Available online at www.sciencedirect.com
SCIENCE DIRECT®
Biochemical Pharmacology 69 (2005) 1325–1331
Biochemical Pharmacology
www.elsevier.com/locate/biochempharm

Severity of pancreatitis-associated gut barrier dysfunction is reduced following treatment with the PAF inhibitor lexipafant

Per Leveau, Xiangdong Wang *, Zhengwu Sun, Anna Börjesson, Ellen Andersson, Roland Andersson

Department of Surgery, Lund University Hospital, SE-22185 Lund, Sweden

Received 16 November 2004; accepted 31 January 2005

Abstract

The aim of the present study was to investigate the potential effect of treatment with a platelet-activating factor (PAF) antagonist, lexipafant (BB-882), on gut endothelial and epithelial barrier dysfunction and leukocyte recruitment in rats with acute pancreatitis. Severe acute pancreatitis was induced by the intraductal administration of 5% sodium taurodeoxycholate and pancreatitis-associated gut barrier dysfunction was characterized by increased exudation of radiolabelled albumin into the interstitium and alterations in bidirectional (over both the endothelial and epithelial barrier components) permeability of the intestine at the early stage of bile salt-induced acute pancreatitis. Levels of interleukin 1β and 6, ileal and colonic myeloperoxidase (MPO) content, clearance of radiolabelled albumin from blood to the gut lumen or gut lumen to blood, and leakage of radiolabelled albumin to the ileum or colon were measured 3 and 12 h after induction of acute pancreatitis. Treatment with lexipafant 30 min and 6 h after pancreatitis reduced severity of pancreatitis-associated intestinal dysfunction, associated with a diminish in systemic concentrations of IL-1 and local leukocyte recruitment. The findings imply that PAF plays a critical role in the development of pancreatitis-associated gut barrier dysfunction and that PAF antagonist in some forms may represent potential candidates for future therapeutic intervention.

Keywords: Pancreatitis; Intestine; Permeability; Cytokines; Platelet-activating factor antagonist

1. Introduction

Pancreatitis-associated complications originating in the gastrointestinal tract [7] have been considered to contribute to mortality. Intestinal reactions during and after acute pancreatitis include inflammatory alterations, disturbed motility, stenosis, ulcerations, bleeding, ischemia, necrosis, and perforation [3,6,15]. Furthermore, pancreatitis-associated failure of the gastrointestinal tract, characterized by translocation of enteric bacteria to extraintestinal sites and the systemic circulation (gut origin sepsis) [13,17], might explain the pathogenesis of pancreatic and peripancreatic infections, considered responsible for up to 80% of deaths in patients with acute pancreatitis [2,12]. Potential mechanisms by which enteric bacteria pass through the intestinal barrier have been suggested to be associated with impaired intestinal motility, disturbed enteric bacterial ecology, increased adhesion of pathogenic bacteria onto the surface of mucosal epithelial cells, mucosal endothelial barrier dysfunction, overactivation of local macrophages together with decreased systemic immune defense function, mucosal ischemia and poor oxygen extraction [11,16–19]. Inflammatory mediators produced and released during acute pancreatitis have been suggested as potential early markers of disease severity and as critical links in the pathogenesis and pathophysiology of the disease. These mediators include interleukins (IL), polymorphonuclear enzymes, oxygen free radicals, platelet-activating factor (PAF), and adhesion molecules [8]. The aim of the present study was to investigate pancreatitis-associated gut barrier dysfunction, leukocyte recruitment and systemic levels of interleukins in experimental acute pancreatitis and the potential effect of treatment with a platelet-activating factor antagonist.

* Corresponding author. Tel.: +46 46 337 883; fax: +46 46 336 624.
E-mail address: xiangdong.wang@telia.com (X. Wang).

0006-2952/$ – see front matter © 2005 Elsevier Inc. All rights reserved.
doi:10.1016/j.bcp.2005.01.023

2. Methods and materials

2.1. Animals

Adult male Sprague–Dawley rats, weighing about 250 g, were fed standard rat chow (R3, Astra-Ewos, Södertälje, Sweden) and water ad libitum. The rats were allowed to acclimatize to our laboratory conditions for 4–6 days and were subjected to a regime of 12 h day/night cycle living in mesh stainless-steel cages (3 rats/cage) at constant temperature (22 °C). The protocol was approved by the Animal Ethics Committee at Lund University. All animals were handled in accordance with the guidelines set forth by the Swedish Physiological Society.

2.2. Induction of acute pancreatitis

The rats were operated on under aseptic conditions using ether anesthesia. Acute hemorrhagic pancreatitis was induced by the intraductal administration of 0.2 ml 0.025 M glycylglycin–NaOH buffer, pH 8.0, containing 5% sodium taurodeoxycholate, by the use of an infusion pump at a speed of 0.04 ml/min, following clamping of the proximal end of the common bile duct, and cannulation of the biliary-pancreatic duct by a thin polyethylene catheter (0.66 mm o.d., Portex Ltd., Hythe, Kent, UK), after sterilizing the solution of sodium taurodeoxycholate at 100 °C for 20 min. Sham operation (controls) included laparotomy and isolation of the common bile duct similar to what was performed in the experimental group, though without bile injection.

The animals were randomly allocated into four groups: (1) sham operation with saline; (2) sham operation with lexipafant, a (S)-4methyl-2-{methyl-[4-(2-methyl-imidazo-{4,5-c}pyridin-1-ylmethyl)-benzenesulphonyl]-amino}-pentanoic acid ethyl ester (BB-882, British Biotech Ltd., Oxford, UK); (3) acute pancreatitis with saline; and (4) acute pancreatitis with lexipafant. Measurements were performed 3 and 12 h after induction of acute pancreatitis or sham operation ($n = 12$ rats/group and $n = 6$ rats/time point). Sterile saline or lexipafant (5 mg/kg) in a volume of 0.2 ml was injected intraperitoneally 30 min and 6 h after sham operation or induction of pancreatitis.

2.3. Measurements of capillary endothelial permeability

Red blood cells (RBC) were labeled with ^{51}Cr (New England Nuclear, Boston, MA, USA) during 20 min incubation at room temperature and then washed twice with physiological saline. The radioactivity was about 1.5×10^6 cpm/ml. Endothelial permeability was assessed by the passage of ^{125}I-labeled human serum albumin (HSA, Institutt for engergiteknikk, Kjeller, Norway) from blood to the intestine. One milliliter ^{125}I-HSA (10^6 cpm radioactivity) was injected into the femoral vein. After 1-h equilibration, 1 ml blood was drawn from the femoral vein, followed by injection of ^{51}Cr-RBC (10^5 cpm/ml). The animals were sacrificed by an over-dose of ether 2 min after the RBC injection. The intestine was harvested and cleared of external blood by blotting dry. The radioactivity of ^{125}I and ^{51}Cr in blood and tissue samples was measured in a gamma-counter (1272 Clinigamma, LKB, Wallac OY, Finland) after that the samples were weighed for determining wet tissue weight.

Capillary endothelial permeability was assessed by leakage of the radioisotope from blood into the tissues and expressed as isotopic flux, defined as the proportion of ^{125}I-radioactivity per gram tissue sample compared with per gram blood as described previously [12]. To assay possible redistribution of tissue blood, tissue blood content (TBC) was calculated by the proportion of counts ^{51}Cr reference per gram tissue sample and counts ^{51}Cr reference per gram blood sample. In order to correct for potential differences in the vascular surface area available for exchange of albumin, the albumin leakage index (ALI) was calculated by dividing the extravascular protein accumulation in each tissue by assuming that all ^{51}Cr-labeled RBC remained intravascularly, using the formula: albumin leakage index = (extravascular tissue ^{125}I counts/^{125}I counts per gram blood)/tissue blood content. Tissue blood content was calculated as ^{51}Cr-RBC counts per gram tissue/^{51}Cr-RBC counts per ml blood. Extravascular tissue ^{125}I counts was obtained by the subtraction of counts ^{125}I reference per gram blood sample multiplied by tissue blood content from counts ^{125}I reference per gram tissue.

2.4. Assays of IL-1β and IL-6

Serum levels of IL-1β and IL-6 were determined by an enzyme linked-immuno-sorbent assay. Antibodies specific for rat IL-1β and IL-6 were coated onto the wells of the microtiter strips provided (BioSource International, Camarillo, CA, USA). After that the samples, including standards of known rat IL-1β and IL-6 content (BioSource), were pipetted into the wells, incubated, and washed, the intensity of the colored product, using the streptavidin–peroxidase system, was measured by a spectrophotometer with 450 nm lengthwave, directly proportional to the concentration of IL-1β and IL-6.

2.5. Leukocyte recruitment

Following harvest of the intestinal samples, they were immediately frozen in liquid nitrogen, and stored at −70 °C until measurements. The samples (100–200 mg) were weighed, put in ice-cold potassium phosphate buffer (20 mM, pH 7.4), and homogenized for 30 s. The suspension was centrifuged at 10,000 rpm for 15 min at 4 °C and the supernatant discharged in order to avoid influence caused by hemoglobin. The precipitates were rehomogenized with 50 mM PBS with 0.5% hexadecyltrimetylammoniumbromide and 10 mM EDTA, followed by

sonicating, freezing, thawing and homogenizing twice in order to permeate cellular membranes. The reaction was terminated by 0.2 M acetate sodium (pH 3.0) after 3-min-incubation at 37 °C with the reaction solution containing 0.5% hexadecyltrimetylammoniumbromide, 1.6 mM 3,3′,5,5′-tetrametylbenzidine and 0.3 mM H202 in 80 mM PBS (pH 5.4) [13]. Tissue leukocyte recruitment was reflected by units of myeloperoxidase (MPO) activity [4], defined as change in one-min absorbance at 655 nm.

2.6. Intestinal barrier permeability

Bidirectional permeability of an isolated ileal loop was measured as previously described [14] in another 48 animals subjected to sham operation or pancreatitis treated with saline or lexipafant. All animals were anesthetized by the intraperitoneal injection of pentobarbital (45 mg/kg). Cannulae were inserted into the left femoral artery for measuring the arterial pressure and for the sampling of blood (0.2 ml). The femoral vein was catheterized for administering drugs and fluids. The body temperature was maintained at 37 °C by use of a heating pad and lamp. A loop of distal ileum (10–12 cm proximal from the cecum), approximately 12–15 cm in length, was isolated and cannulated at both ends with soft polyethylene tubes. The intestinal continuity was restored by anastomosing the remaining parts of the intestine. The ileal loop was gently flushed with physiological saline and returned into the abdominal cavity. The tubes extending from the loop were brought out through two openings in the right and left lower abdominal wall and fixed there. The abdominal wall was closed in order to minimize evaporation of fluid and to avoid hypothermia. The proximal catheter was connected to an injection pump and the loop was perfused with warm lactated Ringer's solution at the rate of 0.5 ml/min. The perfusate included ^{131}I-labeled human serum albumin (^{131}I-HSA) at about 2.5 × 105 counts per milliliter (cpm). One ml of ^{125}I-labeled HSA, containing 10^6 cpm, was injected through the femoral vein catheter. Blood samples (0.2 ml) and perfusate (1 ml) from the effluent ileal cannula were harvested at 30 min intervals for 90 min. An equivalent volume of warmed lactated Ringer's solution was injected to maintain the blood volume. The radioactivity in plasma and the perfusate was measured in a gamma scintillation counter (Model 5320; Packard Instruments Co., Legona Hills, CA, USA). At the end of the study period, the animals were killed by an overdose of pentobarbital intravenously and the ileal loop was removed and weighed. Isotopic clearance of ^{125}I-HSA or ^{131}I-HSA from either blood to the intestinal lumen or from the intestinal lumen to blood at the various time points measured were calculated using the formula: Clearance_{BL} (ml/min/l00 g tissue; from blood to lumen) = [perfusate (cpm/ml) × perfusion rate (ml/min) × 100]/[plasma (cpm/ml) × sample weight (g)]. Clearance_{LB} (ml/min/l00 g tissue; from intestinal lumen to blood) = [plasma (cpm/ml) × perfusion rate (ml/min) × 100]/[perfusate (cpm/ml) × sample weight (g)]. Mucosal and intestinal edema was assessed by the wet and dry tissue weight ratios. Permeability of the capillary endothelial barrier was evaluated by the leakage of ^{125}I-HSA into the interstitial space, expressed by the HSA leakage index and the permeability of the intestinal barrier from the circulation to the intestinal lumen, expressed by the Clearance_{BL}. Permeability of the mucosal epithelial barrier was assayed by isotopic clearance from the intestinal lumen to the systemic circulation, expressed by the Clearance_{LB}.

2.7. Statistics

Unpaired Student's *t*-test or non-parametric test (Mann–Whitney rank sum test) was used after ANOVA. A probability of <0.05 was considered as significant. Values are expressed as mean ± S.E.M.

3. Results

An increase in HSA leakage index from the systemic circulation to the interstitial tissues was noted in the ileum

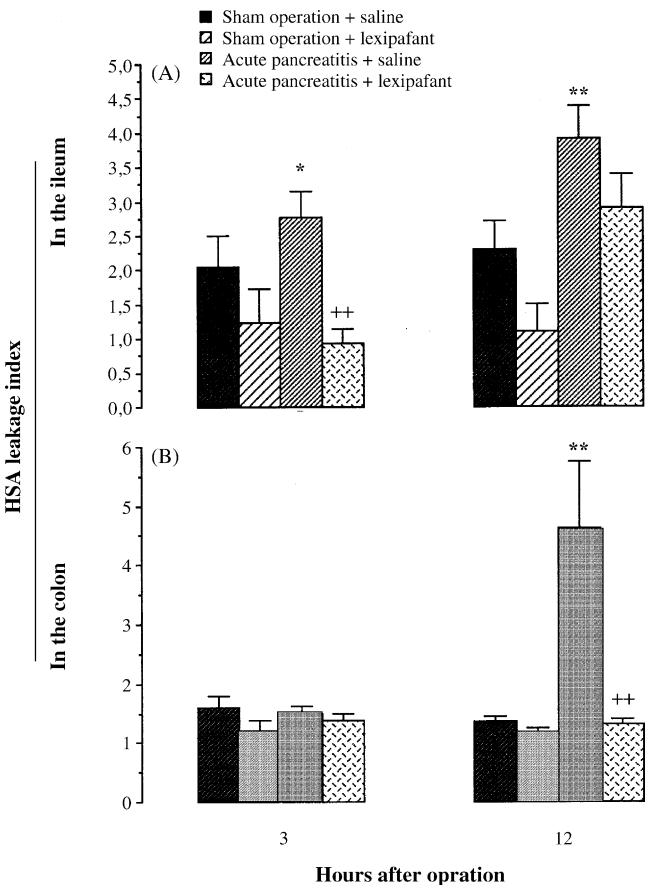

Fig. 1. Human serum albumin (HSA) leakage index in the ileum (A) and colon (B) 3 and 12 h after sham operation or induction of acute pancreatitis and treatment with either saline or lexipafant: (*) and (**) stand for $p < 0.05$ and 0.01, respectively, as compared to sham operation and saline; (++) stands for $p < 0.01$ as compared to acute pancreatitis and saline.

after both 3 and 12 h ($p < 0.05$, respectively, Fig. 1A) and in the colon at 12 h ($p < 0.01$, Fig. 1B) in animals subjected to pancreatitis and treatment with saline, as compared to animals treated with saline or lexipafant and challenged with sham operation or pancreatitis animals treated with lexipafant (Fig. 1). There was no statistical difference of the leakage index in the colon between lexipafant-treated animals challenged with sham operation or acute pancreatitis. Clearance of plasma albumin from the gut lumen to blood or from blood to the gut lumen significantly increased 3 and 12 h after induction of pancreatitis in animals treated with saline, as compared to those treated with saline or lexipafant and challenged with sham operation ($p < 0.01$, Figs. 2 and 3, respectively). Treatment with lexipafant significantly reduced pancreatitis-induced increased clearance from gut lumen to blood (Fig. 2A) or from blood to lumen (Fig. 2B) at 3 h, but the level still was significantly higher than sham-operated animals with saline or lexipafant ($p < 0.05$, respectively, as pancreatitis with saline or sham operation with saline or lexipafant).

At 12 h, clearance of radiolabelled albumin from gut lumen to blood in animals with pancreatitis and lexipafant treatment was significantly lower than in those with pancreatitis and saline treatment ($p < 0.01$), but still higher than sham-operated animals ($p < 0.05$, Fig. 3A). Treatment with lexipafant significantly reduced pancreatitis-increased clearance of albumin from blood to gut lumen till levels of sham-operated animals with saline or lexipafant (Fig. 3B). The levels of clearance_{LB} at 12 h (0.0148–0.0174 at 90 min) was higher as compared to those noted at 3 h (0.0108–0.0131 at 90 min) in animals with pancreatitis and saline, while the levels at 12 h (0.0082–0.0115 at 90 min) in animals subjected to pancreatitis and treatment with lexipafant was similar to those that could be seen at 3 h (0.0084–0.0100 at 90 min).

Serum levels of IL-1β in animals with pancreatitis, treated either with saline or lexipafant, were significantly higher than seen in sham operation at both 3 and 12 h (Fig. 4A). Serum levels of IL-1β in animals with pancreatitis and lexipafant were significantly lower than in those with pancreatitis and saline ($p < 0.05$ and 0.01 at 3 and 12 h, respectively). Similar changes of serum levels of IL-6 were noted in acute pancreatitis treated with saline or lexipafant, levels in all animals varied from 30 to 75 pg/ml (Fig. 4B). Ileal MPO content significantly increased ($p < 0.05$ and 0.01, respectively) 3 and 12 h after induction of pancreatitis and saline-treatment, as compared to animals with sham operation or pancreatitis with lexipafant (Fig. 5A). The ileal MPO content in pancreatitis animals with lexipafant was significantly higher ($p < 0.05$) than in sham-operated animals at 12 h. A significant increase in colonic MPO content was noted 3 ($p < 0.05$) and 12 h ($p < 0.01$) after induction of pancreatitis in saline treated animals as compared with animals with sham operation or pancreatitis animals with lexipafant (Fig. 5B).

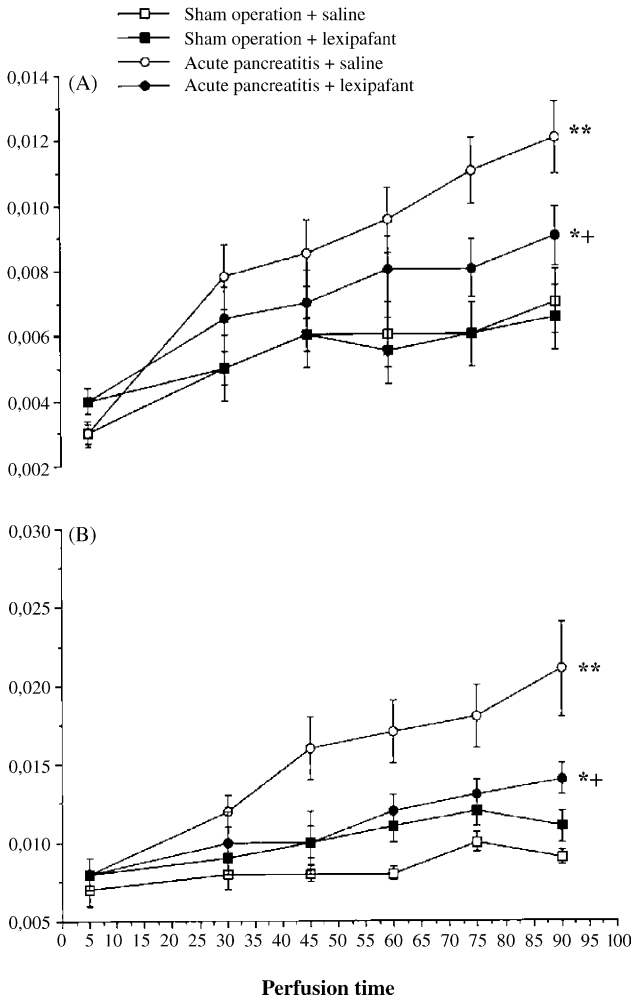

Fig. 2. Clearance of radiolabeled albumin from the gut lumen to blood (A) or from blood to the gut lumen (B) 3 h after sham operation or induction of acute pancreatitis and treatment with saline or lexipafant. Total perfusion time 90 min: (*) and (**) stand for $p < 0.05$ and 0.01, respectively, as compared to sham operation and saline; (+) stands for $p < 0.05$ as compared to acute pancreatitis and saline.

No difference was seen between sham operation and pancreatitis animals treated with lexipafant.

4. Discussion

Platelet-activating factor is a biologically active phospholipid stored in its precursor form within cell membranes, representing an intercellular signal responsible for cell communications, and an inflammatory mediator in the pathogenesis of inflammation. The involvement of PAF in the pathophysiology of pancreatitis-associated distant organ dysfunction is implied by the increase in pulmonary tissue levels of PAF as noted 12 h after induction of experimental pancreatitis, accompanied by progression of the pulmonary injury [21]. Pancreatitis-induced release of phospholipase A_2 might be responsible for local pulmonary PAF accumulation [22], resulting in concomitant development of acute lung injury. Clinical studies have

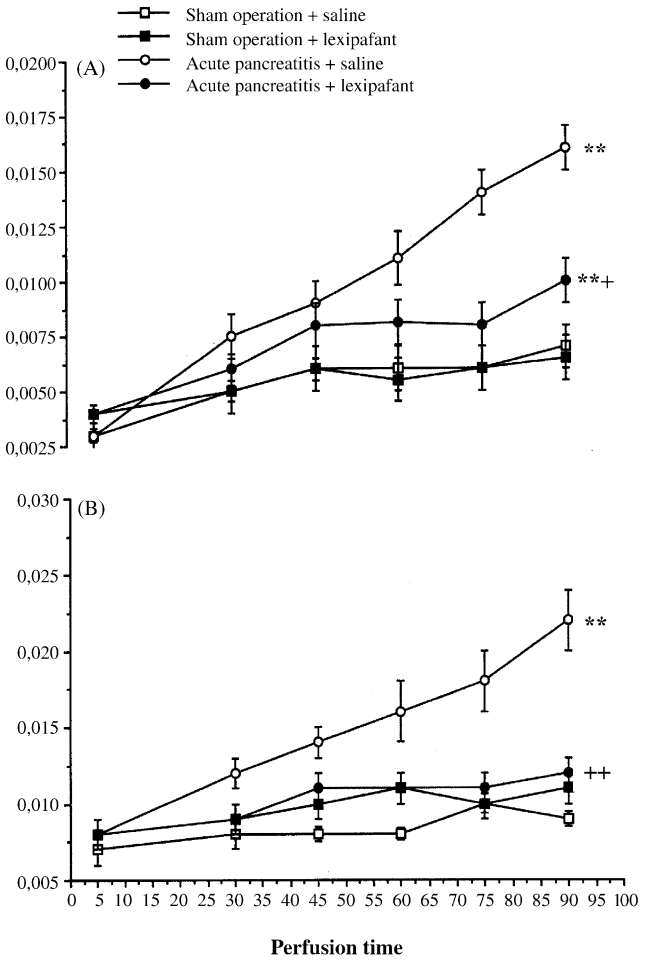

Fig. 3. Clearance of radiolabeled albumin from the gut lumen to blood (A) or from blood to the gut lumen (B) 12 h after sham operation or induction of acute pancreatitis and treatment with saline or lexipafant: (*) and (**) stand for $p < 0.05$ and 0.01, respectively, as compared to sham operation and saline; (++) stands for $p < 0.01$ as compared to acute pancreatitis and saline.

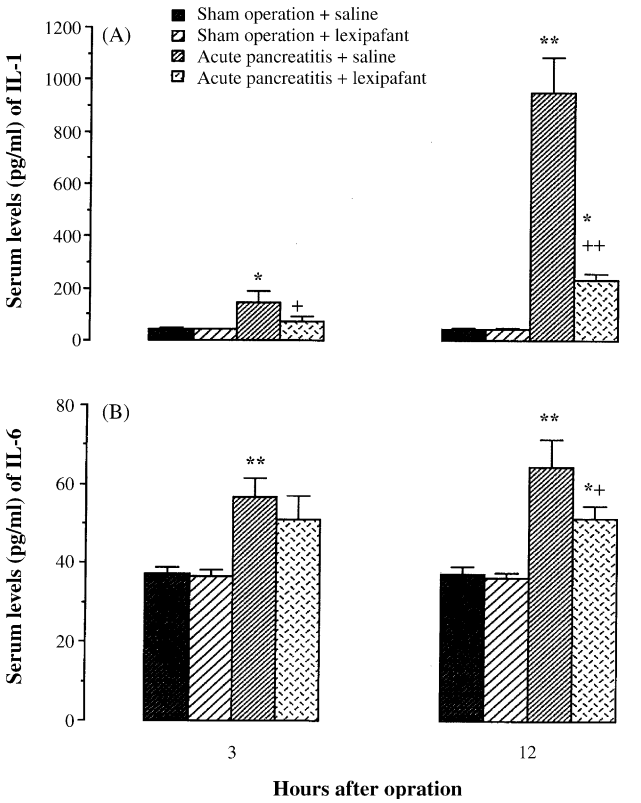

Fig. 4. Serum levels of interleukin-1β (IL-1) (A) and interleukin-6 (IL-6) (B) 3 and 12 h after sham operation or induction of acute pancreatitis and treatment with saline or lexipafant: (*) and (**) stand for $p < 0.05$ and 0.01, respectively, as compared to sham operation and saline; (+) and (++) stand for $p < 0.05$ and 0.01, respectively, as compared to acute pancreatitis and saline.

reported that lexipafant treatment reduced serum levels of interleukin-8, interleukin-6, and polymorphonuclear elastase-alphal-antitrypsin, and decreased organ failure and the incidence of organ failure in severe acute pancreatitis [10,20]. The present experimental study further supports the beneficial effects of treatment with the PAF antagonist lexipafant (BB-882), as severity of pancreatitis-associated intestinal barrier dysfunction ameliorated.

Pancreatitis-associated gut barrier dysfunction has been characterized by the passage of enteric bacteria through the mucosal barrier to extraintestinal sites (bacterial translocation), impaired intestinal motility, and increased intestinal permeability [11,13,17,20]. Alterations in intestinal permeability represent a characteristic of mucosal barrier dysfunction in a variety of clinical conditions, e.g. ischemia and reperfusion, inflammation, sepsis, and others, associated with the development of the multiple organ dysfunction syndrome. Intestinal permeability in experimental acute pancreatitis has been measured by the in vivo leakage of radio-isotopic-labeled albumin and the in vitro passage of small molecular markers (sodium fluorescein and EDTA) and a macromolucular marker (ovalbumin) through the distal ileum and colon from the mucosal to the serosal site in Ussing chambers [20]. An increase in the transport of small markers was followed by increased passage of the macromolecular marker. The early occurrence of increased intestinal permeability has also been reported in humans with severe acute pancreatitis [9]. In the present study, pancreatitis-associated intestinal barrier dysfunction was characterized by an increase in both mucosal endothelial barrier permeability, measured by the exudation of plasma albumin as referred with tissue blood content and the passage of plasma albumin into the intestinal lumen, and mucosal epithelial barrier permeability as reflected by the transport of labeled albumin from the gut lumen to the systemic circulation.

Our data demonstrates that (1) both endothelial and epithelial permeability increased after induction of pancreatitis; (2) an increase in epithelial permeability seemed to depend on time after induction of pancreatitis; and (3) treatment with lexipafant reduced severity of pancreatitis-associated endothelial and epithelial barrier dysfunction. It seems that lexipafant improved pancreatitis-associated gut injury by normalizing mucosal endothelial barrier function at 12 h, while the reversed gut barrier permeability by lexipafant was still higher in animals with

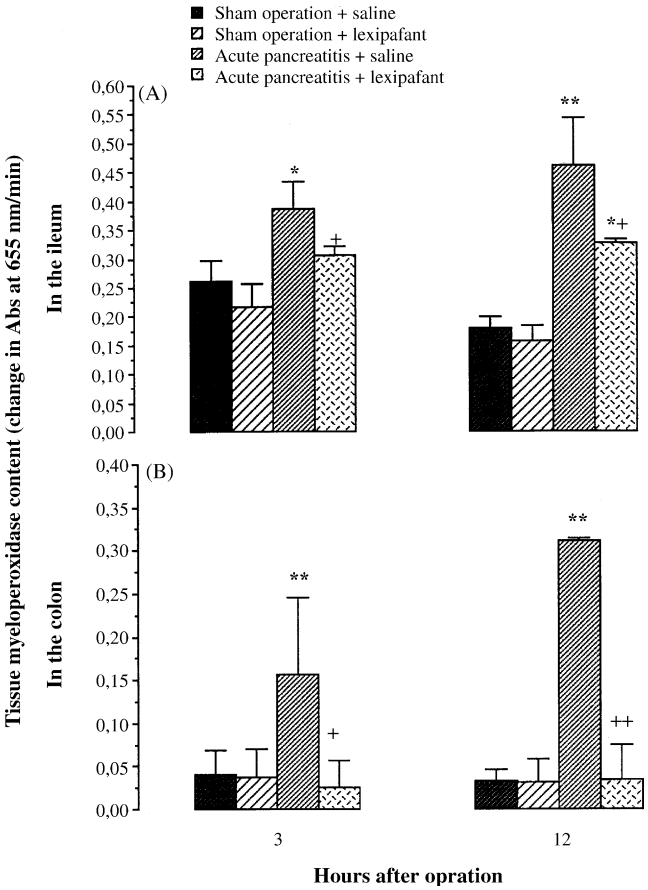

Fig. 5. Tissue myeloperoxidase content in the ileum (A) and colon (B) 3 and 12 h after sham operation or induction of acute pancreatitis and treatment with saline or lexipafant: (*) and (**) stand for $p < 0.05$ and 0.01, respectively, as compared to sham operation and saline; (+) and (++) stand for $p < 0.05$ and 0.01, respectively, as compared to acute pancreatitis and saline.

pancreatitis than whose with sham operation. It indicates that other factors may be involved in the development of pancreatitis-associated gut barrier dusfunction. Comparing to our previous study on the effect of pretreatment with lexipafant on prevention of pancreatitis-associated gut barrier dysfunction [1], it seems that pretreatment with lexipafant was more efficient in reducing pancreatitis-associated gut barrier dysfunction than the presently early inserted treatment. This implies that PAF may not only be a primary mediator to trigger the initiation of pancreatitis-associated mucosal injury, continuously participates in the development and further aggravation of the disease. Major contributions of PAF during inflammatory reaction include the role as an inflammatory stimulator activating the leukocyte system which then can produce and release a variety of secondary inflammatory mediators, and to provoke the exposure of adhesion molecules on the surface of endothelial cells and leukocytes which can result in leukocyte rolling and adhesion on the endothelium and migration between the endothelial cells to the interstitium [5,24]. It is possible that lexipafant may neutralize PAF receptors on the surface of endothelial cells, and reduce intercellular signaling, down-regulate the activation of leukocytes and macrophages and the interaction between leukocytes and endothelial cells, and maintain integrity of the endothelial barrier.

Clinical studies have reported that lexipafant treatment reduced serum levels of IL-8 in patients with severe acute pancreatitis, associated an improvement of the prognosis from pancreatitis-associated multiple organ failure, together with a decline in IL-6, E-selectin, and polymorphonuclear elastase-alphal-antitrypsin [10]. Comparing the levels of cytokines in acute pancreatitis, levels of the IL-1-receptor-antagonist (anticytokine system) were 15 times higher than the levels of IL-6 and 150 times higher than IL-8 at the early stage of severe acute pancreatitis [14], indicating that alterations in the IL-1 and anti-IL-1 patterns may represent a critical link in the pathophysiology of pancreatitis. IL-1 can compromise endothelial barrier integrity probably by over-activating leukocytes, initiating the release of inflammatory mediators (e.g. oxygen free radicals and nitric oxide) from leukocytes and endothelial cells, or enhancing the interaction between these cells. On the other hand, compromised endothelial cells can increase the synthesis and secretion of IL-1. The endothelial-dependent effects of IL-1 have been suggested to have an important role in the pathophysiological process [12]. Data from the present study demonstrated a strong correlation between plasma levels of IL-1 and the exudation of plasma albumin into the interstitium. Treatment with lexipafant, a PAF antagonist, reduced the otherwise occurring pancreatitis-induced increase in plasma levels of IL-1, especially at 12 h after induction, although IL-1 appeared positive in both pancreatitis animals treated with saline and lexipafant as compared to sham-operated animals. Lexipafant has partial effects on reduction of pancreatitis-induced increase of circulating IL-1 and IL-6 at 12 h, indicating that the PAF receptor may be partially involved in the production of these cytokines, probably through the reduction of leukocyte activation and infiltration into the local tissue. Although the exact mechanisms by which PAF receptors are involved in the production of cytokines, PAF was found to directly stimulate the synthesis and production of IL-1, TNFα, and IL-6 in human neutrophils in the in vitro condition, and inhibitory effects of PAF antagonists highly varied among their specificities acting with receptors [23].

In conclusion, pancreatitis-associated gut barrier dysfunction was characterized by increased exudation of plasma albumin into the interstitium and alterations in bidirectional permeability of the intestine at the early stage of bile-induced acute pancreatitis. Treatment with lexipafant, a PAF antagonist, early during the course of disease, reduced the severity of pancreatitis-associated intestinal dysfunction, associated with a diminish in systemic concentrations of IL-1 and local leukocyte recruitment.

Acknowledgments

The present study was supported by grants from the Swedish Medical Research Council (grant no. K99-73X-11236), the Crafoord Foundation, Forsman's Foundation, Ake Wiberg's Foundation, Magnus Bergvall's Foundation and Clas Groschinskys Memorial Foundation.

References

[1] Andersson R, Sun ZW, Wang XD, Deng XM, Soltesz V, Ihse I. Effect of a platelet-activating factor antagonist on pancreatitis-associated gut barrier dysfunction in rats. Pancreas 1998;17:107–19.
[2] Beger HG, Bittner R, Block S, Buchler M. Bacterial contamination of pancreatic necrosis: a prospective clinical study. Gastroenterol 1986;91:433–41.
[3] Bouilot JL, Alexandre JH, Vuong NP. Colonic involvement in acute necrotizing pancreatitis: results of surgical treatment. World J Surg 1989;13:84–91.
[4] Bradley PP, Priebat DA, Christensen RD, Rothstein G. Measurement of cutaneous inflammation: estimation of neutrophil content with an enzyme marker. J Invest Dermatol 1982;78:206–9.
[5] Chao W, Olson MS. Platelet-activating factor: receptors and signal transduction. Biochem J 1993;292:617–27.
[6] Chartrand-Lefebvre C, Clemont RJ, Heppell J, Bernard EJ, Prosmanne O. Necrotic stenosis of the right colon secondary to acute pancreatitis. Can J Surg 1994;37:140–50.
[7] Fernandez-Cruz L, Navarro S, Castells A, Saenz AA. Late outcome after acute pancreatitis: functional impairment and gastrointestinal tract complications. World J Surg 1997;21:169–72.
[8] Formela LJ, Galloway SW, Kingsnorth AN. Inflammatory mediators in acute pancreatitis. Br J Surg 1995;82:6–13.
[9] Juvonen PO, Alhava EM, Takala JA. Gut permeability in patients with acute pancreatitis. Scand J Gastroenterol 2000;35:1314–8.
[10] Kingsnorth AN, Galloway SW, Formela LJ. Randomized, double-blind phase II trial of lexipafant, a platelet-activating factor antagonist, in human acute pancreatitis. Br J Surg 1995;82:1414–20.
[11] Leveau P, Wang XD, Soltesz V, Ihse I, Andersson R. Alterations in intestinal motility and microflora in experimental acute pancreatitis. Int J Pancreatol 1996;20:119–25.
[12] Lumsden A, Bradley III EL. Secondary pancreatic infections. Surg Gynecol Obstet 1990;170:459–65.
[13] Runkel NF, Moody FG, Smith FG, Rodriguez LF, LaRocco MT, Miller TA. The role of the gut in the development of sepsis in acute pancreatitis. J Surg Res 1991;51:18–27.
[14] Schölmerich J. Interleukins in acute pancreatitis. Scand J Gastroenterol 1996;31:S37–42.
[15] Thompson WM, Kelvin FM, Rice RP. Inflammation and necrosis of the transverse colon secondary to pancreatitis. AJR 1977;128:943–9.
[16] Wang XD, Andersson R, Kruse P, Ihse I. Carbon dioxide transport in rats with acute pancreatitis. Int J Pancreatol 1996;19:103–12.
[17] Wang XD, Andersson R, Soltesz V, Leveau P, Ihse I. Gut origin sepsis, macrophage function, and oxygen extraction associated with acute pancreatitis in the rat. World J Surg 1996;20:299–308.
[18] Wang XD, Deng XM, Haraldsen P, Andersson R, Ihse I. Antioxidant and calcium channel blockers counteract endothelial barrier injury induced by acute pancreatitis in the rat. Scand J Gastroenterol 1995;30:1129–36.
[19] Wang XD, Sun ZW, Soltesz V, Deng XD, Andersson R. The role of intravenous administration of dextran 70 in enteric bacterial translocation after partial hepatectomy in rats. Eur J Clin Invest 1997;27:936–42.
[20] Wang XD, Wang Q, Andersson R, Ihse I. Alterations in intestinal function in acute pancreatitis in an experimental model. Br J Surg 1996;83:1537–42.
[21] Zhou W, McCollum MO, Levine BA, Olson MS. Role of plateletactivating factor in pancreatitis-associated acute lung injury in the rat. Am J Physiol 1992;140:971–9.
[22] Zhou W, McCollum MO, Levine BA, Olson MS. Role of platelet-activating factor in pancreatitis-associated acute lung injury in the rat. Am J Pathol 1992;140:971–9.
[23] Herbert JM, Castro-Faria-Neto HC, Barbosa-Filho JM, Cordeiro RS, Tibirica EJ. Pharmacological evidence for the putative existence of two different subtypes of PAF receptors on platelets and leukocytes; studies with yangambin. Lipid Mediat Cell Signal 1997;17:1–14.
[24] Bussolino F, Camussi G, Tetta C, Garbarino G, Bosia A, Baglioni C. Selected cytokines promote the synthesis of platelet-activating factor in vascular endothelial cells: comparison between tumor necrosis factor alpha and beta and interleukin-1. J Lipid Mediat 1990;2: S15–S22.