ISSN: 0975-8585

Research Journal of Pharmaceutical, Biological and Chemical Sciences

Improvement of antibiotic therapy efficacy in an experimental clinical trial in acute pancreatitis.

A.M. Zainutdinov[1]*, I.S. Malkov[2], and A.U. Ziganshin[3].

[1]Assistant professor of the Department of fundamentals of clinical medicine of Kazan Federal University, assistant professor of Department of surgery SBEI CPE KSMA. 420141, Kazan, Fuchika Str.
[2]Professor, Head of Department of surgery SBEI CPE KSMA, 420139, Kazan, Zorge Str.
[3]Professor, Head of the Department of Pharmakology SBEI HPE KSMU, 420012, Kazan, Butlerova Str. 49,

ABSTRACT

This paper presents a brief overview of the methods of treatment of acute pancreatitis and describes an experimentally-clinical research of improvement of antibiotic therapy efficacy. Particular emphasis is placed on the effectiveness of intensive conservative treatment of aseptic pancreatic necroses, particularly, antibiotic therapy. The main causative agents of infection in the infected pancreatic tissue are presented in this paper.
The research was conducted at the Department of Surgery of SBEI CPE KSMA and the Department of Pharmacology of SBEI CHE KSMU on 40 male and female rats weighing 150 to 250 grams used to study the concentration of Cefotaxime by high performance chromatography with different methods of its administration. Application of galvanization at administration of antibiotics in experimental pancreatitis is more efficient as compared with conventional antibiotic administration after 24 hours of reproduction of acute pancreatitis. The clinical part of the study involved comparison of a group of patients (n-27) undergoing an interstitial electrophoresis time on the background of antibacterial therapy, and a group of patients (n-26) receiving conventional antibiotic treatment. Complications in the main group were observed in 13% of patients with a predominance of localized infections (3 cases of acute fluid pockets, 1 case of retroperitoneal abscess) as compared with the control group - 34.6% of patients with a predominance of extensive forms of infection (15% had omental and retroperitoneal abscesses, 19% had retroperitoneal phlegmon). Accordingly, the main group underwent predominantly non-invasive procedures, and the control group underwent laparotomy, marsupialization, and lumbotomy. The mortality rate in the study group was 10%, and in the control group - 15%. The use of interstitial electrophoresis in addition to antibiotic therapy contributes to reduction in the frequency of septic complications, to predominance of localized infections, and reduces mortality in patients with acute destructive pancreatitis.
Keywords: Acute pancreatitis - Antibiotic therapy - Galvanization - Chromatography-Electrophoresis.

Corresponding author

ISSN: 0975-8585

INTRODUCTION

A large number of both the domestic and the foreign publications deals with the issues of treatment of acute pancreatitis [7,8,9,12,14,18,19,22]. A recent characteristic feature is a significant revaluation of thought-to-be "classical" methods of conservative therapy and surgical interventions. Unlike many acute surgical diseases, the leading method of treatment of acute aseptic pancreatitis today is a conservative intense and pathogenetically grounded treatment with minimally invasive technologies.

In recent years, most surgeons adhere to the tactics of combination treatment of this disease that includes active conservative measures, and conduct surgical treatment only in the advanced stages of acute pancreatitis with symptoms of pancreatic necrosis and peritonitis [3,31]. A complex of conservative therapy involves the deactivation of pancreatic enzymes, detoxification, forced diuresis, the correction of both acid-base and electrolyte balance. High mortality in the early stages of acute pancreatitis indicates the lack of effectiveness of conservative treatment and calls for further improvement of both the conservative and surgical therapy [5,13,14]. The difficulty of treatment is due to the complex pathogenesis and course of acute pancreatitis, starting with the intracellular and interstitial activation of pancreatic enzymes and ending with circulatory hypoxia [2,10,13,14,15].

Studies by European surgeons have shown that most patients with aseptic pancreatic necrosis can safely do without operations, using instead the appropriate antibiotics and enzyme inhibitors [10,27,30]. Many domestic authors keep safely only to laparoscopic debridement in aseptic pancreatic necrosis [4,6,7,10]. The effectiveness of conservative treatment of sterile pancreatic necrosis has led to an increase in the number of supporters of minimally invasive and conservative treatment of patients with non-infected pancreatic necrosis [1,4,11,18,19,29].

The leading role in the treatment of pancreatic necrosis belongs to antibiotic treatment [32]. Destructive pancreatitis presents a high risk of infection of the pancreas, resulting in significantly worsened prognosis [16,17,24]. In this case, an important role belongs to the right choice of antibiotics and their mode of administration [28]. In clinical practice, especially in the early stages of treatment of acute pancreatitis, an empirical therapy is used, which is based on the prescription of these drugs, covering the flora of both pancreatogenic and, possibly, nosocomial infection [26].

Currently, many authors found that the main causative agents of pancreatic infections are gram-negative microorganisms, in particular *Echerihia Coli*, the opportunistic enterobacteria (Klebsiella, Proteus), and *Candida* [21, 23].

It was experimentally shown that the administration of antibiotics does not reduce mortality. However, the administration of Cefotaxime [33], piperacillin, and imipenem [20] significantly reduces the incidence of infection of the pancreas (pancreatic). Mithofer et al. [25] have managed to reduce the mortality of test animals with the use of antibiotics.

In connection with this, the problem was set to study the effect of the changing Cefotaxime concentration when using the method of galavanization on the concentration in the pancreatic tissue of experimental animals.

MATERIALS AND RESEARCH METHODS

Experimental studies were conducted on 2 groups of animals at the Department of Surgery of SBEI CPE KSMA of the Federal Service on Surveillance in Healthcare. The first group of 20 rats of both sexes, weighing 150 to 250 grams, was used to study the concentrations of Talcef in pancreatic tissue after its introduction into the femoral vein by high-performance chromatography after 3 and 6 hours of simulation of acute pancreatitis. The second group of 20 rats was studied for the concentration of Talcef in pancreatic tissue by flash chromatography after 3 and 6 hours of simulation of acute pancreatitis after its introduction into the femoral vein and after interstitial electrophoresis.

The sample was centrifuged to remove tissue particles, and acetonitrile was added to the filtrate to precipitate all protein compounds and centrifuged again. The supernatant was investigated by HPLC (high

performance liquid chromatography). The 0.01 M solution of H_3PO_4 and acetonitrile at a ratio of 60:40 (pH=3.0) was used as the mobile phase. The chromatographic unit consisted of a Rainbow injector (USA), coaxial with a Shimadzu SPD10A spectrophotometer (Japan). A Spherisorb column (Hichrom, UK) of 25 cm long and 0.46 cm in diameter was used for nucleotides separation. At the preparatory stage, the column was washed during 24 hours with the mobile phase, at a flow rate of 0.2 ml/min. Chromatography was performed at a mobile phase flow rate of 1 ml/min, at a wavelength of 260 nm, the recording speed of 1 cm/min and a sample volume of 20 µl. Talcef concentration was determined by comparing the peak heights of the experimental samples with the respective peak heights obtained from the standard Talcef solutions. Data were recorded on a peak recorder with different amplitudes. The chromatographic system consisted of a SPD-10A injector, coupled with a LD-10AC spectrophotometer and RPD-10R recorder (Shimadzu, Japan) using Srherisorb column (Hichrom, Great Britain), of 25 cm long and 0.46 mm in inner diameter. The column was washed for 24 hours at the rate of 0.2 ml/min with the mobile phase consisting of the solution of acetonitrile and 0.01 M ortho-phosphoric acid (H_3PO_4) at a ratio of 40:60, pH 3.0. Chromatography was performed at a mobile phase flow rate of 1 ml/min, at a wavelength of 260 nm. The recording speed was 1 cm/min, and a sample volume - 20 µl. Tissue homogenates were deproteinized several times and added with equal amounts of acetonitrile. The concentration of the drug was determined by comparing the area under the peaks in the experimental samples with the corresponding areas of the standard Cefotaxime (Talcef) samples. Statistical analysis of the experimental and clinical results of the study was carried out by variance analysis with the calculation of the arithmetic mean (M) and the arithmetic mean error (m). The significance of differences of averages was determined by Student's t-test on “Pentium” computer using the “Statistica” programs (version 5.0.).

RESULTS

The concentration of Cefotaxime in rat pancreatic tissue at different stages of experimental acute pancreatitis in the study and control groups is shown in Table 1. As can be seen from Table 1, the antibiotic concentration in the pancreatic tissue of animals of study group, depending on the duration of the experimental pancreatitis, changed after EAP simulation (experimental acute pancreatitis) as follows: the concentration of the antibiotic in the pancreas in case of combining an intravenous infusion of Cefotaxime with electrophoresis increased by 1.2 times (in the study group - 0.0224±0.1 mg/ml (P<0.01), in the control - 0.0105±0.1 mg/ml (P<0.05)); one can see 3 hours after EAP that the increase in concentrations in both groups is less in the study group, where the concentration was 0.166±0.1 mg/ml (P<0.01) as compared with control - 0.221±0.1 mg/ml (P<0.01). After 6 hours of simulation of experimental pancreatitis, the antibiotic concentration in the pancreatic tissue of the study group decreases to a lesser extent to 0.0283±0.1 mg/ml (P<0.01), and its decrease in the control group is 2 times more than in the study group and is 0.0108±0.1 mg/ml (P<0.01). After 24 hours of simulation of experimental acute pancreatitis, there is a significant difference in antibiotic concentrations in these groups. Thus, the antibiotic concentration in the study group during galvanization rises to 0.294±0.1 mg/ml (P<0.01), as compared with control group, which only received intravenous Cefotaxime 1 day after EAP simulation, and amounts to 0.037±0.1 mg/ml (P<0.05). The difference in the antibiotic concentration at different stages of acute experimental pancreatitis in the study and control groups are shown in Figure 1. As can be seen from Figure 1, using galvanization in acute experimental pancreatitis is more efficient after 24 hours of simulation of acute pancreatitis, as compared with conventional antibiotic administration.

Twenty-four patients hospitalized with the symptoms of aseptic pancreatic necrosis without early complications (18 men, 6 women; 42±12 years) with moderate severity, and 3 (3 men, 44±7 years) with high severity according to Dzhanelidze St. Petersburg Research Institute scale underwent interstitial electrophoresis in addition to the traditional conservative therapy at the time of antibiotic therapy for 8±4 days. The control group consisted of 26 patients (22 men, 2 women; 44±14 years) with moderate severity, and 2 (2 men, 43±6 years) with high severity according to Dzhanelidze St. Petersburg Research Institute scale, who received traditional conservative treatment. In these groups, both laboratory and clinical parameters, the conducted surgeries, complications during treatment, and mortality were analyzed. Laboratory findings (signs of systemic inflammatory response syndrome, such as fever, leukocytosis, heart rate, respiratory rate, leukocyte intoxication index, and biochemical parameters such as glucose, urea, blood, urine amylase) were compared at the time of admission and at the completion of the conservative therapy. If the laboratory parameters do not differ at the time of treatment, then, ten days after, there is a considerable improvement in the values of body temperature, white blood cells, Kalf-Kalif LII with respect to the values of control group ($p \leq 0.05$) (Table 2).

Four patients (13%) of study group had complications observed, predominantly - localized infections (3 patients had acute fluid pockets, which further transformed into omental abscess, 1 patient had retroperitoneal abscess that required the percutaneous drainage under ultrasound guidance). Septic complications occurred in 9 patients (34.6%) of control group. In this group of patients, the extensive forms of infection were predominant (5 patients - 19% - had retroperitoneal abscess, 4 patients had omental abscess with retroperitoneal abscesses). According to clinical results, the conducted antibiotic therapy combined with interstitial electrophoresis contributed in the study group to a decrease in the incidence of purulent complications with a tendency to process delimitation. All patients with purulent complications underwent surgery. The omental and retroperitoneal abscesses were drained by using minimally invasive techniques (puncture drainage under ultrasound guidance), and the extensive forms of infection were operated by conventional "open" methods such as laparotomy, marsupialization, and lumbotomy. In connection with this, the study group underwent mostly minimally invasive procedures, and the control groupunderwent open surgery through laparotomy.

The total and postoperative mortality was the same in the study and control groups - 10% (3 patients), and 15% (4 patients), respectively. One death in the study group was caused by the development of intestinal fistula and erosive bleeding that required a laparotomy. This complication occurred due to the revaluation of the capabilities of the minimally invasive treatment.

SUMMARY

Abnormal penetration of the antibiotic into the pancreatic tissue can be due to inflammatory infiltrations in the pancreatic tissue. The development of the destructive process leads to difficult penetration of antibiotics into the pancreas. Therefore, the objective of this study was to investigate the effect of different methods of antibiotic therapy on the inflamed pancreatic tissue.

Our experimental study showed that the most efficient transportation of antibacterial drug to the pathological focus occurred through interstitial electrophoresis having maximum effect after 24 hours of simulation of experimental acute pancreatitis.

The authors explain this phenomenon by the influence of the electric field generated between the electrodes, resulting in the varying ionic conjuncture in cells and tissues, an increased permeability of cell membranes, the activated redox processes, intensified action of enzyme systems, improved microcirculation, blood and lymph circulation, which promotes deeper penetration of the drug.

Using the method of interstitial electrophoresis during antibiotic therapy in patients with acute pancreatitis of moderate and high severity reduced the number of septic complications from 34.6% (control group) to 13% (study group). At the same time, the dominant infections in the control group were extensive forms (15% of cases with both omental and retroperitoneal abscesses, 19% of cases with retroperitoneal phlegmon). Accordingly, the main group underwent predominantly non-invasive procedures, and the control group underwent laparotomy, marsupialization, and lumbotomy. The mortality rate in the study group was 10%, and in the control group - 15%.

Based on the obtained experimental data on and clinical results of the use of interstitial electrophoresis as a part of a combination treatment of patients with moderate and severe acute pancreatitis contributes to reduction in the frequency of septic complications, to predominance of localized infections, which requires minimally invasive methods of treatment and reduces mortality in patients with acute destructive pancreatitis.

ACKNOWLEDGEMENTS

The work is performed according to the Russian Government Program of Competitive Growth of Kazan Federal University.

REFERENCES

[1] Briskin B.S., Rybakov G.S., Khalidov O.Kh. The place and role of minimally invasive surgery in the treatment of patients with pancreatic necrosis at different phases of development. The second congress of Moscow surgeons. Emergency and specialized surgical care, 2007, Pp. 77-78.

[2] Vashetko R.V., Tolstoi A.D., Kurygin A.A, Stoiko Iu.M., Krasnorogov V.B. - Acute pancreatitis and pancreatic injuries. St.P.: “Piter", 2000. Pp. 185-191.

[3] Galimzianov F.V. - Diagnosis and debridement of retroperitoneal fat lesions in necrotizing pancreatitis // International Surgical Congress. 2003, - 32 p.

[4] Galperin E.I., Diuzheva T.G., Dokuchaev K.V. - Minimally invasive interventions into omental and retroperitoneal fat for the prevention and treatment of infected pancreatic necrosis // International Surgical Congress. 2003, - 56 p.

[5] Gelfand B.R., Antibiotics in abdominal surgery. // Antibiotics and chemotherapeutic agents, 2001, V.46, No. 11, Pp. 11-18.

[6] Malkov I.S., Alukaev M.I., Zainutdinov A.M., Sharafislamov I.F., Kirshin A.P. A diagnostic method for diseases of the abdominal cavity and retroperitoneal space // A favorable decision on granting a patent for the invention of the Russian Federation No. 2003120741 / 021929 of 07.07.2003.

[7] Kriger A.G., Gorskii V.A., Vladimirov V.G., Seregin R.V. - Laparoscopy in the diagnosis and treatment of pancreatic necrosis // 6th International Congress on endoscopic surgery. 2002 - Pp. 177-178.

[8] Kriger A.G., Kokov L.S., Karmazanovskii G.G., Kuntsevich G.I., Fedorov V.D., Barbin P.B., Tarbaeva N.V. False aneurysm of the celiac trunk arteries in patients with chronic pancreatitis // Surgery. - 2008. - No. 12. Pp. 17-23.

[9] Kubyshkin V.A., Kriger A.G., Tsygankov V.N., Varava A.B. Treatment of patients with chronic pancreatitis complicated by false aneurysm of the celiac trunk artery and the superior mesenteric artery // Journal of Experimental and Clinical Surgery. - 2012. - v.5.- Pp. 12-21.

[10] Malkov I.S., Shaimardanov R.Sh. Acute pancreatitis.-Kazan. 2001,- 60 p.

[11] Mumladze R.B., Chechenin G.M., Lebedev S.S., Barinov Iu.V., Ivanova N.A. New possibilities of minimally invasive surgery in the complex treatment of patients with destructive forms of acute pancreatitis. The second congress of Moscow surgeons. Emergency and specialized surgical care, 2007, Pp. 98-99

[12] Popova E.Iu., Kuznetsov N.A., Vladimirov V.G., Zarinskaia S.A., Andreitsev A.N., Brontvein A.T., Kuzin A.N. The retroperitoneal fat lesion in destructive pancreatitis // Surgery - 2004.- No. 8.- Pp. 52-55.

[13] Rudakov A.A., Duberman B.L. - Pancreatic infection // Annals of Surgical Hepatology, 1998, Vol.3. No.2. Pp. 94-99.

[14] Saveliev V.S., Gelfand B.R., Gologorskii V.A., Filimonov M.I., Burnevich S.Z. The destructive pancreatitis in the light of modern ideas about sepsis // Annals of surgery. - 1999. - No.5. - Pp. 26-29.

[15] Saveliev V.S., Filimonov M.I., Gelfand B.R., Burnevich S.Z. Destructive pancreatitis: diagnosis and treatment algorithm (The project is based on the proceedings of IX All-Russian Congress of Surgeons, held on September 20-22, 2000, in Volgograd.) // Consilium Medicum, 2000, Vol.2, No. 6.- OnLine.

[16] Iakovlev S.V. The modern view of antibacterial therapy of intra-abdominal infections. Consilium medicum, 2003, Volume 05. No. 8.-OnLine.

[17] Beger H. G, Isenmann R. Surgical manangement of necrotizing pancreatitis // Surg. Clin. N. Amer. 1999. Vol., 79 - No. 4 – P. 793 –800.

[18] Bradley E.L., Dexter N.D. Management of severe acute pancreatitis: a surgical odyssey//Ann. Surg. – 2010.-№ 251. P. 6–17.

[19] Buchler MW, Gloor B, Muller CA, et al. Acute necrotizing pancreatitis: treatment strategy according to the status of infection. // Ann Surg 2000; № 232. P. 627-629.

[20] Foitzik T, Fernandez-del Castillo C, Ferraro MJ, et al: Pathogenesis and prevention of early pancreatic infection in experimental acute necrotizing pancreatitis. // Ann Surg. 1995. № 222. P. 179.

[21] Gloor B, Milller CA, Worni M, et al: Pancreatic infection in severe pancreatitis: The role of fungus and multiresistant organisms. // Arch.Surg. 2001. №136 P. 592-596.

[22] Hans G. Beger and Rainer Isenmann. Acute pancreatitis: who needs an operation?//J. Hepatobiliary Pancreat. Surg.- 2002. № 9. P. 436–442.

[23] Isenmann R, Schwarz M, Rau B, Trautmann M, Schober W, Beger HG Characteristics of infection with Candida species in patients with necrotizing pancreatitis. // World J Surg. 2002. - V. 26. - P.372–376

[24] Johnson C.D., Imrie C.W. Pancreatic disease.London, Springer.2004. P. 383-399.

[25] Mithofer F, Fernandez-del Castillo C, Ferraro MJ, et al: Antibiotic treatment improves survival in experimental acute necrotizing pancreatitis.// Gastroenterology.- 1996. № 110. P.232.

[26] Nordback 1, Sand J, Saaristo R, Paajanen H: Early treatment with antibiotics reduces the need for surgery in acute necrotizing pancreatitis - A single.center randomized study. // J. Gastrointest. Surg.- 2001. №5 P..113-120

[27] Paye F, Rotman N, Radier C, Nouira R, Fagniez P-L. Percutaneous aspiration for bacteriological studies in patients with necrotizing pancreatitis//Br. J. Surg. 1998.V. 85. P.755–759.

[28] Ratschko M., Fenner T., Lankisch P. // The role of antibiotic prophylaxis in the treatment of acute pancreatitis. Gastroenterology Clinics. 1999.-№28 – P. 641.

[29] Rau B., Pralle U., Uhl W., Schoenberg M.H., Beger H.G. Management of sterile necrosis in instances of severe acute pancreatitis//J. Am. Coll. Surg.1995 V.181.P.279–288

[30] Rünzi M., Layer P. Nonsurgical management of acute pancreatitis.Use of antibiotics// Surg. Clin. North. Am.1999 -V. 79.- P. 759–765

[31] Runzi M, Layer P, Buchler MW, et al: The therapy of acute pancreatitis. General guidelines. Working group of the Society for Scientific-Medical Specialties. // Z. Gastroenterol. – 2000- № 38. P.571-581.

[32] Sharma VK, Howden CW: Prophylactic antibiotic administration reduces sepsis and mortality in acute necrotizing pancreatitis: a metaanalysis. Pancreas -2001- № 22. P.28-31.

[33] Widdison AL, Karanjia ND, Reber HA: Antimicrobial treatment of pancreatic infection in cats. Br. J. Surg. 1994. № 81. - P886.