Effects of (−)-cis-2,3-Dihydro-3-(4-methylpiperazinylmethyl)-2-phenyl-1,5-benzothiazepin-4-(5H)-one Hydrochloride (BTM-1086) on Ulceration, Gastric Secretion and Mucosal Blood Flow in Experimental Animals

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Anti-ulcerous and anti-secretory effects of (−)-cis-2,3-dihydro-3-(4-methylpiperazinylmethyl)-2-phenyl-1,5-benzothiazepin-4-(5H)-one hydrochloride (BTM-1086), a new peptic-ulcer therapeutic agent, were studied in rats. BTM-1086 effectively prevented the ulceration of pylorus-ligated rats. A remarkable inhibition of gastric secretion was found at a dose of 0.1 mg/kg s.c. within a short period (6 h) after pylorus ligation. In the stomach-perfused rats, BTM-1086 (0.02 to 0.04 mg/kg i.v.) distinctly inhibited the teragastrin- and carbachol-induced gastric acid secretion, but only weakly inhibited the histamine-induced secretion. BTM-1086 also had a depressive effect on the gastric secretion stimulated by such secretagogues as tetragastrin, carbachol and histamine in rats with acute gastric fistulae. BTM-1086 increased the gastric mucosal blood flow in normal and indomethacin-induced ischemic rats. It also increased the gastric mucosal blood flow in water-immersion stress mice at a dose equivalent to that used to prevent the development of ulcers.

These results suggest that the antiulcer effects of BTM-1086 are mainly due to the inhibition of gastric acid secretion and to the increase of gastric mucosal blood flow.

Keywords——(−)-cis-2,3-dihydro-3-(4-methylpiperazinylmethyl)-2-phenyl-1,5-benzothiazepin-4-(5H)-one hydrochloride (BTM-1086); antiulcerogenic activity; gastric secretion; gastric mucosal blood flow

(−)-cis-2,3-Dihydro-3-(4-methylpiperazinylmethyl)-2-phenyl-1,5-benzothiazepin-4-(5H)-one hydrochloride (BTM-1086), a synthetic derivative of benzothiazepin,[1−3] was shown to prevent the formation of acute gastric ulcers and to accelerate the healing of chronic ulcers produced by acetic acid.[4] Although this compound has been shown by Takayanagi *et al.*[5] to inhibit acetylcholine liberation from the parasympathetic nerve endings, little is known about the mechanism of its anti-ulcerogenic actions.

In the present study, we investigated the effect of BTM-1086 on stress-induced ulcers and pylorus-ligation ulcers. The gastric secretion and the gastric local blood flow were determined in rats and mice by using the hydrogen gas clearance method, in order to elucidate the mechanism of the antiulcer effects.

Materials and Methods

BTM-1086 has the chemical structure shown in Fig. 1 (M_r 403.97), and occurs as white crystals that are easily soluble in water.

Other drugs used were atropine sulfate (Wako Pure Chemical Ind., Ltd.), tetragastrin (Nissui Seiyaku Co.), carbachol (Tokyo Kasei Co.), bethanechol chloride (Yoshitomi Pharm. Ind., Ltd.), histamine dihydrochloride (Wako) and insulin (Shimizu Pharm. Co.).

Stress Ulcer——1) Stress-Induced Ulcer in Mice: The stress ulcers were produced following the method described by Watanabe *et al.*[6] Male ddY mice weighing about 20 g were fasted for about 18 h before being immersed in water. Drugs were administered orally or subcutaneously 30 min before the stress. These mice were placed in a
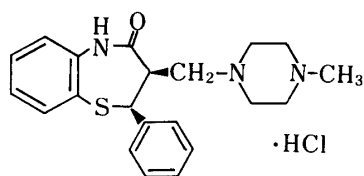
Fig. 1. Chemical Structure of BTM-1086

stress cage and immersed in a water bath (15 °C) to the depth of the xiphoid. At the end of the stress, the animal was sacrificed by dislocation of the cervix, then the stomach was removed, inflated with 1% formalin solution and placed in the same solution for 5 min. The stomach was cut open along the greater curvature and examined grossly for lesions in the glandular portion. The "Ulcer Index" was calculated as the sum of the length (mm) of each lesion in the stomach. Inhibition ratio was calculated as follows;

$$\text{inhibition ratio (\%)} = \frac{\text{ulcer index (control)} - \text{ulcer index (drug)}}{\text{ulcer index (control)}} \times 100$$

2) Stress-Induced Ulcer in Rats: The stress ulcers were produced following the method described by Takagi et al.[7] and Takagi and Okabe.[8] Male Wistar rats weighing 200 to 250 g were fasted for 24 h before being immersed in water. Each drug was given orally 30 min before the stress. These rats were fixed by the limbs on wire-netting and immersed to the depth of the xiphoid in a water bath at 23 °C. The rats were autopsied after 7 h of the stress. The stomach of each rat was removed, inflated by injecting 7.5 ml of 1% formalin solution, and immersed in 1% formalin solution for 20 min to fix the inner and outer layers of the gastric walls. The stomach was then incised along the greater curvature and examined for erosions developed in the glandular portion. The "Ulcer Index" was calculated as the sum of the length (mm) of each lesion in the stomach.

Pyloric Ligation Ulcer: Male Wistar rats weighing about 200 g were fasted for 48 h but provided with water ad libitum. The pylorus was ligated according to the method of Shay et al.,[9] and 5 ml/kg of a test drug dissolved or suspended in 0.5% Gum Arabic solution was given p.o. immediately after ligation. Eighteen hours later, the rats were sacrificed and the stomachs were removed. The severity of the gastric ulcers was scored according to an arbitrary scale and expressed as an "Ulcer Index". 0 = no lesion, 1 = one to three small ulcers (3 mm or smaller), 2 = more than three small ulcers or one large ulcer, 3 = one large and several small ulcers, 4 = several large ulcers, and 5 = perforated ulcers.

Gastric Secretion in Rats——Gastric Secretion in Pylorus-Ligated Rats: Male Wistar rats weighing about 200 g, fasted for 24 h, were anesthetized with ethyl ether and then pylorus-ligated. Drugs were dissolved in saline and administered s.c. immediately after pylorus-ligation. Six hours after the pylorus-ligation, the inner contents of the stomach were collected and the volume of gastric juice, the acidity and the peptic activity were examined. The acidity was determined by the titration with 0.02 N NaOH using phenolphthalein and dimethyl yellow as indicators. Peptic activity was determined according to the method described by Anson.[10] Bovine hemoglobin (Wako reagent) was used as the substrate, and 1 unit of enzyme activity was defined as the amount liberating 1 mg of tyrosine per 1 ml of gastric juice per hour.

Acute Gastric Fistula Rats: Male Wistar rats weighing 150 to 200 g were used. After fasting for 18 h, the animals were anesthetized with urethane (1.2 g/kg, i.p.). After ligation of the esophagus and pylorus, a double tube cannula was inserted into the forestomach. The stomach was perfused with warm saline solution (5 ml) through the cannula at intervals of 30 min. The perfusate was titrated for acid content with 0.01 N NaOH solution using phenolphthalein as an indicator. The acid secretory rate was expressed in terms of μeq of HCl per 30 min. The basal secretory responses were measured for 60 min, and then the test drugs or saline were administered 30 min before the injection of each secretagogue.

Acid Secretion in Schild's Rats: The methods of Ghosh and Schild[11] and Sawada et al.[12] were used for continuous recording of acid gastric secretion in the rat. Male Wistar rats weighing 250 to 300 g were used. Briefly, this method is based on perfusion of the stomach of the rat anesthetized with urethane (1.2 g/kg, i.p.), with graphical recording of the pH of the emerging fluid. All drugs were administered intravenously, usually by single injections, in a volume of 0.2 to 0.4 ml. Agonists were usually injected immediately after the antagonists. The following drugs were used: tetragastrin (2 μg/kg i.v. or 5 and 10 μg/kg/h i.v.), carbachol (2 μg/kg i.v. or 2 μg/kg/h i.v.) and histamine dihydrochloride (1 mg/kg i.v.).

Gastric Mucosal Blood Flow in Rats and Mice——1) Normal Rats: Gastric mucosal blood flow was measured according to the method of Stosseck et al.[13] and Koshu et al.[14,15] Male Wistar rats weighing 250 to 350 g were fasted for 15 h, but allowed free access to water. Under anesthesia with urethane (1.2 g/kg i.p.) the animals were fixed in a supine position and the body temperature was kept around 37 °C with a heating lamp, then the animals were laparotomized. Needle-type platinum electrodes (0.2 mm diameter, 30 mm length), connected to a blood flow meter (electrochemically generated hydrogen gas clearance, type RBF-2, Bio Medical Science Co., Ltd.), were advanced into and out of the serosa through the gastric stomach wall along the mucosal layer without penetrating into the

lumen, and were positioned in the antral and corporal areas. The calomel-electrode (10 mm diameter) was introduced subcutaneously through an incision in the femoral wall. Local blood flow was obtained from the hydrogen gas clearance curve of the tissue. Blood flow was calculated according to the theoretical formula for Kety's[16,17] tissue clearance, based on Fich's principle.

$$F \text{ (ml/min/100 g tissue)} = \frac{0.693}{T1/2}$$

During the determination of blood flow, blood pressure recorded from the carotid artery with an electronic manometer (Nihon Kohden RP-5A) did not vary significantly. For intravenous administration, drugs were dissolved in saline and infused through a canula inserted into the femoral vein at doses of 10 and 50 μg/kg.

2) Indomethacin-Induced Ischemia in Rats: Male Wistar rats weighing 220 to 290 g were studied. The animals were anesthetized with urethan (1.0 g/kg i.p.) then laparotomized, and a needle-type platinum electrode (0.2 mm diameter, 30 mm length) was inserted carefully into the mucosal layer of the anterior wall of the corpus. For measurement of gastric mucosal blood flow, the electrochemically generated hydrogen gas clearance instrument (type RBF-2) was used. Indomethacin (20 mg/kg) was injected into the forestomach and a test drug or saline was administered subcutaneously 15 min before the injection of indomethacin.

3) Ischemia Induced by Water-Immersion Stress in Mice: Male ddY mice weighing about 20 g were used. These mice were fasted but allowed water for about 15—16 h before experiment. Mice were restrained in plastic capsules with holes and then immersed in a water bath (15 °C) for 3 h. The test drugs were administered orally 30 min before the stress. After the restraint, these mice were anesthetized with urethane (1 g/kg i.p.), then the animals were laparotomized and the needle-type platinum electrode was inserted into the gastric mucosa in the corpus and the gastric blood flow was measured by the hydrogen gas clearance method as described above. After measurement of gastric blood flow, the stomach of each mouse was removed, inflated by injecting 1 ml of 1% formalin solution and examined grossly for lesions in the glandular portion. The "Ulcer Index" was calculated as the sum of the length (mm) of each lesion in the stomach.

Erosion and Analysis of Data——The gastric erosions induced by water-immersion stress and the inner surface of damaged mucosa in pylorus-ligation ulcers were examined with a stereoscopic microscope (10 ×). The person measuring the lesions had no knowledge of which treatment an animal had received.

Results are shown as the mean ± standard error. Statistical significance was determined by using Student's t-test. ED_{50} were determined from a plot of log dose against percentage inhibition compared with a control group.

Results

Acute Gastric Ulcers

Water-Immersion Stress Ulcer——1) Stress Ulcers in Mice: As shown in Table I, BTM-1086 at doses of 0.05, 0.2 and 1 mg/kg p.o. showed significant inhibitory effects on the ulcer index; the inhibition ratios were 59.3% ($p < 0.01$), 80.6% ($p < 0.01$) and 91.7% ($p < 0.001$), respectively. The ED_{50} value of BTM-1086 by the oral route was estimated to be 0.02 mg/kg from the dose–response curve of the inhibition rate. Atropine sulfate at 10 mg/kg p.o. also exhibited a significant inhibitory effect; 89.1% ($p < 0.001$).

2) Stress Ulcer in Rats: BTM-1086 at doses of 0.2, 1, 5 and 10 mg/kg p.o. showed

TABLE I. Effects of BTM-1086 and Atropine Sulfate on Restraint and Water-Immersion Stress-Induced Ulcers in Mice

Treatment	Dose mg/kg p.o.	No. of mice	Ulcer index (mm) mean ± S.E.	Inhibition (%)
Control (Vehicle)	—	5	49.6 ± 2.9	—
BTM-1086	0.05	5	20.2 ± 7.7[a]	59.3
	0.2	5	9.6 ± 4.4[b]	80.6
	1	5	4.1 ± 1.7[b]	91.7
Atropine sulfate	10	5	5.4 ± 1.9[b]	89.1

Drugs were administered p.o. 30 min before restraint and water-immersion stress (15 °C, 5 h). a) Significantly different from the control, $p < 0.01$. b) Significantly different from the control, $p < 0.001$.

TABLE II. Effects of BTM-1086 and Atropine Sulfate on Restraint and
Water-Immersion Stress-Induced Ulcers in Rats

Treatment	Dose mg/kg p.o.	No. of rats	Ulcer index (mm) mean ± S.E.	Inhibition (%)
Control (Vehicle)	—	6	127.7 ± 18.1	—
BTM-1086	0.2	6	78.2 ± 6.6[a]	38.8
	1	6	42.1 ± 8.4[b]	67.0
	5	6	8.8 ± 4.8[c]	93.1
	10	6	6.8 ± 2.7[c]	94.7
Atropine sulfate	1	6	96.0 ± 4.9	24.8
	5	6	37.1 ± 7.7[c]	70.9

Drugs were administered p.o. 30 min before restraint and water-immersion stress (23 °C, 7 h). a) Significantly different from the control, $p < 0.05$. b) Significantly different from the control, $p < 0.01$. c) Significantly different from the control, $p < 0.001$.

TABLE III. Effects of BTM-1086 and Atropine Sulfate on Gastric Ulceration
Owing to Pylorus-Ligation in Rats

Treatment	Dose mg/kg p.o.	No. of rats	Ulcer index mean ± S.E.	Inhibition (%)
Control (Vehicle)	—	7	4.0 ± 0.5	—
BTM-1086	1	6	2.7 ± 0.7	32.5
	5	5	1.6 ± 0.6[a]	60.0
	10	6	0.8 ± 0.5[b]	80.0
	20	5	0.2 ± 0.2[c]	95.0
Atropine sulfate	20	5	1.2 ± 0.7[b]	70.0

Drugs were administered p.o. immediately after pylorus-ligation. a) Significantly different from the control, $p < 0.05$. b) Significantly different from the control, $p < 0.01$. c) Significantly different from the control, $p < 0.001$.
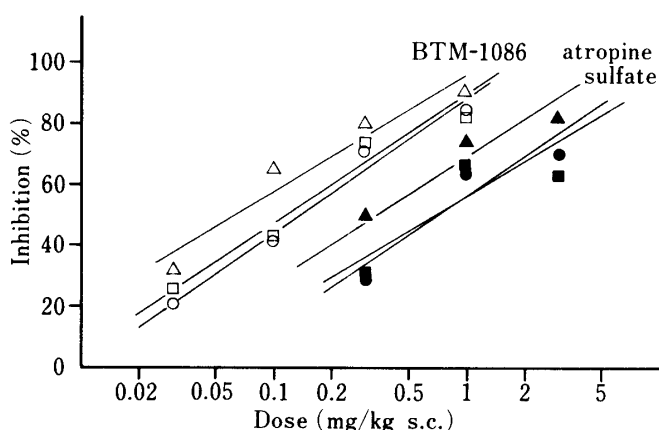

Fig. 2. Dose–Response Relationships for the Effect of BTM-1086 on the Gastric Juice Secretion, Acid Output and Pepsin Activity in Pylorus-Ligated (6 h) Rats

Each point represents the mean of 9—10 experiments.

○, ●, volume; □, ■, acid output; △, ▲, pepsin activity.

significant inhibitory effects on ulcer formation: the inhibition ratios were 38.8% ($p < 0.05$), 67.0% ($p < 0.01$), 93.1% ($p < 0.001$) and 94.7% ($p < 0.001$), respectively. The ED_{50} value of BTM-1086 was estimated to be 0.27 mg/kg p.o. Atropine sulfate (5 mg/kg p.o.) also exhibited a significant inhibitory effect (Table II).

Pyloric Ligation Ulcer——As shown in Table III, BTM-1086 at doses of 5, 10 and 20 mg/kg p.o. showed significant inhibitory effects; 60.0% ($p < 0.05$), 80.0% ($p < 0.01$) and
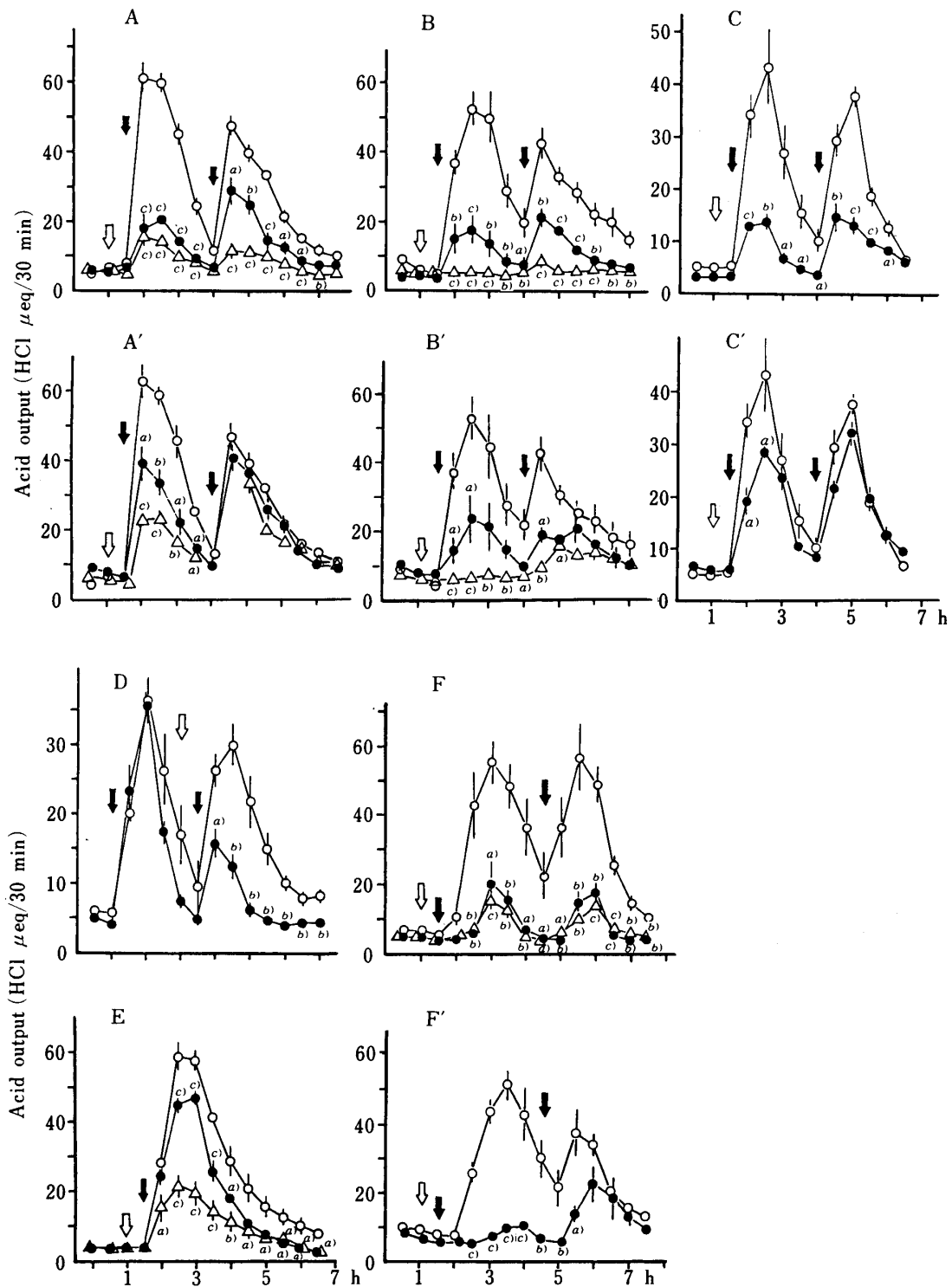
Fig. 3. Inhibitory Effect of BTM-1086 on Gastric Acid Secretion Stimulated by Various Secretagogues in Anesthetized Rats with Acute Fistula

Tetragastrin (A, A'), bethanechol (B, B'), histamine-2HCl (C, C', D), carbachol (E) or insulin (F, F') was injected at the black arrow at the dose of 300, 1000, 5000, 100 μg/kg or 1 U/kg, respectively.

A :   At the white arrow, saline (○—○ 1 ml/kg) or BTM-1086 (●—● 5, △—△ 10 mg/kg) was administered subcutaneously 30 min before the first stimulation.

A' :   At the white arrow, saline (○—○ 1 ml/kg) or atropine sulfate (●—● 5, △—△ 10 mg/kg) was administered subcutaneously 30 min before the first stimulation.

B :   At the white arrow, saline (○—○ 1 ml/kg) or BTM-1086 (●—● 0.1, △—△ 0.3 mg/kg) was administered intraperitoneally 30 min before the first stimulation.

95.0% ($p < 0.001$) inhibition, respectively. The ED_{50} value of BTM-1086 was estimated to be 2.80 mg/kg p.o. Atropine sulfate at 20 mg/kg p.o. also exhibited a significant inhibition: 70.0% ($p < 0.01$).

Gastric Secretion in Rats

Gastric Secretion in Pylorus-Ligated Rats——The volume of gastric secretion, the acid output and the pepsin activity were measured (Fig. 2). The treatment with BTM-1086 (0.03— 1 mg/kg s.c.) or atropine sulfate (0.3—3 mg/kg s.c.) significantly decreased the gastric secretion volume, acid output and pepsin activity, but did not affect the pH of the gastric juice. The ED_{50} values for decreasing the volume were 0.13 and 0.68 mg/kg for BTM-1086 and atropine sulfate, respectively. For decreasing the acid output, similar ED_{50} values were obtained. However, the ED_{50} values for decreasing pepsin activity were 0.06 and 0.34 mg/kg BTM-1086 and atropine sulfate, respectively, which were 2-fold lower than those for the secretion volume and acid output.

Acute Gastric Fistula Rats——The administration of tetragastrin (300 μg/kg s.c.), bethanechol (1 mg/kg s.c.), histamine hydrochloride (5 mg/kg s.c.), carbachol (100 μg/kg s.c.) and insulin (1 U/kg i.v.) resulted in gastric hypersecretion in acute fistula rats. Gastric secretion patterns are shown in Fig. 3 (A, A', B, B', C, C', D, E, F, F'). In these experiments, the test drug was administered subcutaneously or intraperitoneally 30 min before the stimulation. BTM-1086 at a dose of 5 mg/kg significantly inhibited the tetragastrin-induced increase in gastric acid secretion (Fig. 3A). BTM-1086 at a dose of 0.3 mg/kg completely blocked the bethanechol-induced hypersecretion of acid (Fig. 3B). Similarly, carbachol-stimulated secretion (Fig. 3E), histamine-stimulated secretion (Fig. 3C, D) and insulin-stimulated secretion (Fig. 3F) were all reduced by BTM-1086. Atropine sulfate was also found to suppress the effects of all 4 secretagogues (Fig. 3A', B', C', F').

Acid Secretion in Schild's Rats——The intravenous administration of tetragastrin (2 μg/kg i.v. or 5 or 10 μg/kg/h i.v.) and carbachol (2 μg/kg i.v. or 2 μg/kg/h i.v.) resulted in gastric hypersecretion in anesthetized Schild's rats. The treatment with BTM-1086 (10, 30, 50, and 100 μg/kg i.v.) antagonized the action of these drugs dose-dependently by decreasing the acid secretion. Typical results of such experiments are shown in Fig. 4-a, A, Fig. 4-b, A, or Fig. 4-c. The ED_{50} value for the inhibition of tetragastrin-stimulated acid secretion was 40 μg/kg i.v., while that for the inhibition of carbachol-stimulated acid secretion was 24 μg/kg i.v., and that for the inhibition of histamine-stimulated acid secretion was ca. 3 mg/kg i.v. Acid secretion stimulated submaximally by continuous i.v. infusion of tetragastrin (5 or 10 μg/kg/h) or carbachol was also inhibited by i.v. BTM-1086 (100 μg/kg) (Fig. 4-a, B and Fig. 4-b, B).

Gastric Mucosal Blood Flow in Rats and Mice

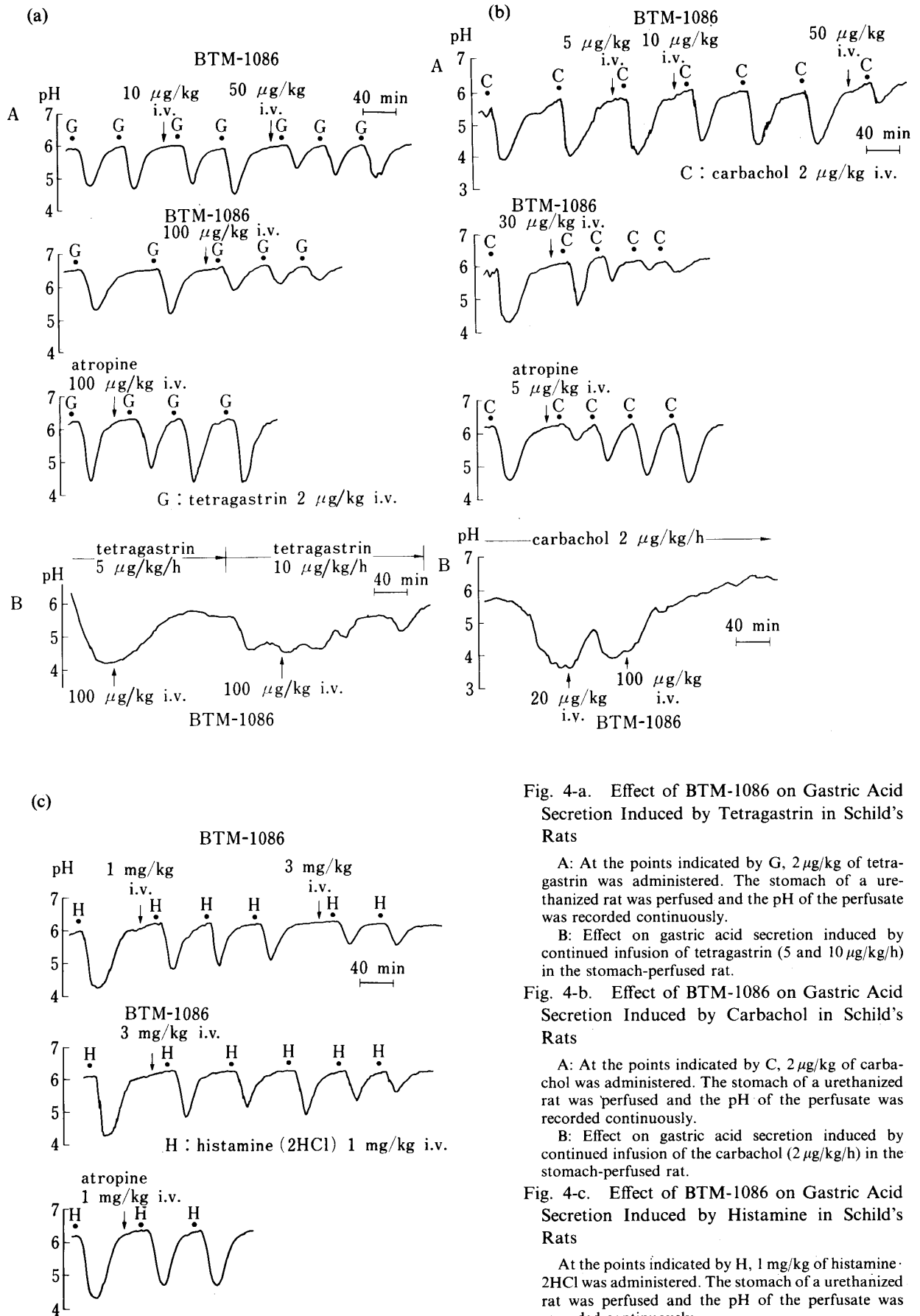1) **Normal Rats**——BTM-1086 at doses of 10 and 50 μg/kg i.v. increased the gastric

(a)

(b)

(c)

Fig. 4-a. Effect of BTM-1086 on Gastric Acid Secretion Induced by Tetragastrin in Schild's Rats

A: At the points indicated by G, 2 µg/kg of tetragastrin was administered. The stomach of a urethanized rat was perfused and the pH of the perfusate was recorded continuously.

B: Effect on gastric acid secretion induced by continued infusion of tetragastrin (5 and 10 µg/kg/h) in the stomach-perfused rat.

Fig. 4-b. Effect of BTM-1086 on Gastric Acid Secretion Induced by Carbachol in Schild's Rats

A: At the points indicated by C, 2 µg/kg of carbachol was administered. The stomach of a urethanized rat was perfused and the pH of the perfusate was recorded continuously.

B: Effect on gastric acid secretion induced by continued infusion of the carbachol (2 µg/kg/h) in the stomach-perfused rat.

Fig. 4-c. Effect of BTM-1086 on Gastric Acid Secretion Induced by Histamine in Schild's Rats

At the points indicated by H, 1 mg/kg of histamine·2HCl was administered. The stomach of a urethanized rat was perfused and the pH of the perfusate was recorded continuously.
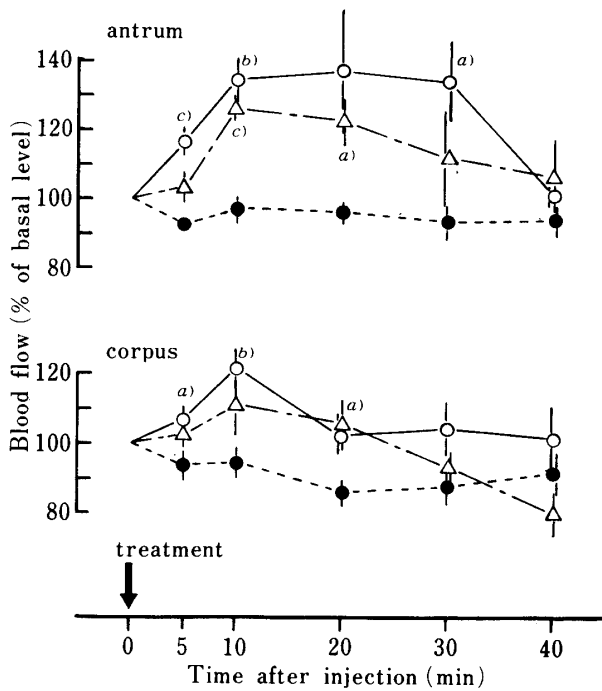
Fig. 5.   Effect of BTM-1086 on Gastric Muco-
sal Blood Flow in Anesthetized Rats

Platinum electrodes were inserted into the mucosal
layer of the antral and corporal areas. Gastric blood
flow was determined by the H_2-gas clearance method.
Each point represents the mean $\pm$ S.E. $n =$ Number
of observations. a) Significantly different from the
control, $p < 0.05$. b) Significantly different from the
control, $p < 0.01$. c) Significantly different from the
control, $p < 0.001$.
●---●, control ($n = 5$); △---△, BTM-1086 10
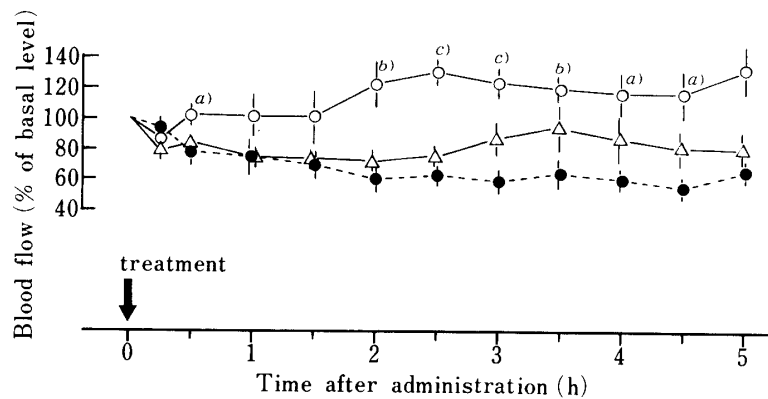$\mu g/kg$ i.v. ($n = 6$); ○—○, BTM-1086 50 $\mu g/kg$ i.v.
($n = 5$).

Fig. 6.   Effect of BTM-1086 on Gastric Mucosal Blood Flow in Anesthetized Rats

A platinum electrode was inserted into the mucosal layer of the corpus. Gastric blood
flow was determined by the H_2-gas clearance method. BTM-1086 was given subcutaneously
before the injection of indomethacin (20 mg/kg i.g.). Each point represents the mean $\pm$ S.E.
$n =$ number of observations. a) Significantly different from the control, $p < 0.05$. b)
Significantly different from the control, $p < 0.01$. c) Significantly different from the control,
$p < 0.001$.
●---●, saline + indomethacin ($n = 6$); △—△, BTM-1086 (0.1 mg/kg s.c.) + indomethacin
($n = 5$); ○—○, BTM-1086 (0.5 mg/kg s.c.) + indomethacin ($n = 5$).

mucosal blood flow both in the antrum and in the corpus (Fig. 5). The effect was dose-
dependent, exhibiting maximal increases of 27—36% and 12—21% at 10 and 50 $\mu g/kg$,
respectively.

2) **Indomethacin-Induced Ischemia in Rats**——The administration of indomethacin
caused a decrease in the regional blood flow for several hours. The administration of BTM-
1086 (0.1 mg/kg s.c.) plus indomethacin caused a similar decrease for 2 h. After 2 to 3 h,
however, the blood flow recovered gradually to the initial level in the BTM-1086 administered
group (Fig. 6). At a higher dose of BTM-1086 (0.5 mg/kg s.c.), the decreasing effect of
indomethacin was completely overcome and little change in mucosal blood flow was observed
over 2 h. After 2 h, the blood flow in this group was significantly higher than that in the

TABLE IV. Effects of BTM-1086 and Atropine Sulfate on Gastric Mucosal Blood Flow
and Ulceration in the Mouse after Restraint and Water-Immersion Stress

Treatment	Dose mg/kg p.o.	No. of mice	Gastric blood flow (ml/min/100 g) mean ± S.E.	Gastric ulcer Ulcer index (mm) mean ± S.E.	Inhibition (%)
Non treated	—	10	161.4 ± 15.0	0	—
Control	—	6	66.7 ± 8.4	46.1 ± 2.9	—
BTM-1086	0.2	6	79.7 ± 3.3	42.5 ± 5.3	7.9
Control	—	5	67.2 ± 9.8	67.3 ± 3.6	—
BTM-1086	1.0	5	$152.6 \pm 9.1^{a)}$	$18.3 \pm 5.0^{a)}$	72.8
Control	—	5	64.6 ± 4.9	53.9 ± 5.6	—
Atropine sulfate	1.0	5	69.3 ± 9.9	36.8 ± 5.3	31.7
	10.0	5	56.5 ± 2.5	$6.3 \pm 5.0^{a)}$	88.3

a) Significantly different from the control, $p < 0.001$.

control group.

3) Ischemia Induced by Water-Immersion Stress in Mice——The restraint and water-immersion stress decreased the gastric blood flow by approximately 60% (Table IV). There was a significant, dose-related recovery of mucosal blood flow in the corpus ventriculi of mice following administration of BTM-1086 at 1 mg/kg p.o. ($p < 0.01$). The incidence of gastro-duodenal ulceration was slightly reduced in these treated mice (7.9%) at 0.2 mg/kg and significantly reduced (by 72.9%) at 1 mg/kg ($p < 0.001$). Thus, the administration of BTM-1086 caused a dose-related increase in the regional blood flow and suppression of ulceration. Atropine sulfate at 10 mg/kg inhibited the ulceration, but the gastric blood flow was little changed.

Discussion

The newly synthesized compound, BTM-1086, was highly active in preventing gastric lesions induced by restraint and water-immersion stress and pylorus-ligation. It is known that the gastric juice plays an important role in the pathogenesis of stress ulcers. Since BTM-1086 inhibited the pylorus-ligation ulceration, which is known to be caused by gastric secretion of acid and pepsin, it may be postulated that the antiulcer effect of BTM-1086 is related to the suppression of gastric secretion. In fact, BTM-1086 markedly reduced dose-dependently the volume, acid and pepsin activity of spontaneous gastric secretion in pylorus-ligated rats.

To clarify the mechanism of the antisecretory effects, the compound was tested against gastric hypersecretions induced by tetragastrin, choline ester, histamine and insulin, which are considered to play major roles in the physiological gastric secretion. BTM-1086 inhibited the gastric acid output induced by all secretagogues (peripherally and centrally mediated secretagogues). In this study, we observed the selective action of BTM-1086, inhibiting the gastric output induced by peripheral site-stimulating secretagogues (gastrin, bethanechol, and carbachol) more than that induced by a vagus nerve-stimulating secretagogue (insulin).

BTM-1086 increased the gastric mucosal blood flow in normal rats, indomethacin-induced ischemic rats and water-immersion stress mice at a dose equivalent to that able to prevent the development of ulcers. As regards the role of gastric acid as an aggressive factor in stress ulcer, greater importance has been attached recently to the diminution of regional blood flow, which is considered to be a protective factor.[18-20] It has been demonstrated by analysis of the gastric mucosal reflex spectrum that impairment of regional blood flow plays a part in

the pathogenesis of stress-induced lesions.[21] Thus, it is considered that the gastric mucosal resistance is also related to gastric blood flow.

It has been shown by Takayanagi and coworkers[5] that BTM-1086 inhibits acetylcholine liberation from the parasympathetic nerve endings. In connection with this result, we examined the effects of the compound on the actions of stimulators of gastric secretion (tetragastrin, choline ester, histamine and insulin), to investigate the mechanism by which the compound inhibits acid secretion. BTM-1086 produced a significant suppression of acid secretion stimulated by these drugs, the effect being particularly conspicuous against tetragastrin and carbachol. These findings seem to imply that the compound inhibits the gastrin secretion from the pyloric antrum and exerts an antipeptic action associated with the direct inhibition of acid secretion. In fact, it showed a marked suppression of acid secretion stimulated by exogenous gastrin.

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