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Action of Gastrin on the Isolated Gastric Mucosa of the Bullfrog.* (30826)

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Gregory and Tracy isolated from hog antral mucosa 2 polypeptides, gastrins I and II, which proved to be the most potent of all known stimuli of gastric secretion(1). They and their colleagues later reported the complete characterization of the 2 heptadecapeptides and the total synthesis of both (2,3). Gastrins I and II are, on a molecular basis, about 500 times more potent gastric stimulants than histamine in the dog (denervated fundic pouch)(1).

Because the secretion of acid in the whole animal is subject to complex neural and humoral influences, it was desirable to attempt the study of the action of gastrin on isolated tissue.

Materials and methods. The bullfrog (*Rana catesbiana*) was chosen for study because the gastric mucosa is separated readily from the muscular layers as an intact sheet; also, the thin mucosal layer is easily oxygenated(4). The studies were carried out during June, July and August. Bullfrogs were stored in a dilute saline solution at 4°C until 24 hours prior to use. They were then fasted at room temperature until they were killed by decapitation and pithing. The mucosa was immediately mounted between 2 chambers containing the following solutions: a) *nutrient* or submucosal side—sodium chloride, 85.3 mM/l; potassium chloride 3.4 mM/l; calcium chloride 1.8 mM/l; magnesium sulfate 0.8 mM/l; potassium dihydrogen phosphate 0.8 mM/l; sodium bicarbonate 17.8 mM/l; and glucose 11.0 mM/l; b) *secretory* side—a similar solution except that the sodium bicarbonate, magnesium sulfate and potassium dihydrogen phosphate were omitted(5). A mixture of 95% O_2 and 5% CO_2 was bubbled through the nutrient solution, while 100% O_2 was passed through the fluid on the secretory side(6). All incubations were carried out at room temperature (22-24°C). The exposed area of mucosa was 4.5 cm^2. The acid formed on the secretory side was titrated automatically and continuously to pH 5.5 with 0.1 N NaOH using a Radiometer pH meter and automatic titrator(6).

Gastrin was prepared by the method of

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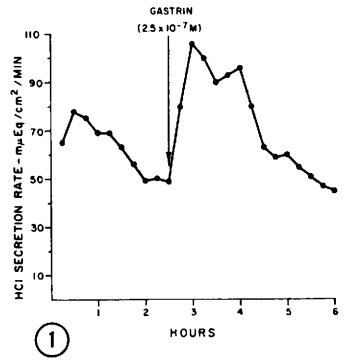

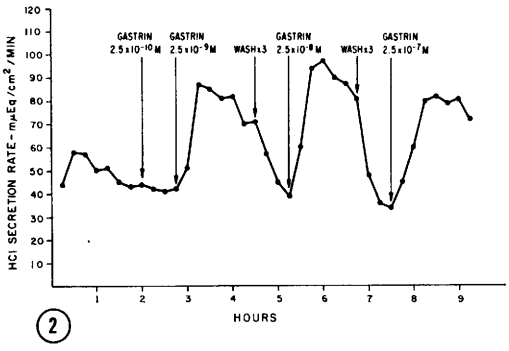

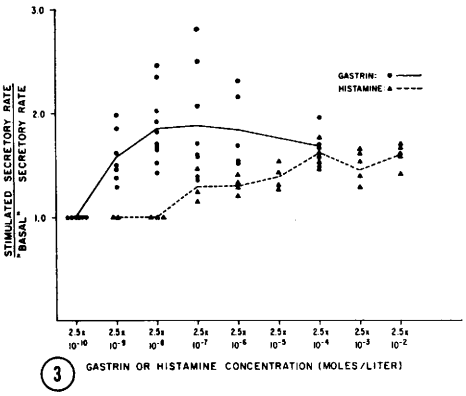

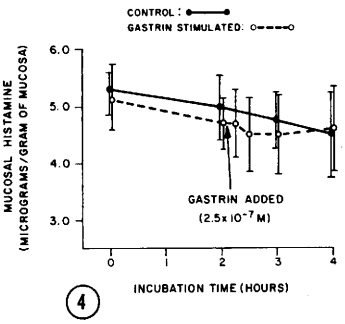

FIG. 1. Response of mucosal preparation to a single dose of gastrin (2.5 x 10^{-7} M) added to the nutrient side.

FIG. 2. Response of a single mucosal preparation to a series of increasing doses of gastrin. The nutrient solution was removed after the peak response to each dose had been established. The nutrient chamber was washed 3 times with nutrient solution, then refilled with fresh nutrient solution.

FIG. 3. Response of 30 mucosae to 71 doses of gastrin and 12 mucosae to 31 doses of histamine. Each dot or triangle represents an individual response at indicated dose level. The continuous lines connect mean responses at each dosage level.

FIG. 4. Mucosal histamine content in fragments of gastric mucosa incubated in nutrient solution with and without gastrin. Each point represents mean of 6 separate mucosae. Vertical lines are standard deviations.

Gregory and Tracy(1) except that the final product was not separated into gastrins I and II. For purposes of calculation, the gastrin was assumed to consist of equal parts of gastrins I and II and to have a molecular weight of 2154, *i.e.*, an average of the molecular weights of gastrins I and II.

To determine whether gastrin affected endogenous mucosal histamine content, gastrin was added to the nutrient solution containing small fragments of gastric mucosa. Histamine determinations were performed on the fragments before and after addition of gastrin and in paired control fragments incubated without gastrin. Mucosal histamine content was determined by the method of Shore, Burkhalter and Cohn(7).

Results and discussion. It was noted that all mucosal sheets incubated under the conditions described exhibited a spontaneous or basal secretion of acid. This initial secretory response declined gradually and reached a plateau at 90 to 120 minutes at which time gastrin or histamine was added to the nutrient side.

A typical response to gastrin is shown in Fig. 1. After addition of 2.5 $\times$ 10^{-7} M gastrin, the first measurable response occurred in 5 to 10 minutes. The peak response occurred at 30 minutes and secretion declined slowly thereafter.

In determining the threshold and peak

secretory responses to gastrin, increasing amounts of gastrin were added to the nutrient solution. After the maximal response to each dose had been recorded, the nutrient solution was exchanged. The results of a typical experiment are shown in Fig. 2. In no case did a dose of 2.5×10^{-10} M elicit a response. In the experiment illustrated, the threshold dose was 2.5×10^{-9} M and the peak response occurred at 2.5×10^{-8} M.

The secretory responses to 71 doses of gastrin in 30 bullfrog mucosae are shown in Fig. 3. The response to each dose is expressed as the ratio of the stimulated rate of secretion to the basal rate just prior to addition of gastrin. The threshold dose in all cases was 2.5×10^{-9} M and the maximal response occurred at 2.5×10^{-8} M or 2.5×10^{-7} M. By comparison, the response of 12 mucosae to 31 doses of histamine indicated that the threshold dose of histamine was 2.5×10^{-7} M and the maximal secretory dose was 2.5×10^{-4}. Therefore, the threshold dose of gastrin is 1/100 the threshold dose of histamine, and the dose of gastrin required to achieve a maximal secretory response is 1/1000 or 1/10000 the corresponding dose of histamine.

Addition of supramaximal doses of gastrin, *i.e.*, 2.5×10^{-4} M, stimulated acid secretion and did not inhibit basal or previously established gastrin- or histamine-stimulated secretion. Secretory inhibition following supramaximal doses of gastrin has been observed in the intact dog(1).

The mucosal content of histamine declined gradually during the course of 6 experiments (Fig. 4). No significant differences in histamine content were observed between the gastrin-stimulated and the non-stimulated mucosae.

Summary. Gastrin stimulates the secretion of acid by the isolated gastric mucosa of the bullfrog. The threshold dose is 2.5×10^{-9} M and the maximal secretory response occurs at 2.5×10^{-8} or 2.5×10^{-7} M. Supramaximal doses of gastrin failed to inhibit gastric secretion. Gastrin has no effect on the levels of endogenous mucosal histamine.

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Temperature Gradients Between Arterial Blood and Brain in the Monkey.* (30827)

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Blood serves as a common carrier of heat from one area of the body to another. In the homeotherm it provides the thermal stimulus for specialized thermosensitive receptors in the preoptic-anterior hypothalamic region and additionally removes the heat produced by neuronal and glial metabolism(1,2). While such generalizations are well established, a number of specific aspects of the thermal relationships between the brain and blood are

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