importance of serum NEFA in modulating binding of drugs to albumin. Urate, bilirubin and cholesterol all produce similar degrees of displacement although very much less than that of palmitate.

Binding constants for ibuprofen with the various additives are shown in Table 4. Also included are values for the parameter Σnk which is useful as an index of binding overall, combining values for both the capacity and strength of the association.

Palmitate and, to a lesser extent, cholesterol alter the number of binding sites available in each class. Direct competition for common binding sites is expected to diminish the association constant of both drug and competitor; however, the number of sites of association is not expected to change. Certainly, small variations in values of n_1 and n_2 are likely to result from iteration on data having varying amounts of experimental error. These are indicated by the coefficients of variation. However, with palmitate the changes in n_1 (0.8 to 2.37) and n_2 (6.27 to 2.95) are much too large to be explained in this way. It seems more likely that the changes in the numbers of sites in addition to the 20-fold and 5-fold reduction in k_1 and k_2, respectively, indicate a conformational change in the albumin molecule. Other workers have also proposed conformational changes in albumin caused by binding of NEFA [31, 32].

The purpose of studying the influence of endogenous materials is to establish the possible significance that fluctuations in their serum concentrations may have on drug disposition. When the data are adjusted for a physiological serum albumin concentration of 4%, the percentage of free ibuprofen is 0.1% for a total serum concentration of 20 $\mu g\ ml^{-1}$. Based on the data in Table 4, the displacing effects of urate, bilirubin and cholesterol are negligible at this drug concentration. At the high albumin concentration present in serum, less than 17 per cent of available primary sites are occupied; thus, the likelihood that both drug and displacer will compete for the same sites is remote. In the unlikely event that competition occurs, the displaced drug molecule will immediately associate with one of the excess free sites. In contrast, palmitate (118 $\mu g\ ml^{-1}$) would increase the percentage of free ibuprofen 5-fold (from 0.1 to 0.5%).

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Effects of adrenergic agonists on gastric secretion in the rat

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Many organ systems, the gastrointestinal tract among them, are controlled in opposite directions by the sympathetic and parasympathetic nervous systems. However, little investigation has been done on gastric secretion under the control of the sympathetic nervous system, because it is usually thought that gastric secretion is controlled mainly by hormones such as gastrin and the parasympathetic nervous system.

Some contradictory results about gastric secretion by adrenergic agonists have been reported [1, 2]. Curwain and Holton [3] mentioned that isoproterenol and norepinephrine inhibited gastric acid secretion stimulated by pentagastrin in the dog, and Daly and Stables [4] obtained the same results; however, Hsu and Cooper [5] reported that epinephrine and isoproterenol raised the serum gastrin level in the rat.

In the present work, we examined how the adrenergic agonists act on gastric secretion in the rat, paying particular attention to whether α- or β-adrenergic activation causes gastric secretion and whether α- and β-adrenergic agonists have the same effects on the responses of gastric secretion induced by tetragastrin, histamine and carbamylcholine.

Male Wistar rats weighing about 200 g were fasted for 1 day and used for the perfused rat stomach preparation according to the method of Ghosh and Schild [6], under urethan anesthesia. The stomach cavity was perfused with 0.2×10^{-3} N NaOH at a constant rate of 0.5 ml/min and the gastric effluent was collected for 10-min intervals. The acidity of the effluent was measured by titration with 0.02 N NaOH up to pH 7.0, using a pH-stat (Toa Electronics Ltd., Tokyo). Peptic activity was determined by the following method, which is a modification of the method described by Anson [7]. A mixture of 1.0 ml of 1.0% casein (pH 1.8) and 1.0 ml of the effluent was incubated for 30 min at 37°, and 2.0 ml of 1.7 M perchloric acid was then added. Absorbance of the supernatant fluid obtained after centrifugation of the reaction mixture was measured at 280 nm.

Benzyloxycarbonyl-Trp-Met-Asp-Phe-NH_2 (tetragastrin) was purchased from San-a Chemical Co., Tokyo. *l*-Epinephrine bitartrate, *l*-norepinephrine bitartrate, *dl*-isoproterenol hydrochloride, carbamylcholine chloride and *dl*-propranolol hydrochloride (Sigma Chemical Co., Saint Louis, MO), histamine dihydrochloride and atropine sulfate (Wako Pure Chemical Industries, Osaka) were used. Chemicals were freshly dissolved before use.

The effects of single intravenous injections of adrenergic agonists were examined. Each chemical was injected into the rat three times at 90-min intervals. Unexpected side effects were not seen with doses of 12.5 to50 μg/kg of epinephrine and 25–100 μg/kg of norepinephrine and isoproterenol. As shown in Table 1, epinephrine and isoproterenol stimulated acid and pepsin secretion; norepinephrine did not. Epinephrine and isoproterenol stimulated acid secretion in a dose-dependent manner, with isoproterenol especially showing a linear dose–response relationship to acid secretion in the dose range of 25–200 μg/kg (Fig. 1). The effective dose of isoproterenol with respect to acid secretion was more than about 10 μg/kg from the intercept obtained by the dose–response curve.

The effects of single intravenous injections of adrenergic agonists on gastric secretion during continuous stimulation with tetragastrin were also examined in the same manner. Table 2 shows that epinephrine and isoproterenol caused a further increase in acid secretion, but norepinephrine reduced acid secretion stimulated by tetragastrin. These adrenergic agonists did not affect pepsin secretion significantly. During tetragastrin stimulation, the dose-dependent response of acid secretion to epinephrine and isoproterenol was not seen. Although tetragastrin was infused continuously and intravenously, the basal level for the calculation of pepsin secretion was not stable.

The effects of adrenergic agonists on gastric secretion were examined in combination with α- and β-adrenoceptor blockers. As shown in Fig. 2, acid secretion in response to isoproterenol was completely prevented by propranolol (5 mg/kg/hr), a β-adrenoceptor blocker. A slight increase in acid and pepsin secretion caused by propranolol was observed for 30 min. No significant change in pepsin secretion was seen in this experiment. Phenoxybenzamine, an α-adrenoceptor blocker, slightly reversed the reduction of tetragastrin-induced gastric secretion by norepinephrine (Fig. 3). Using the anticholinergic agent, atropine, we examined whether gastric secretion induced by β-adrenergic action is a result of rebound of the cholinergic mechanism. Gastric secretion in response to isoproterenol was not reduced by atropine (1 mg/kg/hr) (Fig. 4).

Effects of adrenergic agonists on gastric secretion induced by histamine or carbamylcholine rather than by tetragastrin were examined (Figs. 5 and 6). Isoproterenol (100 μg/kg/hr) and norepinephrine (100 μg/kg/hr) were infused for 60 min at 90-min intervals during continuous injection of histamine (500 μg/kg/hr) or carbamylcholine (2 μg/kg/hr). Histamine-

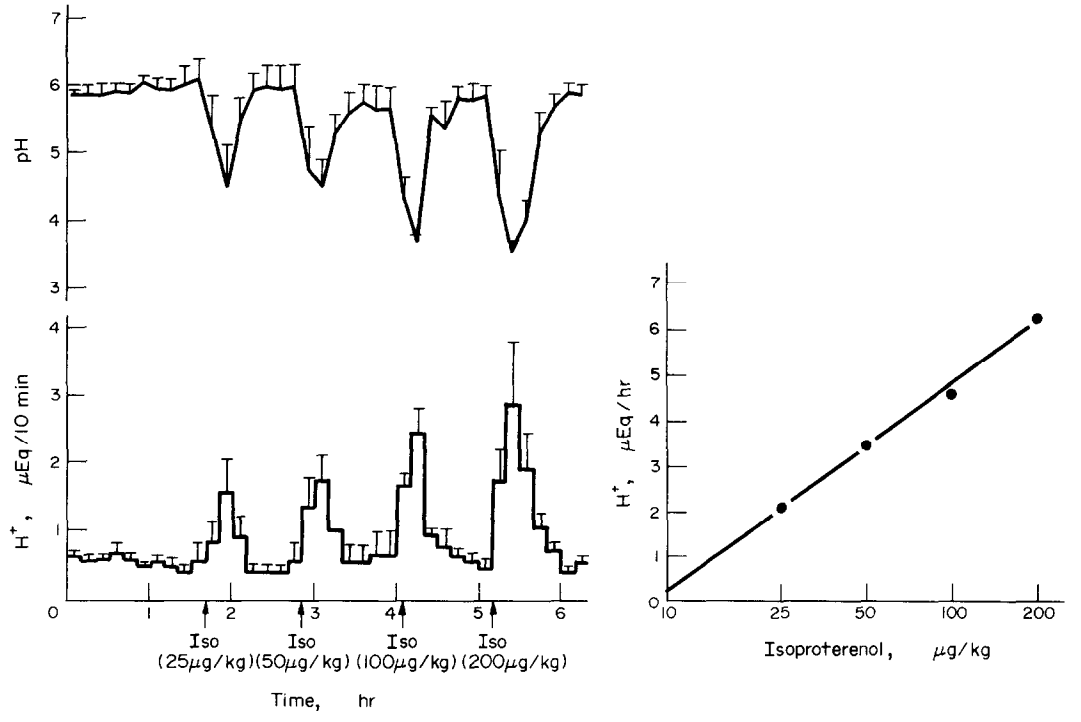

Fig. 1. Dose–response relationship between isoproterenol and acid secretion. Isoproterenol (Iso) was injected intravenously at the arrow and the values represent the mean ± S.E. from three rats.

Table 1. Effects of single injections of adrenergic agonists on gastric secretion*

Chemicals	Dose (μg/kg)	Acid secretion (μ-equiv./1 hr)	Pepsin secretion (%)†
Epinephrine	12.5	0.80 ± 0.23 (3)	28.1 ± 16.1 (4)
	25	1.50 ± 0.55 (3)	38.7 ± 27.4 (4)
	50	1.95 ± 0.47 (4)	36.6 ± 31.0 (4)
Isoproterenol	25	2.03 ± 0.78 (3)	40.0 ± 8.4 (4)
	50	3.89 ± 0.95 (4)	68.3 ± 19.9 (4)
	100	5.48 ± 1.01 (4)	69.9 ± 16.8 (4)
Norepinephrine	25	1.08 ± 0.55 (3)	−12.2 ± 8.5 (3)
	50	0.00 ± 0.00 (3)	20.3 ± 27.1 (4)
	100	0.65 ± 0.43 (3)	21.9 ± 26.2 (4)

* Chemicals were injected intravenously and values represent the mean ± S.M.; the number of experiments is given in parentheses.

† Per cent increase over basal pepsin level [about 15 μg/10 min converted into the weight of hog pepsin (3200 units/mg)].

induced gastric secretion was increased further by isoproterenol, and was reduced by norepinephrine in the same manner as the secretion induced by tetragastrin had been inhibited. The response to histamine infusion gradually became weaker with time, that is, tachyphylaxis occurred (Fig. 5). During carbamylcholine stimulation, isoproterenol stimulated and norepinephrine reduced acid secretion, but both isoproterenol and norepinephrine tended to reduce pepsin secretion. Gastric secretion gradually increased during carbamylcholine infusion, in contrast to the decrease seen with histamine-stimulated secretion.

The present results with adrenergic agonists and blockers

Table 2. Effects of single injections of adrenergic agonists on gastric secretion under tetragastrin stimulation*

Chemicals	Dose (μg/kg)	Acid secretion (%)†	Pepsin secretion (%)‡
Epinephrine	12.5	55.2 ± 54.9 (3)	5.9 ± 10.5 (3)
	25	55.7 ± 41.9 (3)	39.5 ± 14.4 (3)
	50	38.4 ± 26.2 (3)	44.0 ± 27.5 (3)
Isoproterenol	25	34.0 ± 8.1 (4)	18.7 ± 3.7 (4)
	50	65.8 ± 23.8 (4)	1.4 ± 4.4 (4)
	100	50.4 ± 12.9 (4)	15.6 ± 18.6 (3)
Norepinephrine	25	−2.4 ± 8.9 (6)	21.0 ± 13.4 (6)
	50	−18.0 ± 7.1 (7)	6.2 ± 9.6 (7)
	100	−19.4 ± 9.2 (6)	10.0 ± 11.5 (6)

* Chemicals were injected intravenously during continuous stimulation by tetragastrin (10 μg/kg/hr). Values represent the mean ± S.E.; the number of experiments is given in parentheses.

† Per cent increase over stimulated acid level (about 5 μ-equiv./10 min).

‡ Per cent increase over stimulated pepsin level [about 75 μg/10 min converted into the weight of hog pepsin (3200 units/mg)].

have shown that β-adrenergic action increased acid secretion without cholinergic rebounding and α-adrenergic action reduced acid secretion stimulated by secretagogues such as tetragastrin, histamine and carbamylcholine in the rat. The mechanism of stimulation of gastric acid secretion by these secretagogues has not been well established; they may act directly on parietal cells. If these three secretagogues stimulate gastric acid secretion through different pathways, norepinephrine must inhibit at least three different receptors on parietal cells. These results support the theory that these secretagogues all act in the same manner on parietal cells to cause acid secretion.

Many investigators [8–12] have suggested recently that adenylate cyclase is strongly associated with gastric acid secretion. Bieck *et al.* [13] found that gastrin and histamine stimulated adenylate cyclase activity. In contrast, Thompson *et al.* [14, 15] showed that the inhibitors of gastric acid secretion including epinephrine stimulated adenylate cyclase activity. It is thought that the reason for these contrasting results lies in the adenylate cyclase they used. The population of parietal cells in gastric mucosa is relatively small. We can not distinguish what kind of cell is the source of adenylate cyclase in gastric mucosal membrane fraction. There is also the question of the specificity of adenylate cyclase, because we must think about guanylate cyclase. The study by Ruoff [16] is helpful in considering this point. He found that epinephrine elevated the gastric mucosal c-AMP and c-GMP levels in a dose-dependent manner and, from the results using adrenoceptor blockers, he concluded that the effect of epinephrine on the gastric mucosal c-AMP level was mediated by β-adrenoceptors and that on the c-GMP level by α-adrenoceptors.

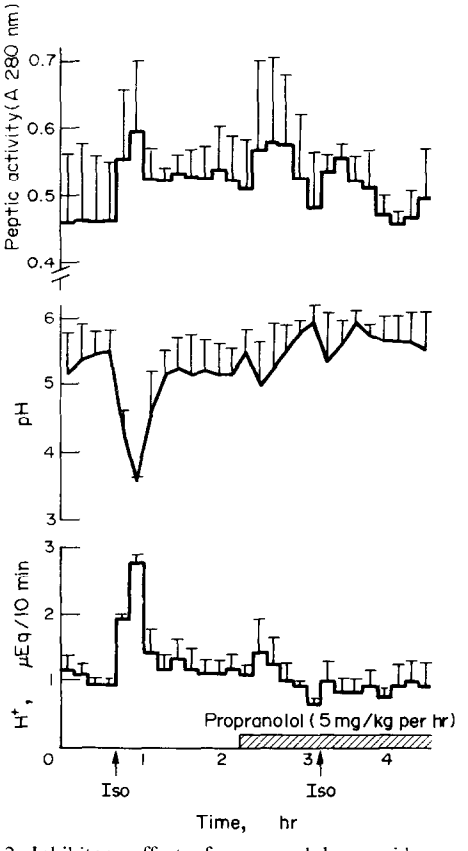

Fig. 2. Inhibitory effect of propranolol on acid secretion stimulated by isoproterenol. Saline was infused at the rate of 1 ml/hr; then saline was changed to propranolol (5 mg/kg/hr) during the period indicated by hatched block. Isoproterenol (100 μg/kg) was injected at the arrow. The values represent the mean ± S.E. from three rats.

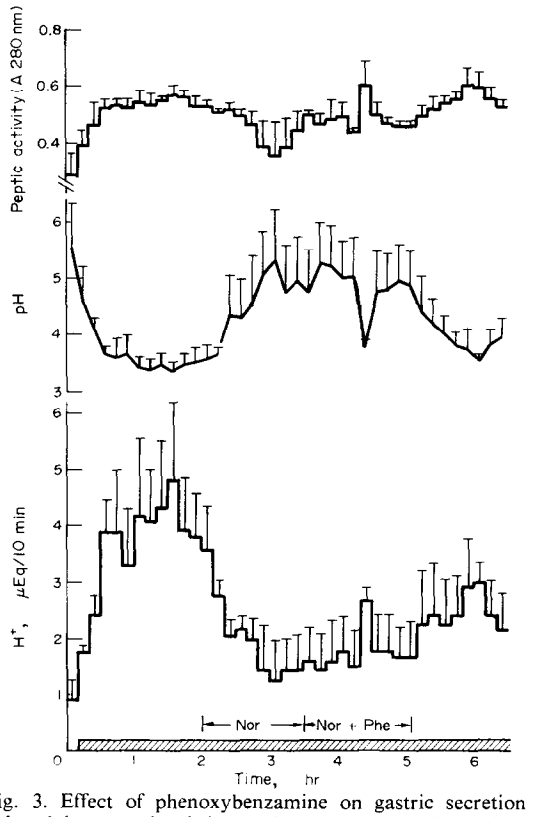

Fig. 3. Effect of phenoxybenzamine on gastric secretion reduced by norepinephrine under tetragastrin stimulation. Tetragastrin (10 μg/kg/hr) was infused during the time indicated by the hatched block. Norepinephrine (100 μg/kg/hr) and norepinephrine (100 μg/kg/hr) plus phenoxybenzamine (5 mg/kg/hr) were infused during the time indicated by Nor and Nor + Phe respectively. The values represent the mean ± S.E. from three rats.

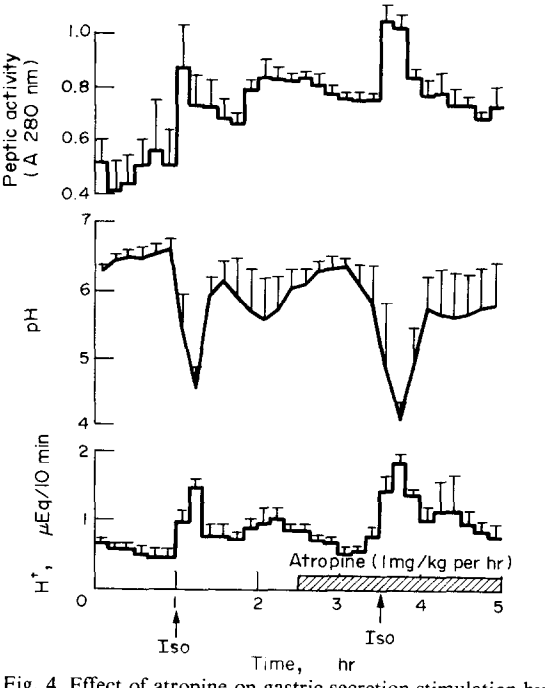

Fig. 4. Effect of atropine on gastric secretion stimulation by isoproterenol. Atropine (1 mg/kg/hr) was infused during the period indicated by the hatched block. Isoproterenol (100 μg/kg)was injected at the arrow. The values represent the mean ± S.E. from four rats.

Taking his report into consideration, our results suggest that β-adrenergic agonists such as isoproterenol raise the mucosal c-AMP level which causes stimulation of acid secretion and α-adrenergic agonists such as norepinephrine raise the c-GMP level which causes inhibition of acid secretion.

We could not obtain conclusive results regarding pepsin secretion from our experiments. The mechanism of pepsin secretion must be different from that of acid secretion. Pepsin secretion depends on two steps, one is synthesis of pepsinogen and the other is secretion of pepsinogen from chief cells. If a certain secretagogue stimulates only the secretory step, pepsin secretion would become weaker with time. In contrast, if a certain secretagogue stimulates only the synthesis step, pepsin secretion would become stronger with time. In order to understand pepsin secretion, it is necessary to understand the relationship between synthesis and secretion of pepsinogen. The effect of isoproterenol on pepsin secretion stimulated by carbamylcholine was unlike its effect on that stimulated by tetragastrin and histamine. Similar results were observed by Magee [1]; the effect of isoproterenol on acid secretion induced by pentagastrin differed from that on methacholine-induced secretion. These observations might reflect the fact that gastric secretion is under the control of many factors, including the nervous system, hormones, and amines, each in a different way.

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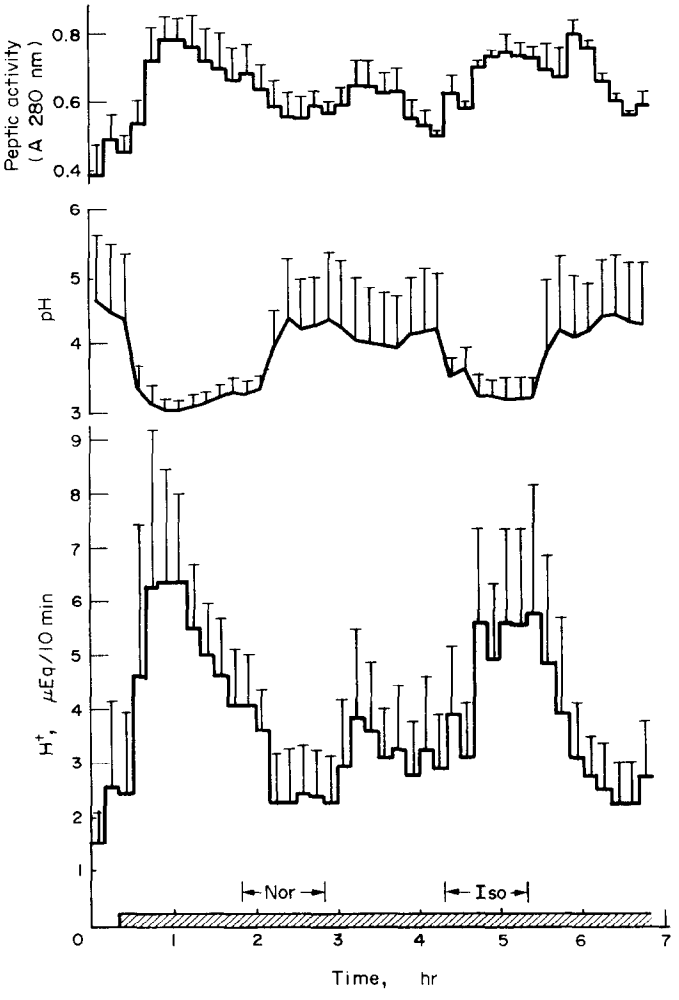

Fig. 5. Effect of norepinephrine and isoproterenol on gastric secretion under histamine stimulation. Histamine (500 μg/kg/hr) was infused during the time indicated by the hatched block. Norepinephrine (100 μg/kg/hr) and isoproterenol (100 μg/kg/hr) were infused during the time indicated by Nor and Iso respectively. The values represent the mean ± S.E. from three rats.

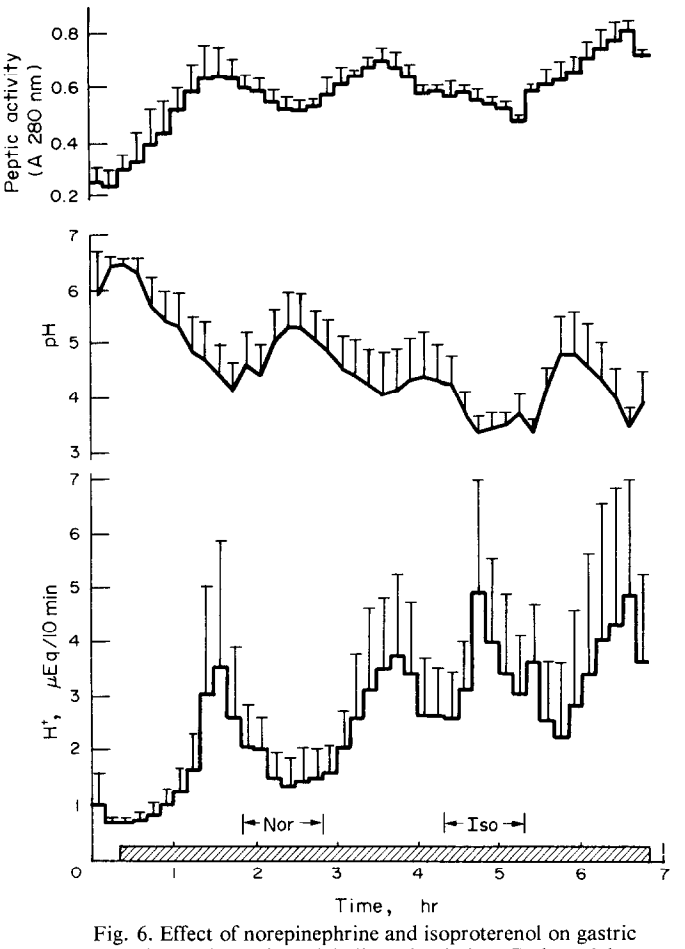

Fig. 6. Effect of norepinephrine and isoproterenol on gastric secretion under carbamylcholine stimulation. Carbamylcholine (2 μg/kg/hr) was infused during the period indicated by the hatched block. Norepinephrine (100 μg/kg/hr) and isoproterenol (100 μg/kg/hr) were infused during the time indicated by Nor and Iso respectively. The values represent the mean ± S.E. from four rats.

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Lack of correlation between cortisol-induced precocious maturation of the fetal rabbit lung and drug metabolism

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Precocious lung maturation produced by prenatal administration of glucocorticoids has received considerable attention since it appears that these steroids may play an important role in the prevention of respiratory distress syndrome (hyaline membrane disease) in premature infants [1–3]. The syndrome is thought to be the result of a deficiency in pulmonary surfactant which is vital for stabilizing the lungs against collapse during expiration [3]. Morphological and biochemi-