hour after an exchange session by the following formula: 

\[
\text{(soluble IL-6 receptor)}_B - \text{(soluble IL-6 receptor)}_A / \text{hrs between A and B}, \] 

where \( \text{(soluble IL-6 receptor)}_A \) is the concentration of soluble IL-6 receptor in ng/mL measured just after an exchange session, and \( \text{(soluble IL-6 receptor)}_B \) is the concentration of soluble IL-6 receptor in ng/mL measured in the next sample after A. Negative changes, therefore, mean that the soluble IL-6 receptor concentration decreases after an exchange session. Positive changes indicate that the soluble IL-6 receptor concentration increases after the exchange session.

Figure 4 shows the calculated changes in relation to the IL-6 concentration at point A. The Spearman correlation coefficient is \(-0.81 \ (p < .0001)\).

**DISCUSSION**

Our data confirm previous studies showing increasing IL-6 concentrations with increasing severity of disease (group C > B > A) \( (3) \). In contrast, increasing severity of meningococcal disease is negatively correlated with soluble IL-6 receptor concentrations. A strong negative correlation is found between IL-6 and soluble IL-6 receptor concentrations, indicating a close relation of these two parameters.

Recently, we \( (18) \) reported that the soluble IL-6 receptor concentration was decreased in patients with sepsis compared with healthy humans. Here, we confirm this finding for patients with meningococcal infection. Patients from group A without septic symptoms also showed low concentrations of soluble IL-6 receptor, indicating that sepsis is not obligatory for soluble IL-6 receptor values to be decreased. The concentration of IL-6 appears to be the main determinant for the soluble IL-6 receptor concentration, which is
in agreement with the negative correlation between concentrations of IL-6 and soluble IL-6 receptor (Figs. 2 and 3).

The supranormal concentrations of soluble IL-6 receptor that we see in patients with undetectable concentrations of IL-6 could be explained by a delay in the regulation of soluble IL-6 receptor concentrations. Assuming that under conditions with high IL-6 concentrations, soluble IL-6 receptor or IL-6/soluble IL-6 receptor complexes are rapidly cleared from the circulation, a counter regulation starts leading to increased production or shedding of soluble IL-6 receptor. Temporarily increased soluble IL-6 receptor concentrations may be the result of continuing high IL-6 production and reduced clearance of soluble IL-6 receptor.

The concentrations of soluble IL-6 receptor in the patients undergoing plasma or whole blood exchange are markedly increased after an exchange session. This finding can be explained by the fact that plasma with a relatively low concentration of soluble IL-6 receptor is partially replaced with plasma containing normal concentrations of soluble IL-6 receptor. Plasma soluble IL-6 receptor concentration of one patient, for instance, was 5 ng/mL before and 30 ng/mL after the first plasma exchange session with 2400 mL of fresh frozen plasma. If the mixture of donor blood with the patient’s blood after each portion is taken into account, and the plasma soluble IL-6 receptor concentration of the donor blood is estimated to be 80 ng/mL, the concentration in the patients plasma would be ±65 ng/mL at the end of the session. The difference with the actual concentration could be the result of ongoing clearance of soluble IL-6 receptors in either free or complexed form with IL-6. Plasma or whole blood exchange did not significantly influence the IL-6 concentration. It needs to be determined whether the increase in soluble IL-6 receptor concentrations is of benefit for the patient, because an increase of the biological activity of IL-6 by binding to soluble IL-6 receptors may be harmful to the patient. However, beneficial effects of plasma or whole blood exchange have been reported. High concentrations of cytokines may not be the only and only determinant for the outcome of sepsis patients, as suggested by lack of efficacy of cytokine antagonists in clinical trials (19).

After an exchange session, the soluble IL-6 receptor concentration rapidly decreases, which indicates that the turnover of soluble IL-6 receptor is high. The rate of decrease is dependent on the IL-6 concentration. In an early phase of the illness with high IL-6 concentrations, soluble IL-6 receptors are more quickly cleared from the exchanged plasma than in a later phase when plasma IL-6 concentrations are low. At low IL-6 concentrations, soluble IL-6 receptor concentrations do not further decrease but increase after an exchange session. This finding indicates that IL-6 is an important factor for the clearance of soluble IL-6 receptor. In vitro studies, however, are not conclusive about the effect of IL-6 on the IL-6 receptor expression or soluble IL-6 receptor production. Some reports (20, 21) indicated a decrease of the expression of IL-6 receptor, but another report (22) showed no effect. These studies (20–22) were performed on different cell types under different experimental conditions. Very little is known about the mode of clearance of the soluble IL-6 receptor. Minute amounts of soluble IL-6 receptor are found in the urine, and moderate renal impairment does not influence the soluble IL-6 receptor concentration (17). However, preliminary results in patients with almost absent renal function do indicate a role for the kidneys in maintaining soluble IL-6 receptor concentrations (Frieling et al., unpublished observation).

To our knowledge, this study is the first report to strongly point to an important role for IL-6 in maintaining soluble IL-6 receptor concentrations and in clearance of the soluble IL-6 receptor in patients. Studies on the underlying mechanisms, however, still have to be done.

REFERENCES


