Reactions in 0.2 N aq. NaOH at 70°, 80°, and 90°: A solution of Ib (92.3 mg) or Ih (100.0 mg) in 0.2 N aq. NaOH (6.00 ml) was allowed to react at the requisite temperature. At intervals a portion (0.50 ml) of the solution was mixed with acetic acid (0.20 ml) to give a sample solution for paper chromatography. Concentrations of IIIm and IVm were determined as described above.

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Significance of Stomach-Emptying-Controlled Rabbits for GI Absorption Studies of Water-Soluble Drugs

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The usefulness of stomach-emptying- (SE-) controlled rabbits for GI absorption studies of relatively water-soluble drugs was investigated, employing aminopyrine and sulfisoxazole as model drugs.

When aminopyrine was administered, similar plasma level-time curves were obtained in both SE-controlled and conventionally fasted rabbits. In the case of sulfisoxazole, the time required to reach the peak plasma level was much longer in conventionally fasted rabbits than in the SE-controlled group.

It was concluded that use of conventionally fasted rabbits for GI absorption studies should be limited to work on water-soluble drugs.

Keywords——stomach-emptying-controlled rabbit; bioavailability; water-soluble drug; aminopyrine; sulfisoxazole

There is considerable evidence that drug bioavailability may be influenced by the presence of food in the GI tract.[2,3] The idea that prohibiting the coprophagy of a rabbit may avoid the delay of gastric emptying rate (GER) by gastric contents led to a series of studies using stomach-emptying-(SE-) controlled rabbits,[4–8] employing practically insoluble drugs such as griseofulvin,[4,5] indomethacin,[4] nalidixic acid,[4] indoxole,[6] and chloramphenicol palmitate[8] as models.

On the other hand, using radioactive tracer techniques the GERs of both the solid and the liquid components of a meal were measured simultaneously; it was demonstrated that liquid left the stomach more rapidly than solids.[9,10] This suggests that conventionally fasted rabbits may also be available for GI absorption studies of water-soluble drugs.

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In this paper, relatively water-soluble drugs, such as aminopyrine and sulfisoxazole, were employed as models, and the usefulness of SE-controlled rabbits for GI absorption studies of water-soluble drugs was investigated.

Experimental

Materials——Aminopyrine and sulfisoxazole used were of JP IX grade. Other reagents were of reagent grade.

Animal Experiments——Male albino rabbits of 2.8—3.2 kg body weight were employed in all experiments. The stomach-emptying time of rabbits was controlled before the absorption study following the procedure described in our previous paper.[7] Conventionally fasted rabbits, which had been fasted for 20 hr with free access to water, were employed as the reference. Aminopyrine (20 mg/kg body weight) and sulfisoxazole (50 mg/kg body weight), both contained in gelatin capsules, were administered to rabbits.

Blood specimens were taken by cardiac puncture using a heparinized syringe at predetermined times up to 8 or 9 hours. Plasma levels of aminopyrine were assayed by a slight modification of the method of Ono *et al.*[11] Plasma levels of sulfisoxazole were assayed by diazotization.[12]

Measurement of Water Solubility——Water solubilities of griseofulvin, indomethacin, nalidixic acid, aminopyrine and sulfisoxazole were determined as follows. Excess amounts of drugs were mixed with 0.1 N HCl (*ca.* pH 1.0) or 0.05 M phosphate buffer (pH 6.0) and shaken vigorously at 24° for 6 hr, then centrifuged at 2000 ***g*** for 15 min. The saturation was checked by periodic sampling of the supernatant. The ultraviolet (UV) absorbance of the supernatant was measured.

Results and Discussion

Aminopyrine was employed as a model water-soluble drug; it had a solubility of 76.5 mg/ml at pH 1.0 and 49.2 mg/ml at pH 6.0, and was not absorbed from the stomach in man or rats.[13,14] Naito *et al.* administered 200 mg of aminopyrine per kg body weight as a 1% solution to a conventionally fasted rabbit.[15] Goto *et al.* administered 255 mg of aminopyrine per kg body weight as a 0.9% solution to a rabbit whose stomach had been lavaged after 24 hours' fasting.[16] Plasma level-time curves in these experiments suggested that there was little delay in the absorption of aminopyrine.

Aminopyrine is usually administered to humans as solid dosage forms such as capsules, granules, and so on. In this investigation, therefore, a comparative bioavailability study of aminopyrine capsules was carried out using SE-controlled and conventionally fasted rabbits. Both were administered 60 mg of aminopyrine (capsule) per rabbit with 20 ml of water, and the plasma aminopyrine level was assayed. As shown in Fig. 1, the extent of bioavailability was not significantly different ($p>0.1$) between the two (mean AUC values ±S.E. were 47.6± 4.8 and 39.9±2.9 μg·hr/ml for SE-controlled and conventionally fasted rabbits, respectively), though the plasma level-time curves are different in detail. In the case of conventionally fasted fabbits, the peak plasma level of aminopyrine was low (5.7 μg/ml) and the time required to reach the peak plasma level was 3 hr after oral administration. On the other hand, the SE-controlled rabbit shows a higher plasma peak level (8.5 μg/ml) at 1 hr. This is probably attributable to the retention of gastric contents in conventionally fasted rabbits, which was 80—100 g after overnight fasting.[4]

The above results suggest that SE-control is not always essential in studies of water-soluble drugs such as aminopyrine.

Next, sulfisoxazole was selected as a model of a slightly soluble drug (0.6 mg/ml at pH 1.0 and 1.0 mg/ml at pH 6.0); it is, however, more soluble than griseofulvin, indomethacin, and

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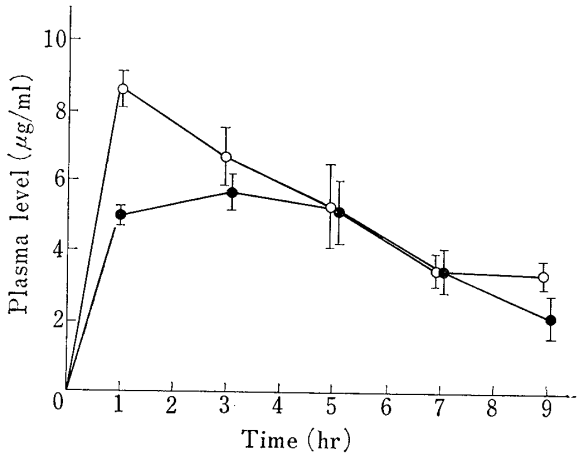

Fig. 1. Average Plasma Aminopyrine Levels Following Oral Administration of 60 mg of Aminopyrine (Capsule) to Rabbits

Each point represents the average±S.E. in 4 animals.
Key: ○, SE-controlled; ●, conventionally fasted.

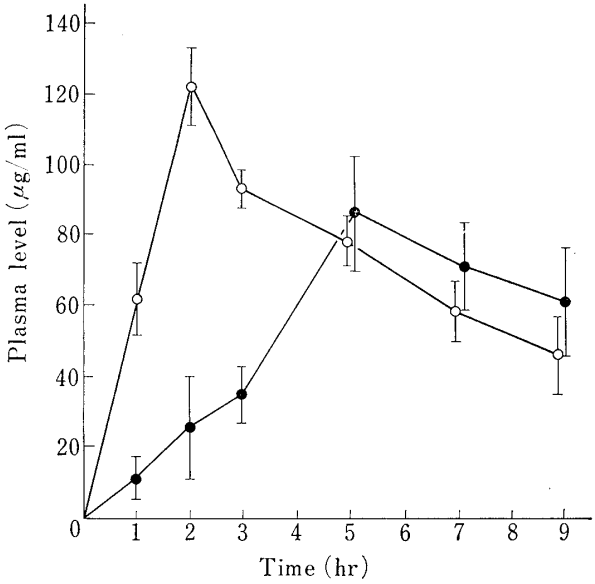

Fig. 2. Average Plasma Sulfisoxazole Levels Following Oral Administration of 150 mg of Sulfisoxazole (Capsule) to Rabbits

Each point represents the average±S.E. in 4 animals.
Key: ○, SE- controlled; ●, conventionally fasted.

Table I. Solubilities and Bioavailability Indices of Drugs employed in the Present Series of GI Absorption Studies using SE–Controlled Rabbits

Drug	Water solubility at 24° (μg/ml)		SE-controlled/Conventionally fasted	
	pH 1.0	pH 6.0	AUC	Peak plasma level[a]
Griseofulvin	1.20×10	1.24×10	1.4	2.1
Indomethacin	1.38	8.20×10	1.5	1.9
Nalidixic acid	4.60×10	5.32×10	1.6	2.7
Aminopyrine	7.65×10^4	4.92×10^4	1.2	1.4
Sulfisoxazole	5.69×10^2	1.03×10^3	1.4	1.4

a) Average of individual peak plasma levels.

so on (Table I). Both SE-controlled and conventionally fasted rabbits were administered 150 mg of sulfisoxazole (capsule) per rabbit with 20 ml of water, and the plasma sulfisoxazole level was assayed. As is clearly shown in Fig. 2, the time required to reach the peak plasma level was very long in conventionally fasted rabbits (5 hr) compared with SE-controlled rabbits (2 hr). The AUC value, which is an index of extent of bioavailability, is significantly different ($p<0.05$) between the two, that is, 651.1±7.4 μg·hr/ml for SE-controlled and 466.3±44.2 μg·hr/ml for conventionally fasted rabbits (mean ±S.E.).

About five drugs employed in a series of GI absorption studies using SE-controlled rabbits, the ratios of AUC and the averages of individual peak plasma levels of SE-controlled rabbits relative to those of conventionally fasted rabbits are listed in Table I, together with the water solubilities. These results suggest that SE-control is essential for studies of poorly water-soluble drugs, and the use of conventionally fasted rabbits for GI absorption studies should be limited to work on water-soluble drugs.