Serum protease inhibitor - Ulinastatin in the complex therapy of severe acute pancreatitis complicated by sepsis

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ABSTRACT

Acute pancreatitis is an acute inflammation of the pancreas that often leads to pancreatic necrosis and is the cause of death in 40% to 70% of patients. [1]
Given the severity of this disease and the insufficient effectiveness of generally accepted methods of treatment, we studied the effectiveness of the drug Ulinastatin (serum protease inhibitor) in the complex therapy of acute pancreatic necrosis, complicated by diffuse purulent peritonitis and sepsis.

Keywords: acute pancreatitis, pancreatic necrosis, sepsis, peritonitis, trypsin, serum protease inhibitor, enzymatic autolysis, SOFA, SAPS II, APACHE II, interleukins.

Acute pancreatic necrosis is an acute inflammatory process in the pancreas (pancreas), capturing the surrounding tissues, accompanied by a syndrome of systemic inflammatory response with its transition to multiple organ failure with a high probability of death. (Berger H. Y., 2018, A. Y. Xiao et al, 2016).
The incidence of acute pancreatictitis (OP) worldwide ranges from 4.9 to 73.4 cases per 100,000 population and has a pronounced upward trend [2-3].
In recent decades, the problem of acute pancreatitis remains one of the most urgent in emergency abdominal surgery [4-6]. Among the emergency diseases of the abdominal organs, OP accounts for 3 to 10% of cases. In the structure of acute pancreatitis, the share of patients with pancreatic necrosis accounts for an average of 15 – 30% [7, 8]. Pathology has become a medicosocial problem in the structure of urgent surgery, mortality in OP ranges from 1 to 75%, depending on the form of the disease [3, 7-10].
Pancreatic necrosis is one of the most severe manifestations of acute pancreatitis and a formidable surgical disease. Mortality in pancreatic necrosis, even in specialized clinics, according to various authors, is from 11 to 30%, with large-focal pancreatic necrosis - more than 70% [3, 7, 11-13]. The fate of a patient with acute pancreatic necrosis is largely determined by the volume of pancreatic necrosis and the

addition of infection. The absence of any surgical intervention in this situation brings the mortality rate closer to 100 [7].

The pathogenesis of acute pancreatic necrosis is based on "pancreatic self-digestion" [14].

Normally, the protection of the pancreas from self-digestion occurs because its enzymes are released in an inactive form and are activated in the duodenum. Intestinal enterokinase activates the trypsinogen proenzyme, converting it to trypsin, which catalyzes the activation of other enzymes. It is known that normally the pancreas produces trypsin inhibitors, which prevent its spontaneous activation in acinar cells. In acute pancreatitis, spontaneous activation of trypsin causes activation of other enzymes, the coagulation cascade and fibrinolysis, which contributes to pancreatic necrosis and the spread of the process beyond the pancreas [15].

Enzymatic autolysis of the pancreas triggers the syndrome of a systemic inflammatory response involving the immune system, which leads to thrombosis of the vessels of microcirculation, ischemia and necrosis. [16].

The study of the effect of trypsin on the hemocoagulation system showed that even a negligible concentration of trypsin in plasma (0.5 μg / ml) has a pronounced procoagulant activity, while an increase in the level of trypsin in the blood causes localized (in the pancreas) and diffuse intravascular coagulation of blood, on the degree of which the stage, form and clinical symptomatology of acute pancreatictitis depends.

The main factors linking the pathogenesis of pancreatic necrosis and disorders in the hemostasis and fibrinolysis system are:

- direct influence of pancreatic enzymes on coagulation cascades, activation of factors and endothelial damage;
- pathological pain stimulation (vegetative, sympathetic) and chemo stimulation during the "enzyme explosion" forms a "vicious circle" and contributes to the additional release of inflammatory mediators and biologically active substances that affect the coagulation status;
- metabolic, endocrine and inflammatory reactions-constituent syndrome of systemic inflammatory response;
- intestinal paresis and intra-abdominal hypertension lead to a slowdown in blood flow in the system of the inferior vena cava (stasis) and ischemia of the abdominal organs;
- the phenomena of pancreatic shock cause capillary leakage syndrome, microcirculation disorders throughout the body, leading to the pathology of hemostasis.

It was shown that patients with severe acute pancreatitis were characterized by a higher level of D-dimer and a lower concentration of protein C compared to patients with mild pancreatitis [6]. [17], [5], after analyzing multiple logistic regression, they showed that D-dimer levels >400 ng/ ml and AT-III <71% at admission were associated with higher mortality (OR 11.2, AUROC 0.70 and OR16.6 AUROC 0.82, respectively). Thus, the authors conclude, the levels of D-dimer and antithrombin III can be used to assess the severity of the disease and predict the outcome.

To date, there is no specific therapy for acute pancreatitis and pancreatic necrosis that would reduce the cascade of inflammation, prevent coagulation disorders and inhibit trypsin, which is the most important trigger of acute pancreatitis.

As a therapy that could stop the ongoing digestion of the pancreas itself, reduce the cascade of inflammation and disseminated intravascular coagulation syndrome, the use of protease inhibitors, one of which is ulinastatin (trypsin inhibitor isolated from human urine), is considered. [18].

The precursor of ulinastatin - inter - a - trypsin inhibitor, the liver is normally synthesized [19].

Inflammation stimulates the release of serum proteases from neutrophils, macrophages, lymphocytes, mast, endothelial and epithelial cells. Activated neutrophilic elastase breaks down inter-a - trypsin inhibitor, turning it into ulinastatin.

Prior tothis, it is a fact that the level of endogenous ulinastatin in the blood plasma increases during inflammation [20].

Ulinastatin inhibits various serum proteases (trypsin, thrombin, chymotrypsin, plasmin, elastase and others), blood coagulation factors (IXa, Xa, XIa, XIIa)[21], inhibits the production of pro-inflammatory interleukins (IL-1, IL-6, IL-8),

stimulating the release of anti-inflammatory interleukin 10 and T cells [22].

All of the above prompted us to study the effectiveness of Ulinastatin in acute pancreatic necrosis, complicated by peritonitis and sepsis.

Purpose of the study:

Improve the results of treatment of patients with acute pancreatic necrosis, complicated by diffuse purulent peritonitis and sepsis.

Clinical materials and research methods:

In the intensive care units of TMA clinics, we examined 12 patients with acute pancreatic necrosis complicated by diffuse purulent peritonitis and sepsis (8 men and 4 women), whose average age was 56.1 to 2.4 years. These patients underwent surgery. All patients were divided into 2 groups: a control group, which included 6 patients, received standard therapy (antibacterial±, infusion-transfusion, detoxification, anticoagulant (low molecular weight heparins), sedating, analgesic, anti-enzymatic and symptomatic therapies) and the study group, which included the remaining 6 who, in addition to this therapy, received ulinastatin (serum protease inhibitor) in individually selected dosages.

Both groups were randomized by us according to gender and age, the nature of the standard examination and surgical treatment.

All patients underwent clinical and biochemical studies, radiography, computed tomography (CT), in the process of therapy, blood pressure (BP), average blood pressure SrAD, central venous pressure (CVD), blood glucose, thermometry and saturation of venous (jugular) blood were monitored.

In addition to general clinical methods of blood and urine research, 6 patients of the study group monitored coagulogram indicators, biochemical blood parameters, blood tests for pro-inflammatory interleukins (IL-1, IL-6, IL-8) and anti-inflammatory interleukins (IL-10), C-reactive protein, markers of kidney function (urea, creatinine), liver (total protein content, bilirubin, serum transaminases), CSC and blood gas composition, procalcitonin test.

С целью прогнозирования исходов заболевания через каждые трое суток мы использовали шкалы SOFA (Sequential Organ Failure Assessment), SAPS II (Simplified Acute Physiology Score), APACHE II

(The Acute Physiology and Chronic Health Evaluation). Уровень сознания определяли по шкале Глазго.

According to ultrasound data, the volume of fluid in the abdominal cavity was determined, the excreted exudate from the drains of the abdominal cavity and retroperitoneal space was monitored and counted.

Such indicators of inflammation as leukocytosis, leukocyte index of intoxication, the level of alpha-amylase of blood and urine diastase were also monitored, and determined by unified methods.

The duration of stay of patients in the ICU and in the multidisciplinary clinic TMA as a whole was studied.

The results of our own research:

During the course of our study, out of 6 selected patients in the study group, only one patient had a fatal outcome, on the example of which we would like to demonstrate the data obtained by the study group.

Patient Gafurov A.Kh. 68 years old, medical history No., was taken by ambulance to the multidisciplinary clinic of the Tashkent Medical Academy on November 6, 2021, in extremely serious condition with a diagnosis of acute pancreatitis (pancreatic necrosis?), diffuse peritonitis.

After the diagnostic laparoscopy on November 6, 2021, an operation was performed - median laparotomy, drainage of the omentum bag, abdominal cavity, retroperitoneal tissue.

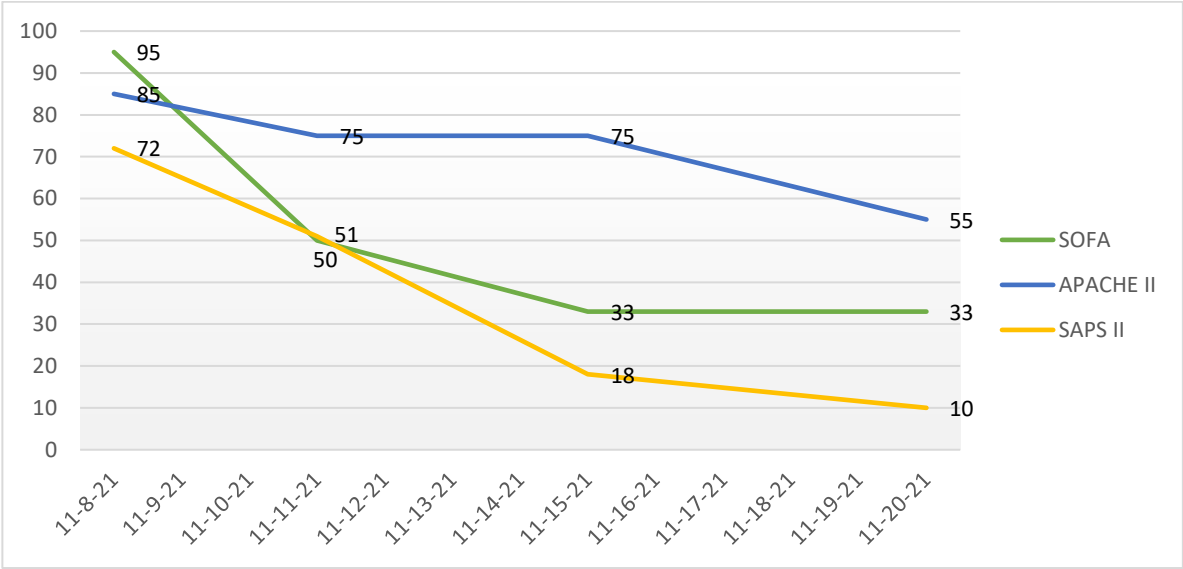
Postoperative diagnosis: total infected pancreatic necrosis with the spread of the process into the retroperitoneal tissue. Sepsis. Complications: diffuse fibrinous-purulent peritonitis. Concomitant pathology: cholelithiasis, chronic calculous cholecystitis, bilateral hydrothorax.

In the intensive care unit, the patient was continued with a ventilator, catheterization v was performed. Subclavia dextra according to Seldinger. Antibacterial, infusion-transfusion,

detoxifying therapy with early nutritional support was carried out - a complete parenteral nutrition with a total callocillation of 3,000 kcal, but with a high (up to 1.6 g / kg body weight) protein content.

This patient underwent all the research in full. Stages of the study: initial data, 3, 7, 12 days after the surgical intervention.

Chart No. 1. Dynamics of outcome prediction data on the three scales described above at the study stages.



From the data presented, it is easy to see that on all three scales, the initial percentages of 95 - 72%) of the predicted mortality rate became lower every three days in the process of the therapy, reaching 10 - 55% by 11 - 12 days, which indicated its effectiveness.

Table 2. Dynamics of leukocyte index of intoxication, alpha amylase, C reactive protein and procalcitonin at the stages of the study.

Indicator/ Date	09. 11. 2021	12. 11. 2021	15. 11. 2021	20. 11. 2021
LEE	12,7 (3)	10,9	8,4	5,1
Alpha/amimanhole, E/L	48 (13-53)	19,9	34,9	33,8
C-reactive protein, IU/ml	18.9 (<14)	24,8	21,1	15,4
Procalcitonin, mg/ml	2.17 (<0.05)	1,38	0,94	0,51

Note: the normal values of the studied indicators are given in parentheses.

The indicated effectiveness of thetherapy was indicated by the dynamics of the indicators of inflammation given in this table.

So LII by 11 days decreased by 60.1%,C-active protein - by 8.5%.

The procalcitonin test continued to indicate a persistent systemic inflammatory response syndrome (SSS), but with a clear tendency to reduce its intensity.

Table No. 3 Dynamics of pro- and anti-inflammatory interleukins.

Interleukins	09. 11. 2021	12.11. 2021	15.11. 2021	20.11. 2021
IL-1 бета Pg/ml	19 (<14)	25	20	18
IL – 6 Pg/ml	79.3 (<7)	64	53	44,9
IL – 8 Pg/ml	25.9 (<12)	29,35	26,4	21,0
IL – 10 Pg/ml	9,9 (,8)	17,33	14,88	13,7

Note: the normal values of the studied indicators are given in parentheses.

The most indicative was the dynamics of pro- and anti-inflammatory interleukins in the process of complex therapy with the inclusion of Ulinastatin.

Pro-inflammatory interleukins (cytokines) IL-1, IL-6, IL-8 by day 11 decreased by 5.3%, 43.4%, 19%, respectively, but all of them still exceeded the normal value. The values of anti-inflammatory IL-10 (anticytokine) at all stages were higher than the initial values.

All of the above clearly confirmed the positive role of Ulinastatin in blocking the cytokine cascade.

Table 4. Dynamics of coagulogram data at the stages of the study.

INDEX /DATE	09. 11. 2021	12. 11. 2021	15. 11. 2021	20. 11. 2021
Prothrombin. time, sec	16,2 (12-15)	12,9	13,4	16,5
PTI ,%	63 (75-120)	90	84	61
MNO	1,38 (0,8-1,25)	1,08	1,13	1,32
APTT, sec	32 (25-43)	35	39,5	38,3
Fibrinogen, mg/dl	580 (200-400)	587	434	404
D-dimer, ng/ml	978 (<285)	1025	649	306

Note: the normal values of the studied indicators are given in parentheses.

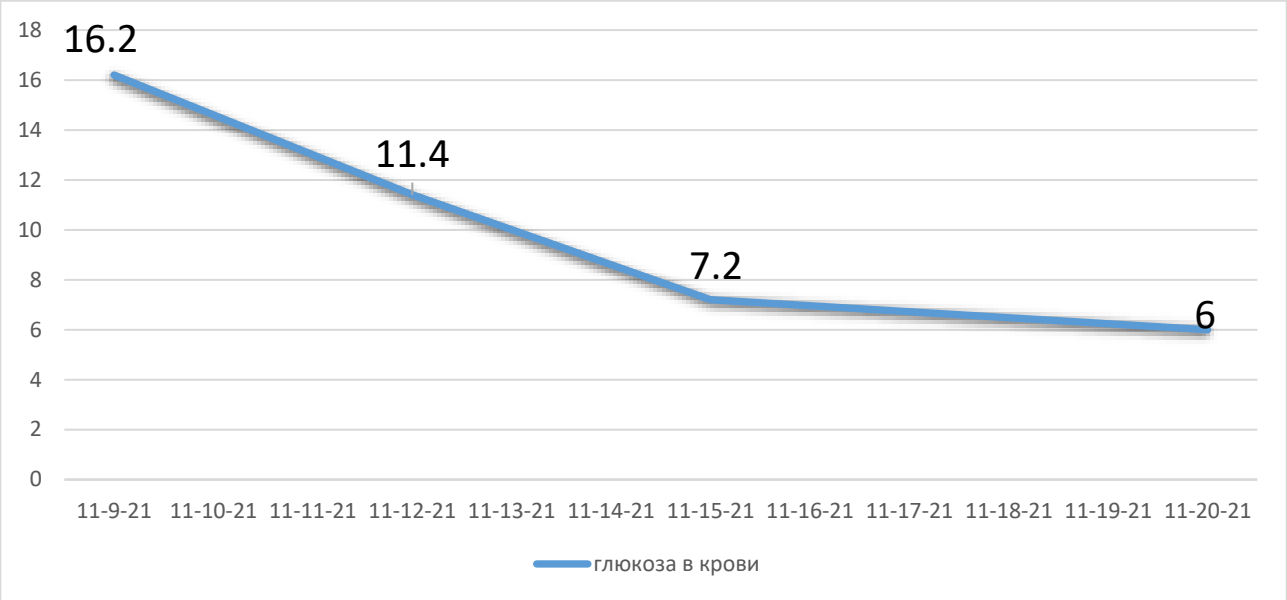
Interesting was the dynamics of the coagulogram indicators, which throughout the therapy indicated the presence of DIC syndrome of the second stage, since they were multidirectional in nature.

So, if the prothrombin time, PTI indicated an increase in the activity of the anticoagulation

system of the blood, then the values of fibrinogen, D-dimer indicated the continuing intravascular thrombosis.

These circumstances were the reason to abandon heparin therapy due to the danger of bleeding from necrotic tissues of the retroperitoneal space.

Graph No. 2 : Dynamics of glucose in the blood.



As for the dynamics of changes in the concentration of glucose in the blood, after the initial hyperglycemia, by the sixth day it reached subnormal values. By day 11, plasma glucose values normalized without further reduction.

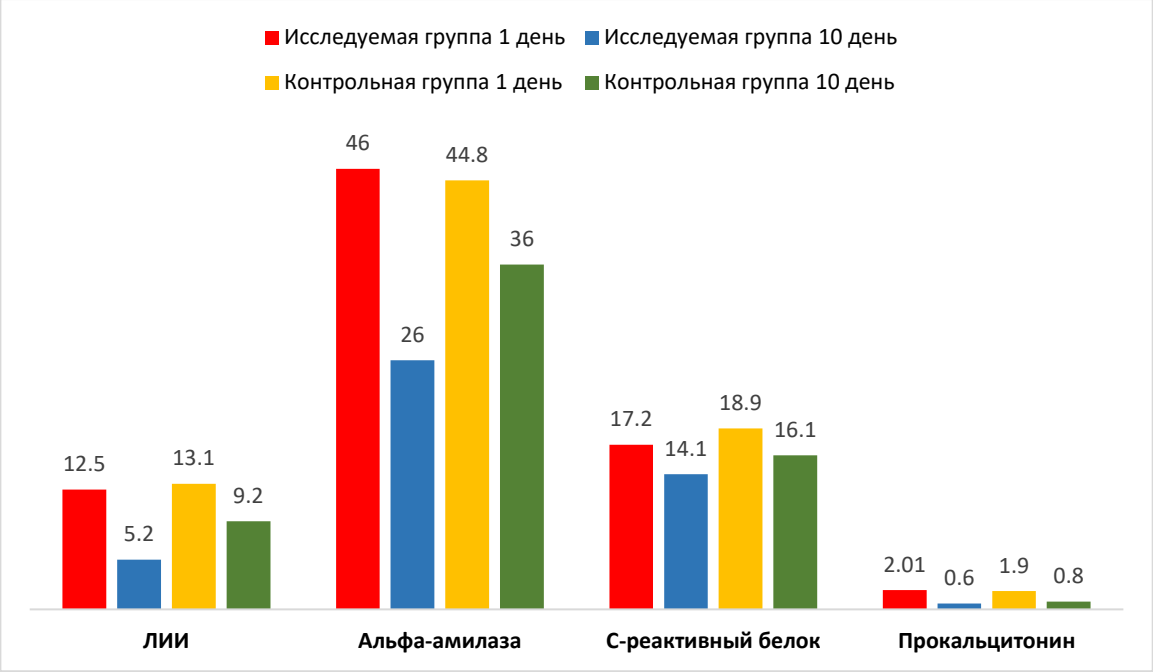
The initial dosage of ulinastatin was 100,000 units three times a day intravenously in saline.

Due to the remaining high titers of pro-inflammatory interleukins, the value of C-reactive protein and procalcitonin test scores, as well as abundant cheesy discharge from drains on the third day, the dosage of ulinastatin was increased to 200,000 units twice a day, which subsequently had a positive effect on the studied indicators (see 7 days).

A three-day break in the administration of the drug again increased the concentration of IL 6, 8 and the amount discharged from the drains with a weakening of intestinal motility, which forced to resume the administration of the drug at a dose of 200,000 units three times a day intravenously drip. Over the next three days, discharge from the drains was significantly reduced, good intestinal motility resumed with a positive overall clinical picture. With ultrasound examination of organs abdominal cavity, fluid in the abdominal cavity was practically not noted.

On the 11th 12th day, the drains were removed, the patient was verticalized, activated, transferred to independent breathing through the tracheostomy hole (the tracheostomy was performed on the third day after the operation). The patient was transferred to the surgical department, where on the third day he died of acute respiratory failure caused by submassive thromboembolism of the pulmonary artery, despite the fact that upon admission and on the fourth day, duplex scanning of the deep veins of the lower extremities did not reveal the presence of thrombotic changes. Apparently, PE originated from the injured veins of the pelvis and retroperitoneal space. The description suggests that in these conditions, elastic bandaging of the lower extremities, their elevated position is not enough to prevent deep vein thrombosis of the lower extremities and pelvis. It is necessary to prescribe low-molecular heparins despite the possibility of bleeding from self-digestible tissues.

Chart No. 3. Comparison of the dynamics of inflammation in both groups.



This graph shows a more positive dynamics of normalization of inflammation indicators in the study group compared to the control group. Reliability: p*- <0.05.

Chart #4. Comparison of the stay of patients of both groups in the ICU.

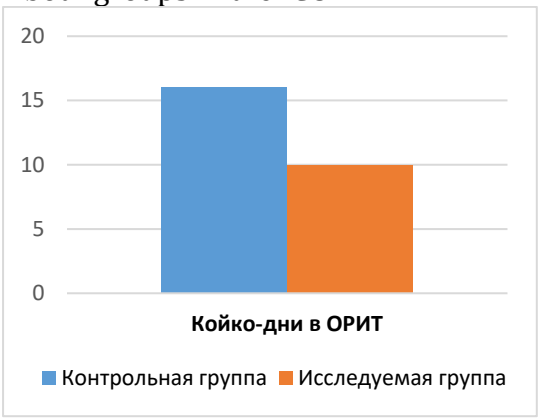
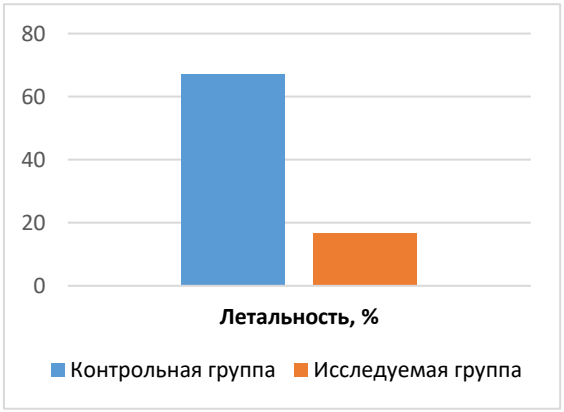


Chart #5. Comparison of the mortality of patients in both groups.



When comparing the final values of the studied indicators in the control and study groups, the following data were obtained: the duration of stay of patients in the ICU was 16 and 10 days, and the mortality rate was 67.2% and 16.6%, respectively.

Findings:

This study clearly indicates the effectiveness of Ulinastatin in severeacute pancreatitis and sepsis, providing atogenetically directed effect.

The use of Ulinastatin (serum protease inhibitor) in the early post-napedietary period with pancreatic necrosis increases the survival rate of patients, reducing their stay in the ICU and in the hospital.

We consider it necessary in patients with pancreatic necrosis to carry out comprehensive prevention of thromboembolic complications with the mandatory use of LMWH , monitoring of hemostasis, despite the high risk of bleeding from destructive tissues of the pancreas, retroperitoneal tissue, since only physical

methods of preventing thrombofor deep veins of the lower extremities, pelvis and retroperitoneal tissue are ineffective.

Ongoing research in our clinic on the use of ulinastatin in patients with acute panctreatitis and sepsis of various etiologies made it possible to include it in the algorithm for treating such patients.

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