Healing effect of heparin in the course of acute cerulein-induced pancreatitis

Leczniczy efekt heparyny w przebiegu ostrego zapalenia trzustki wywołanego ceruleiną

Piotr Ceranowicz[1], Marcin Dembiński[2], Zygmunt Warzecha[1], Jakub Cieszkowski[1], Beata Kuśnierz-Cabala[3], Romana Tomaszewska[4], Artur Dembiński[1]

[1]Department of Physiology, Jagiellonian University Medical College, Kraków
[2]Second Department of Surgery, Jagiellonian University Medical College, Kraków
[3]Department of Clinical Biochemistry, Jagiellonian University Medical College, Kraków
[4]Department of Pathology, Jagiellonian University Medical College, Kraków

Przegląd Gastroenterologiczny 2009; 4 (4): 199–205

Key words: pancreatitis, interleukin-1β, activated partial thromboplastin time.
Słowa kluczowe: zapalenie trzustki, interleukina-1β, czas kaolinowo-kefalinowy.

Address for correspondence: Prof. Artur Dembiński, MD, PhD, Department of Physiology, Jagiellonian University Medical College, 16 Grzegórzecka Street, 31-531 Kraków, phone +48 12 421 10 06, fax +48 12 422 54 78, e-mail: mpdembin@cyf-kr.edu.pl

Abstract

Introduction: Previous experimental studies have shown that pretreatment with heparin inhibits the development of acute pancreatitis, and administration of heparin after development of ischaemia/reperfusion-induced pancreatitis exhibits a therapeutic effect in this disease.
Aim: The aim of this study was to determine the influence of heparin administration on the course of acute pancreatitis evoked by a primary non-vascular mechanism.
Material and methods: The study was performed on Wistar rats. Acute pancreatitis was induced by cerulein. The severity of acute pancreatitis was evaluated between the first and tenth day of inflammation. Heparin was administered subcutaneously twice a day at the dose of 150 U/kg, starting 24 h after cerulein administration.
Results: Treatment with heparin, after the development of cerulein-induced acute pancreatitis, significantly reduced plasma activity of lipase and plasma concentration of pro-inflammatory interleukin-1β. These effects were associated with a partial reversion of the pancreatitis-evoked drop in pancreatic DNA synthesis and the improvement of pancreatic blood flow. The activated partial thromboplastin time was prolonged, whereas plasma level of D-dimer was reduced. Histological features showed faster normalization of pancreatic morphology.
Conclusions: Heparin exhibits a healing effect in the course of oedematous pancreatitis, leading to faster normalization of biochemical markers of acute pancreatitis severity, as well as accelerating the pancreatic regeneration.

Streszczenie

Wprowadzenie: Wyniki wcześniejszych badań eksperymentalnych wykazały, że podanie heparyny przed wywołaniem ostrego zapalenia trzustki hamuje rozwój tego zapalenia oraz że podawanie heparyny w trakcie przebiegu ostrego zapalenia trzustki wywołanego niedotlenieniem z reperfuzją wywołuje działanie lecznicze.
Cel: Celem badań było określenie wpływu podawania heparyny na przebieg ostrego zapalenia trzustki wywołanego czynnikiem pierwotnie pozanaczyniowym.
Materiał i metody: Badania przeprowadzono na szczurach rasy Wistar. Ostre zapalenie trzustki wywołano przy użyciu ceruleiny. Ciężkość ostrego zapalenia trzustki określano między 1. a 10. dniem zapalenia. Heparynę podawano podskórnie 2 razy dziennie w dawce 150 U/kg m.c., zaczynając dzień po podaniu ceruleiny.
Wyniki: Przyjmowanie heparyny w przebiegu ostrego zapalenia trzustki wywołanego ceruleiną znamiennie zmniejszało aktywność lipazy i stężenie prozapalnej interleukiny 1β w osoczu. Efekty te występowały wspólnie z częściowym odwróceniem, wywołanego zapaleniem trzustki, spadku trzustkowej syntezy DNA oraz poprawą trzustkowego przepływu krwi. Czas kaolinowo-kefalinowy przedłużył się, podczas gdy osoczowe stężenie D-dimeru się zmniejszyło. Nastąpiła także wcześniejsza normalizacja morfologii trzustki w ocenie histologicznej.
Wnioski: Heparyna wykazuje działanie lecznicze w ostrym obrzękowym zapaleniu trzustki, prowadząc do wcześniejszej normalizacji biochemicznych wskaźników ciężkości ostrego zapalenia trzustki, oraz przyspiesza regenerację trzustki w przebiegu tej choroby.

Introduction

There is evidence that initiation and progression of acute pancreatitis are associated with disturbance in the pancreatic microcirculation, leading to formation of thrombi in capillaries, activation of leukocytes, release of proteolytic enzymes, and formation of oxygen-derived free radicals and pro-inflammatory cytokines. Coagulative disorders are related to the severity of acute pancreatitis [1, 2]. In this disease, activation of the haemostatic system may range from scattered intravascular thrombosis to severe disseminated intravascular coagulation (DIC) [3]. Inflammation and coagulation are closely linked processes [4] and inflammatory cytokines activate coagulation by increasing expression of tissue factor on monocytes and endothelium, leading to thrombin formation [5].

Heparin, a heteroglycan containing alternating sulphated units of glucuronic acid and glucosamine, exhibits numerous biological activities [6]. Heparin, in complex with antithrombin III, prevents coagulation and, in large doses, may inhibit platelet aggregation [6]. Beside anticoagulative properties related to direct or indirect inhibition of protease involved in the coagulation cascade, heparin also inhibits other proteases present in plasma and tissues, including pancreatic enzymes. Heparin directly or indirectly reduces activity of trypsin [7, 8] and chymotrypsin [9], and inhibits conversion of trypsinogen to trypsin [10, 11].

Previous experimental and some clinical studies have shown that pretreatment or early treatment with heparin during induction of acute pancreatitis exhibits a protective effect on the pancreas, inhibiting the development of acute pancreatitis evoked by cerulein [12], bile [13], taurocholate [14], pancreatic ischaemia followed by reperfusion [15] or endoscopic retrograde cholangiopancreatography (ERCP) [16]. This effect of heparin may be useful in the prevention of acute pancreatitis, but its clinical value is considerably limited. Clinically patients are usually seen several hours or days after the onset of acute pancreatitis and therapy is started after admission to hospital. For this reason, it is more important to answer the question whether treatment with heparin after development of acute pancreatitis can affect the course of acute pancreatitis. Our previous study [15] has shown that administration of heparin after development of ischaemia/reperfusion-induced pancreatitis exhibits a therapeutic effect in this disease. However, this experimental model of acute pancreatitis depends on a primary vascular mechanism related to severe pancreatic ischaemia followed by reperfusion. The effect of heparin administration on the course of acute pancreatitis evoked by a primary non-vascular mechanism is unknown. For this reason, the present study was designed to determine the influence of treatment with unfractionated heparin on the course of acute cerulein-induced pancreatitis.

Material and methods

Studies were performed on male Wistar rats weighing 160-180 g. The experimental protocol is in agreement with the "European Convention for the Protection of Vertebrate Animals used for Experimental and other Scientific Purpose" and was approved by the Local Commission of Ethics for the Care and Use of Laboratory Animals. Animals were housed in cages with wire mesh bottoms, with normal room temperature and a 12-h light-dark cycle.

Acute pancreatitis was induced by cerulein (Sigma-Aldrich, GmbH, Steinheim, Germany) administered intraperitoneally (*i.p.*) 5 times with 1 h intervals at a dose of 50 μg/kg/dose.

Unfractionated heparin (Heparinum, Polfa, Warszawa, Poland) was administered subcutaneously at the dose of 150 U/kg, twice a day, starting 24 h after the last injection of cerulein. The dose of heparin, 150 U/kg, was chosen because this dose caused a two-fold increase in activated partial thromboplastin time (aPTT) in a preliminary study (data not shown).

Pancreatic blood flow was measured using a laser Doppler flowmeter (PeriFlux 4001 Master monitor, Perimed AB, J rf lla, Sweden).

Plasma lipase activity was determined with a Kodak Ectachem DT II System analyzer (Eastman Kodak Company, Rochester, NY, USA) using Lipa DT Slides (Vitros DT Chemistry System, Johnson & Johnson Clinical Diagnostic, Inc., Rochester, NY, USA). Plasma concentration of interleukin-1β (IL-1β) was measured using the BioSource Cytoscreen rat IL-1β kit (BioSource International, Camarillo, California, USA) based on ELISA. Activated partial thromboplastin time (aPTT) was determined in fresh plasma, using Plastelin LS (Organon Teknika Corporation, Dirham, NC, USA). Plasma D-dimer concentration was determined using a latex-enhanced immunoturbidimetric assay (D-dimer test, Roche Diagnostics).

Pancreatic DNA synthesis was measured by incubation of minced pancreatic tissue at 37°C for 45 min in 2 ml of medium containing 8 μCi /ml of [^{3}H] thymidine ([6-^{3}H]-thymidine, 20-30 Ci/mmol, Institute for Research, Production and Application of Radioisotopes, Prague, Czech Republic), as described previously [17]. DNA synthesis was expressed as [^{3}H] thymidine disintegrations per minute per microgram DNA (dpm/μg DNA).

Morphological examination of pancreatic tissue was performed in haematoxylin and eosin stained slides as described previously in detail [17]. The histological grading of oedema, leukocytic inflammatory infiltration, vacuolization of acinar cells, haemorrhages and necrosis was made using a scale raging from 0 (absent) to 3 for maximal alteration. Results of histological examination were expressed as a predominant histological grading in each experimental group of animals.

Statistical analysis, except histological data, was made by analysis of variance followed by Tukey's multiple comparison test. A difference with a *p* value of less than 0.05 was considered significant.

Results

Treatment with heparin after induction of acute pancreatitis reduced the severity of this disease and accelerated pancreatic regeneration. In histological examination, a reduction in pancreatic oedema, inflammatory infiltration, vacuolization of acinar cells, and haemorrhages was observed (Table I). Pancreases of animals treated with heparin after induction of acute pancreatitis recovered during 7 days from induction of acute pancreatitis, whereas pancreases of animals without treatment with heparin reached normal morphology after 10 days (Table I). Also, treatment with heparin reduced biochemical indices of the severity

Table I. Influence of heparin administration on morphological signs of pancreatic damage in the course of cerulein-induced acute pancreatitis

Tabela I. *Wpływ podawania heparyny na morfologiczne objawy uszkodzenia trzustki w przebiegu ostrego zapalenia trzustki wywołanego ceruleiną*

	Edema (0-3)	Inflammatory infiltration (0-3)	Vacuolization (0-3)	Necrosis (0-3)	Haemorrhages (0-3)
Control	0	0	0	0	0
Heparin	0	0	0	0	0
Cerulein 0 h	2-3	2	2	0	0
Cerulein 1 day	1-2	1-2	1-2	0	1-2
Cerulein 2 days	1	1	1-2	0	0-1
Cerulein + heparin 2 days	1	0-1	1	0	0-1
Cerulein 3 days	1	1	1	0	0-1
Cerulein + heparin 3 days	0-1	0-1	0-1	0	0
Cerulein 4 days	1	0-1	1	0	0
Cerulein + heparin 4 days	0-1	0	0-1	0	0
Cerulein 5 days	1	0	1	0	0
Cerulein + heparin 5 days	0-1	0	0	0	0
Cerulein 7 days	0-1	0	0	0	0
Cerulein + heparin 7 days	0	0	0	0	0
Cerulein 10 days	0	0	0	0	0
Cerulein + heparin 10 days	0	0	0	0	0

Numbers represent the predominant histological grading in each group

of acute pancreatitis. Treatment with heparin significantly reduced the pancreatitis-evoked increase in plasma activity of lipase (Fig. 1) and plasma concentration of pro-inflammatory IL-1β (Fig. 2). Pancreatic DNA synthesis (Fig. 3) and pancreatic blood flow (Fig. 4) were increased in animals with acute pancreatitis and treated with heparin. Treatment with heparin after induction of acute pancreatitis prolonged aPTT (Fig. 5) and this effect was associated with a significant reduction in plasma concentration of D-dimer (Fig. 6).

Discussion

Previous studies have shown that pretreatment with heparin inhibits the development of acute pancreatitis in different experimental models of this disease [12-15], as well as protecting the pancreas against acute pancreatitis evoked by ERCP in a clinical study in humans [16]. Our present study confirms and extends these observations. In a previously used model of acute pancreatitis, acute pancreatitis was evoked by severe pancreatic ischaemia followed by reperfusion, where a vascular mechanism with extensive intravascular coagulation was the primary cause of this disease. In our present study we induced acute pancreatitis using an experimental model of this disease with a primary non-vascular aetiology. We have found that administration of heparin demonstrates a healing effect in the course of cerulein-induced acute pancreatitis. This observation suggests that heparin exhibits a therapeutic effect in acute pancreatitis independently of the primary aetiology of this disease. The therapeutic effect of heparin in the course of cerulein-induced pancreatitis was manifested as a reduction in the severity of acute pancreatitis and a faster pancreatic recovery. Morphological features of pancreatic tissue have shown that treatment with heparin reduces pancreatic oedema, necrosis, haemorrhages and leukocyte infiltration. Reduction in pancreatic leukocyte infiltration was in harmony with the reduction in plasma interleukin-1β observed by us. Activation of leukocytes and release of pro-inflammatory cytokines are responsible for local pancreatic damage and development of systemic inflammatory response syndrome (SIRS) and multiple organ failure (MOF) in the course of acute pancreatitis [18]. Pro-inflammatory cytokines, such as IL-1β, IL-6 and tumour necrosis factor-α (TNF-α), are produced within the pancreas and subsequently within distant organs, leading to

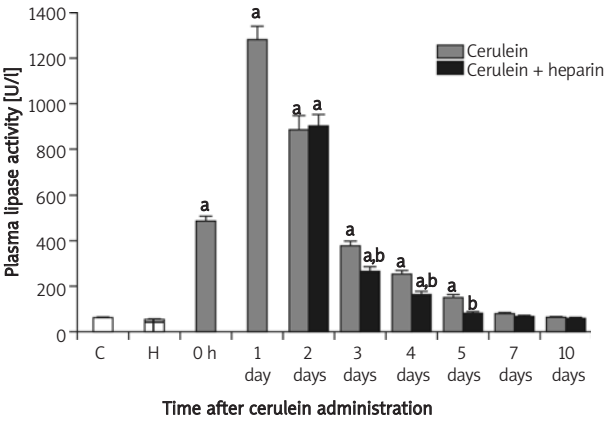

Fig. 1. Influence of heparin (H) administration on plasma activity of lipase in the course of cerulein-induced pancreatitis

Mean ± SEM, N = 8 in each group of rats, $^{a}p < 0.05$ compared to control (C), $^{b}p < 0.05$ compared to cerulein alone at the same time of observation

Ryc. 1. *Wpływ podawania heparyny (H) na aktywność lipazy w osoczu w przebiegu zapalenia trzustki wywołanego ceruleiną*

Wartość średnia ± błąd standardowy, osiem obserwacji w grupie, $^{a}p < 0,05$ w porównaniu z grupą kontrolną (C), $^{b}p < 0,05$ w porównaniu z samą ceruleiną w tym samym czasie obserwacji

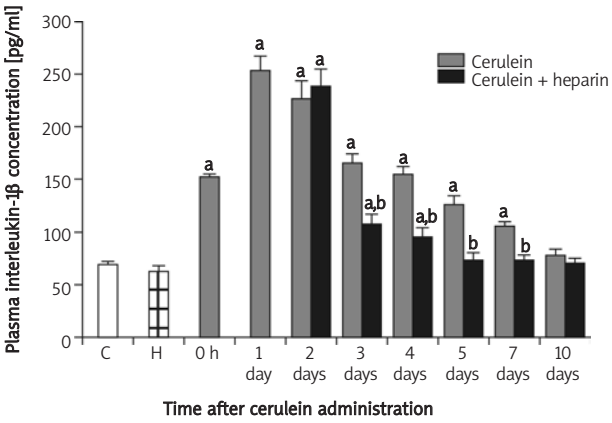

Fig. 2. Influence of heparin (H) administration on plasma concentration of pro-inflammatory interleukin-1β in the course of cerulein-induced pancreatitis

Mean ± SEM, N = 8 in each group of rats, $^{a}p < 0.05$ compared to control (C), $^{b}p < 0.05$ compared to cerulein alone at the same time of observation

Ryc. 2. *Wpływ podawania heparyny (H) na stężenie prozapalnej interleukiny 1β w osoczu w przebiegu zapalenia trzustki wywołanego ceruleiną*

Wartość średnia ± błąd standardowy, osiem obserwacji w grupie, $^{a}p < 0,05$ w porównaniu z grupą kontrolną (C), $^{b}p < 0,05$ w porównaniu z samą ceruleiną w tym samym czasie obserwacji

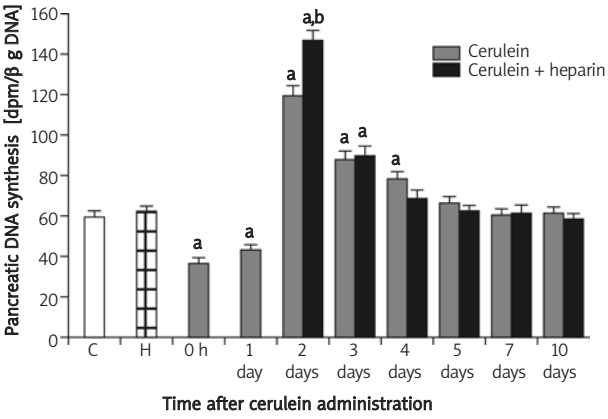

Fig. 3. Influence of heparin (H) administration on pancreatic DNA synthesis in the course of cerulein-induced pancreatitis

Mean ± SEM, N = 8 in each group of rats, [a]p < 0.05 compared to control (C), [b]p < 0.05 compared to cerulein alone at the same time of observation

Ryc. 3. *Wpływ podawania heparyny (H) na syntezę DNA w trzustce w przebiegu zapalenia trzustki wywołanego ceruleiną*

Wartość średnia ± błąd standardowy, osiem obserwacji w grupie, [a]p < 0,05 w porównaniu z grupą kontrolną (C), [b]p < 0,05 w porównaniu z samą ceruleiną w tym samym czasie obserwacji

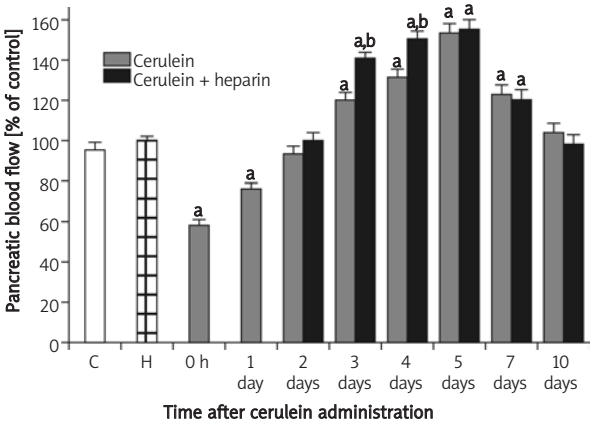

Fig. 4. Influence of heparin (H) administration on pancreatic blood flow in the course of cerulein-induced pancreatitis

Mean ± SEM, N = 8 in each group of rats, [a]p < 0.05 compared to control (C), [b]p < 0.05 compared to cerulein alone at the same time of observation

Ryc. 4. *Wpływ podawania heparyny (H) na trzustkowy przepływ krwi w przebiegu zapalenia trzustki wywołanego ceruleiną*

Wartość średnia ± błąd standardowy, osiem obserwacji w grupie, [a]p < 0,05 w porównaniu z grupą kontrolną (C), [b]p < 0,05 w porównaniu z samą ceruleiną w tym samym czasie obserwacji

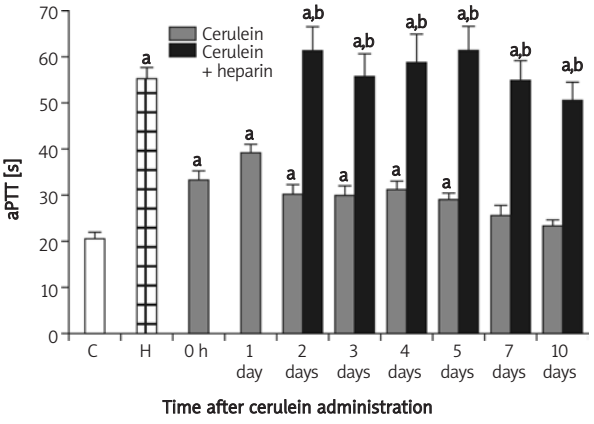

Fig. 5. Influence of heparin (H) administration on activated partial thromboplastin time (aPTT) in the course of cerulein-induced pancreatitis

Mean ± SEM, N = 8 in each group of rats, [a]p < 0.05 compared to control (C), [b]p < 0.05 compared to cerulein alone at the same time of observation

Ryc. 5. *Wpływ podawania heparyny (H) na czas koalinowo-kefalinowy w przebiegu zapalenia trzustki wywołanego ceruleiną*

Wartość średnia ± błąd standardowy, osiem obserwacji w grupie, [a]p < 0,05 w porównaniu z grupą kontrolną (C), [b]p < 0,05 w porównaniu z samą ceruleiną w tym samym czasie obserwacji

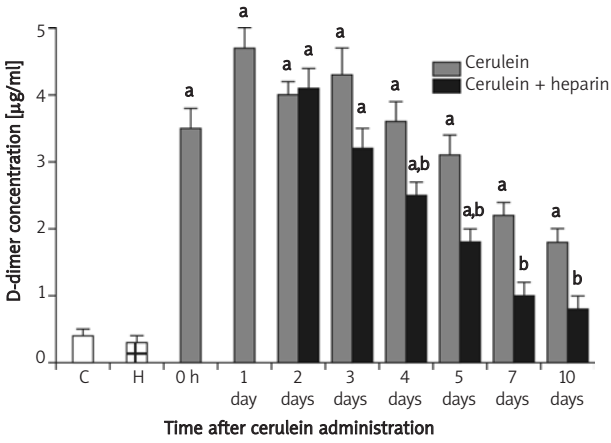

Fig. 6. Influence of heparin (H) administration on plasma D-dimer concentration in the course of cerulein-induced pancreatitis

Mean ± SEM, N = 8 in each group of rats, [a]p < 0.05 compared to control (C), [b]p < 0.05 compared to cerulein alone at the same time of observation

Ryc. 6. *Wpływ podawania heparyny (H) na stężenie D-dimeru w osoczu w przebiegu zapalenia trzustki wywołanego ceruleiną*

Wartość średnia ± błąd standardowy, osiem obserwacji w grupie, [a]p < 0,05 w porównaniu z grupą kontrolną (C), [b]p < 0,05 w porównaniu z samą ceruleiną w tym samym czasie obserwacji

the development of MOF in severe acute pancreatitis [19]. IL-1β plays the most important role in the induction of the systemic acute phase response and in the release of other members of the pro-inflammatory cytokine cascade [20]. Pro-inflammatory cytokine production is well correlated with severity of acute pancreatitis [21].

This anti-inflammatory effect of heparin in the course of cerulein-induced acute pancreatitis seems to be dependent on direct and indirect mechanisms. The concept of direct anti-inflammatory action of heparin is supported by numerous studies. Already in 1984, Laghi Pasini *et al*. [22] showed in an in vitro study that heparin inhibits granulocyte aggregation and degranulation stimulated by chemotactic factor FMLP (N-formyl-methionyl-leucyl-phenylalanine) or by zymosan activated serum, and reduces the FMLP-dependent superoxide anion generation. In agreement with these data is the observation that heparin inhibits neutrophil aggregation stimulated by FMLP or PAF (platelet-activating factor), as well as reducing the release of elastase by these cells [23].

Our present study has shown that the anti-inflammatory effect of heparin in the course of acute cerulein-induced pancreatitis is also related to its influence on the plasma activity of pancreatic digestive enzymes. The increase in plasma activity of lipase and amylase is a well established index of acute pancreatitis severity with high sensitivity and specificity [24]. In our present study, pretreatment with heparin reduced the pancreatitis-evoked increase in serum activity of lipase. This observation is further evidence of the therapeutic effect of heparin in acute pancreatitis. A reduction in plasma activity of pancreatic enzymes may be a result of direct action of heparin on these enzymes or a result of improvement of pancreatic condition, or both these effects. The concept of direct inhibitory action of heparin on pancreatic enzymes is supported by the observation that heparin directly reduces activity of pancreatic enzymes [7-11]. On the other hand, there is a study showing interaction between pancreatic enzymes and intravascular activation of leukocytes. Keck *et al*. [25] showed that presence of active trypsin and elastase in the circulation up-regulates the expression of adhesion molecules on leukocytes and endothelial cells. This effect leads to an increase in leukocyte-endothelial interaction, promoting pancreatic microcirculatory failure. For this reason, the heparin-evoked reduction in plasma activity of pancreatic enzymes can be a result and/or cause of its therapeutic effect in acute pancreatitis.

Another important mechanism of the therapeutic effect of heparin in acute cerulein-induced pancreatitis seems to be dependent on its influence on pancreatic blood flow. Pancreatic ischaemia plays an important role in the development of acute pancreatitis [26] and the severity of acute pancreatitis is closely correlated with the disturbance of pancreatic circulation. Our present study has shown that treatment with heparin improves pancreatic blood flow and reduces the severity of acute pancreatitis. This effect seems be a result and cause of the therapeutic effect of heparin because the improvement of pancreatic blood flow, during induction of acute pancreatitis, reduces the severity of pancreatic damage [27], but simultaneously a reduction in pancreatic damage improves pancreatic blood flow.

It is most likely that the circulatory effect of heparin is, at least in part, related to its well-known anticoagulant activity. In our present study, induction of acute pancreatitis by cerulein led to a reduction in pancreatic blood flow and this effect was associated with an almost two-fold increase in aPTT and a twelve-fold increase in D-dimer concentration. These data indicate that development of acute pancreatitis, also with primary non-vascular aetiology, is associated with formation of thrombi within the pancreatic and systemic circulation. Changes in aPTT are a result of consumption of factors involved in coagulation, whereas the increase in D-dimer concentration indicates subsequent activation of coagulation. Our present study has shown that treatment with heparin accelerated reduction in plasma D-dimer concentration in animals with acute pancreatitis. This observation indicates that treatment with heparin inhibits coagulation and for this reason reduces consumption of coagulation factors and creation of products of fibrinolysis.

Pancreatic DNA synthesis is an index of cell proliferation in the pancreas and the reduction in pancreatic DNA synthesis is well correlated with pancreatic damage in acute pancreatitis [27-28]. Our present study has shown that administration of heparin increases pancreatic DNA synthesis in the early stage of acute pancreatitis. This observation is additional evidence of the beneficial effect of heparin in acute pancreatitis and may explain our observation that heparin accelerates pancreatic regeneration in this disease.

Conclusions

Heparin, a safe and well-known medicine, exhibits a strong therapeutic effect in the course of acute pancreatitis independently of the primary cause of this disease. This observation suggests that heparin may be useful in routine clinical management of acute pancreatitis.

References

1. Lasson A, Ohlsson K. Consumptive coagulopathy, fibrinolysis and protease-antiprotease interactions during acute human pancreatitis. Thromb Res 1986; 41: 167-83.
2. Salomone T, Tosi P, Palareti G, et al. Coagulative disorders in human acute pancreatitis: role for the D-dimer. Pancreas 2003; 26: 111-6.
3. Agarwal N, Pitchumoni CS. Acute pancreatitis: a multisystem disease. Gastroenterologist 1993; 1: 115-28.
4. Esmon CT, Taylor FB Jr, Snow TR. Inflammation and coagulation: linked processes potentially regulated through a common pathway mediated by protein C. Thromb Haemost 1991; 66: 160-5.
5. Esmon CT. Possible involvement of cytokines in diffuse intravascular coagulation and thrombosis. Baillieres Best Pract Res Clin Haematol 1999; 12: 343-59.
6. Bowman WC, Rand MJ. The blood: drugs affecting coagulation, fibrinolysis, haematopoiesis and functioning of blood cells. In: Textbook of pharmacology. Bowman WC, Rand MJ (eds). Oxford, Blackwell Scientific Publication, 1980; 21.15-21.16.
7. Finotti P, Manente S. Heparin-induced structural and functional alteration s of bovine trypsin. Biochim Biophys Acta 1994; 1207: 80-7.
8. Nobar SM, Guy-Crotte O, Rabaud M, Bieth JG. Inhibition of human pancreatic proteinases by human plasma alpha2-antiplasmin and antithrombin. Biol Chem 2004; 385: 423-7.
9. Struss D, Storck J, Zimmermann RE. The inhibition of thrombin and chymotrypsin by heparin-cofactor II. Trombin Res 1992; 68: 45-56.
10. Wolosowicz N, Prokopowicz J, Gabryelewicz A. The inhibitory effect of heparin on trypsinogen activation with enterokinase. Acta Hepatogastroenterol (Stuttg) 1977; 24: 368-71.
11. Gabryelewicz A, Kosidlo S, Prokopowicz J, Podkowicz K. Does heparin modify protease-antiprotease balance in acute experimental pancreatitis in rats. Hepatogastroenterology 1986; 33: 79-82.
12. Dobosz M, Mionskowska L, Hac S, et al. Heparin improves organ microcirculatory disturbances in caerulein-induced acute pancreatitis in rats. World J Gastroenterol 2004; 10: 2553-6.
13. Gabryelewicz A, Niewiarowski S, Prokopowicz J, Clebowski J. Heparin and protease inhibitors in the prevention of experimental acute pancreatic necrosis in dogs. Digestion 1969; 2: 7-16.
14. Qiu F, Lü XS, Huang YK. Effect of low molecular weight heparin on pancreatic micro-circulation in severe acute pancreatitis in a rodent model. Chin Med J 2007; 120: 2260-3.
15. Ceranowicz P, Dembinski A, Warzecha Z, et al. Protective and therapeutic effect of heparin in acute pancreatitis. J Physiol Pharmacol 2008; 59 (Suppl 4): 103-25.
16. Rabenstein T, Roggenbuck S, Framke B, et al. Complications of endoscopic sphincterotomy: can heparin prevent acute pancreatitis after ERCP? Gastrointest Endosc 2002; 55: 476-83.
17. Warzecha Z, Dembiński A, Ceranowicz P, et al. Influence of ischemic preconditioning on blood coagulation, fibrinolytic activity and pancreatic repair in the course of caerulein-induced acute pancreatitis in rats. J Physiol Pharmacol 2007; 58: 303-19.
18. Frossard JL, Past CM. Experimental acute pancreatitis: new insight into the pathophysiology. Front Biosci 2002; 7: d275-87.
19. Norman JG, Fink GW, Denham W, et al. Tissue-specific cytokine production during experimental acute pancreatitis. A probable mechanism for distant organ dysfunction. Dig Dis Sci 1997; 42: 1783-8.
20. Dinarello CA. Interleukin-1 and interleukin-1 antagonism. Blood 1991; 77: 1627-52.
21. Norman J, Franz M, Messina J, et al. Interleukin-1 receptor antagonist decreases severity of experimental acute pancreatitis. Surgery 1995; 117: 648-55.
22. Laghi Pasini F, Pasqui Al, Ceccatelli L, et al. Heparin inhibition of polymorphonuclear leukocyte activation in vitro. A possible pharmacological approach to granulocyte-mediated vascular damage. Thromb Res 1984; 35: 527-37.
23. Brown RA, Lever R, Jones NA, Page CP. Effects of heparin and related molecules upon neutrophil aggregation and elastase release in vitro. Br J Pharmacol 2003; 139: 845-53.
24. Dervenis C, Johnson CD, Bassi C, et al. Diagnosis, objective assessment of severity, and management of acute pancreatitis. Santorini consensus conference. Int J Pancreatol 1999; 25: 195-210.
25. Keck T, Friebe V, Warshaw AL, et al. Pancreatic proteases in serum induce leukocyte-endothelial adhesion and pancreatic microcirculatory failure. Pancreatology 2005; 5: 241-50.
26. Vollmar B, Menger MD. Microcirculatory dysfunction in acute pancreatitis. A new concept of pathogenesis involving vasomotion-associated arteriolar constriction and dilation. Pancreatology 2003; 3: 181-90.
27. Warzecha Z, Dembiński A, Ceranowicz P, et al. Protective effect of calcitonin gene-related peptide against caerulein-induced pancreatitis in rats. J Physiol Pharmacol 1997; 48: 775-87.
28. Dembiński A, Warzecha Z, Ceranowicz P, et al. Dual, time-dependent deleterious and protective effect of anandamide on the course of cerulein-induced acute pancreatitis. Role of sensory nerves. Eur J Pharmacol 2008; 591: 284-92.