Melatonin, produced from L-tryptophan, protects the pancreas against acute damage by improving the antioxidative status of tissue. Melatonin receptors have been detected in the brain, but the contribution of these receptors to the pancreatic protection is unknown. The aim of our study was to compare the effects of melatonin precursor; L-tryptophan given intracerebroventricularly (i.c.v.) or intraperitoneally (i.p.) on the course of acute pancreatitis. Acute pancreatitis was induced by subcutaneous infusion of caerulein (5μg/kg-h x 5h). L-tryptophan was given i.p. (2.5, 25 or 250 mg/kg) or administered into right cerebral ventricle (0.02, 0.2 or 2.0 mg/rat) 30 min prior to the start of caerulein infusion. Plasma amylase, lipase and TNFα activities were measured to determine the severity of caerulein-induced pancreatitis (CIP). The lipid peroxidation products: malonyldialdehyde and 4-hydroxynonenal (MDA + 4-HNE) and activity of superoxide dismutase (SOD) were measured in the pancreas of intact or CIP rats with or without L-tryptophan pretreatment. Melatonin blood level was measured by RIA. CIP was confirmed by histological examination and manifested as an edema and rises of plasma levels of amylase, lipase and TNFα (by 550%, 1000% and 600%), MDA + 4-HNE was increased by 600%, whereas SOD activity was reduced by 75% in the pancreas of CIP rats. All manifestations of CIP were significantly reduced by pretreatment of the rats with L-tryptophan given i.c.v. at doses of 0.2 or 2.0 mg/rat, or by peripheral administration of this amino acid used at dose of 250 mg/kg i.p. In control rats plasma level of melatonin averaged about 40 ± 2 pg/ml and was not significantly affected by CIP, by central application of L-tryptophan (0.02, 0.2 or 2.0 mg/rat) or by peripheral administration of this melatonin precursor used at doses of 2.5 or 25 mg/kg i.p. Plasma melatonin level was markedly increased by pretreatment of the rats with L-tryptophan given i.p. at dose of 250 mg/kg. We conclude that central administration of melatonin precursor; L-tryptophan, as well as peripheral application of high dose of this melatonin precursor prevented the pancreatic damage produced by CIP. The favorable effect of peripherally administered L-tryptophan
could be related to the rise of melatonin plasma level and to pancreatoprotective action of this indoleamine. The beneficial effect of centrally administered L-tryptophan could be mediated through activation of central receptors for locally produced melatonin.

**Key words:** L-tryptophan, melatonin, reactive oxygen species (ROS), lipid peroxidation, superoxide dismutase (SOD).

**INTRODUCTION**

Melatonin (5-methoxy-N-acetyltryptamine) is a pineal hormone, which is produced from L-tryptophan in four-steps process, involving specific enzymes, two of them are: N-acetyltransferase of serotonin (NAT) and hydroxyindolo-O-methyltransferase (HIOMT) (1-2). Melatonin has been isolated primarily from the pineal gland at 1958, but following studies have shown that this hormone is present in many tissues including retina, Harderian gland, ciliary bodies and intestinal mucosa (3 - 4). Recent studies have reported that total amount of melatonin in gastrointestinal tract (g.i. tract) was almost 400 times higher than that detected in the pineal gland (5). This indoleamine binds to the specific membrane receptors, which are widely distributed in the brain (6 - 7) and in the other tissues such as lymphoid organs, gonads, kidneys and g.i. tract (8 - 10). Melatonin receptors have been also characterized in the pancreas but the physiological role of this substance in the pancreas is unknown (11). Previous reports have shown that melatonin could take a part in the regulation of pancreatic endocrine function and that activation of mt1 melatonin receptor in β-cells and in INS-1 insulinoma cells decreased cAMP in these cells and supressed insulin secretion (12, 13).

Besides melatonin's involvement in the regulation of the biological rhythms of the organism, this substance is also capable to scavenge of reactive oxygen species (ROS) (14) and to activate antioxidative enzymes such as superoxide dismutase (SOD), catalase (CAT) or glutathione peroxidase (GSH-Px) (15). The intensity of oxidative stress in the pancreas is estimated by an increase of lipid peroxidation products such as: malondialdehyde (MDA) and 4-hydroxynonenal (4-HNE) as well as by the decreases of SOD activity in pancreatic tissue (16). The involvement of ROS in the pancreatic damage has been studied in different experimental models of acute pancreatitis, including that produced by caerulein overstimulation (17, 19 - 21). Pretreatment of animals subjected to caeruleine-induced pancreatitis (CIP) with the antioxidative enzymes such as SOD or CAT resulted in the significant attenuation of all inflammatory changes in the pancreas of these animals (22).

Our earlier studies have shown that melatonin exerted pancreatoprotective effects following the peripheral, but not central administration of this
substance (17). We have found that above protection is related to the significant improvement of pancreatic blood flow (PBF) and to the marked reduction of ROS generation in the pancreatic tissue of rats with acute pancreatitis (17, 21).

The aim of this study was: 1) to compare the effects of intracerebroventricular (i.c.v.) or intraperitoneal (i.p.) administration of melatonin precursor; L-tryptophan on the course of CIP, 2) to assess whether the generation of ROS and activity of SOD could be affected by this amino acid and 3) to determine the effect of L-tryptophan on proinflammatory cytokine; TNFα production in animals subjected to CIP.

MATERIALS AND METHODS

Animals and drugs

Studies were performed on male Wistar rats weighing 180-220 g. Animals were housed in cages under standard conditions, on commercial pellet chow, at room temperature with a 12-hr light: 12-hr dark cycle. Rats were deprived of food 17 h prior to the start of experiment, while drinking water was available ad libitum.

Following items were purchased: caerulein (Takus) from Pharmacia GmbH (Erlangen, Germany), L-tryptophan from Sigma Co (St Louis, MO, USA), TNFα solid phase enzyme-linked immunosorbent assay (ELISA) kit from BioSource International INC. (Camarillo, CA, USA), LPO-586 and SOD-525 commercial kits from OXIS Research (Portland, OR, USA) and rat melatonin immunoassay (RIA) was purchased from IBL (Immuno-Biological Laboratory, Hamburg, Germany).

Experimental protocol

The experimental protocol has been approved by the Jagiellonian University Ethical Committee for Animal Experimentations.

All experiments were carried out at the same time in the morning. During the experiments rats were placed in individual Bollman cages. Acute caerulein-induced pancreatitis was produced by subcutaneous (s.c.) infusion of caerulein at a total dose of 25 μg/kg (5 μg/kg/h for 5 hr). Caerulein was diluted in saline and infused at a rate 1 ml/h. L-tryptophan was dissolved in a drop of 0.1 N HCl and then in 0.9% saline. For the first part of the study various doses of L-tryptophan (2.5, 25 or 250 mg/kg) were diluted in 0.5 ml of solution and given i.p. to the rats as a bolus injection 30 min prior to the start of caerulein or saline (control experiments) infusion. In the second part of the study L-tryptophan (0.02, 0.2, or 2.0 mg/rat) dissolved in 20 µl of vehicle was administered i.c.v. 30 min prior to the start of CIP or saline infusion (control test) to the rats.

The study consists of two parts (A and B).

In part A L-tryptophan was given i.p. as a bolus injection 30 min prior to the start of caerulein infusion to induce CIP. Part B was concerned with the effects of centrally administered L-tryptophan on the course of CIP. Each part of the study consisted of several experimental groups of animals, 6-8 fasted rats in each single group.
PART A

The study on the effects of peripheral (i.p.) administration of L-tryptophan on CIP

The following study groups, each consisted of 6-8 animals, were employed including: 1) Control (0.5 ml of vehicle saline) injected i.p., followed 30 min later by s.c. infusion of 0.9% saline for 5 hr, 2) Vehicle (0.5 ml) injected i.p., followed 30 min later by s.c. infusion of caerulein at a total dose of 25 µg/kg (5 µg/kg/h for 5 hr) to induce CIP, 3) L-tryptophan (2.5, 25, or 250 mg/kg i.p.) dissolved in 0.5 ml of vehicle followed by s.c. infusion of caerulein at a total dose of 25 µg/kg for 5 hr, 4) L-tryptophan (2.5, 25, or 250 mg/kg i.p.) dissolved in 0.5 ml of vehicle followed by s.c. infusion of 0.9% saline for 5 h.

PART B

The study on the effects of central (i.c.v.) administration of L-tryptophan on CIP

For this part of the study L-tryptophan was dissolved in 20 µl of vehicle and administered into right cerebral ventricle (i.c.v.) of the rats as described previously (19). Briefly, under the ether light anaesthesia, an incision was made along the midline of the skull, the skull bones were cleaned of connective tissues and intersection between the sagittal and coronary sutures was visualized. A point at a distance of 2.5 mm from both sutures was found and at this point small hole was made in the skull using a needle with a sharp end. The hole was made with a rotary movement of the needle and the head wound was closed by a clip. The effectiveness of i.c.v. administration was verified by injecting 20 µl of 0.1% toluidine blue.

The following study groups, each consisted of 6-8 animals, were employed including: 1) Control (20 µl of vehicle saline) injected i.c.v. followed 30 min later by s.c. infusion of 0.9% saline for 5 hr, 2) Vehicle (20 µl) injected i.c.v. followed 30 min later by s.c. infusion of caerulein at a dose of 5 µg/kg/h for 5 hr to induce CIP, 3) L-tryptophan (0.02, 0.2 or 2.0 mg/rat i.c.v.) dissolved in 20 µl of vehicle followed by s.c. infusion of caerulein for 5 hr, 4) L-tryptophan (0.02, 0.2 or 2.0 mg/rat i.c.v.) dissolved in 20 µl of vehicle followed by s.c. infusion of 0.9% saline for 5 h.

Determination of plasma levels of amylase, lipase, TNFα and melatonin

Following 5-hr infusion of caerulein or vehicle (in control tests) animals were anaesthetized with Morbital (0.2 ml/rat) and then the abdominal cavity was opened. The vena cava was exposed and blood was withdrawn into ethylenediaminetetraacetic acid (EDTA)-containing tubes for determination of plasma amylase, lipase and TNFα activities. Plasma amylase was measured with the modified scharogenic method using Alpha Diagnostics kit as described previously (18). Lipase was determined using an automatic analyser Kodak Ektachem chemistry slides (LIPA, Rochester, NY, USA) (20). Plasma TNFα was determined using an ELISA according to the manufacturer’s protocol as described previously (18). Plasma melatonin concentration was measured by RIA using rat melatonin RIA kit, according to the manufacture’s procedure (23).

Pancreatic weight and histological examination

The pancreas was carefully dissected from its attachment to the stomach, duodenum and spleen, rinsed and weighted. Pieces of the pancreas were excised from the body portion, fixed in 10% formaline and stained with hematoxylin and eosin (H&E). Pancreatic samples were examined by a professional histologist without knowledge of the treatment given. The histologic grading of edema, leukocyte infiltration and vacuolisation was made using a scale ranging from 0 to 3 as described previously (18).
Determination of lipid peroxidation products in the pancreatic tissue

The samples of fresh pancreatic tissue were taken for measurement of lipid peroxidation products malondialdehyde (MDA) and 4-hydroxynonenal (4-HNE), using LPO-586 commercial kit, according to the manufacturer's protocol. Briefly, samples of pancreatic tissue weighing about 300 mg were homogenized in the phosphate buffer (20mM, pH 7.4). Then, 10 µl of 0.5 M butylated hydroxytoluene in acetonitrile was added to each sample to prevent tissue oxidation. Samples were centrifuged and the supernatant were immediately frozen at 70°C until assay. MDA + 4-HNE was measured in duplicate and expressed as nM/g of tissue.

Determination superoxide dismutase (SOD) activity in the pancreas

To determine the activity of superoxide dismutase (SOD), a sample of pancreatic tissue was obtained and perfused with 0.9% NaCl containing 0.16 mg/ml heparin to remove red blood cells, followed by homogenization and centrifugation, as described previously (24). The colorimetric assay for assessment of SOD activity (Bioxytech SOD-525, Oxis, Portland, USA) was used. SOD activity was measured in duplicate and expressed as units per gram of tissue (U/g).

Statistical analysis

Comparison of the differences between the mean values of various groups of experiments were made using an ANOVA and the Student's t-test for unpaired data. A difference with a P value of < 0.05 was considered statistically significant. Results were expressed as means ± S.E.M.

![Graph](image)

Fig. 1. Pancreatic weight in rat subjected to caerulein-induced pancreatitis (CIP) pretreated with increasing doses of L-tryptophan given intracerebroventricularly (i.c.v.) or intraperitoneally (i.p.). (To induce CIP caerulein was given isubcutaneously at dose of 5 µg/kg x 5 h). Asterisk (*) indicates significant ($P < 0.05$) change, as compared to the value obtained from the rats subjected to CIP alone. Control = value obtained from the rats pretreated with vehicle saline alone. Means ± SEM of 6-8 rats in each experimental group.
**Fig. 2.** Effect of central (i.c.v) or peripheral (i.p.) application of L-tryptophan on plasma amylase activity in rats subjected to caerulein-induced pancreatitis (CIP). Asterisk (*) indicates significant ($P > 0.05$) change, as compared to the value obtained from the rats subjected to CIP alone. Control = value obtained from the rats pretreated with vehicle saline alone. Means ± SEM of 6-8 rats in each experimental group.

**Fig. 3.** Effect of central (i.c.v) or peripheral (i.p.) application of L-tryptophan on plasma lipase activity in rats subjected to caerulein-induced pancreatitis (CIP). Asterisk (*) indicates significant ($P > 0.05$) change, as compared to the value obtained from the rats subjected to CIP alone. Control = value obtained from the rats pretreated with vehicle saline alone. Means ± of 6-8 rats in each experimental group.
RESULTS

Effects of L-tryptophan given i.p. or i.c.v. on pancreatic weight, plasma amylase, lipase and TNFα activities

Subcutaneous infusion of caerulein (5 µg/kg/h during 5 hr) to the rats produced CIP in all animals tested. CIP was manifested by an increase of pancreatic weight, plasma amylase and lipase activities (by 200%, 550% and 1000% respectively) (Figs 1-3). Infusion of caerulein resulted also in marked rise in the plasma level of proinflammatory cytokine; TNFα (by about 600%) in the CIP rats as compared to the vehicle-treated control animals (Fig. 4).

Pretreatment of CIP rats with L-tryptophan given centrally at doses of 0.2 or 2.0 mg/rat i.c.v., as well as peripheral application of this amino acid at dose of 250 mg/kg i.p., resulted in the significant reduction of pancreatic weight, plasma amylase, and TNFα activities, as compared to the values obtained from the CIP rats, without L-tryptophan pretreatment (Figs 1, 2, 4). Plasma lipase level was markedly decreased by L-tryptophan given at dose of 2.0 mg/rat i.c.v. or by 250 mg/kg i.p. (Fig. 3). Intraperitoneal application of L-tryptophan (2.5, or 25 mg/kg i.p.) given to the rats 30 min prior to the start of CIP failed to affect significantly pancreatic edema as well as plasma amylase, lipase and TNFα activities in these animals. (Figs 1-4).

![Graph](image)

*Fig. 4.* Effect of i.c.v. or i.p. administration of L-tryptophan on TNFα plasma activity in rats subjected to caerulein-induced pancreatitis (CIP). Asterisk (*) indicates significant \( P > 0.05 \) change, as compared to the value obtained from the rats subjected to CIP alone. Control = value obtained from the rats pretreated with vehicle saline alone. Means ± of 6-8 rats in each experimental group.
Effects of L-tryptophan given i.p. or i.c.v. MDA + 4-HNE content and SOD activity in pancreatic tissue

The content of MDA + 4-HNE in the pancreatic tissue obtained from vehicle-treated rats was about 6.0 ± 1.4 nM/g of tissue. Caerulein overstimulation markedly increased MDA + 4-HNE in the pancreas to about 27.0 ± 2.0 nM/g of tissue (Fig. 5).

L-tryptophan given into the right cerebral ventricle resulted in the significant and dose-dependent decrease of MDA + 4-HNE generation in the pancreatic tissue, as compared to the value obtained in the rats with CIP alone (Fig. 5). Peripheral application of this melatonin precursor at dose of 250 mg/kg also markedly reduced the content of lipid peroxidation products in the pancreas of rat subjected to caerulein overstimulation (Fig. 5).

Following caerulein infusion to produce CIP, the activity of superoxide dismutase (SOD) in the pancreas decreased by about 60%, comparing to control value. Central administration of higher doses of L-tryptophan (0.2 or 2.0 mg/rat i.c.v.), or intraperitoneal application of this amino acid at dose of 250 mg/kg significantly increased SOD level in pancreatic tissue (by about 40%, 105% or 94%, respectively) comparing to the values obtained from rats with CIP alone (Fig. 6). Pretreatment of the CIP rats with L-tryptophan given at a dose of 0.02 mg/rat i.c.v. or peripheral administration of L-tryptophan (2.5 or 25 mg/kg i.p.)

Fig. 5. Lipid peroxidation products (MDA+4-HNE) in pancreatic tissue obtained from the rats subjected to caerulein-induced pancreatitis (CIP) pretreated with L-tryptophan given i.c.v. or i.p. Asterisk (*) indicates significant ($P > 0.05$) change, as compared to the value obtained from the rats subjected to CIP alone. Control = value obtained from the rats pretreated with vehicle saline alone. Means ± of 6-8 rats in each experimental group.

Effects of L-tryptophan given i.p. or i.c.v. MDA + 4-HNE content and SOD activity in pancreatic tissue

The content of MDA + 4-HNE in the pancreatic tissue obtained from vehicle-treated rats was about 6.0 ± 1.4 nM/g of tissue. Caerulein overstimulation markedly increased MDA + 4-HNE in the pancreas to about 27.0 ± 2.0 nM/g of tissue (Fig. 5).

L-tryptophan given into the right cerebral ventricle resulted in the significant and dose-dependent decrease of MDA + 4-HNE generation in the pancreatic tissue, as compared to the value obtained in the rats with CIP alone (Fig. 5). Peripheral application of this melatonin precursor at dose of 250 mg/kg also markedly reduced the content of lipid peroxidation products in the pancreas of rat subjected to caerulein overstimulation (Fig. 5).

Following caerulein infusion to produce CIP, the activity of superoxide dismutase (SOD) in the pancreas decreased by about 60%, comparing to control value. Central administration of higher doses of L-tryptophan (0.2 or 2.0 mg/rat i.c.v.), or intraperitoneal application of this amino acid at dose of 250 mg/kg significantly increased SOD level in pancreatic tissue (by about 40%, 105% or 94%, respectively) comparing to the values obtained from rats with CIP alone (Fig. 6). Pretreatment of the CIP rats with L-tryptophan given at a dose of 0.02 mg/rat i.c.v. or peripheral administration of L-tryptophan (2.5 or 25 mg/kg i.p.)
Fig. 6. Superoxide dismutase (SOD) activity in pancreatic tissue obtained from the rats subjected to caerulein-induced pancreatitis (CIP) pretreated with L-tryptophan given i.c.v. or i.p. Asterisk (*) indicates significant \((P > 0.05)\) change, as compared to the value obtained from the rats subjected to CIP alone. Control = value obtained from the rats pretreated with vehicle saline alone. Means ± of 6-8 rats in each experimental group.

Fig. 7. Plasma melatonin immunoreactivity in CIP rats pretreated with increasing doses of L-tryptophan given intracerebroventricularly (i.c.v.) or intraperitoneally (i.p.). Asterisk (*) indicates significant \((P > 0.05)\) change, as compared to the value obtained from the rats subjected to CIP alone. Control = value obtained from the rats pretreated with vehicle saline alone. Means ± of 6-8 rats in each experimental group.
failed to affect significantly pancreatic content of MDA + 4-HNE or SOD activity in the pancreas of these animals (Figs 5, 6).

In the control rats subjected to infusion of vehicle instead of caerulein, administration of L-tryptophan i.p (2.5, 25 or 250 mg/kg) as well as i.c.v (0.02, 0.2 or 2.0 mg/rat) failed to affect significantly all pancreatic parameters tested and these results were omitted for the sake of clarity.

Effects of L-tryptophan given i.p. or i.c.v. on melatonin plasma level

In control rats plasma melatonin immunoreactivity averaged 40 ± 2 pg/ml (Fig. 7). Caerulein overstimulation did not influence significantly this plasma melatonin level (Fig. 7). Central administration of L-tryptophan at graded doses (0.02, 0.2 or 2.0 mg/rat i.c.v.) as well as peripheral application of this amino acid at lower doses (2.5 or 25 mg/kg i.p.) failed to affect significantly this plasma melatonin

---

**Fig. 8.** Histologic section of pancreas taken from control rats (A), from the rats subjected to caerulein-induced pancreatitis (CIP) alone (B), from the CIP animals pretreated with L-tryptophan (250 mg/kg) given i.p. (C) and from the rats pretreated with L-tryptophan (2 mg/rat) applied i.c.v. (D). Hematoxylin and eosin (H&E) stain; magnification 165x.
immunoreactivity, whereas pretreatment of the rats with 250 mg/kg of L-tryptophan resulted in the increase of plasma melatonin level up to 400 ± 85 pg/ml (Fig. 7).

Histological examination

Subcutaneous infusion of caerulein (5 µg/kg/h for 5 hr) produced typical pancreatic lesions in all tested rats (Fig. 8, Table 1). The pancreas was grossly swollen and enlarged, peritoneal fluid was present in all animals, and edema was accompanied by perivascular infiltration of leukocytes and the vacuolization of acinar cells. In CIP rats pretreated with L-tryptophan given centrally (i.c.v.) at doses of 0.2 or 2 mg/rat, or peripherally (i.p.) at dose of 250 mg/kg a significant reduction of edema and neutrophil infiltration was observed. These changes were accompanied by the decrease of total amount of vacuolized cells (Fig. 8, Table 1). Central application of L-tryptophan at lowest dose (0.02 mg/kg i.c.v.) as well as intraperitoneal administration of this amino acid at doses of 2.5 or 25 mg/kg i.p. to the CIP animals failed to affect significantly pancreatic inflammatory changes produced by CIP (Fig. 8 and Table 1).
DISCUSSION

The results of present study provides the evidence that administration of melatonin precursor; L-tryptophan given i.c.v., effectively protects the pancreas against acute inflammation provoked by caerulein overstimulation. In this study we have compared the pancreatoprotective influence of central application of L-tryptophan with the effect of intraperitoneal administration of this melatonin precursor. Herein we confirm the results of our previous study showing that L-tryptophan given i.p. at high doses is able to protect effectively the pancreas from acute damage, and that above beneficial effect of L-tryptophan is related to the increase of melatonin content in the organism (17). Our previous paper have documented, that exogenous melatonin as well as that produced endogenously from L-tryptophan protects pancreatic tissue from the acute damage by the reduction of ROS generation in the pancreas. Melatonin is also able to modulate the inflammatory cytokine production. This indoleamine reduced plasma level of proinflammatory TNFα, while increasing anti-inflammatory interleukin 10 (IL-10) (17, 20, 21). However, administration of melatonin into right cerebral ventricle of rats subjected to caerulein overstimulation failed to affect inflammatory changes in pancreatic tissue produced by CIP (17).

Previous reports have revealed that increased generation of ROS in pancreatic tissue is responsible for pancreatic damage in acute pancreatitis (25 - 26). Melatonin is a potent scavenger of ROS, and this indoleamine is able to scavenge the most dangerous hydroxyl radical (OH), which is produced in Fenton reaction (27). Since melatonin can easily cross cell membranes it was concluded that this indoleamine is able to pass through the blood-brain barrier and thus melatonin could regulate some processes in peripheral tissues (28). Besides melatonin's ability to scavenge of ROS, this substance has been demonstrated to activate the antioxidative enzymes such as catalase (CAT) and glutathione peroxidase (GSH-Px) (25 - 26). Recent reports have shown that also melatonin precursor; L-tryptophan exhibits a strong, antioxidative effect that was comparable to that of mannitol or to DMSO (29). At 1999 Tan and Reiter have reported that oxidative stress is involved in the pathogenesis of neurodegenerative diseases such as Alzheimer's disease or Parkinson's disease, and that this neurodegeneration could be attenuated by melatonin (30). It was also observed that melatonin prevented the animals from alloxan-induced diabetes by scavenging of hydroxyl radical produced in pancreatic β-cells (31-33).

Our studies have confirmed that content of lipid peroxidation products in the pancreas is dramatically enhanced by acute pancreatitis caused by caerulein overstimulation or ischemia-reperfusion (19 - 21). Peripheral administration of melatonin or L-tryptophan resulted in significant and dose-dependent reduction of MDA + 4-HNE in pancreatic tissue (21). Studies on acute pancreatitis have shown that the antioxidative enzymes such as SOD and CAT given to the animals subjected to experimental pancreatitis attenuated inflammatory changes in the
pancreatic tissue (25, 34). This observation suggests that ROS play a pivotal role in pathomechanism of AP. Administration of allopurinol; inhibitor of xanthine oxidase, which is involved in ROS production, reduced pancreatic damage caused by acute pancreatitis provoked by caerulein or L-arginine application (22). Melatonin or its precursor; L-tryptophan were shown to prevent from the development of gastric ulcers induced by ischemia-reperfusion, stress or application of aspirin by scavenging of ROS and by producing an improvement in gastric blood flow (16, 23). Recent studies have evidenced that melatonin exerts a gastroprotective effect following its administration to cerebral ventricles at a dose several timers lower than that, which produced attenuation of gastric ulcerations following peripheral application this indolamine (35).

Acute pancreatitis is associated with increased expression and production of pro-inflammatory cytokines, such as TNFα, IL-1β or IL-6 (19 - 21). Our earlier study has shown that administration of melatonin or L-tryptophan before the onset of CIP caused a significant reduction in plasma level of TNFα while increasing that of anti-inflammatory cytokine IL-10 (21). It has been observed that melatonin improved a functional and energetic status of the liver subjected to ischemia-reperfusion by decreasing TNFα level (36). Above results are in agreement with earlier observations of Lissoni who have revealed that melatonin is able to decrease TNFα production (37). To the contrary, other investigators have reported that this pineal indoleamine failed to influence TNFα production (38 - 39). The effect of melatonin on cytokine production is not completely clear, because it has been also demonstrated that melatonin have increased gene expression and production of IL-2, IL-6 and IL-12 while decreasing plasma level of IL-10 (21, 37, 40).

Our present study have shown that administration of low doses of L-tryptophan centrally (0.02, 0.2, or 2.0 mg/rat) as well as peripherally (2.5 or 25 mg/kg i.p.) failed to affect significantly melatonin plasma level. Previous reports have demonstrated that L-tryptophan given to the rats intraperitoneally or intraduodenally resulted in the dose-dependent rises of melatonin plasma activities (21, 23). In present study we confirm that peripheral application of high doses of L-tryptophan (250 mg/kg i.p.) resulted in the marked rise of plasma level of melatonin. It is likely that L-tryptophan given intracerebroventricularly was converted into melatonin in the brain. This locally produced melatonin could influence its central receptors to exert the pancreatoprotective effect. The mechanism of the beneficial effect of L-tryptophan after its central administration is unclear and could be related to the activation of efferent nerves or others indirect mechanisms, but it requires further study.

In conclusion: central administration of L-tryptophan to the CIP rats is able to attenuate pancreatic inflammatory changes thought activation of central, yet unknown, mechanisms. The protective effect of this melatonin precursor on the pancreas could be related, at least in part to the modulation of immune defence
of an organism and to the activation of antioxidative properties of the pancreatic tissue.

Acknowledgement: This study was supported by State Committee of Research Grant no 4 p05 B 061 19.

REFERENCES


Received: February 10, 2004
Accepted: March 1, 2004

Address correspondence to: Jolanta Jaworek MD PhD, Chair of Physiology Jagiellonian University CM, Grzegórzecka Street 16, 31-531 Kraków, Poland, phone: +48+12 424-72-30, fax: +48+12 421-15-78
E-mail: mpjawore@cyf-kr.edu.pl