Differential Prognosis of Gram-Negative Versus Gram-Positive Infected and Sterile Pancreatic Necrosis: Results of a Randomized Trial in Patients with Severe Acute Pancreatitis Treated with Adjuvant Selective Decontamination

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Results of a previous randomized multicenter trial involving 102 patients with severe acute pancreatitis treated with or without adjuvant selective decontamination (SD) were analyzed additionally with regard to the bacteriologic status of (peri)pancreatic necrosis. The incidence of gram-negative pancreatic infection was significantly reduced in patients treated with SD (P = .004). Once such an infection develops, mortality increases 15-fold (P < .001) in comparison with that for patients with sterile necrosis. Among patients in whom only gram-positive infection of pancreatic necrosis was found, there was no significant increase in mortality. These results were similar in both treatment groups. In addition, the hospital stay was significantly longer in cases of gram-negative infected necrosis. The incidence of gram-positive infected necrosis in patients treated with SD did not increase. Gram-negative pancreatic infection can be prevented with adjuvant SD, thereby reducing mortality among patients with severe acute pancreatitis.

As infectious complications have become the leading cause of mortality and morbidity in cases of acute necrotizing pancreatitis, patients in whom devitalized pancreatic and peripancreatic tissues remain sterile have to be distinguished from others in whom secondary infection of pancreatic necrosis develops (i.e., those with sterile vs. infected necrosis). Sterile pancreatic necrosis, especially in the absence of systemic complications, has a favorable prognosis, with a reported mortality rate of zero to 11% [1–4]. Mortality, which increases sharply with increasing Ranson or Imrie scores on admission, is related to systemic complications that result in multiple organ failure, occurring most frequently during the first 2 weeks of illness [5–10].

Infected necrosis, on the other hand, occurs later during the course of the disease, often proves fatal, and is generally agreed to represent an absolute indication for surgery in an effort to reduce mortality [2, 11–14].

 Cultures in cases of infected necrosis most frequently yield a polymicrobial flora, with a preponderance of gram-negative aerobic bacteria in 50%–70% of cultures, suggesting an enteric origin [2, 5, 12, 15–19]. Gram-positive aerobes (mainly enterococci and staphylococci) are isolated in only 5%–20% of cultures [2, 5, 12, 15–19].

 Patients with severe acute pancreatitis in whom infected necrosis exists are usually dealt with as one group, irrespective of the specific flora cultured. However, because of varying intrinsic pathogenic potential, the prognosis for patients with infected pancreatic necrosis may differ according to the bacteria cultured. In accordance with results of bacteriological analyses of infected pancreatic necrosis, two major groups of patients can be distinguished: those with gram-positive infected necrosis and those with gram-negative infected necrosis. This distinction between patients with infected pancreatic necrosis has not been studied prospectively to date.

 In a recent controlled clinical trial, selective decontamination (SD) was shown to effectively reduce mortality among patients with objective signs of severe acute pancreatitis [5]. However, SD does not prevent gram-positive infection. Subsequent intestinal overgrowth with Enterococcus species, resulting in an increase in gram-positive infections, has been suggested to be a limitation of SD [20].

 An additional analysis of the results of this prospective, controlled clinical study was performed to evaluate for both treatment groups (i.e., the SD group and the control group) a
possible difference concerning mortality between (1) patients with either gram-positive infected necrosis or gram-negative pancreatic infection during the course of the disease and (2) patients in whom pancreatic necrosis remained sterile.

**Patients and Methods**

Between 22 April 1990 and 19 April 1993, 102 patients with objective signs of severe acute pancreatitis were admitted to 16 participating hospitals. The diagnosis of acute pancreatitis had been established on the basis of clinical examination and elevated plasma levels of serum amylase (>1,000 IU/L; normal range, 0–300 IU/L [Phadebas]) or at diagnostic laparotomy (10 patients). All patients had severe acute pancreatitis, according to a multiple-laboratory-criteria score (Imrie score, ≥3) and/or a disease-severity grade of D or E (Balthazar grades) determined by contrast-enhanced CT [7, 21]. Bacteriologically proven infected necrosis at the time of randomization was defined as an exclusion criterion.

The patients were randomly assigned to receive standard treatment (control group: n = 52) or the same treatment plus SD (SD group: n = 50). A 24-hour randomization service was available to randomize patients, with stratification per center. Informed consent was obtained from the patient or relatives by the attending clinician.

The SD regimen consisted of oral administration of colistin sulfate (200 mg), amphotericin (500 mg), and norfloxacin (Noroxin, Merck & Co., West Point, PA; 50 mg) every 6 hours. A sticky paste containing 2% of the three SD drugs was smeared along the upper and lower gums every 6 hours and at the tracheostomy, if present. The aforementioned daily dose also was given in a rectal enema every day.

Short-term systemic prophylaxis with cefotaxime sodium (Claforan, Hoechst-Roussel Pharmaceuticals, Somerville, NJ; 500 mg) was given every 8 hours until gram-negative bacteria were eliminated from the oral cavity and rectum (average, 7.4 days). SD was discontinued as soon as the risk of acquiring a new infection was absent, i.e., the patient was extubated, receiving no supplementary oxygen therapy or infusions, on a regular oral diet, and ambulatory on the ward. A more elaborate outline has been reported previously [5].

**Microbiology**

An ultrasonography or CT-guided fine-needle aspiration (FNA) with subsequent culture was performed if there was clinical suspicion of infected pancreatic necrosis [11]. Clinical suspicion of pancreatic infection was based on the occurrence of fever and leukocytosis (usually lasting at least 2–3 days, during which other sources of infection were excluded), associated with CT findings demonstrating pancreatic necrosis.

The microbial flora of the infected pancreas was carefully monitored. Culture specimens of pancreatic and peripancreatic devitalized tissues (i.e., necrosis) were obtained at every laparotomy and from drainage. They were sent directly to the laboratory and cultured semiquantitatively. If fever (temperature of ≥39°C) was present, blood cultures were performed. Identifications were made following routine microbiological procedures.

Pancreatic necrosis, peripancreatic devitalized tissues, and fluid collections were considered sterile in those patients with a nonseptic course and in those with negative cultures.

**Surgery**

Besides severe intraabdominal hemorrhage or presence of enteric fistulas, reasons for surgery included (1) aspirate cultures that demonstrated development of infected necrosis and (2) rapid deterioration of the patient’s condition toward multiple-organ failure that was resistant to exhaustive intensive treatment (SD group, 16 patients; controls, 24). Results of surveillance cultures of the digestive tract were not taken into account in the decision to perform a reintervention. The decision to perform either percutaneous drainage or relaparotomy was based on ultrasonographic and CT findings as well as findings from the previous laparotomy.

Access to the pancreas was obtained through a median or (preferably) transverse laparotomy. If repeated laparotomies were foreseen, a laparostomy (i.e., ventral open packing of the abdominal cavity) was created, ensuring rapid and easy access to the upper abdominal cavity [12]. Removal of necrotic tissue was mainly performed by means of finger or clamp fraction (i.e., necrosectomy). Infected necrosis is mostly solid, in contrast with a pancreatic abscess, which represents a localized collection of fluid, often encapsulated, that occurs after the pancreatitis has subsided.

**Statistical Analysis**

Percentages and continuous data were compared between groups by means of Fisher’s exact test and Mann-Whitney’s test, respectively. Cumulative percentages of patients developing gram-negative or gram-positive infected necrosis, taking into account the length of survival, were assessed by the actuarial Kaplan-Meier method and log-rank test. Cox regression was used to evaluate various factors simultaneously with regard to mortality [22]. This method was also used to assess the relationship between the occurrence of pancreatic infections (only gram-negative, only gram-positive, or mixed gram-negative/gram-positive) and mortality [23]. P values given are two-sided, and P = .05 was considered the limit of significance.

**Results**

Of 102 patients with objective signs of severe acute pancreatitis (Imrie score of ≥3 and/or Balthazar CT score grade of D or E), 50 patients were assigned to the SD group and 52 to
Figure 1. Cumulative percentages over time of patients with infection due to gram-negative bacteria (left) and with gram-positive infection of pancreatic necrosis (right), according to treatment group. In parentheses are numbers of patients at risk. Control group (dotted line), n = 52; selective decontamination group (solid line), n = 50.

the control group. The groups were well matched with regard to Imrie score (mean for both, 3.2) and Balthazar grade [5].

Microbiology

Twenty-nine of 102 patients (28%) developed infected pancreatic necrosis. Pseudomonas aeruginosa (13 patients), Escherichia coli (13), Staphylococcus epidermidis (21), and enterococci (19) were most frequently isolated; anaerobes (3) were found to play only a minor role and were not further analyzed.

Infected necrosis occurred in 9 of 50 patients (18%) of the SD group, in comparison with 20 of 52 patients (37%) of the control group (P = .03), because of a significant reduction of gram-negative infected necrosis (SD group, 4 of 50 [8%]; controls, 17 of 52 [33%]). Three patients who developed infected necrosis had not undergone surgery. Infected necrosis was demonstrated at autopsy (two patients) or by percutaneous drainage (one).

Figure 1 shows the increasing percentage of patients over time who developed gram-negative pancreatic infection. The incidence of gram-positive infection of pancreatic necrosis did not significantly differ between treatment groups (SD group, 9 of 50 [18%]; controls, 16 of 52 [31%]) (figure 1).

The Imrie score at enrollment in the study appeared to correlate very strongly with the incidence of gram-negative pancreatic necrosis over time, especially in the control group (P < .001) (figure 2). The Balthazar grade at enrollment in the study also correlated with the incidence of gram-negative pancreatic infection, although this correlation was less pronounced (grade C/D, 6 of 48 [12%]; grade E, 14 of 53 [26%]) (with adjustment for treatment group, P = .03).

Forty FNAs were performed in 25 patients (range, 1–4 per patient). Twenty-one of these FNAs (17 patients) were done without treatment with intravenous antibiotics at the time of aspiration. Only 4 of these aspirations (19%) showed presence of bacteria (E. coli, 1; S. epidermidis, 3). Nineteen FNAs (16 patients) were done during simultaneous treatment with intravenous antibiotics. Eight of these (42%) showed infection of pancreatic necrosis (Enterobacter species, 1; Klebsiella species, 1; S. epidermidis, 4; enterococci, 4; Staphylococcus aureus, 1). Some aspirates contained more than one type of bacteria.

Mortality

Mortality is significantly reduced among patients treated with adjuvant SD [5]. Among patients who survived (n = 73), the percentage in whom infected necrosis had occurred was 19% (14 patients), which is significantly (P = .002) lower than the 52% (15) of nonsurvivors (n = 29) (table 1). To evaluate the impact on mortality of infected pancreatic necrosis developing during treatment, patients were classified each day according to whether pancreatic necrosis was still sterile or whether only a gram-positive, only a gram-negative, or a mixed gram-negative/gram-positive pancreatic infection had occurred.

All patients started in the sterile condition, in accordance with the enrollment criteria. With the use of Cox regression for both groups, it emerged that in comparison with patients with sterile necrosis, those who acquired only a gram-positive pancreatic infection had a 1.6-fold increased death rate (P = .52). Patients who developed only a gram-negative pancreatic infection had a 14.4-fold increased death rate (P < .001) in comparison with the rate for those with sterile necrosis. A similarly increased mortality of 15.8-fold (P < 0.001) was found for those with mixed gram-negative/gram-positive infected necrosis.
Table 2 shows results of multivariate analysis of the relationship between mortality and the type of pancreatic infection, taking into account the Imrie score, Balthazar grade, and randomized treatment. This analysis demonstrates that development of a gram-negative infection of pancreatic necrosis during the course of the disease is an important and ominous sign, while there was no significant increase in mortality due to gram-positive pancreatic infection. With these infections taken into account, there is still an increased death rate among patients with a higher Imrie multifactorial initial assessment. No additional prognostic value was found for the Balthazar grade. The data in parentheses (table 2) also show that SD decreases mortality when analyzed without consideration of infectious status.

There was no mortality difference between treatment groups for patients without gram-negative infected necrosis (i.e., sterile or only gram-positive), as demonstrated in figure 3 (left panel). After occurrence of a gram-negative pancreatic infection, mortality was high (13/21; 62%). As shown in figure 3 (right panel), survival among these patients did not significantly differ between treatment groups. However, a gram-negative pancreatic infection occurred in only four patients in the SD group.

### Hospital Stay

The average hospital stay of survivors with gram-negative necrosis (8 patients) was 135 days (range, 56–241 days), which is significantly higher than the mean values of 55 days (range, 26–82 days) \((P = .01)\) and 30 days (range, 10–71 days) \((P < .001)\) for survivors with only gram-positive (6 patients) or sterile necrosis (59), respectively. Although smaller, the difference between hospital stay of survivors with only gram-positive infected and sterile necrosis is also significant \((P = .004)\). These results were similar in both treatment groups (SD group and controls).

### Discussion

This study demonstrates that mortality increases dramatically once gram-negative infection of pancreatic necrosis occurs in patients with severe acute pancreatitis. However, if pancreatic necrosis becomes infected with only gram-positive aerobic bacteria, mortality is not significantly increased and is comparable with that among patients in whom pancreatic necrosis remains sterile throughout the course of the disease. This is probably because *S. epidermidis* or enterococci, most frequently isolated in cases of infected necrosis due to a solitary gram-positive organism, are less pathogenic in these patients.

The overall incidence of secondary infection of pancreatic necrosis is 28%. In the control group infected necrosis occurred at a rate of 38%, which has also been described by others [1, 15, 24]. In patients treated with SD, the overall incidence of infected necrosis (18%) was significantly reduced because of a marked reduction in incidence of gram-negative infected necrosis (only 8%, vs. 33% in the control group).

### Table 1. Bacteriologic classification of pancreatic necrosis during the course of the disease in survivors and nonsurvivors.

<table>
<thead>
<tr>
<th>Classification</th>
<th>Survivors ((n = 73))</th>
<th>Nonsurvivors ((n = 29))</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sterile</td>
<td>59 (81)</td>
<td>14 (48)</td>
</tr>
<tr>
<td>Only gram-positive</td>
<td>6 (8)</td>
<td>2 (7)</td>
</tr>
<tr>
<td>Only gram-negative</td>
<td>0</td>
<td>4 (14)</td>
</tr>
<tr>
<td>Mixed gram-positive/gram-negative</td>
<td>8 (11)</td>
<td>9 (31)</td>
</tr>
</tbody>
</table>

NOTE. The percentage of patients with infected pancreatic necrosis among nonsurvivors (15 of 29; 52%) is significantly higher than that among survivors (14 of 73; 19%) \((P = .002)\).
Table 2. Results of multivariate analysis of mortality in relation to development of infection of pancreatic necrosis during treatment, baseline Imrie score, baseline Balthazar grade, and randomized treatment. All patients started in the sterile category, in accordance with the enrollment criteria.

<table>
<thead>
<tr>
<th>Factor</th>
<th>No. of deaths*</th>
<th>Relative death rate</th>
<th>Significance²</th>
<th>95% Confidence limits of relative death rate</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Infected pancreatic necrosis</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sterile</td>
<td>14/5,209</td>
<td>1.0</td>
<td>...</td>
<td>...</td>
</tr>
<tr>
<td>Only G+</td>
<td>2/615</td>
<td>1.2</td>
<td>.86</td>
<td>0.2, 5.5</td>
</tr>
<tr>
<td>Only G−</td>
<td>4/103</td>
<td>8.0†</td>
<td>.001</td>
<td>2.4, 27.1</td>
</tr>
<tr>
<td>Mixed G+/G−</td>
<td>9/508</td>
<td>7.0†</td>
<td>.002</td>
<td>2.1, 24.5</td>
</tr>
<tr>
<td><strong>Imrie score</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0–2</td>
<td>1/36</td>
<td>1.0 (1)</td>
<td>... (... )</td>
<td>...</td>
</tr>
<tr>
<td>3–4</td>
<td>9/41</td>
<td>7.4 (10.6)</td>
<td>.06 (0.03)</td>
<td>0.9, 60.3</td>
</tr>
<tr>
<td>5–7</td>
<td>19/25</td>
<td>31.2 (56.8)</td>
<td>.001 (.001)</td>
<td>3.9, &gt;100</td>
</tr>
<tr>
<td><strong>Balthazar grade</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C/D</td>
<td>9/48</td>
<td>1.0 (1)</td>
<td>... (... )</td>
<td>...</td>
</tr>
<tr>
<td>E</td>
<td>20/53</td>
<td>0.9 (1.3)</td>
<td>.76 (.38)</td>
<td>0.3, 2.3</td>
</tr>
<tr>
<td><strong>Treatment group</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Controls</td>
<td>18/52</td>
<td>1.0 (1)</td>
<td>... (... )</td>
<td>...</td>
</tr>
<tr>
<td>SD</td>
<td>11/50</td>
<td>0.8 (0.4)</td>
<td>.56 (.03)</td>
<td>0.3, 1.8</td>
</tr>
</tbody>
</table>

NOTE. G+ = gram-positive; G− = gram-negative; SD = selective decontamination. Parentheses around data denote results obtained without allowance for the factor infections (infectious status). Upon enrollment, CT (Balthazer grade) was not performed for one patient.

* Per no. of patient-days (up to day 80, i.e., the day number of the last death) after the first occurrence of the infection specified, or per no. of patients for Imrie score, Balthazar grade, or treatment group.

† In comparison with reference category.

‡ Reference category.

§ Not significantly different from each other, but both significantly greater in comparison with “Only G+” category value.

The occurrence of a gram-negative infection of pancreatic necrosis is an ominous sign. Mortality among these patients increases significantly, irrespective of coexistence of a gram-positive infection, as shown in this study. Mortality with regard to the bacteriologic status of pancreatic necrosis was comparable for the SD group and the control group, i.e., once gram-negative infection of pancreatic necrosis occurred, mortality increased considerably in both treatment groups. However,
mortality in the treatment group was significantly reduced among patients treated with adjuvant SD (table 2), a finding that was also published previously [5]. Consequently, SD reduces mortality among patients with severe acute pancreatitis because of a significant reduction in the development of gram-negative infection of pancreatic necrosis. This is accomplished by reduction of gram-negative intestinal translocation into the pancreatic necrosis. However, SD is not useful for patients in whom gram-negative pancreatic infection already exists or develops during SD administration, as is demonstrated in this study.

It has been suggested that overgrowth and translocation of gram-positive bacteria, i.e., enterococci or staphylococci, may be a drawback of SD [20, 25]. Our results do not support this hypothesis. Neither intestinal overgrowth nor increased incidence of gram-positive infected necrosis has been found in our study. Deaths due to unexplained gram-positive sepsis along with positive blood cultures (two patients) and otherwise-documented sterile necrosis at time of death were equally divided among the SD group and controls. However, these possible hazards demand strict indications and careful bacteriologic surveillance, as is the case for any kind of antibiotic regimen.

The development of gram-negative infection of devitalized tissues in and around the pancreas is, apart from the Imrie score, the most important parameter determining outcome. Gram-negative pancreatic infection can be minimized with adjuvant SD, thereby reducing mortality among patients with severe acute pancreatitis.

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References