ORIGINAL ARTICLE

Neutrophil CD64 (FcγRI) expression is a specific marker of bacterial infection: A study on the kinetics and the impact of major surgery

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Abstract

Neutrophil CD64 expression is a diagnostic marker for the early detection of bacterial infections. The aim was to investigate the kinetics of neutrophil CD64 expression during bacterial infection and the possible impact of surgical trauma. Blood samples were collected daily during 3 d after admission for analysis by flow cytometry of the surface expressions on neutrophils and monocytes of CD64, CD16, CD32, CD11b/CD18 and CD35, and analysis of serum CRP and blood WBC. Serum concentrations of IFNγ, G-CSF, IL-6 and IL-8 were also analysed in adults. Eight children and 19 adult patients with bacterial infections, 12 patients admitted for hip-arthroplasty because of coxarthrosis and 30 healthy adults were studied. Neutrophil CD64 was increased all 3 d after start of treatment (p<0.0001) in children and adults with bacterial infections. The postoperative increase after surgery was less than the increase seen during bacterial infections (p<0.0001). CRP, G-CSF, IL-6 and IL-8 were raised both in bacterial infections and after surgery. Our results indicate that the expression of CD64 on neutrophils is a specific sign of bacterial infections. Neutrophil expression of CD64, therefore, seems to be a promising tool for the early detection of bacterial infections even during surgery.

Introduction

Fcγ receptor I (FcγRI, CD64) is a high affinity receptor normally expressed by monocytes and only to a small extent by neutrophils [1]. During bacterial infections, however, the neutrophil expression of CD64 is markedly increased [2–4]. In a previous study we found that neutrophils from even very preterm, newborn infants, during bacterial infections increase the expression of CD64 to the same extent as do neutrophils from children and adult patients [5]. In addition, this increased expression seemed to be particularly prominent in bacterial infections, although minor increases are also be seen in other conditions [6–11]. According to these findings we hypothesized that the up-regulation of CD64 on neutrophils is caused specifically by bacterial infections, rendering the measurement of this receptor expression a possible diagnostic test for early detection of severe bacterial infections. Such a test would be especially useful in clinical situations where today’s tests are unable to discriminate between bacterial infections and inflammation caused by other mechanisms, infectious as well as non-infectious.

The aim of this study was to investigate the kinetics of the expression of CD64 on neutrophils during the course of bacterial infections. Mainly for comparison, the expressions of the 2 other Fcγ-receptors, CD16 (FcγRIII) and CD32 (FcγRII), as well as the complement receptors CD11b/CD18 (CR3) and CD35 (CR1), on neutrophils and monocytes were also investigated, as was the expression of CD64 on monocytes. To further investigate the hypothesized specificity of markedly increased neutrophil CD64 expression as a response to bacterial infections alone, we also investigated the expression of CD64 as well as the other receptors in response to a well standardized surgical trauma i.e. hip-arthroplasty, in which an inflammatory response is known to occur [12]. IFNγ and G-CSF are the 2 cytokines shown to be able to induce the expression of CD64 on neutrophils.
To further elucidate the mechanisms involved in the up-regulation of CD64 during bacterial infections, the serum concentrations of IFNγ, G-CSF, IL-8 and IL-6 were also analysed. Our results indicate that the expression of CD64 on neutrophils is a more specific sign of bacterial infections than conventionally used markers such as CRP in plasma and blood neutrophil counts.

**Material and methods**

**Subjects**

Three groups of patients and 1 group of healthy subjects were studied. Group 1 consisted of 8 children, aged 8 d to 7 y who were hospitalized because of bacterial infection (Table I). The diagnosis was based on typical clinical signs, signs of focal infections, positive culture in urine and/or blood, and chest X-ray examination. None of the children was blood culture positive. Blood samples for analysis of the receptor expression were collected within 20 h after admittance (the following morning) and the 2 following d.

Group 2 consisted of 19 adult patients, median age 62 y (24–89 y) who were hospitalized because of bacterial infection (Table II). The diagnosis was based on typical clinical signs, signs of focal infections, positive culture in relevant biological material including blood and chest X-ray examination. Blood samples for analysis of receptor expression and serum markers were collected within 20 h after admittance (the following morning) and the 2 following d.

Group 3 consisted of 12 adult patients (8F, 4M), mean age 58.4 y (48–76 y), admitted to Uppsala University Hospital, the Department of Orthopaedics, for total hip-joint replacement due to coxarthrosis. Blood samples for analysis of receptor expression were collected preoperatively and the 3 d following surgery. In addition a serum sample was collected 6 h after start of surgery. None of these patients had a former history of inflammatory joint disease or malignancy, and none of them had ongoing treatment with steroids or non-steroid anti-inflammatory drug. The surgical procedure was uniform for all patients. The hip was dissected through an anterolateral approach according to Hardinge [16]. After resection of the femoral head, the acetabulum and femur were prepared, respectively, for their components, the acetabular cup and the femoral stem. In 9 patients (6F, 3M), the components were fixated without cement by ‘press-fitting’. Duration of surgery for these patients was measured to mean 81 min (60–100 min).

In 3 patients (2F, 1M), the components were fixated with cement, Palaco-gentamicin TM, (Schering-Plough, Europe). For these patients duration of surgery was mean 106 min (100–120 min). The acetabular and femoral components were cemented consecutively and the total time of hardening was about 30 min per operation. While hardening the cement, polymethylmethacrylate (PMMA) has a thermal influence on the surrounding tissues with a temperature about 60 °C as a peak. Routinely 3 doses of Ekvacillin TM, (Astra, Sweden), 1 g each, is given on the d of surgery.

Group 4 comprised 30 healthy adults (blood donors and laboratory technicians) from whom blood was drawn by venous puncture.

**Ethics**

The study was performed with permission from the Ethics Committee, Faculty of Medicine, Uppsala University. The blood samples were collected after informed consent from the parents and patients, respectively.

**Methods**

**Preparation of leukocytes.** Leukocytes were prepared as previously described [17,18]. Briefly, 1 ml heparinized blood was mixed with an equal volume of 0.4% paraformaldehyde in phosphate-buffered saline (PBS) and incubated for 4 min at 37 °C. The erythrocytes were then lysed by incubation with 40 ml 0.85% (w/v) NH₄Cl in Tris-HCl-buffer (Tris(hydroxymethyl)-aminomethan 0.01 mol/l, pH 7.4) for another 15 min. Finally the cells were washed twice with PBS containing sodium citrate (0.012 mol/l) and human serum albumin (HSA) (0.1%, w/v) and diluted to the concentration of 1.7–2.5 × 10⁶/ml.

**Labelling of leukocytes with antibodies to cell surface antigens.** 50-μl samples of the leukocyte suspension were mixed with optimally titrated FITC- or PE-labelled mouse monoclonal antibodies (mab) against CD11b, CD64, (Immunotech S.A., Marseille, France), CD16, CD14, CD18 (Dakopatts,
Glostrup, Denmark), CD32 (Becton-Dickinson, San Diego, CA, USA) and CD35 (Serotec Inc, Raleigh, NC, USA) and incubated for 30 min at 4°C. After incubation, the cells were washed twice with PBS and thereafter diluted with 200 μl PBS with sodium citrate and HSA. Leukocytes were also labelled by an identical procedure with negative isotype controls for mouse IgG1 and IgG2 (Dako-patts, Glostrup, Denmark). After labelling, the cells were kept on ice until analysis.

Flow cytometry. Flow cytometric analysis was performed on an EPICS II Profile or EPIC MCL-XL flow cytometer (Coulter Company Inc., Hialeah, FL, USA). Neutrophils were identified based on their forward scatter/side scatter (FSC/SSC) dot-plot profile and monocytes based on the forward scatter/side scatter (FSC/SSC) dot-plot profile and positive staining with anti-CD14. The neutrophil and monocyte populations were gated and the FITC-fluorescence measured. The intensity of fluorescence above background of neutrophils and monocytes was determined and expressed as mean fluorescence intensity (MFI) of CD64 expression, i.e. mean +2 SD, and also given as relative number of positive cells (%), defined as the relative number of cells that expressed CD64 to a higher extent than the isotype-defined background (Figure 1a, b).

Table II. Adult patients with bacterial infections.

<table>
<thead>
<tr>
<th>Patient</th>
<th>Gender (m/f)</th>
<th>Age (y)</th>
<th>Diagnosis</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>m</td>
<td>59</td>
<td>Pneumonia/septicaemia (S. pneumoniae)</td>
</tr>
<tr>
<td>2</td>
<td>m</td>
<td>79</td>
<td>Pneumonia and pyelonephritis (β-strept. Group B)</td>
</tr>
<tr>
<td>3</td>
<td>f</td>
<td>67</td>
<td>Pyelonephritis (Klebsiella pneumoniae)</td>
</tr>
<tr>
<td>4</td>
<td>m</td>
<td>79</td>
<td>Pneumonia</td>
</tr>
<tr>
<td>5</td>
<td>m</td>
<td>78</td>
<td>Erysipelas</td>
</tr>
<tr>
<td>6</td>
<td>f</td>
<td>50</td>
<td>Erysipelas/Phlegmone</td>
</tr>
<tr>
<td>7</td>
<td>m</td>
<td>55</td>
<td>Pyelonephritis (E. coli)</td>
</tr>
<tr>
<td>8</td>
<td>m</td>
<td>35</td>
<td>Meningitis/septicaemia (N. meningitides type B)</td>
</tr>
<tr>
<td>9</td>
<td>m</td>
<td>24</td>
<td>Pneumonia</td>
</tr>
<tr>
<td>10</td>
<td>m</td>
<td>57</td>
<td>Pneumonia/septicaemia (S. pneumoniae)</td>
</tr>
<tr>
<td>11</td>
<td>f</td>
<td>75</td>
<td>Pneumonia (H. influenzae / S. pneumoniae)</td>
</tr>
<tr>
<td>12</td>
<td>f</td>
<td>62</td>
<td>Pyelonephritis (E. coli)</td>
</tr>
<tr>
<td>13</td>
<td>m</td>
<td>31</td>
<td>Erysipelas, bursitis (prepatellar)</td>
</tr>
<tr>
<td>14</td>
<td>m</td>
<td>24</td>
<td>Meningitis/septicaemia (N. meningitides type B)</td>
</tr>
<tr>
<td>15</td>
<td>m</td>
<td>72</td>
<td>Erysipelas (β-strept. Group A)</td>
</tr>
<tr>
<td>16</td>
<td>f</td>
<td>89</td>
<td>Pneumonia, COPD</td>
</tr>
<tr>
<td>17</td>
<td>f</td>
<td>55</td>
<td>Dental abscess/septicaemia (Acinetobacter species)</td>
</tr>
<tr>
<td>18</td>
<td>m</td>
<td>83</td>
<td>Cholangitis/septicaemia (Cl. perfringens)</td>
</tr>
<tr>
<td>19</td>
<td>m</td>
<td>73</td>
<td>Erysipelas and pyelonephritis (Proteus species)</td>
</tr>
</tbody>
</table>

Figure 1. Analysis of receptor expression as illustrated by measurement of neutrophil expression of CD64 and CD32. A. Isotype control antibodies are used to establish the background level of fluorescence. The level of positive cells is set to 1.0% and the MFI of the whole population of cells is determined. B. Expression of CD64 above background is measured with the same settings, which gives the relative number of positive cells. The MFI of CD64 expression is calculated as MFI of the whole populations of cells subtracted by background MFI obtained under A. C. Expression of CD32 as an example of receptors expressed by the whole population of neutrophils. The MFI of the 100% positive cells is given.
Serum concentrations of CRP, G-CSF, IFNγ, IL-6 and IL-8. Measurements of the serum concentrations of G-CSF, IFNγ, IL-6 and IL-8 were performed using commercial enzyme-linked immunosorbent assay (ELISA) kits (R&D Systems, Abingdon, UK). The detection limits of the respective assays were 20 ng/l (G-CSF), 8 ng/l (IFNγ), 0.7 ng/l (IL-6) and 0.8 ng/l (IL-8). The reference limits as determined by the manufacturer were <39 ng/l (G-CSF), <8 ng/l (IFNγ), <12.5 ng/l (IL-6), and <25 ng/l (IL-8). CRP was measured by an immunonephelometric assay at the Department of Clinical Chemistry, University Hospital, Uppsala, Sweden.

Statistics

Non-parametric statistics were used throughout the paper. For comparisons between groups the Mann-Whitney U-test was used and for paired comparisons the Wilcoxon test was used. All calculations were performed by means of the statistical softwares, Statistica for Windows (Statsoft Inc., Tulsa, OK, USA) or Medcalc (Medcalc Software, Mariakerke, Belgium).

Results

Neutrophil expression of CD64 in bacterial infections and after surgical trauma

The expression of CD64 on neutrophils from children as well as from adult patients with bacterial infections was significantly higher compared with neutrophils from healthy adults \((p < 0.001)\). The neutrophil expression of CD64 in adult patients undergoing surgery was also increased compared with healthy adults \((p < 0.001)\). The postoperative increase was, however, markedly less than the increase seen during bacterial infections \((p < 0.001)\) (Figure 2). Neutrophils from children with bacterial infection expressed CD64 to a higher extent than neutrophils from adult patients with a bacterial infection \((p < 0.001)\). In 3 of the 4 subjects with CD64 expressions below 60%, the expressions approached 100% the day after.

Kinetics of the neutrophil expression of CD64 in bacterial infections and after surgical trauma

Although declining somewhat over time, the expression of CD64 on neutrophils from both adults and children with bacterial infections was significantly increased all 3 d after start of antibiotic treatment compared with the same expression on neutrophils from healthy individuals \((p < 0.0001)\). The result was the same measured both as percentage of neutrophils expressing CD64 (Table III) and as density of receptor expression per cell (Figure 3). In the adult patients the decline in neutrophil CD64 expression roughly paralleled the decline in the values for CRP and neutrophil count, respectively. In the children the CD64 expression as well as the neutrophil count tended to normalize more rapidly compared with CRP.

After surgical trauma the CD64 expression on neutrophils was significantly increased on d 1 \((p < 0.006)\) as well as d 2 and 3 \((p < 0.0001)\), with maximum increase on d 2, compared with healthy
adults (Figure 3). Also the percentage of CD64 positive cells was significantly increased on d 1 and 2 (Table III). The values for CRP and blood neutrophil counts were also increased all 3 d, with maximum increase on d 2.

For comparison we also examined the neutrophil expression of CD16, CD32, CD11b, CD18 and CD35 during the first 3 d after start of antibiotic treatment in patients with bacterial infections and after surgical trauma. The receptors were not expressed to a level markedly above the 97.5 percentile for those in healthy adults, except for the CD35 expression on d 1 and 3 on neutrophils from children, and d 2 on neutrophils from adults with a bacterial infection. In children and adults with bacterial infections the neutrophil expression of CD32 was significantly increased during the first 2–3 d after start of treatment, with the greatest

<table>
<thead>
<tr>
<th>D after admission</th>
<th>n</th>
<th>CD32 MFI Median (interquartile range)</th>
<th>CD64 Positive cells (%) Median (interquartile range)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>7</td>
<td>77.8** (66.2–90.2)</td>
<td>99.5*** (97.7–100)</td>
</tr>
<tr>
<td>2</td>
<td>7</td>
<td>87.3*** (82.4–87.5)</td>
<td>92.4*** (78.8–97.5)</td>
</tr>
<tr>
<td>3</td>
<td>5</td>
<td>69.0 (68.1–91.0)</td>
<td>81.1*** (77–92.1)</td>
</tr>
</tbody>
</table>

Significant changes compared with references (A,B) or d 0 (C) are indicated, ** p <0.01, *** p <0.001.
increase on d 2 in both groups (p < 0.001) (Table III). No significant changes in CD32 expression were seen after surgery.

**Kinetics of the monocyte expression of CD64 in bacterial infections and after surgical trauma**

In children with bacterial infections the monocyte expression of CD64 was significantly increased on d 1 (p < 0.001) and then gradually declined (Table IV). On d 3 the expression of CD64 had normalized. In adult patients with bacterial infections the monocyte expression of CD64 was significantly increased on d 1 and still elevated on d 3 (p < 0.01). After surgery the expression of CD64 on monocytes was significantly increased on d 1 to 3 (p < 0.01).

For comparison we also examined the monocyte expression of CD11b, CD18, CD32 and CD35 during the first 3 d after start of antibiotic treatment in patients with bacterial infections and after surgical trauma. In children with bacterial infections the monocyte expression of CD11b and CD35 were both significantly increased on d 1 (p < 0.001) and still significantly elevated on d 3 (p < 0.05) (Table IV). In adult patients with bacterial infections the monocyte expression of CD35 was significantly increased on d 1 and still on d 3 (p < 0.05). The only significant change of the expression of CD11b was a slight increase on d 1 (p < 0.05). After surgery the expression of CD35 was slightly elevated 24 h postoperatively (p < 0.05), but then normalized. Regarding the CD11b expression we found a slight increase in samples collected before start of surgery but no increase postoperatively.

**Serum concentrations of IFNγ, G-CSF, IL-6 and IL-8 in bacterial infections**

The serum concentrations of G-CSF and IL-6 were markedly increased the first d after start of antibiotic treatment and declined rapidly the 2 following d. For IL-8 and IFNγ no increases were found (results not shown).

**Serum concentrations of IFNγ, G-CSF, IL-6 and IL-8 after surgical trauma**

The serum concentrations of G-CSF, IL-6 and IL-8 were increased as soon as 6 h after start of surgery, and stayed elevated for 48 h (G-CSF and IL-8 ) and 72 h (IL-6), respectively (Figure 4). The increase in IL-8 did, however, not exceed the upper reference limit. For IFNγ no increase in the serum concentrations was found (results not shown).

**Example: Patient with postoperative bacterial infection**

To illustrate the possible usefulness of neutrophil CD64 expression in the diagnosis of bacterial infections after surgery, data are presented from the only patient in this study, a 55-yr-old male patient, who suffered from a postoperative infection (a wound infection culture positive for S. epidermidis). The infection was first diagnosed based on clinical

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**Table IV. Cell surface expression of CD11b, CD35 and CD64 on monocytes.**

| A. Infection, children. | | | |
|---|---|---|
| **D after admission** | **n** | **CD11b MFI Median (interquartile range)** | **CD35 MFI Median (interquartile range)** | **CD64 MFI Median (interquartile range)** |
| 1 | 7 | 40.1*** (27.4–40.8) | 15.8*** (13.1–20.1) | 56.2*** (47.7–64.5) |
| 2 | 7 | 31.0** (17.0–52.8) | 8.7*** (5.4–27.4) | 45.2** (38.8–53.3) |
| 3 | 5 | 52.1* (33.7–56.2) | 4.8* (4.3–21.9) | 45.2** (25.7–45.8) |

| B. Infection, adults. | | | |
|---|---|---|
| **D after admission** | **n** | **CD11b MFI Median (interquartile range)** | **CD35 MFI Median (interquartile range)** | **CD64 MFI Median (interquartile range)** |
| 1 | 19 | 28.3* (15.2–37.6) | 10.6*** (7.5–18.9) | 49.0*** (41.0–75.6) |
| 2 | 19 | 21.7 (18.6–29.1) | 8.6*** (7.2–11.8) | 47.1*** (34.2–60.4) |
| 3 | 5 | 19.0 (13.7–31.0) | 6.6* (5.9–10.2) | 44.8** (29.6–54.0) |

| C. Surgery, adults. | | | |
|---|---|---|
| **D post-surgery** | **n** | **CD11b MFI Median (interquartile range)** | **CD35 MFI Median (interquartile range)** | **CD64 MFI Median (interquartile range)** |
| 0 | 11 | 32.9* (23.7–51.5) | 6.4 (4.9–17.5) | 32.1 (29.0–42.2) |
| 1 | 11 | 28.7 (20.1–41.9) | 14.6* (11.4–18.8) | 44.0** (40.5–53.9) |
| 2 | 11 | 22.3 (17.4–32.3) | 13.5 (10.8–19.3) | 46.7** (40.7–54.7) |
| 3 | 11 | 17.7 (15.2–30.1) | 11.4 (8.0–14.6) | 44.9** (37.4–50.8) |

Significant changes compared with references (A,B) or day 0 (C) are indicated, *p < 0.05, **p < 0.01, ***p < 0.001.
symptoms approximately 48 h after start of surgery, and treatment was initiated with flucloxacillin p.o. for 3 weeks. As shown in Figure 5a, marked elevation in neutrophil CD64 expression was seen compared with the small increase caused by the surgery itself 24 h earlier. The elevation became even more pronounced at the time the clinical diagnosis was first suspected. In contrast, the CRP values did not differ from the levels caused by the operation, and consequently were of no use for the diagnosis. As illustrated in Figure 5b, also some additional increase in the serum concentrations of G-CSF and

Figure 4. Changes in serum concentrations of IL-6, IL-8 and G-CSF after surgery. Serum concentrations of interleukin-6, interleukin-8 and G-CSF in patients before, and 6, 24, 48 and 72 h after surgery. Results are shown as medians and upper or lower quartiles. Statistically significant differences between values after as compared with before surgery are indicated, *p < 0.05, **p < 0.01, ***p < 0.001.

Figure 5. Changes in inflammation markers in a patient with post-surgery infection Comparison of the changes in a) neutrophil CD64 expression, serum concentration of CRP, white blood cell count; b) serum concentration of IL-6, IL-8 and G-CSF in a patient with post-surgery infection (closed symbols), with the pattern induced by surgery only (open symbols).
IL-6 was noted, while the levels did not differ at all from what was seen after uncomplicated surgery.

Discussion

A pronounced increase in neutrophil expression of CD64 is a typical finding in patients with bacterial infections [2,19,20]. The specificity of this expression was further indicated in this report by our study on patients undergoing surgery, since the changes seen under these conditions were significantly less than those seen in bacterial infections. Taken together with the findings of minor up-regulation of CD64 on neutrophils in serious viral diseases such as those caused by influenza A [10], these findings show that the expression of CD64 on neutrophils is a specific indicator of bacterial infections and also a promising marker for the detection of bacterial infections during the first d after surgery. From this study, however, we cannot exclude that other major surgical trauma would affect the CD64 expression on neutrophils. Nor can we exclude that large haematoma could affect the expression. The rationale for choosing hip surgery patients as the study group and not other patient groups undergoing major surgery such as cardiac surgery, hepatic surgery or large debulking surgery for malignancy, was the fact that this group of patients was a reasonably homogeneous group with regard to the complicating disorders, but also the fact that this group of patients was a reasonably homogeneous group with few other complications. The pattern is seen in spite of the fact that the adult patients with bacterial infections in our study had higher levels of CRP. These results therefore indicate that CD64 expression may be particularly useful as a diagnostic means in children with bacterial infections. This conclusion is further supported by our previous findings of highly up-regulated expressions on cells from preterm infants [28].

The second aim of the study was to investigate the kinetics of neutrophil CD64 expression during bacterial infections as well as after surgery. To fulfil the criteria of a clinically useful diagnostic tool for diagnosis of bacterial infections, the changes in concentration or expression of the actual marker caused by the infection should be clearly recognizable for at least 48 h [29]. The increased neutrophil expression of CD64 during bacterial infections lasted for the whole observation period, which was more than 2 d, rendering this receptor expression a suitable for diagnostic purposes also in this aspect. The importance of this time aspect is best illustrated by the rapid and transient increase of the serum concentrations of IL-6 and G-CSF, which reduces their usefulness as diagnostic tests for bacterial infections.

The expression of CD16 showed no significant changes during bacterial infections or after surgery, which is in accordance with some earlier reports [30,31]. During the course of more serious bacterial infections, however, CD16 is shed from the cell surface, and soluble CD16 (s-CD16) can then be measured in serum, the level of which has been shown to correspond well to the severity of the disease [32]. In such situations a corresponding decreased value for neutrophil CD16 expression is to be expected.

Neutrophil expression of CD11b has also been investigated as a potential marker for the diagnosis of bacterial infections [33–35]. There are, however, diverging results on whether CD11b is up-regulated or not by a bacterial infection, at least in adults. In the present study the neutrophil expression of
CD11b remained unaltered during bacterial infection and after surgery. This might, however, be due to the fact that a special laboratory procedure is needed to detect changes of CD11b expression on neutrophils and monocytes [36]. Neutrophil expression of CD35 usually parallels that of CD11b. We found CD35 to be slightly more sensitive than the CD11b expression, since a small increase was found on d 1 and 3 in children, and on d 2 in adults with bacterial infections.

The increased expression on monocytes of CD64, CD11b and CD35 during the first 2–3 d after start of treatment of a bacterial infection is in accordance with previous results [37]. The significant increase in monocyte expression of CD64 after surgery was reported previously [38], and indicates that activation of CD64 on monocytes is a more unspecific phenomenon than activation of this receptor on neutrophils. It also leads to the conclusion that up-regulation of CD64 on neutrophils and on monocytes at least partly is mediated by different mechanisms. In adult patients with bacterial infection we found G-CSF and IL-6 to be elevated on d 1, which was expected [39,40]. In the postoperative patients G-CSF, IL-6 and IL-8 were all elevated from the first postoperative sample collected 6 h after start of surgery, to 48 h, and for IL-6 to 72 h, after initiation of operation. These results correspond well with earlier reports [41,42]. Despite elevated serum concentrations of G-CSF, IL-6 and IL-8, neutrophils from patients undergoing surgery expressed CD64 only to a limited extent. Consequently, none of these cytokines is likely to be responsible for the significantly higher increase in the CD64 expression seen during bacterial infections. In our patients with bacterial infections, as in patients undergoing surgery, we found no elevation in serum concentrations of IFN-γ, at least not from d 1 and forward. According to this it seems less likely that the powerful stimulation of the neutrophil CD64 expression seen during bacterial infections is caused by IFN-γ alone. IFN-γ and G-CSF are the only 2 cytokines known to induce the expression of CD64 on neutrophils. When administrated in vivo in pharmacological doses both cytokines cause a marked neutrophil CD64 expression [43–45]. In vitro, however, only stimulation of neutrophils with IFN-γ causes a CD64 expression, probably illustrating that the in vivo effect of G-CSF on the CD64 expression is indirect [46,47]. Thus, the mechanisms regulating CD64 expression on neutrophils and monocytes in vivo are still far from understood.

In conclusion, we found neutrophil CD64 expression to be a specific marker for the detection of bacterial infections after a surgical trauma. Other investigations have shown this receptor expression to represent a promising diagnostic test for early detection of severe bacterial infections in the neonatal period as well as in patients with inflammatory diseases. In these clinical situations also a specific test for early detection of severe bacterial infection is much needed. Such a test would improve the possibility for early start of effective anti-microbial treatment and also reduce the unnecessary use of antibiotic treatment, thereby reducing the risk of further development of antimicrobial resistance.

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