

Proinflammatory cytokines and Mannheim index in predicting mortality rates in patients with peritonitis caused by perforation

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Abstract

Acute diffuse peritonitis caused by perforation represents a complex entity, which, beside the surgery itself, represents the understanding of the physiopathological mechanisms designed to anticipate the unfavorable evolution. Material and methods: A prospective study was undergone on a lot of 100 patients, 71 males and 29 females, who were hospitalized in the General Surgery Clinic of "Bagdasar Arseni" Clinical Emergency Hospital, while being diagnosed with peritonitis caused by perforation, for which the value of proinflammatory cytokines (IL1 β , IL6 and TNF α) and also of Mannheim Index was analyzed in predicting the mortality rate. Cytokines dosage was preoperative performed and also on the 3rd and 7th day postoperative and Mannheim Index was immediately calculated postoperative. Results: The results for IL1- β were insignificant and had no statistical value. However, from the serial determination in the dynamics of the other proinflammatory cytokines, IL6 and TNF α , it resulted that these could be important in the quantification of the mortality rate risk. Moreover, the Mannheim Index was calculated, thus achieving a higher score or equal to 26 points, a sensitivity of 0.7895 and a specificity of 0.7284 in the prediction of the mortality rate. Conclusions: Both IL6 and TNF α have an important value in predicting the mortality rates in patients with acute diffuse peritonitis caused by digestive tract perforations, the patients with elevated serum concentrations both at admission in hospital and postoperative evolving more frequently to death. The calculation of Mannheim Index is useful in patients with peritonitis caused by perforation, the patients with a higher score or equal to 26 points having a higher mortality rate risk.

Keywords: proinflammatory cytokines, Mannheim Index, peritonitis, mortality rate, survival

1. Introduction

Acute diffuse peritonitis caused by perforation represents a very complex entity, which, beside the surgery itself, also needs the understanding of the physiopathological mechanisms in order to foresee the unfavorable evolution and death [35]. Starting from the data in the specialty literature, we have directed towards the serum dosing of some important mediators represented by proinflammatory cytokines and we have also tried to study the usefulness of Mannheim Index in predicting the mortality rate in patients with this condition[36].

2. Materials and methods

A prospective study was undergone on a group of 100 patients, 71 males and 29 females, emergency hospitalized in the General Surgery Clinic of "Bagdasar Arseni" Clinical Emergency Hospital, in Bucharest, with the diagnosis of peritonitis caused by perforation, for which the value of proinflammatory cytokines (IL-1 β , IL-6 and TNF α) and the Mannheim Index respectively, were analyzed in order to predict the mortality rate. The serum levels of IL-1 β , IL-6, and TNF α were determined in the laboratory of the Department of Physiopathology and Immunology of "Carol Davila" University of Medicine and Pharmacy in Bucharest by using ELISA (Enzyme Linked Immunosorbent Assay), Biosource and Invitrogen commercial kits. The concentration of these cytokines was determined based on a standard curve graph achieved through the simultaneous determinations of the concentrations in the working kits, as well as through the blood samples by ELISA method. The cytokines dosage was performed preoperatory and respectively on the 3rd and 7th days postoperatory. The Mannheim Index was calculated immediately postoperatory. Moreover, Mannheim Index is a score system used to evaluate the prognosis of patients with peritonitis. It was easily applied and was based on the evaluation of the clinical and humoral parameters, being useful in choosing the optimal therapeutic options in patients with peritonitis. Mannheim Index was calculated according to the literature data as it follows:

Table 1. Mannheim Index

Age > 50 years old	5 P
Females	5 P
Organ deficiency (organ insufficiency*)	7 P
Neoplastic disease	4 P
Preoperatory period >24h	4 P
Non-colonic origin of the peritonitis	4 P
Generalized peritonitis	6 P
Peritoneal citrine serous fluid	0 P
Turbid peritoneal fluid	6 P
Sterocoral peritonitis	12 P

*1. renal: creatinine > 177 mmol/ l, urea > 16,7 mmol/ l, oliguria < 20 ml/ h;

2. pulmonary: PaO₂ < 50 mmHg, Pa CO₂ > 50 mmHg;

3. hypo/ hyperdynamic shock;

4. paralytic bowel obstruction > 24h or complete mechanical bowel obstruction.

The statistical analysis was undergone by using the applications of SPSS 16 software and Microsoft Excel. The predictive values, which were less than 0.05, were considered statistically significant.

3. Results and discussions

The mortality rates in the 100 patients included in the study was of 19%, in 15 cases the death resulting after the 7th day from the surgery due to septic complications in patients with severe sepsis, sepsis shock and/ or Multiple Organs Dysfunction Syndrome (MODS), in 4 cases the death occurring early postoperatory due to other causes (2 patients had a myocardial infarction, 1 patient had a pulmonary embolism and 1 patient had a stroke).

The results achieved in the 3 periods of the study for the 100 patients were the following:

- a) Preoperative (on the 1st day)
 - IL-6: values between 10 and 4986 pg/ ml; medium value - 698.27 pg/ ml
 - TNF α : 23,5 - 800 pg/ ml; medium value - 263.21 pg/ ml
- b) On the 3rd day postoperative
 - IL-6 varied between 8,4 and 4399 pg/ ml; medium value - 467.25 pg/ ml
 - TNF α : 4 - 1040 pg/ ml; medium value - 192.16 pg/ ml
- c) On the 7th day postoperative
 - IL-6: 0 - 3718 pg/ ml; medium value - 297.66 pg/ ml
 - TNF α : 8,7 - 740 pg/ ml; medium value - 138.14 pg/ ml

In deceased patients, the serum values of the studied cytokines varied between the following values during the 3 periods of the study:

- a) Preoperative (on the 1st day)
 - IL-6: 338 – 4836 pg/ ml; medium value - 1722.04 pg/ ml
 - TNF α : 145 – 800 pg/ ml; medium value - 386.94 pg/ ml
- b) On the 3rd day postoperative
 - IL-6: 200 – 4399 pg/ ml; medium value - 1755.46 pg/ ml
 - TNF α : 133,16 – 570 pg/ ml; medium value - 348.53 pg/ ml
- c) On the 7th day postoperative
 - IL-6: 110 – 3718 pg/ ml; medium value - 1912,71 pg/ ml
 - TNF α : 40 – 740 pg/ ml; medium value - 431.32 pg/ ml

In the control group we have presented (15 patients who were operated for non-infectious pathologies) both preoperative and postoperative, the IL-6 values were 0 - 48,8 pg/ ml (with a medium value of 29,49 pg/ ml) and the TNF α values were 84,11 – 162,4 pg/ ml (with a medium value of 130.33 pg/ ml). Unfortunately, the results for IL-1 β were way below our expectations. IL-1 β was identified in the serum of only 8 patients with peritonitis caused by perforation, being determined only once, having values below 100pg/ ml at the same time being unable to present a statistical significance in the performed analysis. Moreover, by calculating the Mannheim Index for the 100 patients, a medium value of 22,41 points regarding the survival rate was achieved and for the ones who died, the medium value was of 29,6 points. The Mannheim Index value in the patients who died was between 21 and 35 points. In the patients with a Mannheim Index of ≥ 26 points, an amount of 15 deaths were registered, the rest of 4 coming from patients with values between 21 and 25 points. Among the patients who survived, an amount of 21 presented a Mannheim Index value of ≥ 26 points (the data in literature underlines a high risk of death, the MI scores being ≥ 26 points). In order to analyze the capacity of proinflammatory cytokines in the three study periods of the mortality prediction, non-parametric Mann-Whitney U and Wilcoxon tests were applied obtaining the following results:

Table 2. Mortality prediction: non-parametric Mann-Whitney U and Wilcoxon tests

Death			Z ₁ IL-1 β	Z ₁ IL-6	Z ₁ TNF α
Predictive value (P)			0,481	<0,01	< 0,01
Z ₃ IL-1 β	Z ₃ IL-6	Z ₃ TNF α	Z ₇ IL-1 β	Z ₇ IL-6	Z ₇ TNF α
0,263	<0,01	< 0,01	0,724	< 0,01	< 0,01

It could be noticed that beside IL-1 β , for which no statistical value existed, all the other elements presented an important statistical significance in predicting mortality in the 100 patients with peritonitis caused by perforation ($P < 0.01$). In order to comparatively observe the value of proinflammatory cytokines dosing pre and postoperatory and of Mannheim Index in predicting the mortality rate, the graphic representation was performed in the form of ROC curves. The ROC curve (Receiver Operating Characteristics) is a bidimensional curve, which has as characteristics sensitivity on the Y-axis and specificity on the X-axis. This curve helps measuring the efficiency of a model. The more the area under the curve is bigger (the maximum value is 1), the better the model (when it has values which are lower than 0,6 – the model is rejected). In addition our results were the following:

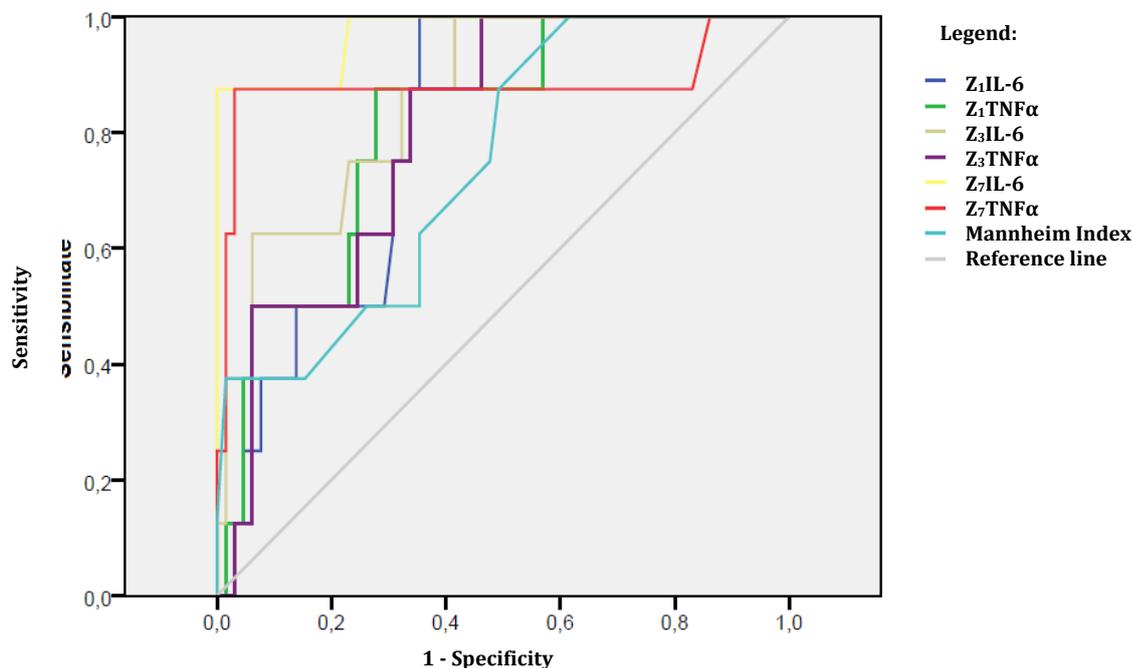


Fig. 1 Value of proinflammatory cytokines dosing pre and postoperatory and of Mannheim Index in predicting the mortality rate

Table 3. Interpretation of ROC curves in Fig. 1

Variables	ASC	Predictive value (P)
Z1IL-6	0,801	0,006
Z1TNF α	0,813	0,004
Z3IL-6	0,861	0,001
Z3TNF α	0,804	0,005
Z7IL-6	0,972	0,000
Z7TNF α	0,881	0,000
Mannheim Index	0,746	0,024

Legend: ASC represents the area below the ROC curve. Z₁IL-6 and Z₁TNF α represent the value of IL-6 and also of TNF α on day 1 (preoperatory), Z₃IL-6, Z₇IL-6 and Z₃TNF α , Z₇TNF α represent the values of IL-6 and TNF α on the 3rd and 7th day postoperatory.

It could be noticed from the table above that all the variables analyzed had a statistical significance in quantifying the death risk, the most valuable in our study being the IL-6 determination on the 7th day postoperatory. Taking this element into account, a graphic representation of IL-6 values having a general-linear model form was achieved by repeated measurements (Graph in Figure 2) in order to notice the variation of its values in survivors and deceased.

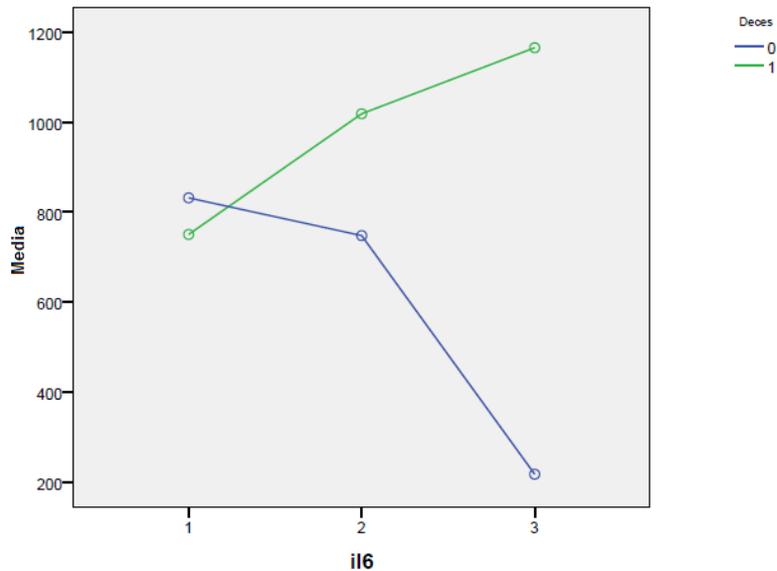


Fig. 2 The variation of IL-6 (pg/ ml) values in survivors and deceased (repeated measurements)
Legend: 0 – survivors;
1 – deceased.

In order to understand the relationship between the sensitivity and specificity of Mannheim Index in predicting the mortality rate and to be able to report to the value of 26 points, a value whereof the literature data indicate a high risk of death, the ROC curve was separately analyzed:

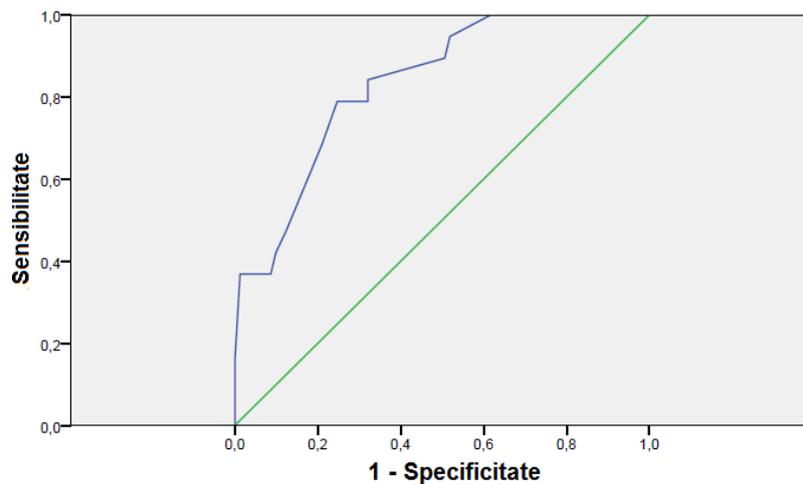


Fig. 3 Relationship between the sensitivity and specificity of Mannheim Index in predicting the mortality rate

For the association Mannheim Index ≥ 26 points and death rate or survival rate, the following values were obtained for sensitivity, specificity, positive predictive value, and negative predictive value:

Table 4. Analysis of the association of Mannheim Index ≥ 26 – mortality rate

Variable	Value	95% confidence interval
Sensitivity	0,7895	0,5443 – 0,9395
Specificity	0,7284	0,6185 – 0,8212
Positive predictive value	0,4054	0,2477 – 0,5795
Negative predictive value	0,9365	0,8453 – 0,9824

As shown before, the results for IL-1 β were unsatisfactory and had no statistical significance [35]. However, from the dynamic serial determination of the other proinflammatory cytokines, respectively IL-6 and TNF α , it was revealed that these could be important in quantifying the death rate risk. The analyzed proinflammatory cytokines (except for IL-1 β) presented a different kinetics in deceased patients compared to the survivors, persistently high medium values or even ascendant dynamics being noticed in patients who had subsequently died due to septic complications. As a result of the statistical analysis presented, that the highest importance in predicting the mortality rate in this study was the determination of IL-6 and TNF α values on the 7th day postoperatory, which presented the highest capacity of discrimination (with ASC=0,972, respectively 0,881). Although it presents a statistical significance (P=0,024 and ASC=0,746), Mannheim Index was the less important among the analyzed variables in predicting the mortality rate. As shown before, for a Mannheim Index ≥ 26 points, a sensitivity of 0.7895 and a specificity of 0.7284 were obtained while predicting the death rate [36]. Beside the constants in the study performed, the data in literature, present in many studies performed on this subject, were also analyzed. Different studies such as: Cytokine profiles as markers of disease severity in sepsis: a multiplex analysis, undergone by Fernando A Bozza et al.; Plasma cytokine measurements augment prognostic scores as indicators of outcome in patients with severe sepsis, undergone by Oberholzer A et al. and Serum cytokine levels in human septic shock – relation to multiple system organ failure and mortality, undergone by Pinsky M.R. et al., have shown that the plasmatic levels of IL-6 were significantly higher than the levels of TNF α and had higher values compared to the initial determination in deceased patients as compared to TNF α [1-3]. This is possibly due to the stable kinetics of IL-6 in the plasma, which also recognizes it as a valuable cytokine in predicting sepsis and its complications [4,5]. In a study performed on patients with severe sepsis, Hamishehkar and col. noticed significantly high plasmatic levels of TNF α and mostly of IL-6 in patients who died and a strong correlation of IL-6 with APACHE II and SOFA scores [6]. Moreover, in a study performed in the intensive therapy unit, although the TNF α plasmatic levels were high in patients with septic shock, a correlation between the TNF α , severity of sepsis and mortality rate could not be established [7]. Other studies showed that the measurement of the constant high level of IL-6 and TNF α through serial determinations can identify patients with a high risk of evolving to organs dysfunction or death, better than the highest levels of these cytokines can do it [3,8]. Reinhart and col. showed that the septic patients with an IL-6 value higher than 1000 pg/ ml, had a mortality rate of 56%, compared to 40% the rate of patients with interleukin 6 serum levels of less than 1000 pg/ ml [9]. Moreover, it was also observed that in laboratory mice, IL-6 measured at 6 hours from the induction of peritonitis, accurately predicted mortality, which resulted from the experimental sepsis [29]. Other studies showed that the laboratory rats with the highest mortality rate have high levels of IL-6 for at least 24 hours [30,31]. Although the proinflammatory mediators correlate with mortality, as it resulted from our study, the raise of

the level of anti-inflammatory cytokines, especially of IL-10 and of IL-4, could also indicate an unfavorable result in sepsis [26-28]. Osuchowski and col. showed that the values of proinflammatory cytokines measured 24 hours before the death are better predictors of mortality than the values obtained 48 hours before the death [10]. The way we were showing in a previous article [35], IL-6 and TNF α proinflammatory cytokines have both a diagnostic and a prognosis value in cases of patients with peritonitis, being able to help in anticipating the evolution to septic complications (severe sepsis, multiple organ dysfunction syndrome, septic shock). The early prophylactic inhibition of proinflammatory cytokines improved the survival rate in animals and in certain experimental models of peritonitis [11-13,25]. Other studies also performed on animals with peritonitis showed that the administration of TNF α inhibitors did not produce any effect as far as the survival rate was concerned [14-16], moreover, the harmful effects were noticed [17]. Likewise, in human patients the results of the studies regarding the therapeutic prophylactic inhibition of TNF α and IL-1 β with antibodies against them were contradictory. Some studies showed that the improvement of the survival rate after these treatments [18], however, most of them do not show any beneficial effects of these treatments [19,20]. Moreover, the idea that in order to improve the survival rate, these cytokines must be prevented from an early expressing but should not be completely eliminated, also emerged [21]. Most probably, there will be many studies in the future regarding the prophylactic inhibition of interleukin 6 in septic patients, taking into account that both our study and the numerous other international studies confirmed the very high serum values of interleukin 6 in patients who evolved towards septic complications or death.

Compared to the results we obtained regarding the connection between the value of Mannheim Index ≥ 26 points and death, the conclusions of another study [22] were also analyzed. They revealed a mortality rate of 76,6% for MI ≥ 26 , a sensitivity of predicting death of 71,8% and a specificity of 94%. The positive predictive value was of 0,76 and the negative predictive value was of 0,92. Similar results were observed in our study and the previously mentioned, as far as sensitivity and negative predictive value is concerned. In the Mannheim study, the prognosis value of Mannheim Index was of 56,7% [32], in the Tilburg study it was of 69,2% [33], and in the one performed in München it was of 55% [34]. Another study showed a mortality rate of 50% at a MI ≥ 30 , of 14% at a MI between 21 and 29 and of 5% at a MI ≤ 20 , with a sensitivity in predicting the death at a MI 25 of 72,09% and a specificity of 71,43%, and at a MI ≥ 25 a sensitivity of 80,65% and a specificity of 57,89% [23]. Malik and col. observed that at a value of the Mannheim Index below 15 points, there was no mortality rate and at an index higher than 25 points, the mortality rate was over 80% [24].

4. Conclusions

Both IL-6 and TNF α have an important value in predicting mortality in patients with acute diffuse peritonitis caused by digestive perforations, the ones with high serum concentrations both in preoperative and in postoperative period more frequently evolving to death. The conclusion is that the utility of proinflammatory cytokines dosage in patients with peritonitis is important in order to set the severity diagnosis and the fast establishment of a sustained intensive therapy, which, associated with surgery, will maximally exploit the chances of survival. Moreover, calculating the Mannheim index is also useful in establishing the vital prognosis of patients with peritonitis caused by perforation, patients with a higher score or equal to 26 points, who present a high risk of death.

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