Effects of Misoprostol and Taurine on Monochloramine Ulcerogenesis in Rats

A. A. Spasov, M. V. Chernikov, A. S. Speranskaya, and S. I. Zaichenko

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 141, No. 3, pp. 309-311, March, 2006
Original article submitted May 30, 2005

We compared gastroprotective characteristics of synthetic prostaglandin E_1 misoprostol and amino acid taurine on rat model of monochloramine injury to the gastric mucosa. Both substances exhibited a pronounced gastroprotective effect.

Key Words: *gastroprotectors; taurine; misoprostol; Helicobacter pylori*

Helicobacter pylori invasion is regarded as the most important pathogenetic factor of peptic ulcer [3,12]. The detrimental effect of *H. pylori* on the gastric mucosa (GM) is realized via a cascade process leading to the formation of active cytotoxic substance monochloramine [5]. High urease activity of *H. pylori* results in the formation of high concentrations of ammonium in the stomach of infected patients [2]. Stress, tobacco smoking, chronic inflammatory diseases of the stomach, *etc.*, cause microcirculatory disorders in GM [6,10]. The resultant ischemia and hypoxia lead to activation of xanthine oxidase and intensive generation of H_2O_2 by neutrophils, which intensifies the formation of hypochlorous acid by neutrophils. Ammonium interacts with hypochlorous acid with the formation of monochloramine. This agent induces damage to GM in far lower concentrations compared to ammonium [5,8,10,11].

We studied the gastroprotective activity of misoprostol on the model of monochloramine ulcer formation (it is believed to be effective towards this mechanism of ulcerogenesis). Misoprostol, a prostaglandin E_1 preparation, produces local antiischemic effect and improves microcirculation in GM [4], thus breaking the pathological chain at the level of xanthine oxidase activation and peroxide formation. For comparison amino acid taurine was used; it is known as a substance neutralizing hypochlorous acid, which stops monochloramine formation. This explains high gastroprotective activity of this amino acid in monochloramine-mediated mechanism of GM damage [7,9].

MATERIALS AND METHODS

Experiments were carried out on 97 male and female outbred albino rats (180-230 g). Before the experiment the animals were subjected to 24-h fasting with free access to water. Monochloramine is an unstable substance difficult for *in vitro* synthesis, and hence, in order to reproduce the specific monochloramine injuries, after administration of ammonium in suberosive doses we induced acute ischemia for stimulating hypochlorous acid release by neutrophils and increasing monochloramine formation directly in GM [11].

Ammonium solution (100 mmol/liter) was introduced into the stomach through a tube directly after acute dosed bleeding (1 ml/100 g) from the carotid artery in narcotized rats [11]. The substances were administered into the stomach through a tube 1 h before the formation of acute ischemia. The dose of 200-400 μg (2.9-5.8 μg/kg) is an effective dose of misoprostol for the treatment of erosive gastritis, gastroduodenal ulcer, and for preven-

Department of Pharmacology, Department of Forensic Medicine, Volgograd State Medical University; Laboratory of Experimental Pharmacology, Volgograd Research Center, Russian Academy of Medical Sciences. ***Address for correspondence:*** farm@interdacom.ru. A. A. Spasov

0007-4888/06/1413-0334

tion of gastroduodenal diseases during treatment with nonsteroid antiinflammatory drugs, and therefore we used the dose of 6 μg/kg in our study. Taurine was administered in the mean effective dose (30 mg/kg) on the basis of published reports on gastroprotective effect of this substance in doses of 10-100 mg/kg [7,9]. The severity of GM lesions was evaluated 1 h after administration of ammonium and expressed as the area (mm^2) of erosive hemorrhagic lesions.

The type of GM injuries was evaluated morphologically on macro- and micropreparations of the stomach wall of experimental animals. Tissue fragments were fixed in 10% neutral formalin and embedded in paraffin. The sections were stained with hematoxylin and eosin, Azur II and eosin, Toluidine Blue at pH 4.8-5.0 for evaluating the morphology and function of mast cells. Analysis of the histological preparations included evaluation of the severity of GM necrosis, type of cell infiltration, degree of edema and vascular plethora, and mast cell degranulation [1]. Polymorphonuclear leukocytes (PMNL), lymphocytes, and plasmacytes were counted in five visual fields under a microscope at ×400 and arithmetic mean per visual field was calculated.

The results were processed using Student's *t* test.

RESULTS

Ammonium treatment in combination with acute ischemia led to the development of numerous pronounced necrotic foci in GM epithelium (erosive ulcerative index 36.8±2.41 mm^2/animal), involving up to $^1/_4$ of mucosa thickness (Fig. 1, *a*). Degenerative and necrobiotic changes in surface epithelium were seen along the perimeter of necrobiotic foci. Changes in the submucosa of the stomach wall were seen as intensive diffuse focal cell infiltration in epithelial necrotic zones presented mainly by neutrophilic PMNL and to a lesser extent by lym-

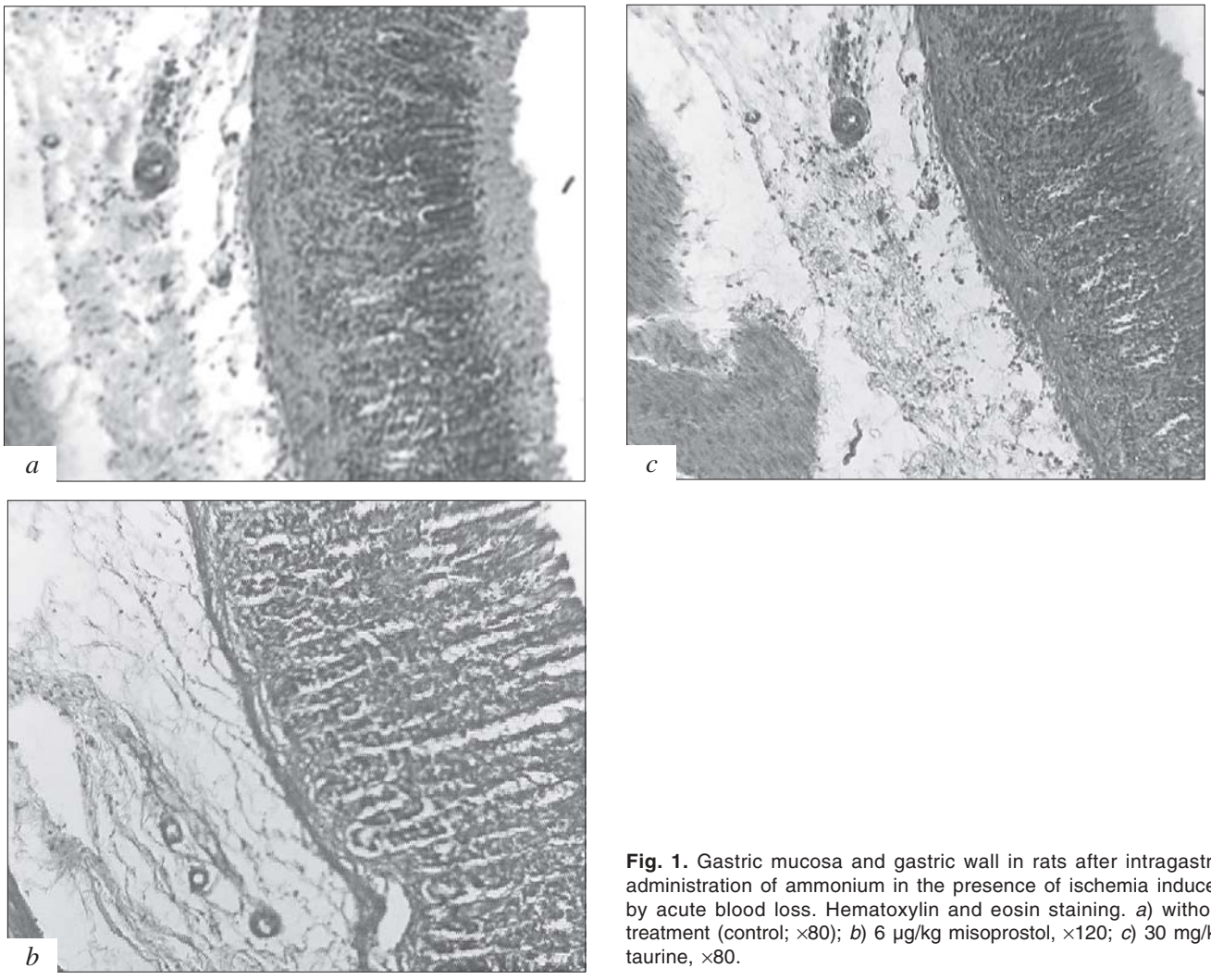

Fig. 1. Gastric mucosa and gastric wall in rats after intragastric administration of ammonium in the presence of ischemia induced by acute blood loss. Hematoxylin and eosin staining. *a*) without treatment (control; ×80); *b*) 6 μg/kg misoprostol, ×120; *c*) 30 mg/kg taurine, ×80.

TABLE 1. Effects of Misoprostol and Taurine on the Degree of Cell Infiltration in GM and Submucosa after Intragastric Treatment with Ammonium in the Presence of Ischemia Induced by Acute Hemorrhage ($M \pm m$)

Substance	Dose, mg/kg	Number of cells/visual field		
		PMNL	lymphocytes	plasma cells
Control		9.40±1.21	6.20±0.86	3.20±0.58
Misoprostol	6×10^{-3}	0.40±0.25*	0.2±0.2*	0.4±0.4*
Taurine	30	1.60±0.51*	0.80±0.37*	0.8±0.2*

Note. *$p<0.01$ compared to the control.

phocytes and plasma cells (Table 1). PMNL:lymphocytes:plasma cells ratio was 3:2:1. Degranulating cells predominated among mast cells; cells with signs of holocrine secretion were detected. Morphological signs of circulatory disorders (pronounced edema of the submucosa, plethoric vessels, leukodiapedesis and marginal stasis of leukocytes in vessels) were detected.

Preventive treatment with misoprostol was significantly decreased the area of GM lesions by more than 90% (erosive ulcerative index 2.5±0.37 mm^2/animal). Microscopy (Fig. 1, *b*) showed that GM necroses were superficial and involved mainly the surface epithelium; slight edema of the submucosa and lamina propria were observed. Cell migration into the mucosa and submucosa was markedly inhibited (Table 1). PMNL:lymphocytes:plasma cell ratio was 1:2:1. Moderate degranulation of mast cells was seen.

Taurine also reduced the area of GM lesions by 70% (erosive ulcerative index 8.23±0.92 mm^2/animal). Morphologically (Fig. 1, *c*) slight lesions of GM and negligible circulatory disorders with moderate plethora were paralleled by more intensive cell infiltration of the mucosa and submucosa (Table 1).

Hence, misoprostol exhibited a pronounced gastroprotective effect superior to that of taurine in rats with monochloramine-induced lesions of GM. Improving microcirculation in GM at the earliest stage of ulceration, misoprostol arrested peroxide hyperproduction, formation of hypochlorous acid, and suppressed leukocyte migration into GM, preventing the synthesis of cytotoxic monochloramine. Presumably, misoprostol not only disrupted the pathological chain of monochloramine formation, but also realized its gastroprotective effect by improving the barrier function of the mucus and suppressing hydrochloric acid production by parietal cells.

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