PREVENTIVE EFFECT OF FUSARIC ACID, A DOPAMINE β-HYDROXYLASE INHIBITOR, ON THE GASTRIC ULCERATION INDUCED BY WATER-IMMERSION STRESS IN RATS

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It is well known that a wide variety of acute stress in experimental animals results in gastric ulceration and reduction of the noradrenaline content in the brain (1). Brodie and Valtiiski (2) and Watanabe (3) have shown the suppressive effects of adrenaline and noradrenaline on the gastric ulceration induced by restraint stress in rats. Vagotomy was found to decrease the incidence of gastric ulcer, while adrenalectomy increased the incidence of the ulcer lesions (4). Release of endogenous monoamines by reserpine and tetrabenazine aggravated the gastric ulcer induced by water-immersion stress (5). Okabe et al. (6) have reported that most (-adrenergic blocking agents aggravate the gastric ulceration, while ß-adrenergic blocking agents significantly inhibit the ulceration. Information concerning the relationship between gastric ulcer and adrenergic mechanism is still, however, obscure.

Fusaric acid (5-butylpicolinic acid) has been described as a potent inhibitor of dopamine β-hydroxylase by Hidaka et al. (7) and Nagatsu et al. (8). This compound showed a consistent hypotensive effect in both experimental animals (7) and humans (9). The present experiments were designed to study the effect of fusaric acid on the gastric ulcer induced by water-immersion stress with special reference to the level of brain noradrenaline and dopamine. Tetrabenazine, a central monoamine depleter, was used as reference.

Male Wistar strain rats, weighing 200-300 g were housed in separate cages at 22-24°C under a constant day-night rhythm. Following the method of Takagi and Okabe (10), the animals were fasted for 16 hr, then confined in a wire-meshed restraint cage which was immersed into water up to the xyphoid and kept at 21 ± 0.5°C. The rats were left in this situation for 3 hr, as ulcerative gastric changes were fairly severe and constant in preliminary experiments. After being sacrificed by decapitation, ulcer index obtained from macroscopic examination of the gastric mucosa was as follows: 0; normal, 1; slight edema and congestion, 2; edema, congestion and bleeding, 3; one or 2 spot erosions, 4; one or 2 linearly arranged erosions, 5; many small-sized and a few large-sized erosions, 6; extensive erosions were visible over the entire mucosa. Brain catecholamines were extracted according to our modified method (11) of Anton and Sayre (12). Noradrenaline (13) and dopamine (14) were assayed fluorometrically. Fusaric acid (kindly provided by Dr. H.
Hidaka) and tetrabenazine were injected intraperitoneally. In the next series of experiment, the rats was anesthetized with pentobarbital sodium and a needle was implanted stereotaxically into the lateral ventricle according to the brain atlas by König and Klippel (15). Five days later, fusaric acid dissolved in 40 μl of 0.15 M phosphate buffer, pH 7.0, was administered into the ventricle, and the result was compared to that after an intraventricular administration of the vehicle.

The water-immersion stress produced different sized erosions with blood clots in the glandular portion of the stomach and the ulcer index was 3.3 ± 0.2 (S.E.), as shown in Table 1. In these animals, the brain noradrenaline was lowered to 76% of the control without affecting the brain dopamine (Table 2). Decrease in noradrenaline content in the brain may be attributed to increased activity of noradrenaline containing neurons with a subsequent release of this amine exceeding the rate of neuronal resynthesis. In the rats treated with 20 mg/kg of fusaric acid 1.5 hr prior to the stress, the ulcer index and brain noradrenaline level were almost the same as those of the animals given only stress. Noradrenaline level in the brain dropped to 52% of the control 4.5 hr after the administration of fusaric acid, 100 mg/kg, i.p., without a concomitant change in the brain dopamine.

### Table 1. Effects of fusaric acid and tetrabenazine on the stomach ulcer index of rat given water-immersion stress for 3 hr.

| Drugs         | Doses    | Route   | Number of rats | Ulcer index* | % change** |
|---------------|----------|---------|----------------|--------------|------------|
| Saline        | i.p.     | 12      | 3.3±0.2        |              |            |
| Fusaric acid  | 20 mg/kg | i.v.    | 8              | 3.1±0.4      | −6         |
| ″             | 100      | i.v.    | 8              | 0.5±0.3###   | −85        |
| Tetrabenazine | 50 i.v.  |         | 8              | 4.4±0.4#     | +33        |
| Vehicle       | intravent.| 8      | 3.6±0.3        |              |            |
| Fusaric acid  | 0.5 mg/animal i.v. | 8 | 1.7±0.4### | −53 |

*: Results are mean±standard errors.
**: Values are compared with respective controls treated via the same route.
Statistical significance against respective control: *=p<0.05, ###=p<0.001

### Table 2. Effects of fusaric acid and tetrabenazine on brain noradrenaline (NA) and dopamine (DA) contents in rats given water-immersion stress for 3 hr.

| Drugs         | Doses (mg/kg i.p.) | Number of rats | Stress | Brain NA (µg/g)* | Brain NA (%)** | Brain DA (µg/g)* | Brain DA (%)** |
|---------------|-------------------|----------------|--------|-----------------|----------------|-----------------|----------------|
| Saline        | 8                 | 0.33±0.02      | 100    | 0.95±0.07       | 100            | 0.95±0.07       | 100            |
| Fusaric acid  | 20                | 0.25±0.01#     | 76     | 1.00±0.05       | 105            | 0.94±0.12       | 99             |
| 100           | 0.26±0.01#        | 79             | 0.94±0.12 | 99             | 0.93±0.05      | 98              |                |
| 100           | 0.17±0.02##       | 52             | 1.13±0.06 | 119            | 0.04±0.01      | 4               |                |
| Tetrabenazine | 0.16±0.02##       | 48             | 1.13±0.06 | 119            | 0.04±0.01      | 4               |                |

*: Results are the mean±standard errors.
**: Values are expressed as percentage of control.
Statistical significance against control: # = p<0.01, ## = p<0.001
These data coincided well with the results described by Nagatsu et al. (8). Pretreatment of rats with fusaric acid 100 mg/kg, i.p., 1.5 hr prior to the stress almost completely prevented the formation of gastric ulcer, and the ulcer index was only 0.5 ± 0.3 (Table 1). Noradrenaline content in these animals did not significantly differ from that of non-stressed rats treated with the same dose of fusaric acid 4.5 hr before sacrifice. Intraventricular administration of fusaric acid, 0.5 mg/animal, 1.5 hr prior to the stress also prevented the gastric ulceration, the ulcer index of these rats being 1.7 ± 0.4 as compared to the control 3.6 ± 0.3 (Table 1). On the other hand, tetrabenazine in a dose of 50 mg/kg, i.p., 30 min prior to the stress produced an aggravation of the gastric ulcer, the ulcer index being 4.4 ± 0.4 (Table 1). Noradrenaline and dopamine contents of these rats decreased to 9 and 4% that of the control, respectively, with increased release of these catecholamines.

The present results suggest that both intraperitoneal and intraventricular administration of fusaric acid prevent the formation of gastric stress ulcer presumably due to a decrease in the release of noradrenaline in the central nervous system. Details of the central and peripheral actions of the compound on gastric function are now being investigated.

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REFERENCES
1) Bliss, E.L., Allion, J. and Zwanziger, J.: J. Pharmacol. exp. Ther. 164, 122 (1968); 2) Brodie, D.A. and Valitski, L.S.: Proc. Soc. exp. Biol. Med. 113, 998 (1963); 3) Watanabe, K.: Chem. Pharm. Bull. 14, 101 (1966); 4) Brodie, D.A. and Hanson, H.M.: Gastroenterol. 38, 353 (1960); 5) Fujihara, M. and Mori, J.: Saishin-Igaku 25, 2058 (1970) (in Japanese); 6) Okabe, S., Sazuki, R. and Takagi, K.: Japan. J. Pharmacol. 20, 10 (1970); 7) Hidaka, H., Nagatsu, T., Takeya, K., Takeuchi, T., Suda, H., Kohri, K., Matsuzaki, M. and Umezawa, H.: J. Antibiotics 22, 228 (1969); 8) Nagatsu, T., Hidaka, H., Kuzuya, H., Takeda, K., Umezawa, H., Takeuchi, T. and Suda, H.: Biochem. Pharmacol. 19, 35 (1970); 9) Terasawa, F. and Kamiyama, M.: Japan. Circul. J. 35, 339 (1971); 10) Takagi, K. and Okabe, S.: Japan. J. Pharmacol. 18, 9 (1968); 11) Osumi, Y., Wada, I. and Fujihara, M.: Japan. J. Pharmacol. 22, 723 (1972); 12) Anton, A.H. and Sayre, D.F.: J. Pharmacol. exp. Ther. 138, 360 (1962); 13) Bertler, A., Carlsson, A. and Rosengren, E.: Acta physiol. scand. 44, 273 (1958); 14) Anton, A.H. and Sayre, D.F.: J. Pharmacol. exp. Ther. 145, 326 (1964); 15) König, J.F.R. and Klippel, B.A.: The Rat Brain, p. 162, Williams and Wilkins, Baltimore (1963)