GASTRIC SECRETION AND DUODENAL ULCER FORMATION INDUCED BY CYSTEAMINE IN RATS

Hajime TAMAKI, Yuichi ONODA and Tatsuo KASHIDA

*Pharmacological Research Laboratory, Tanabe Seiyaku Co., Ltd.,
Toda, Saitama 335, Japan*

Accepted April 19, 1978

Selye and Szabo (1) reported that perforating duodenal ulcers can be developed almost invariably in rats after a single oral or parenteral dose of cysteamine. Later, Robert et al. (2), Groves et al. (3) and Fujii and Ishii (4) confirmed that the cysteamine-induced duodenal ulcers provide a reliable model for the test of anti-ulcer agents and/or for study of the ulcer healing process. Regarding the pathogenesis of duodenal ulcers produced by cysteamine, Groves et al. (3) and Ishii et al. (5) postulated that gastric acid secretion stimulated by cysteamine is involved in the ulcer formation. Robert et al. (2) suggested that the pathogenesis required some acid secretion, although cysteamine by itself is antisecretory. In any case, the passage of acid gastric contents through the duodenum is apparently necessary for the formation of ulcers. While pepsin is considered to be another factor responsible for the ulcerogenic effect of cysteamine, data so far reported indicate that the concentration of pepsin is not affected by cysteamine in pylorus-ligated rats (2, 4). We discuss herein the possibility that secretion of pepsin, as well as gastric acid secretion, is involved in the pathogenesis of cysteamine-induced duodenal ulcers in rats.

Female Sprague-Dawley rats, weighing approx. 220 g, were deprived of solid food but were allowed free access to water for 24 hr prior to experiments. Duodenal ulcers were induced as follows: a single dose of cysteamine HCl dissolved in water was given s.c. and the animals were fasted for 16 hr. Following sacrifice by an overdose of ether, the duodenum was excised and the duodenal lesions investigated. In each rat, the area (mm²) of lesions was measured under the dissecting microscope with a square grid and the sum was used as the ulcer index. In experiments with gastric secretion, animals were anesthetized with urethane (1 g/kg, i.p.) and the stomach was perfused (6) with distilled water at a rate of 2.5 ml/hr. Cysteamine was given s.c. in one dose and the perfusate was collected every hr. An aliquot of the perfusate was titrated with 1/500 N NaOH using autotitrator (Radiometer,

TABLE 1. Duodenal ulcers induced by administration of cysteamine to rats

Dose (mg/kg)	No. of animals	Incidence of ulcer(%)*	Ulcer index ($\bar{x} \pm S.E.$)	Perforating ulcer (%)**	Mortality (%)
200	10	0	0	0	0
300	20	65	8.7 ± 3.1	5	10
400	20	95	20.6 ± 6.7	20	20

* No. of animals with ulcer formation.

** No. of animals with perforating ulcer.

Copenhagen) and the acid output ($\mu\text{Eq/hr}$) was thus determined. Pepsin activity was measured by the method of Anson and Mirsky (7) and was expressed as mg of tyrosine/hr.

Table 1 illustrates the ulcerogenic activity of cysteamine in female rats. Cysteamine in a dose of 200 mg/kg produced no ulcer formation. When the dose of cysteamine was increased to 300 mg/kg, definite duodenal ulcers developed usually just below the pylorus. With 400 mg/kg of cysteamine, duodenal ulcers were found in almost every animal. Cysteamine also produced a dose-related increase in the ulcer index and perforating ulcer.

Fig. 1 shows the effect of cysteamine on gastric secretion in anesthetized rats. Experiments were carried out at two dose levels of cysteamine; one which did not produce an ulceration (100 mg/kg) and the other which produced severe duodenal ulceration (400 mg/kg). The stomach was perfused with distilled water and the contents of gastric acid and pepsin in the perfusate were determined. Cysteamine at a dose of 100 mg/kg produced only a slight increase in the gastric acid output, whereas the pepsin output was significantly increased, being maximum at approx. 4 hr after the administration of cysteamine. On the other hand, the higher dose of cysteamine (400 mg/kg) strongly stimulated the secretion of both gastric acid and pepsin. The acid output was increased linearly with time for the experimental period, while the pepsin output was increased more rapidly than that of the acid output and reached a peak at approx. 4 hr after the administration of cysteamine. Nine hr after the administration of cysteamine, a considerable increase in the pepsin output was still detectable.

Relationship between stimulation of the acid secretion and the ulceration of cysteamine has already been reported by Groves et al. (3) and Fujii and Ishii (4) and our results herein are fundamentally similar. While the concentration of pepsin is reportedly not affected by cysteamine in pylorus-ligated rats (2, 4), the present results demonstrated clearly that cyste-

FIG. 1. Time courses for the effects of cysteamine on the secretion of gastric acid and pepsin in stomach-perfused rats. Cysteamine was given s.c. at the arrow. A, gastric acid output; B, pepsin output. $\cdots\blacktriangle\cdots$: control (saline, $n=8$), $-\circ-$: cysteamine (100 mg/kg, $n=9$), $-\bullet-$: cysteamine (400 mg/kg, $n=14$). Some animals died during dosing with cysteamine (400 mg/kg). The number of animals used is indicated in parentheses. The vertical bars in the figure represent S.E. of the mean.

amine stimulated the pepsin secretion in a dose-dependent manner in stomach-perfused rats.

It has been stated that perfusion of the intestinal loop with hydrochloric acid-pepsin solutions produces peptic ulcers (8, 9). Hydrochloric acid alone in the perfusate was not productive of peptic ulcers (8), while these ulcers were formed more readily with acid-pepsin solutions of increasing acidity (9). In the present experiments, it was demonstrated clearly that subcutaneous administration of a high dose of cysteamine (400 mg/kg) produced duodenal ulcers and stimulated the secretion of both gastric acid and pepsin in rats. Therefore, it is assumed that the formation of cysteamine-induced duodenal ulcers is ascribable to the increased amounts of gastric acid and pepsin, both of which are secreted in response to the stimulation by cysteamine, and later act on the duodenum. In the presence of a low dose of cysteamine (100 mg/kg), no duodenal ulcer was formed, whereas the secretion of gastric acid and pepsin was to some extent, increased by cysteamine. Such may be explained by assuming that the amounts of gastric acid and pepsin secreted after cysteamine dosing were too small to produce duodenal ulcers, or that the amount of acid secreted was insufficient for stimulation of the ulcerogenic activity of pepsin.

Acknowledgements: We express our gratitude to Professor Emeritus H. Kumagai, University of Tokyo, Dr. A. Kiyomoto, Director of Pharmacological Research Laboratory, Tanabe Seiyaku Co., Ltd., and Dr. H. Nakajima for constant encouragement and pertinent suggestions.

REFERENCES

- 1) SELYE, H. AND SZABO, S.: Experimental model for production of perforating duodenal ulcers by cysteamine in the rat. *Nature* **244**, 458-459 (1973);
- 2) ROBERT, A., NEZAMIS, J.E., LANCASTER, C. AND BADALAMENTI, J.N.: Cysteamine-induced duodenal ulcers: A new model to test antiulcer agents. *Digestion* **11**, 199-214 (1974);
- 3) GROVES, W.G., SCHLOSSER, J.H. AND MEAD, F.D.: Acid hypersecretion and duodenal ulcers produced by cysteamine in rats. *Res. Commun. Chem. Pathol. Pharmacol.* **9**, 523-534 (1974);
- 4) FUJII, Y. AND ISHII, Y.: Influence of various factors and drugs on cysteamine-induced duodenal ulcers in the rats. *Japan. J. Pharmacol.* **25**, 663-670 (1975);
- 5) ISHII, Y., FUJII, Y. AND HOMMA, M.: Gastric acid stimulating action of cysteamine in the rat. *Europ. J. Pharmacol.* **36**, 331-336 (1976);
- 6) GHOSH, M.N. AND SCHILD, H.O.: Continuous recording of acid gastric secretion in the rat. *Brit. J. Pharmacol.* **13**, 54-61 (1958);
- 7) ANSON, M.L. AND MIRSKY, A.E.: The estimation of pepsin with hemoglobin. *J. gen. Physiol.* **16**, 59-63 (1932);
- 8) SCHIFFRIN, M.J.: Production of experimental jejunal ulcer. *Gastroenterology* **8**, 592-594 (1947);
- 9) LEVEEN, H.H.: Chemical, physiological, and pathological observations on the role of pepsin and hydrochloric acid in the production of experimental ulcers. *Proc. Soc. exp. Biol. Med.* **45**, 648-661 (1940)