Динаміка змін вмісту білків, що зв'язують жирні кислоти з плазмою, та кишкового зонуліну у пацієнтів із генералізованою внутрішньочеревною інфекцією і абдомінальним сепсисом залежно від тяжкості стану пацієнтів

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Dynamics of changes in plasma fatty acid binding proteins and intestinal zonulin in patients with generalized intra–abdominal infection and abdominal sepsis depending on the severity of patients

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Реферат

Мета. Визначити рівні білків, що зв'язують кишкові жирні кислоти, у плазмі крові у поєднанні із зонуліном у пацієнтів із генералізованою внутрішньочеревною інфекцією та абдомінальним сепсисом, та визначити їх клінічну корисність для оцінки тяжкості пацієнтів.

Матеріали і методи. Це дослідження було засноване на аналізі 59 пацієнтів обох статей віком від 18 до 70 років з генералізованою внутрішньочеревною інфекцією (ГВЧІ) та абдомінальним сепсисом (АС).

Результати. Відповідно до цілей та завдань дослідження пацієнти були розподілені на три групи за критерієм «Сепсис–3»: перша група – 26 пацієнтів із генералізованим перитонітом без сепсису; друга група – 24 хворі на сепсис, а третя група – 9 хворих на септичний шок. Ми виявили статистично значущо вищі рівні I–FABP у всіх групах пацієнтів (p = 0,000). Однакова тенденція спостерігалася у всі періоди дослідження, і найбільш значущі рівні I–FABP були на десяту добу після операції у пацієнтів із септичним шоком: IQR 1567,3– 3876,1 (p = 0 000). Пацієнти з абдомінальним сепсисом не мали статистично значущих змін рівня зонуліну порівняно з пацієнтами з ГВЧІ без сепсису (p = 0,560), і подібна тенденція спостерігалася на 3–й день після операції (p = 0,135). Лише на 7–й та 10–й дні після операції зміни рівня зонуліну були значущими у пацієнтів з ГВЧІ без сепсису, з АС та септичним шоком (p = 0,000 та p = 0,004, відповідно).

Висновки. Рівні I–FABP у сироватці крові були цінними та об'єктивно ранніми предикторами тяжкості травм травного тракту при ГВЧІ. Ми також представили докази підвищення рівня зонуліну в плазмі крові при ГВЧІ порівняно з контрольною групою. Підвищений рівень зонуліну був додатковим показником спостережуваного збільшення проникності кишечника при внутрішньочеревній інфекції, але він, подібно до I–FABP, не був раннім біомаркером тяжкості пошкодження травного тракту.

Ключові слова: генералізована внутрішньочеревна інфекція; абдомінальний сепсис; біомаркери; I–FABP, зонулін; результати.

Abstract

Objective. Determine the levels of plasma intestinal fatty acid binding proteins levels in combination with zonulin in patients with generalized intra–abdominal infection and abdominal sepsis, and define the clinical usefulness them to assess the severity of patients.

Materials ad methods. This study was based on an analysis of 59 patients of both sexes aged 18 to 70 years with generalized intra–abdominal infection (gIAI) and abdominal sepsis (AS).

Results. According to the aims and objectives of the study the patients were divided into three groups: the 1st group – 26 patients with generalized peritonitis without sepsis according to the «Sepsis–3» criterion; the 2nd group – 24 patients with sepsis, and the 3rd group – 9 patients with septic shock. We found statistically significant higher levels of I–FABP in the all groups of patients (p = 0.000). The same tendency was observed in all periods of the study, and the most significant levels of I–FABP were by the tenth day after surgery in patients with septic shock: IQR 1567.3– 3876.1 (p = 0.000). Patients with abdominal sepsis did not have a statistically significant change in zonulin levels compared to patients with gIAI without sepsis (p = 0.560) and a similar trend was observed on the 3rd day after surgery (p = 0.135). Only by the 7th and 10th days after surgery changes in zonulin levels were significant in intra–abdominal infection patients without sepsis, with abdominal sepsis and septic shock (p = 0.000 and P = 0.004, respectively).

Conclusions. Serum I–FABP levels were valuable and objectively early predictors of the severity of gastrointestinal injuries in gIAI. We also presented evidence of increased plasma zonulin levels in generalized intra–abdominal infection compared with the control group. Elevated zonulin levels were an additional indicator of the observed increase in intestinal permeability during gIAI, but zonulin was not an early biomarker of the severity of gastrointestinal damage like I–FABP.

Keywords: generalized intra–abdominal infection; abdominal sepsis; biomarkers; I–FABP; zonulin; results.

Introduction

Generalized intra–abdominal infection (gIAI), abdominal sepsis and septic shock correlate in patients with poor treatment outcomes, which is accompanied by high mortality [1]. The gut is believed to play a key role in the development of systemic inflammatory response syndrome (SIRS), the digestive tract (GI) is quite vulnerable, the occurrence of its dysfunction plays a significant role in the development of complications of the disease, which is also observed in other emergency conditions [2, 3]. There is ample evidence of impaired intestinal barrier function in critically ill patients, disruption of tight junctions of the epithelium, which leads to increased permeability and delayed release of inflammatory cytokines. Thus, pro–inflammatory mediators from the intestine can expand the inflammatory cascade, putting other organs at risk [4, 5].

Often, the severity of GI dysfunction indicates the severity of the condition of critical patients, since a number of publications reported that almost 50% of patients in intensive care units (ICU) have damage to enterocytes at the initial stage of treatment [6], and among critically ill patients with GI dysfunction high mortality rates are more often observed [7, 8].

Acute dysfunction and GI deficiency are increasingly recognized in critically ill patients. The variety of definitions that have been proposed in the past has led to confusion and difficulty in comparing one study with another. In 2012, the Working Group on Abdominal Problems (WGAP) of the European Society of Intensive Care Medicine (ESICM) defined acute GI injury (AGI) as a malfunction in critically ill patients with an acute illness, including surgery, and recommended a classification for assessing the severity of AGI [9].

Over the past 20 years, the diagnostic value of a new clinical and laboratory indicator – fatty acid binding proteins (I–FABP), which is one of the intracellular proteins and is involved in the transport and metabolism of long–chain fatty acids, has been studied. The family of FABP proteins has good diagnostic characteristics: 1) soluble in the cytoplasm; 2) have a high specificity to the tissue from which they originate; 3) are contained in the cell in high concentration; 4) have a low molecular weight (15 kDa) [10]. These characteristics make it possible to use the appearance of these markers in the peripheral blood as sensitive and specific, indicating tissue damage (heart, liver, small intestine, etc.). It is known that I–FABP is contained in the epithelial cells of the small intestine, and the literature is currently actively discussing the diagnostic value of increasing this biomarker in the blood in a severe category of patients, including those with gIAI [11, 12]. Considering the fact that GI problems that arise in a severe category of patients are not equally assessed in patients in surgical departments and ICUs, and there is insufficient data on this in the available literature.

Tight junction proteins, which are the membrane–associated, guanylate kinase – like proteins ZO–1, ZO–2, and ZO–3 as well as the integral membrane proteins occludin and claudin, are critical to maintaining normal physiological processes in the GI, which is confirmed by a number of studies [13–15].

Human zonulin is a 47 kDa protein that increases the intestinal permeability of the epithelium of the small intestine; it is a eukaryotic analogue of the Vibrio cholerae zonula occludens toxin [16]. Zonulin is the only known physiological mediator that increases intestinal permeability by reversibly modulating tight junctions between cells. Elevated plasma zonulin levels have been reported in celiac disease, type 1 diabetes and obesity–related insulin resistance [17, 18].

The exact physiological function of zonulin as a potent modulator of intestinal permeability has not yet been fully understood [19]. In addition, there is no clear evidence that circulating plasma zonulin is a marker of intestinal permeability in the treatment of gIAI and abdominal sepsis, although this indicator has been proposed as a potential marker for the study of intestinal permeability [20, 21].

We hypothesized that patients with gIAI and abdominal sepsis have elevated plasma levels of I–FABP and zonulin compared to different patients severity and different clinical parameters in their plasma levels.

The objective was determined the levels of plasma intestinal fatty acid binding proteins levels in combination with zonulin in patients with generalized intra–abdominal infection and abdominal sepsis, and define the clinical usefulness them to assess the severity of patients.

Materials and methods

Design of study. A prospective single–center controlled study was conducted in 59 patients with generalized peritonitis of both sexes who were treated at the Kharkiv Regional Clinical Hospital (Ukraine). The study was approved by the institutional ethics committee, and is in accordance with the Helsinki Declaration of 1975.

We used the Sepsis–3 classification according to the recommendations of the international Consensus [22]. Patients included in the study were treated according to international guidelines adapted to local resources and procedures [23].

Inclusion criteria. The study included men and women over 18 and under 70 years of age were admitted to hospital for 59 with IAI without and with sepsis or septic shock within 24–72 hours and more of admission to the surgery or intensive care unit. All of these patients were operated on urgently.

Exclusion criterias. Comorbidity with acute myocardial infarction and stroke; IAI caused by the cancer of the hollow organ; post–resuscitation illness due to stopping effective blood circulation; pregnancy; chronic infectious diseases, such as hepatitis, HIV, tuberculosis, and autoimmune diseases, celiac disease or Crohn's disease, cancer in anamnesis, the useless resuscitation status due to refractory shock, and the refusal of patients to participate in the study.

According to the aims and objectives of the study the patients were divided into three groups: the first group – 26 patients with generalized peritonitis without sepsis according to the «Sepsis–3» criterion; the second group – 24 patients with sepsis, and the third group – 9 patients with septic shock. All the patients were done surgery with effective source control, supporting appropriate antibiotics, resuscitation, and organ support therapy.
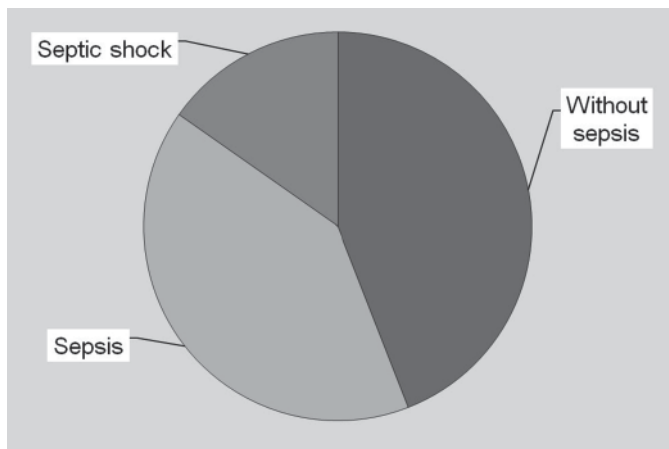
Figure 1.
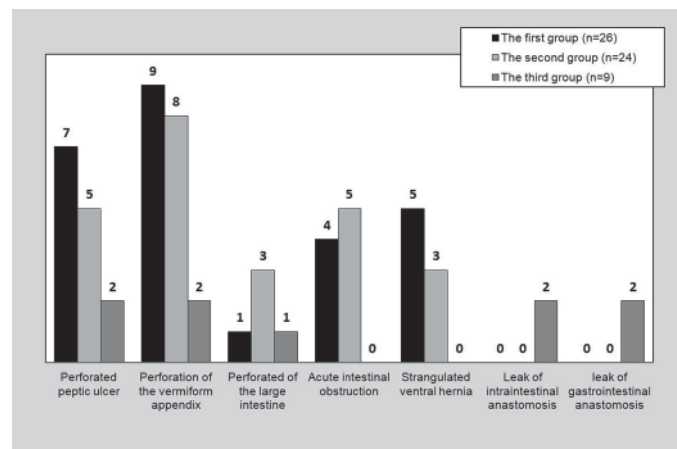*Distribution of patients with intra-abdominal infections.*

Figure 2.
The causes of intra-abdominal infections.

Control laboratory studies were carried out in 15 apparently healthy volunteers people. The study collected data: patient, characteristics (gender, weight, body mass index), assessment of severity according to the APACHE II score and assessment of organ dysfunction according to the SOFA scale, the indicators for which were calculated on the ClinCalc.com website, characteristics and causes of peritonitis, nature of treatment (mechanical ventilation, inotropic support, artificial kidney), results (30–day mortality). The diagnostic program for patients was included clinical blood and urine tests, biochemical blood tests according to unified methods, determination of lactate

in blood plasma as a marker of tissue hypoperfusion and an indicator of the acid–base state of the body using a spectrophotometric method, enzyme immunoassays: the level of I–FABP was determined using commercial kits «I–FABP, Human, ELISA kit», Netherlands; the level of zonulin with using «Zonulin ELISA Kit, Immundiagnostik, Bensheim», Germany. A blood sample for immunoassay researches (4.5 ml venous blood at a 1:10 dilution of 0.11 M trisodium citrate was taken), and centrifuged at 3500 xg for 15 minutes at 4 ° C for subsequent plasma extraction, which was instantly frozen and stored at −80 °C. In patients with generalized peritonitis, blood was taken

Table 1.   **Demographic, clinical and laboratory characteristics of patients with intra-abdominal infection.**

Indicators	The 1st group (n=26)	The 2nd group (n=24)	The 3rd group (n=9)	χ^2 / p value
Age (years), Me [IQR]	54 [36; 62]	53 [45; 66]	56 [42; 69]	7.669/0.022
Male/Famale	18/8	15/9	5/4	0,610/0,737
BMI, kg/m^2, Me [IQR]	24 [21; 28]	25 [22; 27]	28 [22; 31]	7.608/0.022
WBC count, ×10^9/l Me [IQR]	15.7 [13.8; 21.3]	21.8 [14.2; 24.2]	22.6 [15.3; 24.1]	NA
Creatinine, mg/dl, Me [IQR]	0.87 [0.76; 1.109]	0.113 [0.88; 1.543]	1.92 [1.46; 2.21]	NA
Total bilirubin, µmol/l Me [IQR]	24.4 [21.2; 64.6]	26.5 [14.5; 58.2]	59.5 [28.3; 108.2]	NA
Lactate, mmol/l, Me [IQR]	2.6 [1.8; 6.7]	3.1 [1.9; 7.5]	10.7 [5.3; 15.3]	NA
PCT, pg/ml, Me [IQR]	2.434 [0.117; 78.213]	6.871 [2.237; 81.134]	10.115 [3.456; 123.118]	NA
APACHE II sore (points) Me [IQR]	13 [10; 22]	21 [14; 28]	26 [16; 36]	12.783/0.002
SOFA score (points) Me [IQR]	9 [7; 11]	12 [8; 14]	14 [9; 16]	7.720/0.021
Mechanical ventilation, n	3	7	9	9.567/0.008
Inotropic support, n	3	8	9	9.240/0.010
Artificial kidney, n	1	1	3	6.059/0.048
ICU stay, n	4	5	9	9.422/0.009
30-day mortality n (%)	2 (7.7%)	5 (20.9%)	7 (77.8%)	8.919/0.012
Note: NA – not applicable.				

Table 2.   **Plasma levels of I-FABP (pg/ml) and Zonulin (pg/ml) in patients with intra-abdominal infection**

Timing research	The 1st group (n=26): IAI without sepsis Me [IQR]:	The 2nd group (n=24): Abdominal sepsis Me [IQR]:	The 3rd group (n=9): Septic shock Me [IQR]:
The levels of plasma fatty acid binding proteins			
Outcome	490.8 [298.5; 748.8] $P_1/P_3 = 0.000$	992.5 [789.5; 1567.1] $P_1/P_2 = 0.000$	1678.1 [1234.4; 2446.6] $P_2/P_3 = 0.000$
3d day	534.5 [345.2: 1110.4] $P_1/P_3 = 0.000$	1044.3 [658.5; 1876.6] $P_1/P_2 = 0.000$	2345.2 [1234.1; 3123.8] $P_2/P_3 = 0.000$
7th day	456.9 [345.2; 896.4] $P_1/P_3 = 0.000$	998.8 [673.2; 1567.2] $P_1/P_2 = 0.000$	2134.3 [1065.5; 3124.8] $P_2/P_3 = 0.000$
10th day	456.3 [345.2; 671.2] $P_1/P_3 = 0.000$	897.4 [678.8; 1234.6] $P_1/P_2 = 0.000$	2435.2 [1567.3; 3876.1] $P_2/P_3 = 0.000$
Control	89.6 [67.6; 172.2]		
The levels of plasma zonulin			
Outcome	6.66 [3.23; 9.45] $P_1/P_3 = 0.000$	7.06 [3.98; 9.56] $P_1/P_2 = 0.135$	19.34 [14.53; 26.34] $P_2/P_3 = 0.002$
3d day	6.11 [3.12; 7.89] $P_1/P_3 = 0.000$	6.34 [4.32; 11.23] $P_1/P_2 = 0.000$	18.23 [16.14; 21.22] $P_2/P_3 = 0.000$
7th day	4.65 [3.11; 8.78] $P_1/P_3 = 0.000$	13.89 [11.34; 16.12] $P_1/P_2 = 0.000l$	19.56 [17.23; 22.12] $P_2/P_3 = 0.002$
10th day	3.67 [3.12; 4.65] $P_1/P_3 = 0.000$	13.45 [11.56; 16.11] $P_1/P_2 = 0.000$	17.45 [16.11; 19.23] $P_2/P_3 = 0.004$
Control (n=15)	3.46 [2.81; 4.12]		

Note: P – Mann-Whitney test: P_1 – differences between the first and second groups; P_2 – differences between the second and third groups; P_3 – differences between the first and third groups; IAI – intra-abdominal infection; Me – the median; IQR – the interquartile range (IQR).

every 48 hours, in the sepsis group within 24 hours after diagnosis and surgery, within 8–10 days. In the group of healthy people blood was taken once after abstaining from food for at least 6 hours blood for immunoassay research. We also performed a statistical analysis of the results obtained.

Statistical analyses. Statistical data processing was performed using the trial version of STATISTICA 13.3 EN. Since the distribution laws of the studied indicators have differed from the normal (Shapiro–Wilk test), the medians, 25% and 75% quartiles, were calculated. Methods of nonparametric statistics were used in the work: Mann–Whitney, Wilcoxon tests, χ^2, Spearman's correlation coefficient. Continuous data was presented as Me (Q1; Q3), where Me is the median, Q1 and Q3 is the interquartile range (IQR). In all cases, statistical hypotheses were tested with a confidence level greater than 95%. To assess the adequacy of comparisons and the accuracy of the forecast quality, the method of analysis of operating characteristics curves (ROC – Receiver Operating Characteristic curve analysis) was used. The prediction method was chosen based on the Pareto criterion. The predictive efficiency of the models was assessed by discrimination based on the AUC index. The effectiveness of the model was considered limited at AUC ≥ 0.70; good – with AUC ≥ 0.80; remarkable – with AUC≥0.90.

Results

The patients were divided according to the severity of the condition which was determined by the criteria of Sepsis–3: the patients without sepsis were 26 (46.7%), with abdominal sepsis was diagnosed in 24 (43.1%), and septic shock was diagnosed in 9 (10.2%) patients (*Figure 1*).

The causes of IAI were secondary peritonitis with perforated peptic ulcer, perforation of the vermiform appendix, perforation of the large colon in diverticulitis, acute intestinal obstruction with generalized purulent peritonitis, strangulated ventral hernia with generalized purulent peritonitis, leak of intra intestinal and gastrointestinal anastomoses (*Figure 2*). The study was dominated by men (64.4%).

The main characteristics of the patients who entered the study are shown in *Table 1*. In elderly patients (p = 0.022) with a higher BMI (p = 0.022), sepsis and septic shock as a result of generalized intra–abdominal infection were significantly more common (*Table 1*). It was in these patients that higher scores were observed on the APACHE II score (p = 0.002),
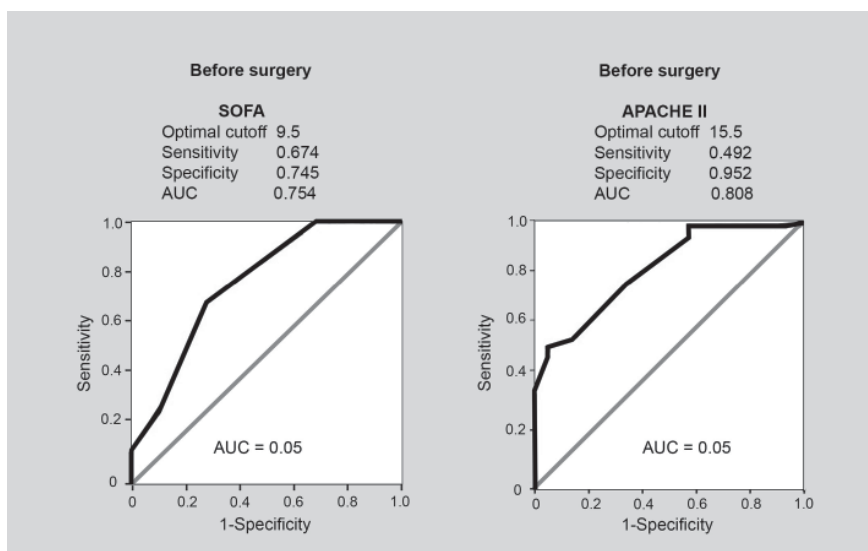
Figure 3.
ROC curves SOFA and APACHE II scores
before surgery.

on the SOFA score (p = 0.021); due to organ failure were used more often than in patients without sepsis such support as mechanical ventilation (p = 0.008), inotropic support (p = 0.010), and artificial kidney (p = 0.048) were used more often than in patients without sepsis. In terms of ICU stay, the differences between the three groups were also significant (p = 0.009), the mortality was dependent on the severity of the patient's condition, and the poor outcome was higher in patients with septic shock (p = 0.012). It was confirmed that the APACHE II score (AUC = 0.808 ± 0.06, 95% CI 0.695–0.918, cut–off value 15.5 with a sensitivity of 0.492 and a specificity of 0.952) had a better predictive value for assessing the degree of patients on admission to hospital than the SOFA score (AUC = 0.754 ± 0.03, 95% CI 0.614–0.878, cut–off value 9.5 with a sensitivity of 0.674 and a specificity of 0.727) (*Figure 3*).

Plasma levels of I–FABP and zonulin of patients which studies are depicted in Figures 3, 4 and in *Table 2*. We found statistically significant higher levels of I–FABP in the IAI group without sepsis: 490.8 ng/mL [IQR 298.5–748.8] as compared to the control group: 89.6 ng/mL [IQR 67.6–172.2]; in the sepsis group: 992.5 ng/mL [IQR 789.5 –1567.1] and in the septic shock group: 1678.1 ng/mL I[QR 1234.4– 2446.6] (P = 0.000).

The same tendency was observed in all periods of the study, and the most significant levels of I–FABP were by the tenth day after surgery in patients with septic shock: 2435.2 ng/mL [IQR 1567.3– 3876.1] (P = 0.000).

We also found statistically significant higher levels of zonulin in the IAI group without sepsis: zonulin in the sepsis group: 6.66 ng/mL [IQR 3.66 –9.45], as compared to the control group: 3.45 ng/mL [IQR 2.81–4.12]; in the sepsis group: 7.06 ng/mL [IQR 3.98–9.56] and in the septic shock group: 19.34 ng/mL [IQR 14.53–26.34] (P = 0.002). Patients with abdominal sepsis (n = 24) did not have a statistically significant change in zonulin levels: 7.06 ng/mL (IQR 3.98–9.56), compared to patients with IAI without sepsis (n = 26): 6.66 ng/mL (IQR 3.23–9.45) (P = 0.560). A similar trend was observed on the third day after surgery (P = 0.135). Only by the seventh and tenth days after surgery, changes in zonulin levels were significant in IAI patients without sepsis, with abdominal sepsis and septic shock: P = 0.000 and P = 0.004, respectively.

As shown in *Table 2*, there was significant alteration of zonulin in patients of septic shock in the all periods: 19.34 ng/mL [IQR 14.53–26.34] before surgery (n = 9); 19.34 ng/mL [IQR 14.53–26.34] before surgery (n = 9); 18.23 ng/mL [IQR
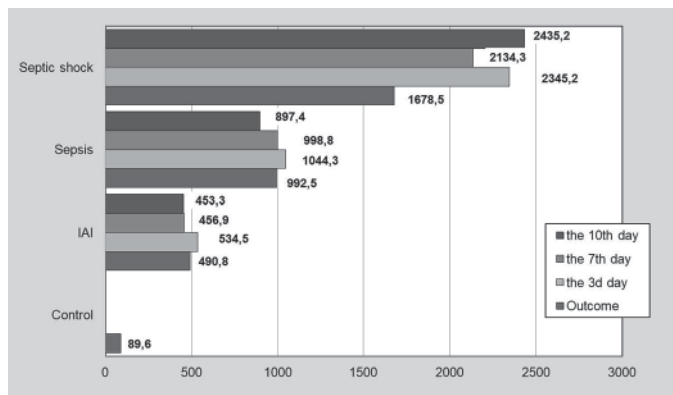

Figure 4.
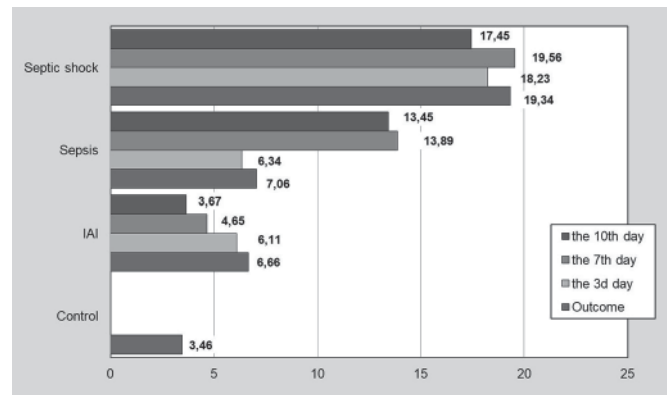*Plasma intestinal fatty acid binding proteins concentrations*
(ng/mL).
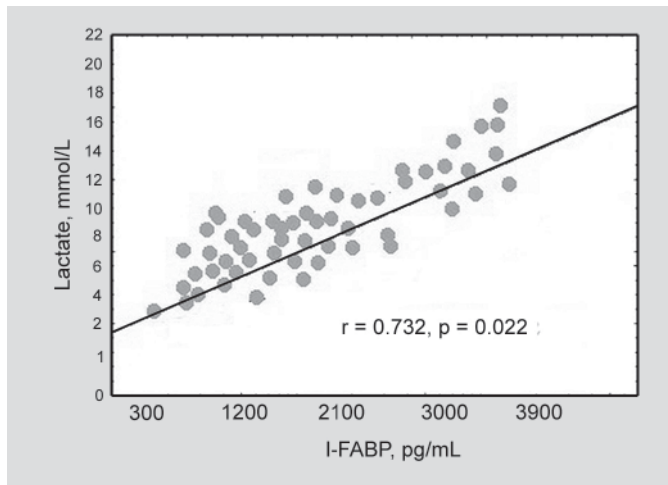

Figure 5.
Plasma zonulin concentrations (pg/mL).

Figure 6.
Correlations between plasma concentrations of Lactate and I-FABP.

Figure 7.
Correlations between plasma concentrations of Lactate and Zonulin.

16.14–21.22]; 19.56 ng/mL [IQR 17.23–22.12]; 17.45 ng/mL [IQR 16.11–19.23] (P = 0.419). It should be noted that there was no statistically significant difference between survivors (n = 21) 7.32 ng/mL (IQR 4.38–11.15) and non–survivors patients (n = 12): 8.23 ng/mL (IQR 4.89–17.17) with sepsis and septic shock (P = 0.185).

There was a positive correlation between Lactate and I–FABP plasma concentrations (r = 0.732; p = 0.022) (*Figure 4, 5*) and was not found to be associated between Lactate with zonulin (r = 0.311; p = 0.215) before surgery at all (*Figure 6, 7*).

Discussion

There are many risk factors for mortality in generalized intra–abdominal infection (early MOF, use of vasopressors, ventilation for more than 24 hours, surgical stress, the development of decompensated septic shock, etc.), and the intestine is the "motor for the development of multiple organ failure" [24]. As the function of the digestive tract is known to be complex, many researchers have tried to develop various assessment systems to assess its severity in the ICU. AGI estimate proposed by ESICM working group [25], which includes abdominal signs and symptoms, assessment of intra–abdominal pressure and organ function, is considered an important indicator for assessing the function of the digestive tract in ICU patients. This classification is now classical and accepted by various medical societies. The barrier function of the digestive tract can be impaired due to severe structural damage to any of the components of the intestinal barrier [26].

According to S.C. Bischoff et al. (2014), the terms "intestinal barrier" and "intestinal permeability" describe two different aspects of the same anatomical structure – the intestinal wall, consisting of mucosa, submucosa, muscular and serous membranes [27] and in their opinion, the intestinal barrier is a functional formation that separates the intestinal lumen and the internal environment of the body, intestinal permeability makes it possible to assess the functioning of the intestinal barrier. Normal intestinal permeability in persons without signs of intoxication, inflammation, intestinal dysfunction is char-
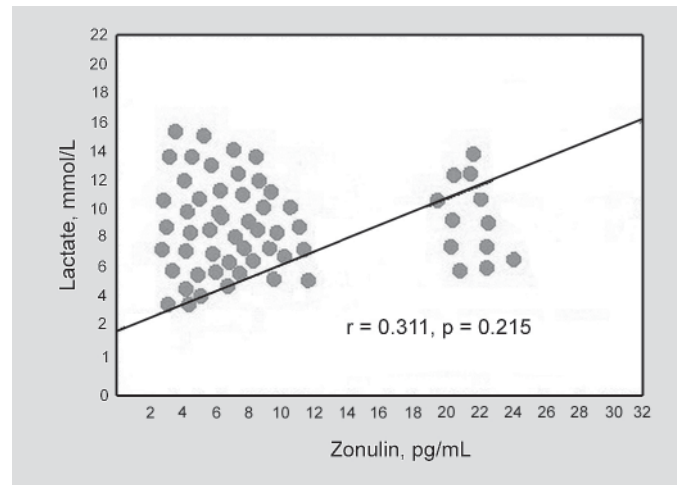
acterized by stability; violation of permeability is periodically unstable and can lead to loss of intestinal homeostasis and the development of functional disorders and organic diseases.

There are two large groups of biomarkers that can be used to assess intestinal permeability. The first group includes markers of epithelial cell damage such as citrulline, fatty acid binding proteins (FABP), and other biomarkers. The second group is markers of intestinal and immune inflammation: fecal calprotectin, zonulin, secretory IgA, and others.

Fatty acid–binding proteins (FABPs) are a family of small cytosolic water–soluble proteins that are present in enterocytes that can be detected as a result of damage to enterocytes, and therefore can act as biomarkers of enterocyte death and mucosal atrophy intestinal lining. These proteins are involved in the transport of fatty acids from the apical membrane of the enterocyte to the endoplasmic reticulum, where the synthesis of complex lipids occurs [28]. Proteins of the FABP family can be used as markers of tissue damage, since they are low molecular weight soluble proteins located in the cytoplasm and having high tissue specificity [29–32]. An important regulator of permeability is zonulin, a peptide with a molecular weight of 47 kDa. Physiological role the zonulin system has not been finally established, however, it is known that it is involved in the regulation of tight junctions, and excessive activation of zonulin production, both in duration and in severity, can lead to an excessive and unregulated increase in the permeability of the epithelial barrier. The available data on the role of zonulin in the regulation of tight junctions made it possible to use its determination in blood serum or feces to assess the state of intestinal permeability [33, 34].

In the studies of Kimball and Kirkpatrik it is noted that infection, trauma, burns lead to a significant activation of a nonspecific link of the immune system [35]. This systemic response leads to massive production of pro–inflammatory cytokines, the consequence of the above phenomena is the accumulation of fluid in the interstitial space and the development of local tissue ischemia [36]. It is this mechanism that underlies the development of acute damage to the intestine

that manifests itself as edema of the intestinal wall and entails the development of other damage [37]. Another marker of bowel function is the level of intra–abdominal pressure, since a number of studies have shown that it is this marker that correlates quite closely with the outcome in critically ill patients [38]. This study has shown that it found statistically significant higher levels of I–FABP in the IAI group without sepsis as compared to the control group, in the sepsis group, and in the septic shock group (P = 0.000). The same tendency was observed in all periods of the study, and the most significant levels of I–FABP were by the tenth day after surgery in patients with septic shock (P = 0.000). It had also found statistically significant higher levels of zonulin in the IAI group without sepsis, in the sepsis group as compared to the control group (P = 0.002). Patients with abdominal sepsis did not have a statistically significant change in zonulin levels compared to patients with IAI without sepsis (P = 0.560). A similar trend was observed on the third day after surgery (P = 0.135). Only by the seventh and tenth days after surgery, changes in zonulin levels were significant in IAI patients without sepsis, with abdominal sepsis and septic shock: P = 0.000 and P = 0.004, respectively. In addition, it was noted that there was a positive correlation between Lactate and I–FABP plasma concentrations (P = 0.022) and it was not found to be associated between Lactate levels with zonulin P = 0.215).

Inspite of the evidence received that serum I–FABP levels were a valuable and objectively early predictor of the severity of gastrointestinal tract injury. However, no similar evidence has been established for zonulin. The study has some limitations, including the lack of the intestinal microflora analysis and the absence of microflora bacterial (ribosomal) DNA identification in plasma samples, and the lack of performance of intestinal permeability tests. Despite the relatively small sample size and single–center design of the presented work, our data indicate that the use of I–FABP is an objectively early predictor of damage to the digestive tract in generalized intra–abdominal infection.

Conclusion

The impact of sepsis on the gut is manifold, e.g., sepsis mediated alteration of the gut–blood barrier and increase in the intestinal permeability, which may correlate with the phenomena of bacterial translocation. Systemic consequences of sepsis are widespread but nevertheless, the therapeutic approaches for modulating the mucosal immune system are still rarely effective in the treatment of these patients. Serum I–FABP levels were avaluable and objectively early predictors of the severity of gastrointestinal tract injury in generalized intra–abdominal infection. We also presented evidence of increased plasma zonulin levels in gIAI compared to control group. Elevated zonulin was been serve as an additional mechanism of observed increased intestinal permeability during abdominal sepsis but it had not been the similar as I–FABP as early predictor of the severity of gastrointestinal tract injury evidence.

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