ANTI-ULCER EFFECTS OF 4'-(2-CARBOXYETHYL) PHENYL TRANS-4-AMINOMETHYL CYCLOHEXANECARBOXYLATE HYDROCHLORIDE (CETRAXATE) ON VARIOUS EXPERIMENTAL GASTRIC ULCERS IN RATS

Yoshio SUZUKI, Mihoko HAYASHI, Mikio ITO
and Ichika YAMAGAMI

*Department of Pharmacology, Faculty of Pharmacy, Meijo University
Nagoya 468, Japan*

Accepted May 11, 1976

Abstract—Anti-ulcer effects of cetraxate, a new compound possessing anti-plasmin, anti-casein and anti-trypsin actions were investigated by using experimental gastric ulcer models in rats. Cetraxate, 300 mg/kg p.o. showed significant inhibitory effects of 65.3%, 70.0%, 30.2% and 67.1% against acute types of ulcers producing by aspirin, phenylbutazone, indomethacin, and pyloric ligation (Shay's ulcer), respectively. These effects were greater than those obtained by gefarnate and aluminum sucrose sulfate and may be mainly attributed to the protecting action of this drug on gastric mucosa. Cetraxate further revealed remarkable inhibitory effects on chronic types of ulcers produced by acetic acid, clamping, and clamping-cortisone. In acetic acid ulcer in particular, cetraxate was found to have a dose-dependent inhibitory effect at doses over 50 mg/kg. Of test drugs including L-glutamine and methylmethionine sulfonium chloride, cetraxate showed the most remarkable inhibitory effect on β -glucuronidase activity in ulcer tissue of these three types of ulcers. These findings suggest that cetraxate may prevent the connective tissue in the ulcer location from decomposition due to lysosomal enzymes such as β -glucuronidase, thereby accelerating the recovery from ulcer.

Cetraxate, 4'-(2-carboxyethyl)phenyl trans-4-aminomethyl cyclohexanecarboxylate hydrochloride (DV-1006), is chemically a *p*-hydroxyphenyl propionic ester of tranexamic acid (t-AMCHA) and a new compound developed by the collaboration of Muramatsu et al. (1) of Tokushima University and the research laboratory of Daiichi Seiyaku Co (2). This drug was proved *in vitro* to have more potent anti-plasmin action than t-AMCHA, and also anti-trypsin and anti-kinin actions which t-AMCHA does not possess. Beneficial effects on inflammatory, allergenic or hemorrhagic disorders can thus be assumed. We previously reported the effect of this drug on experimental nephritis in rats (3).

The aim of the present study was to determine the anti-ulcer effects of cetraxate by using various models of experimental gastric ulcers in rats.

MATERIALS AND METHODS

Cetraxate is an odorless compound of white crystals or crystalline powder which is soluble in water and has a slightly bitter taste. The chemical structure is shown in Fig. 1.

The following drugs were used for comparison: t-AMCHA (Daiichi Seiyaku), gefarnate

(gefanil, Sumitomo Kagaku), aluminum sucrose sulfate (ulcerlmin, Chugai Seiyaku), L-glutamine (Kyowa Hakko) and methylmethionine sulfonium chloride (Katayama Kagaku).

Other chemicals used were aspirin (J.P.), phenylbutazone (Fujisawa Yaku-hin), indomethacin (Merke-Banyu) and acetic acid (special grade, Katayama Kagaku).

Acute gastric ulcers

Drug induced ulcers (aspirin, phenylbutazone and indomethacin) (A): Groups of 5 female Wistar rats, weighing about 150 g were fasted for 20 hr but provided water *ad libitum*. Thereafter, 10 ml/kg of each test drug dissolved or suspended in 1% Gum Arabic solution was given orally. An hour later, gastric ulcers were induced by giving p.o. 300 mg/kg of aspirin, 200 mg/kg of phenylbutazone or 25 mg/kg of indomethacin. Food and water were withheld. Five hours after dosing with aspirin, phenylbutazone or indomethacin, the animals were sacrificed under ether anesthesia. Stomachs were removed and incised along the greater curvature. The number of ulcers was counted under a magnifying glass. Each ulcer was then measured with a caliper to assess the diameter. The extent of each ulcer was expressed as the sum of scores described in the following Shay rat ulcer and used as the ulcer index. The percent inhibition of the ulcer index with each test drug was also calculated.

Shay rat ulcer: Groups of 10 female Wistar rats, weighing about 180 g were fasted for 48 hr but provided water *ad libitum*. The pylorus was ligated according to the method of Shay et al. (4), and 10 ml/kg of each test drug dissolved or suspended in 1% Gum Arabic solution was given p.o. immediately. Eighteen hours later they were sacrificed and the stomachs removed. The number and diameter of ulcers were measured as in (A). The ulcer index was expressed as the sum of scores described below.

- Score 1: maximal diameter of 1 mm.
- Score 2: maximal diameter of 1–2 mm.
- Score 3: maximal diameter of 2–3 mm.
- Score 4: maximal diameter of 3–4 mm.
- Score 5: maximal diameter of 4–5 mm.
- Score 10: an ulcer over 5 mm in diameter.
- Score 25: a perforated ulcer.

In addition to the ulcer index, the volume, total acidity and peptic activity of gastric juice were also determined 18 hr after pylorus ligation. Total acidity was determined by titrating undiluted gastric juice centrifuged (5,000 g for 20 min) with 0.1 N NaOH using phenolphthalein as an indicator. Peptic activity was determined according the method described by Prino et al. (5). Bovine albumin (Katayama Kagaku) was used as substrate, and the unit of enzyme activity was expressed as μg tyrosine liberated per 0.1 ml of gastric juice

FIG. 1. Chemical structure of cetraxate.

for an hour. The percent inhibition of the ulcer index and the volume, total acidity and peptic activity of the gastric juice with each drug was calculated.

Chronic gastric ulcers

Acetic acid ulcer (A): The experiment was carried out according to the method of Takagi et al. (6). Groups of 8 female Wistar rats, weighing about 180 g were anesthetized with ether, and subjected to laparotomy to expose the stomach after which 0.05 ml of 10% acetic acid was injected carefully under the serous membrane of the abdominal side in the glandular stomach. The abdomen was closed after adequate disinfection with 1% acrinol solution. Thereafter, the animals were maintained under normal conditions and administered 10 ml/kg of each test drug dissolved or suspended in 1% Gum Arabic solution p.o. daily $\times 8$ starting from the day of the operation. On the 9th day, they were sacrificed under ether anesthesia, stomachs were removed and incised along the greater curvature. The longitudinal and abscissal lengths of the ulcer areas were quickly measured with a caliper, and the multiplied product was used as the ulcer index. In addition to the ulcer index, the activity of β -glucuronidase (β -Gase) in the gastric tissue was also determined in accordance with the following procedure: As soon as possible after the ulcer index was measured, the glandular stomach was weighed, 10 vol. of physiological saline solution was added and under a cold condition the tissue cut into pieces with scissors. The tissue was homogenized by using Polyton of PT 200 D type under cooling and the homogenate was then centrifuged at 12,000 r.p.m. at 4°C for 20 min. The supernatant was collected, filtrated and diluted with physiological saline solution to make an adequate concentration after which it was used for enzyme assay. β -Gase activity was determined in accordance with the method described in our earlier report (7). For the determination of this enzyme, *p*-nitrophenyl glucuronide (Chugai Seiyaku) was used as substrate. The unit of enzyme activity was expressed as μ g *p*-nitrophenol liberated per mg protein for one hour.

Clamping ulcer (B): The test was carried out in accordance with the method of Umehara et al. (8). Groups of 8 female Wistar rats, weighing about 200 g, were subjected to laparotomy under ether anesthesia following a fast of 24 hr. The stomachs were exposed, and clamping was done at the area 3 mm from the foregut boundary at the greater curvature side toward the pylorus by pressing with a two-fold aluminium plate 4 $\times$ 12 mm in size after two sheets of gauze had been placed. The abdomen was closed after disinfection with 1% acrinol solution. Twenty-four hours later the abdomen was again opened to remove the aluminium plate and gauze. A normal diet was provided to these postoperative animals. For supply of nutriment, 10 ml of 5% glucose solution was given s.c. for 3 consecutive days from the fasting day. Each test drug was dissolved or suspended in 1% Gum Arabic solution and 10 ml/kg p.o. daily was given for 14 days from the day of the removal of aluminium plate. On the 15th day, these animals were sacrificed under ether anesthesia. The stomachs were removed to determine the ulcer index and β -glucuronidase activity in the gastric tissue as in (A). The percent inhibition of the ulcer index and enzyme activity with each drug was calculated.

Clamping-cortisone ulcer (8): Clamping was made by the same procedure as described

in (B). A daily i.m. injection of 5 mg/kg of hydrocortisone was carried out for 7 days starting from the day of removal of aluminium plate. Each test drug was dissolved or suspended in 1% Gum Arabic solution and 10 ml/kg p.o. for 11 consecutive days was given starting the day after the last day of hydrocortisone injections. Twenty-four hours later these rats were sacrificed under ether anesthesia, and the stomachs were removed. The ulcer index and β -Gase activity in the gastric tissue were determined and categorized as in (A). The percent inhibition with each drug was calculated.

The results of the above experiments were statistically evaluated using Student's t-test.

RESULTS

Anti-ulcer effects of cetraxate on acute gastric ulcers

Inhibitory effect on aspirin ulcer: As shown in Table 1, the inhibitory effect of cetraxate on aspirin ulcer was not observed with a dose of 50 mg/kg p.o. while 300 mg/kg p.o. of the drug showed a significant inhibitory effect of 65.3% ($p < 0.01$). Gefarnate 100 mg/kg p.o. and aluminum sucrose sulfate 1,000 mg/kg p.o. also exhibited the inhibitory effect of 42.8% ($p < 0.05$) and 38.6% ($p < 0.01$), respectively.

TABLE 1. Inhibitory effects of cetraxate, gefarnate and aluminum sucrose sulfate on aspirin ulcer in rats

Drugs	Dose mg/kg, p.o.	Ulcer index mean $\pm$ S.E.	% inhibition
Control	(5)	56.0 $\pm$ 8.6	
Cetraxate	50 (5)	52.3 $\pm$ 6.6	6.4
	300 (5)	19.4 $\pm$ 2.8**	65.3
Gefarnate	100 (5)	32.0 $\pm$ 8.0*	42.8
Aluminum sucrose sulfate	1000 (5)	34.4 $\pm$ 1.4**	38.6

(), Number of rats. *, Significant difference from control ($p < 0.05$). **, Significant difference from control ($p < 0.01$).

Inhibitory effect on phenylbutazone ulcer: As shown in Table 2, cetraxate 300 mg/kg resulted in a remarkable inhibitory effect of 70.0% ($p < 0.01$) on phenylbutazone ulcer. Gefarnate 100 mg/kg p.o. and aluminum sucrose sulfate 1,000 mg/kg p.o. showed the inhibitory effects of 31.5% ($p < 0.01$) and 38.0% ($p < 0.01$), respectively.

TABLE 2. Inhibitory effects of cetraxate, gefarnate and aluminum sucrose sulfate on phenylbutazone ulcer in rats

Drugs	Dose mg/kg, p.o.	Ulcer index mean $\pm$ S.E.	% inhibition
Control	(5)	44.0 $\pm$ 2.8	
Cetraxate	50 (5)	40.5 $\pm$ 4.6	8.0
	300 (5)	13.2 $\pm$ 3.3**	70.0
Gefarnate	100 (5)	30.1 $\pm$ 1.4**	31.5
Aluminum sucrose sulfate	1000 (5)	27.3 $\pm$ 1.7**	38.1

(), Number of rats. **, Significant difference from control ($p < 0.01$).

Inhibitory effect on indomethacin ulcer: As shown in Table 3, cetraxate at a dose of 50 mg/kg p.o. had no inhibitory effect on indomethacin ulcer as in other types of drug ulcers, but at a dose of 300 mg/kg p.o., there was a significant inhibitory effect of 30.2% ($p < 0.01$). Gefarnate 100 mg/kg p.o. and aluminum sucrose sulfate 1,000 mg/kg p.o. also exhibited inhibitory effects of 29.0% ($p < 0.05$) and 40.0% ($p < 0.01$), respectively.

TABLE 3. Inhibitory effects of cetraxate, gefarnate and aluminum sucrose sulfate on indomethacin ulcer in rats

Drugs	Dose mg/kg, p.o.	Ulcer index mean $\pm$ S.E.	% inhibition
Control	(5)	29.0 $\pm$ 2.0	
Cetraxate	50 (5)	27.6 $\pm$ 2.7	4.8
	300 (5)	20.2 $\pm$ 1.4**	30.2
Gefarnate	100 (5)	20.6 $\pm$ 5.1*	29.0
Aluminum sucrose sulfate	1000 (5)	17.4 $\pm$ 0.9**	40.0

(), Number of rats. *, Significant difference from control ($p < 0.05$), **, Significant difference from control ($p < 0.01$).

Inhibitory effect on Shay rat ulcer: As shown in Table 4, cetraxate at doses of 50 and 300 mg/kg p.o. showed significant inhibitory effects of 35.0% ($p < 0.05$) and 67.1% ($p < 0.001$) on ulcer index, respectively. Gefarnate 100 mg/kg p.o. and aluminum sucrose sulfate 1,000 mg/kg p.o. also exhibited a strong inhibitory effect over 80% ($p < 0.001$). In contrast to the effect, cetraxate 50 mg/kg or 300 mg/kg p.o. did not affect either the volume or total acidity of gastric juice, but at a dose of 300 mg/kg p.o. there was a slight inhibitory effect of 17.0% ($p < 0.05$) on peptic activity. Gefarnate 100 mg/kg p.o. failed to inhibit these three parameters in gastric juice. However, aluminum sucrose sulfate at a dose of 1,000 mg/kg p.o. had strong inhibitory effects of 65.0% ($p < 0.001$) and 77.9% ($p < 0.001$), respectively, on both the total acidity and peptic activity of gastric juice, although no effect was seen on gastric secretion.

Anti-ulcer effects of cetraxate on chronic gastric ulcers

Inhibitory effects on ulcer index and β -Gase activity in ulcer tissue of acetic acid ulcer: Following daily oral administrations for 8 successive days, cetraxate at doses of 50 mg/kg, 100 mg/kg and 300 mg/kg a day showed inhibitory effects of 22.7%, 47.3% ($p < 0.05$) and 63.7% ($p < 0.01$) on ulcer index, respectively (Table 5). This drug at doses of 100 mg/kg and 300 mg/kg daily brought about significant inhibitory effects of 21.4% ($p < 0.05$) and 41.5% ($p < 0.01$) against β -Gase activity, respectively. Of drugs used for a comparison, t-AMCHA 500 mg/kg/day showed no inhibitory effects on either the ulcer index or β -Gase activity, while gefarnate 200 mg/kg/day showed a significant inhibition of 22.1% ($p < 0.05$) against β -Gase activity. Aluminum sucrose sulfate 1,000 mg/kg/day and L-glutamine 500 mg/kg/day exhibited inhibitions of 47.3% ($p < 0.05$) and 56.6% ($p < 0.01$), respectively, on ulcer index, while only L-glutamine showed a significant inhibition of 18.9% ($p < 0.05$) on β -Gase activity. Methylmethionine sulfonium chloride 50 mg/kg/day caused an inhibition of 49.1% ($p < 0.01$) on ulcer index and 17.3% ($p < 0.05$) on β -Gase activity.

TABLE 4. Inhibitory effects of cetraxate, gefarnate and aluminum sucrose sulfate on shay rat ulcer

Drugs	Dose mg/kg, p.o.	Stomach wall			Gastric juice		
		Ulcer index mean $\pm$ S.E.	% inhibition	Volume ml mean $\pm$ S.E.	% inhibition	Total acidity meq/l mean $\pm$ S.E.	Peptic activity μ g tyrosine/ 0.1 ml/hr mean $\pm$ S.E.
Control	(10)	67.8 $\pm$ 7.8		9.6 $\pm$ 0.7		82.5 $\pm$ 3.5	588.2 $\pm$ 22.1
Cetraxate	50 (10)	44.1 $\pm$ 6.3*	35.0	9.0 $\pm$ 0.5	6.3	88.2 $\pm$ 7.1	527.7 $\pm$ 23.3
	300 (10)	22.3 $\pm$ 3.6***	67.1	9.4 $\pm$ 0.9	2.1	86.2 $\pm$ 5.0	488.5 $\pm$ 29.1*
Gefarnate	100 (10)	9.0 $\pm$ 2.5***	86.7	10.6 $\pm$ 1.1	-10.4	87.6 $\pm$ 3.5	527.0 $\pm$ 41.5
Aluminum sucrose sulfate	1000 (10)	10.1 $\pm$ 1.9***	85.1	10.2 $\pm$ 1.1	-6.3	28.8 $\pm$ 3.9***	130.1 $\pm$ 26.3***
							77.9

(), Number of rats. *, Significant difference from control ($p < 0.05$).***, Significant difference from control ($p < 0.001$).

TABLE 5. Inhibitory effects of cetraxate and several drugs on ulcer index and β -glucuronidase activity in ulcer tissue of acetic acid ulcer in rats

Drugs	Dose mg/kg, p.o. $\times 8$ days	Ulcer index		β -glucuronidase activity	
		Index mean $\pm$ S.E.	% healing rate	μ g <i>p</i> -nitrophenol /mg protein/hr mean $\pm$ S.E.	% inhibition
Control	(8)	25.1 $\pm$ 8.5		38.1 $\pm$ 3.7	
Cetraxate	50 (8)	19.4 $\pm$ 10.3	22.7	32.5 $\pm$ 3.8	14.7
Control	(8)	27.0 $\pm$ 3.4		27.7 $\pm$ 2.3	
Cetraxate	100 (8)	14.2 $\pm$ 1.8*	47.3	21.9 $\pm$ 1.7*	21.4
Control	(8)	25.1 $\pm$ 8.5		38.1 $\pm$ 3.7	
Cetraxate	300 (8)	9.1 $\pm$ 0.1**	63.7	22.3 $\pm$ 3.5**	41.5
Control	(8)	25.7 $\pm$ 2.9		34.2 $\pm$ 2.6	
Gefarnate	100 (8)	22.9 $\pm$ 6.8	10.5	33.9 $\pm$ 4.1	0.8
Control	(8)	23.9 $\pm$ 9.4		27.2 $\pm$ 3.4	
Gefarnate	200 (8)	17.4 $\pm$ 8.1	27.1	21.2 $\pm$ 1.3*	22.1
Control	(8)	28.5 $\pm$ 2.1		33.4 $\pm$ 4.5	
Aluminum sucrose sulfate	1000 (8)	15.0 $\pm$ 0.4*	47.3	30.2 $\pm$ 5.5	9.6
Control	(8)	25.1 $\pm$ 8.5		38.1 $\pm$ 3.7	
L-glutamine	500 (8)	10.9 $\pm$ 0.2**	56.6	30.9 $\pm$ 1.3*	18.9
Control	(8)	27.0 $\pm$ 3.4		27.8 $\pm$ 2.3	
Methylmethionine sulfonium chloride	50 (8)	13.7 $\pm$ 2.6**	49.1	23.0 $\pm$ 1.5*	17.3
Control	(8)	27.0 $\pm$ 3.4		27.8 $\pm$ 2.3	
t-AMCHA	500 (8)	23.6 $\pm$ 2.4	12.7	24.6 $\pm$ 2.1	11.7

(), Number of rats. *, Significant difference from control ($p < 0.05$). **, Significant difference from control ($p < 0.01$).

Inhibitory effects on ulcer index and β -Gase activity in ulcer tissue of clamping ulcer: Following daily oral administrations for 14 consecutive days, cetraxate 200 mg/kg/day caused inhibitory effects of 63.8% ($p < 0.05$) on ulcer index and 35.9% ($p < 0.05$) on β -Gase activity, respectively (Table 6). The daily administrations of gefarnate 100 mg/kg and L-glutamine 500 mg/kg induced significant inhibitory effects of 67.3% ($p < 0.05$) and 44.3% ($p < 0.05$) on ulcer index, respectively. However, aluminum sucrose sulfate 1,000 mg/kg/day and methylmethionine sulfonium chloride 50 mg/kg/day had no significant inhibition. Against β -Gase activity gefarnate 100 mg/kg/day, L-glutamine 500 mg/kg/day and methylmethionine sulfonium chloride 50 mg/kg/day brought about significant inhibitions of 13.2% ($p < 0.05$), 14.7% ($p < 0.05$) and 26.6% ($p < 0.05$), respectively. Only aluminum sucrose sulfate 1,000 mg/kg/day increased this enzyme activity by 48.5% ($p < 0.01$).

Inhibitory effects on ulcer index and β -Gase activity in ulcer tissue of clamping-cortisone ulcer: Following daily oral administrations for 12 consecutive days, cetraxate 200 mg/kg/day showed inhibitory effects of 37.6% ($p < 0.05$) on ulcer index and 32.4% ($p < 0.01$) on β -Gase activity, respectively (Table 7). Among compared drugs, only L-glutamine 500

TABLE 6. Inhibitory effects of cetraxate and several drugs on ulcer index and β -glucuronidase activity in ulcer tissue of clamping ulcer in rats

Drugs	Dose mg/kg, p.o. $\times 14$ days	Ulcer index		β -glucuronidase activity	
		Index mean $\pm$ S.E.	% healing rate	μ g <i>p</i> -nitrophenol /mg protein/hr mean $\pm$ S.E.	% inhibition
Control	(8)	50.0 $\pm$ 15.6		38.7 $\pm$ 6.0	
Cetraxate	200 (8)	18.1 $\pm$ 3.1*	63.8	24.8 $\pm$ 1.2*	35.9
Control	(8)	50.0 $\pm$ 15.6		38.7 $\pm$ 6.0	
Gefarnate	100 (8)	16.4 $\pm$ 2.3*	67.3	33.6 $\pm$ 2.9*	13.2
Control	(8)	47.4 $\pm$ 15.5		38.9 $\pm$ 2.2	
Aluminum sucrose sulfate	1000 (8)	33.6 $\pm$ 19.8	29.1	57.8 $\pm$ 5.2**	-48.5
Control	(8)	47.4 $\pm$ 15.5		38.9 $\pm$ 2.2	
L-glutamine	500 (8)	26.9 $\pm$ 6.0*	44.3	33.2 $\pm$ 0.9*	14.7
Control	(8)	44.0 $\pm$ 12.6		26.7 $\pm$ 1.7	
Methylmethionine sulfonium chloride	50 (8)	28.0 $\pm$ 11.6	36.4	19.6 $\pm$ 1.9**	26.6

(), Number of rats. *, Significant difference from control ($p < 0.05$). **, Significant difference from control ($p < 0.01$).

TABLE 7. Inhibitory effects of cetraxate and several drugs on ulcer index and β -glucuronidase activity in ulcer tissue of clamping cortisone ulcer in rats

Drugs	Dose mg/kg, p.o. $\times 12$ days	Ulcer index		β -glucuronidase activity	
		Index mean $\pm$ S.E.	% healing rate	μ g <i>p</i> -nitrophenol /mg protein/hr mean $\pm$ S.E.	% inhibition
Control	(8)	9.3 $\pm$ 2.4		56.1 $\pm$ 4.4	
Cetraxate	200 (8)	5.8 $\pm$ 0.5*	37.6	37.9 $\pm$ 3.2**	32.4
Gefarnate	100 (8)	6.1 $\pm$ 1.4	34.4	52.2 $\pm$ 5.3	7.0
Aluminum sucrose sulfate	1000 (8)	9.3 $\pm$ 3.4	0	70.8 $\pm$ 9.3	-26.2
L-glutamine	500 (8)	3.5 $\pm$ 0.8**	62.4	41.4 $\pm$ 3.5*	26.2
Methylmethionine sulfonium chloride	50 (8)	7.4 $\pm$ 1.0	20.4	49.7 $\pm$ 4.5	11.4

(), Number of rats. *, Significant difference from control ($p < 0.05$). **, Significant difference from control ($p < 0.01$).

mg/kg/day caused significant inhibitions of 62.4% ($p < 0.01$) and 26.2% ($p < 0.05$) against both ulcer index and β -Gase activity, respectively.

DISCUSSION

The p.o. administration of 300 mg/kg of cetraxate inhibited acute gastric ulcers induced by aspirin, phenylbutazone and indomethacin. The mechanism of development of aspirin ulcer has been claimed to be primarily due to the direct irritation to gastric mucosa and

partially due to the disturbance of the gastric mucosal barrier by the back diffusion of gastric acid (9). In the light of the marked inhibitory effect of cetraxate on aspirin ulcer, it may be assumed that this drug has a protective effect similar to that of L-glutamine on the gastric mucosal barrier.

Shay's ulcer induced by pylorus ligation is considered to be unsuitable as a model of gastric ulcer, since the ulceration develops in the forestomach. However, this method can serve for detecting drugs which are effective in eliminating the aggressive factors such as pepsin and HCl in gastric juice, as the mechanism of ulceration for this model may be attributed mainly to the digestive action of gastric juice. Okabe (10) reported that pepsin is probably a more important factor than gastric juice in Shay's ulcer. Cetraxate 300 mg/kg p.o. significantly inhibited the peptic activity of gastric juice 18 hr after pylorus ligation, while the volume and the total acidity remained unaffected. Fujii et al. found that cetraxate at a concentration of 10^{-7} M remarkably inhibited peptic activity, the decrease of pH and the increase of Cl^- following the perfusion of tetragastrin through the stomach in rats (personal communication). Therefore, cetraxate may partially inhibit this experimental ulcer through its anti-peptic activity. We previously reported that increased activity of mucopolysaccharase such as β -Gase was evident in the ulcer tissue of chronic types of gastric ulcers induced by acetic acid, clamping or clamping-cortisone, but not in acute types of ulcers induced by anti-inflammatory drugs or pylorus ligation (7). We assumed that in these chronic types of ulcers, β -Gase might be liberated from lysosomes of necrotic cells in the gastric tissue subjected to a severe injury by the injection of acetic acid or by clamping with aluminium plate and would bring about the destruction of the normal tissue surrounding the necrotic wound. Therefore, β -Gase activity in ulcer tissue may reflect the extent of the lesion of gastric tissue. In the present experiment, cetraxate was effective in inhibiting both the ulcer index and β -Gase activity of ulcer tissue in models of these chronic ulcers, suggesting that this drug may prevent destruction of gastric tissue due to lysosomal enzyme such as β -Gase. Fukawa et al. (11) reported that drugs such as antacids, anticholinergic agents and anti-pepsin agents which eliminate the aggressive factors were ineffective on acetic acid ulcer, whereas the therapeutic efficacy was observed with drugs such as L-glutamine and methylmethionine sulfonium chloride which are known to promote the regeneration of mucosa and the formation of granuloma tissue. Thus cetraxate also may be active in promoting the acceleration of ulcer healing.

The relationship between gastric ulcer and the fibrinolytic activity is now frequently discussed and Abe et al. (12) reported that fibrinolytic activity was elevated at the ulcer region in the gastric ulcer produced by immersing rats in cold water. Kondo et al. (13) reported that the local fibrinolytic activity in the gastric ulcer induced by the injection of immunocomplex in dogs was elevated up to 9 times the normal level after 2 hr, and 8 and 4 times more than the normal after 12 and 24 hr, respectively. The possible effect of cetraxate *in vivo* on local fibrinolytic activity is under investigation by our team. In view of the results obtained *in vitro*, cetraxate may also have inhibitory effects on the elevation of fibrinolytic activity in ulcer tissue.

Yokoyama suggested that cetraxate might have a strong affinity to gastric mucosa from results obtained by the systemic autoradiographic method in rats which showed that this agent was retained longer in the stomach (personal communication).

From these findings, cetraxate proved to be applicable for clinical therapy as it had not only the promoting effect on the prevention and healing of the ulcer itself, but also a hemostatic effect.

REFERENCES

- 1) MURAMATSU, M. AND FUJII, S.: *Biochim. Biophys. Acta* **242**, 222 (1971)
- 2) OKANO, A., INAOKA, M., FUNABASHI, S., IWAMOTO, M., ISODA, S., MOROI, R., ABIKO, Y. AND HIRATA, M.: *J. med. Chem.* **15**, 247 (1972)
- 3) SUZUKI, Y., INA, K., KOHARA, U. AND YAMAGAMI, I.: *Folia pharmacol. japon.* **68**, 226 (1972) (in Japanese)
- 4) SHAY, H., KOMAROV, S.A. AND FELS, S.S.: *Gastroenterology* **5**, 43 (1945)
- 5) PRINO, G., PAGLIALUNGA, S., NARDI, G. AND LIETTI, A.: *Europ. J. Pharmacol.* **15**, 119 (1971)
- 6) TAKAGI, K., OKABE, S. AND SAZIKI, R.: *Japan. J. Pharmacol.* **19**, 418 (1969)
- 7) SUZUKI, Y., HAYASHI, M., HAMAGUCHI, Y., ITO, M. AND YAMAGAMI, I.: *Pharmacometrics* **12**, No. 1 (1976) (in press)
- 8) UMEHARA, S., TABAYASHI, T., SHIBUYA, E. AND ITO, Y.: *Medicine and Biology* **66**, 7 (1960) (in Japanese)
- 9) DAVENPORT, H.W.: *Gastroenterology* **46**, 245 (1964)
- 10) OKABE, S.: *J. Pr. Ph.* **25**, 54 (1974) (in Japanese)
- 11) FUKAWA, K., IRINO, O., IIO, Y., MISAKI, N. AND NOMURA, F.: *Pharmacometrics* **7**, 1329 (1973) (in Japanese)
- 12) ABE, H., MATSUO, Y. AND YUTANI, M.: *Reports from 10th Meeting of Plasmin*, p. 142 (1970) (in Japanese)
- 13) KONDO, K., KAWAI, K. AND MASUDA, M.: *Japan. J. dig. Dis.* **71**, 41 (1974) (in Japanese)