AGGRAVATING EFFECT OF ERGOMETRINE ON PYLORIC ANTRAL LESIONS IN INDOMETHACIN-TREATED ANIMALS AND STIMULATING EFFECT OF THIS DRUG ON GASTRIC ACID SECRETION

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Abstract—Induction of distinctive gastric antral ulcer by ergometrine in indomethacin-treated rats was investigated together with the gastric secretagogue property of this drug. Influence of several prophylactic drugs on indomethacin ulcer was examined, and among these drugs, ergometrine and methysergide were found to aggravate the gastric lesions. The aggravating effect was exerted exclusively in the pyloric antrum of rats and mice. Methysergide (20 mg/kg s.c.) and ergometrine (5-40 mg/kg s.c.), but not cyproheptadine (20 mg/kg s.c.), produced aggravation in pyloric antral lesions. This aggravating effect of ergometrine was also found in aspirin-induced gastric lesions in rats. However, in cold and restraint stress ulcers of rats, this effect was not apparent. Both ergometrine (0.5-2.5 mg/kg i.v.) and methysergide (2.5-5.0 mg/kg i.v.) stimulated gastric acid secretion, dose dependently, in anesthetized rats.

It is generally accepted that the high incidence of gastric ulcer in humans occurs in the antrum of the lesser curvature. In the rat used extensively in experimental ulcers, gastric lesions rarely occur in the antrum, although some workers have induced hemorrhagic erosions and deep ulcers in the antrum of dogs by repeated administrations of indomethacin (1). In the course of research on the effects of various receptor antagonists on indomethacin-induced gastric lesions, we found that methysergide, serotoninergic antagonist (anti-5HT), aggravated gastric lesions and the effect was exerted exclusively in the pyloric antrum of rats and mice.

The present study was undertaken to determine if other anti-5HT aggravate gastric antral lesions induced by indomethacin and if the drugs affect gastric acid secretion. A preliminary account of some of these data have been published in abstract form (2).

MATERIALS AND METHODS

1) Gastric ulceration: Male ddY mice, weighing 25-30 g, and Wistar rats, weighing 170-220 g, were used. Indomethacin (10 mg/kg p.o. or 40 mg/kg i.p.) or aspirin (200 mg/kg p.o.) was administered after a 48 hr fast and 6 hr later the stomach was removed with the animals under urethane anesthesia (1.25 g/kg i.p.). Pontamine Sky Blue (2% solution) had been injected into the tail vein 10 min previously remove. In the experiment on stress ulcer, the rat in a restraining cage was immersed up to the xyphoid process into water (23°C) for 17 hr, after a 24 hr fast as described previously (3, 4). The excised stomach was inflated with
saline solution and immersed into 5% formalin solution for 10 min to fix lightly the outer surface. The mucosal lesions were observed under the dissection microscope, and the sum of length of each lesion was recorded as the ulcer index. Test drugs were given 30 min before the administration of indomethacin or aspirin, or 10 min before the immersion of animals into water.

2) Gastric secretion: Male Wistar rats, weighing 170–220 g, were anesthetized with urethane (1.25 g/kg i.p.) after a 24 hr fast. The trachea was exposed and cannulated. A dual polyethylene cannula was introduced into the gastric lumen after ligation of the pylorus and the esophagus. The stomach was continuously perfused with saline solution through the gastric cannula at the rate of 2.5 ml/min. The perfusate was titrated with N/200 NaOH at pH 4 using an automatic titrator. The acid output during 2 min period in the perfusate was continuously recorded as described previously (5), except that a zero suppression adaptor (TOA DEN PA) was used.

Drugs: Drugs used were cimetidine (Smith, Kline & French Laboratories), atropine sulfate (Wako), mepyramine (ICN. K & K Laboratories), phenoxybenzamine-HCl (Tokyo-Kasei), propranolol-HCl (Nakarai), bethanechol (Eisai), cyproheptadine-HCl (Merck), ergometrine maleate (Tokyo-Kasei) and methysergide. The tested dose of drug was expressed in mg/kg in terms of the salt.

3) Statistical analysis: All data are presented as mean±S.E. Data were analyzed by the Student’s t-test (gastric lesion) and a paired t-test (gastric secretion). All differences were considered significant at p<0.05 or p<0.01.

RESULTS

1) Gastric ulceration

Mice: Methysergide, an anti-5HT, aggravated indomethacin (40 mg/kg i.p.)-induced gastric lesions, while relatively high doses of various receptor antagonists, including cimetidine, mepyramine, atropine and phenoxybenzamine, reduced the lesions in mice, as summarized in Table 1. Beta-adrenergic blocking agents, propranolol, did not significantly influence the indomethacin-induced gastric lesions. The aggravating effect of methysergide was restricted to the pyloric antrum and the macroscopically observed shape of pyloric antral lesions was not uniform. Ulcer index of indomethacin (10 mg/kg p.o.)-induced gastric lesions in the

| Treatments          | Dose mg/kg s.c. | No. of animals | Ulcer index mm |
|---------------------|-----------------|----------------|---------------|
| Control             |                 | (30)           | 21.9±2.8      |
| Methysergide        | 20              | (15)           | 29.9±2.7*     |
| Cimetidine          | 20              | (5)            | 21.0±3.3      |
|                     | 50              | (10)           | 16.2±0.9*     |
| Mepyramine          | 20              | (5)            | 17.8±3.3      |
|                     | 50              | (10)           | 9.4±2.0*      |
| Atropine            | 5               | (10)           | 14.1±2.5*     |
|                     | 10              | (10)           | 8.5±2.3**     |
| Phenoxybenzamine    | 20              | (10)           | 7.8±2.4**     |
| Propranolol         | 20              | (10)           | 26.7±3.8      |

Indomethacin suspended in 1% CMC solution (40 mg/kg i.p.) was given after a 48 hr fast and 6 hr later animals were sacrificed. Test drugs dissolved in saline were administered 30 min before the injection of indomethacin. *p<0.05, **p<0.01.
pyloric antrum was 6.3±1.3 mm and the severity of the lesion was significantly enhanced not only by methysergide (20 mg/kg s.c.) but also by another ergot alkaloid, ergometrine (40 mg/kg s.c.), as shown in Fig. 1. On the contrary, however, cyproheptadine inhibited pyloric lesions induced by indomethacin (Fig. 1). Perforation in the pyloric antral area was observed when the stomach was inflated with saline solution in the indomethacin (40 mg/kg i.p.) and ergometrine (40 mg/kg s.c.)-treated animals (two out of five).

**Rats:** Indomethacin at 40 mg/kg i.p. produced only a few petechiae in the pyloric antrum in rats and the lesions were aggravated by ergometrine (20 mg/kg s.c.), as shown in Fig. 2. Ergometrine alone did not produce gastric mucosal lesions. Microscopical observation of H.E. stained specimens of the aggravated area of the pyloric antrum by the treatment of ergometrine, indicated severe necrosis of the mucosal layer and the muscularis mucosa. Figure 3 summarizes the effect of ergometrine and methysergide on various experimental ulcers in rats. Ergometrine aggravated pyloric antral lesions, dose-dependently, in indomethacin-treated rats. The aggravating effect of ergometrine was also observed in aspirin-treated rats, but this drug had no influence on stress induced ulcers.

**2) Gastric acid secretion**

Figure 4 shows the influence of ergometrine and methysergide on basal gastric acid secretion in anesthetized rats. Both ergometrine and methysergide stimulated acid secretion, in a dose-related manner. Gastric acid stimulatory activity of ergometrine was 5–10 times of that of methysergide and was long lasting in comparison with that of bethanechol (Table 2).
Fig. 3. Effects of ergometrine and methysergide on various experimental ulcers in the rat.
Indomethacin or aspirin suspended in 1% CMC solution was given after a 48 hr fast and the animals were sacrificed 6 hr later. Stress ulcer was induced by immersing the restrained rats up to the xyphoid process into cold water at 23°C for 17 hr after a 24 hr fast. Test drugs dissolved in saline were administered s.c. 30 min before the immersion of animals into water. The ulcer index (mm) was measured separately in the pyloric antrum and the corpus. Indicates the index of only the antral area. Each group included 6–10 rats. *p<0.05, **p<0.01.

Table 2. Effect on gastric acid secretion in anesthetized rats

| Drugs      | Dose mg/kg i.v. | No. of animals | Increase of acid output μeq/0–30 min | μeq/30–60 min |
|------------|-----------------|----------------|--------------------------------------|---------------|
| Ergometrine| 0.5             | 5              | 13.6±6.5                             | 17.7±6.7      |
|            | 1.0             | 5              | 26.9±7.4                             | 45.1±16.8     |
|            | 2.5             | 5              | 43.3±11.5                            | 82.2±17.1     |
| Methysergide| 5.0            | 5              | 10.0±6.9                             | 10.6±6.3      |
|            | 0.2             | 5              | 30.2±7.3                             | 15.6±6.5      |

Animals were anesthetized with urethane (1.25 g/kg i.p.) after a 24 hr fast. The stomach was continuously perfused with saline solution through the gastric cannula. The perfusate was titrated with N/200 NaOH at pH 4. The mean value of the basal secretion before drug administration is 6.9±1.7 μeq/30 min (n=25). The values are mean±S.E. of the increase of acid output (the acid output after drug administration minus the value before drug administration) during 30 min period. *p<0.05, **p<0.01 (paired t-test).

**DISCUSSION**

Under the experimental conditions described herein, indomethacin alone produced only slight superficial erosion in the pyloric antrum, but methysergide and ergometrine aggravated gastric lesions induced by the administration of indomethacin or aspirin and the exacerbating effect of both drugs was restricted to the pyloric antrum. We also found that anti-5HT, methysergide and ergometrine, stimu-
lated gastric acid secretion in anesthetized rats.

In humans, the high incidence of peptic ulcer is observed in the antral area of the lesser curvature. However, antral lesions rarely occur in most experimental ulcer models, in small animals. Thus the gastric lesions developed in the pyloric antrum of rats in the present study warrant detailed investigations. Some workers have induced hemorrhagic erosions and deep ulcers in the antrum of dogs by the repeated administration of indomethacin (1). It has been also reported that aspirin produced antral gastric ulcers in cats given an intravenous infusion of histamine (6, 7). Although gastric lesions rarely occurred in the pyloric antral area of indomethacin- or aspirin-treated rats and of stress-loaded rats (Fig. 3), ergometrine or methysergide administered concomitantly with nonsteroidal anti-inflammatory drugs, produced aggravation in antral lesions and the effect of ergometrine was markedly stronger than that of methysergide.

Ergotalkoids have three major actions: 1) smooth muscle contraction, particularly in blood vessels and in the uterus, 2) adrenergic blocking effect and 3) central nervous system effects such as 5HT antagonistic and dopamine agonistic activity (8). We recently reported that the vasoactive factor of 5HT may be involved in the aggravating process of gastric lesions induced by thermocautery, under the healing process (9). Ergometrine-induced aggravation of antral lesions may be partly due to one of three effects of ergometrine, that is, smooth muscle contraction of blood vessels.

Endogenous prostaglandins markedly inhibit the ulcerogenic action of aspirin in rats (10) and of a wide variety of other experimental ulcerogenic agents (11). Therefore, effects of methysergide and ergometrine on the activity of prostaglandins should be investigated.

As to anti-5HT activity, Sanyal et al. reported that the administration of cyproheptadine for 6 days, in doses from 1 to 5 mg/kg i.p., caused not only gastric hemorrhagic erosions, but an increase in the incidence of restraint ulcers in rats (12). In the present study, however, cyproheptadine in a dose of 20 mg/kg s.c. inhibited indomethacin-induced gastric lesions in mice. This effect may be due to the potent histamine antagonistic activity of cyproheptadine (13).

In the present study, ergometrine and methysergide stimulated gastric acid secretion in anesthetized rats. Similar increases in gastric secretion with other anti-5HT were seen in pyloric ligated rats (12, 14). Therefore, the aggravating effect of anti-5HT on indomethacin-induced gastric lesions may be due to the gastric acid stimulatory activity of these drugs. However, bethanechol at the gastric acid secretory dosage (1 mg/kg s.c.)
had no effect on indomethacin-induced gastric lesions in the rat and the mouse (data not shown).

Although the mechanisms have not been clarified, ergometrine or methysergide aggravated antral lesions induced by indomethacin in rodents. In addition, the gastric acid secretory activity of ergometrine was evident. Further studies are underway to elucidate the underlying mechanisms of ergometrine on gastric function.

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