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Effect of Tryptophan and its Metabolites on Gastric Emptying of Liquid Meals in Dogs (39461)

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In a previous study (1) we found that tryptophan slowed gastric emptying significantly (60% inhibition) in concentrations about 10× lower than 25 other amino acids including phenylalanine. We concluded that there appeared to be a specific tryptophan receptor which was probably different from the classical osmoreceptor (1-3).

Tryptophan is well absorbed by the small intestine and is largely metabolized in the liver (4). It is possible, therefore, that tryptophan or its metabolites may act, after absorption, directly on the stomach to slow emptying.

The aim of this study was to determine whether tryptophan or some of its metabolites act directly on the stomach or brain rather than via gastrointestinal receptors. This was done by instilling tryptophan metabolites into the gut or infusing tryptophan and some metabolites intravenously (Fig. 1).

Methods and materials. Studies over a period of several months were made in five healthy dogs weighing 14 to 19 kg. Each dog was equipped with a Thomas (5) cannula to form a gastric fistula. The dogs were fasted 18 hr before testing but were allowed water *ad lib.*

Procedures. The dogs were brought to the laboratory and their stomachs were rinsed with tap water via the gastric fistula to clear accumulated secretions. This rinse was allowed to drain by gravity. A plastic adapter with a stop-cock was then connected to the gastric cannula through which the test meals were instilled and drained. Two 25-min test meals were given in succession with 35 min between each meal. The first meal (300 ml at 37°) was a control and consisted of deionized water with phenol red (40 mg liter^{-1}), a nonabsorbable marker dye and was instilled via the gastric fistula into the stomach in less than 45 sec. After 25 min, the contents of the stomach were drained completely by gravity through the gastric fistula. Immediately after the gastric contents were collected, 200 ml of a "wash" solution of deionized water was instilled into the stomach to recover any residual contents and then drained. The gastric fistula was left open and gastric secretions were allowed to drain by gravity in the 35 min interval between tests. The second meal containing the test substance and phenol red was administered and collected in precisely the same fashion as the first.

These techniques were varied slightly in the intravenous studies. For each experiment, a polyethylene catheter (PE 50, Intramedic, Clay Adams Inc., N.Y.) was inserted into a dog's hind leg vein. A syringe pump (Harvard Apparatus, Dover, Mass.) was used to infuse normal saline or the test substance into the vein at 25 ml hr^{-1}. As the control, the dog was infused for 1 hr with normal saline and during the last 25 min a deionized water meal was given. The test substance was then added to the iv infusion and the mixture infused for 1 hr, the last 25 min of which a deionized water meal was given. The collection of the test meals was identical to that described above.

The volumes of the residual gastric contents and wash solutions were measured to the nearest 1 ml and samples were saved for determination of phenol red concentration. Phenol red concentration after alkalinization was measured spectrophotometrically at a wavelength of 558 nm.

Test Meals and Substances. All meals were 300 ml in volume with phenol red (40 mg liter^{-1}). The osmolalities were the same as the individual concentrations. In the first series of studies the test meals contained DL-kynurenine (5 m*M*), 5-hydroxytryptophan (5 m*M*), or 5-hydroxytryptamine (serotonin) in a concentration of 20 m*M* (20

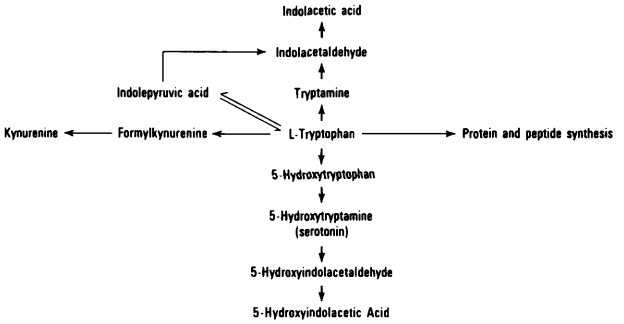

FIG. 1. A diagram to indicate some of the major metabolic pathways of tryptophan.

mOs). For the iv studies the following compounds were infused: L-tryptophan, 5 m*M*, 20 m*M*, and 50 m*M*; DL-kynurenine, 2 m*M*, 5 m*M*, and 10 m*M*; 5-hydroxytryptamine, 2 m*M* and 10 m*M*; and 3-indolepyruvic acid. All test substances were dissolved in saline except 3-indolepyruvic acid which was dissolved in ethanol. It was given as an infusion at a concentration of 2 m*M* and as a single injection at 5 m*M* and 10 m*M* because of high concentrations of ethanol required to dissolve it. All these compounds have a molecular weight of about 200 and the 10 m*M* concentrations calculated to be an iv dose of about 3 mg kg^{-1} hr^{-1}.

Calculations. Concentrations of phenol red were measured in the meal ingested, in the gastric contents recovered, and the wash solutions. The volume of the original meal left in the stomach was used as the index of gastric emptying. This volume was calculated from the formula:

$$V_{25} = [V_n + (V_w P_w / P_n)] (P_n / P_o)$$

in which V_n and V_w are the volumes of the gastric contents collected at 25 min and wash solution (200 ml), respectively. The concentration of phenol red in the solution instilled into the stomach is P_o, in the gastric contents recovered at 25 min is P_n and in the wash solution is P_w.

Statistics. Differences in the volumes of original meal left in the stomach at 25 min during control and test situations were analyzed by a paired *t* test (6).

Results. Effect of instilled metabolites. At the concentrations studied, DL-kynurenine (5 m*M*) and 5-hydroxytryptamine (20 m*M*) did not produce significant slowing of gastric emptying (Table I). The metabolite 5-hydroxytryptophan (5 m*M*) was studied in two dogs only because of toxicity (extreme restlessness).

TABLE I. THE EFFECT OF INSTILLED TRYPTOPHAN METABOLITES ON GASTRIC EMPTYING.

Metabolite	Number of dogs	Mean control (ml ± SE)	Mean Test (ml ± SE)	*P*
5-Hydroxy-tryptamine (20 m*M*)	4	33 ±6	44 ±19	>0.05
DL-Kynurenine (5 m*M*)	4	25 ±5	42 ±12	>0.05
5-Hydroxy-tryptophan (5 m*M*)	2	23 ±3	53 ±18	–

Effect of intravenous tryptophan and metabolites. All concentrations of L-tryptophan, 3-indolepyruvic acid, 5-hydroxytryptamine and DL-kynurenine failed to cause significant slowing of gastric emptying ($p > 0.05$) (Table II).

Discussion. Previous studies have indicated that tryptophan is a powerful inhibitor of gastric emptying and is unique among amino acids for this action (1). Those studies, however, did not determine whether the effect of tryptophan was due to tryptophan itself or some of its metabolites.

In the present study we did not find an effect on gastric emptying with large doses of the tryptophan metabolites 5-hydroxytryptamine, DL-kynurenine, and 5-hydroxytryptophan following instillation into the gut (Table I). Furthermore, when these same metabolites as well as 3-indolepyruvic acid and L-tryptophan itself were infused intravenously, no statistically significant slowing of emptying occurred (Table II). These studies

TABLE II. THE EFFECT OF INTRAVENOUS TRYPTOPHAN AND METABOLITES ON GASTRIC EMPTYING.

Substance	Δ[a] mean volume (ml ± SE) at various concentrations				
	2 m*M*	5 m*M*	10 m*M*	20 m*M*	50 m*M*
L-Tryptophan	–	− 6 ± 17	–	9 ± 19	−13 ± 12
3-Indolepyruvic	−19 ± 8	29 ± 30	13 ± 20	–	–
5-Hydroxytryptamine	65 ± 43	–	65 ± 30	–	–
DL-Kynurenine	20 ± 33	62 ± 23	48 ± 23	–	–

[a] The Δ volume is the difference between control and following iv infusion. Negative values indicate that control was greater than the test situation.

indicate that tryptophan is the agent which delays gastric emptying and not its metabolites and that it works by exciting receptors in the gut and not by direct action on the stomach or brain.

Summary. The effects of tryptophan and its metabolites were studied to determine whether the known inhibiting effect of tryptophan on emptying was due to local or systemic effects or due to tryptophan metabolites. In five dogs with chronic gastric fistulas, instillation of 300 ml of DL-kynurenine (5 m*M*), 5-hydroxytryptophan (5 m*M*), or 5-hydroxytryptamine (20 m*M*) into the gut did not slow gastric emptying. Furthermore, iv infusion of L-tryptophan (5, 20, 50 m*M*), DL-kynurenine (2, 5, 10 m*M*), 5-hydroxytryptamine (2 m*M*, 10 m*M*), and 3-indolepyruvic acid (2, 5, 10 m*M*) also did not slow gastric emptying.

These studies indicate that tryptophan slows gastric emptying by exciting a receptor in the gut and not by a direct effect on the stomach or brain or via its major metabolites.

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