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The Lack of Effects of Somatostatin on Gastric Responses Induced by Electrical Vagal Stimulation

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CHO, C. H., S. W. CHEN, S. M. CHEN AND L. T. HO. *The lack of effects of somatostatin on gastric responses induced by electrical vagal stimulation.* PHARMACOL BIOCHEM BEHAV 19(6) 925–927, 1983.—The effects of somatostatin on ulcer formation, gastric acid secretion and histamine release were assessed during vagus nerve stimulation in rats. Direct electrical vagal stimulation significantly increased histamine release and acid output in gastric secretion but decreased mast cell counts in gastric glandular mucosa. Hemorrhagic ulceration on the gastric glandular mucosa was also observed. Somatostatin pretreatment (10 μg/kg) did not inhibit gastric ulcer formation, gastric acid secretion or histamine release induced by vagal stimulation. Cimetidine (an H_2 blocker) pretreatment, however, significantly decreased gastric acid secretion as well as ulcer formation. The present study indicates the direct vagal stimulation increases gastric acid secretion and ulcer formation. These effects are partially histamine dependent. Somatostatin did not inhibit histamine release induced by vagal stimulation and reflects the inability of the drug to prevent ulcer formation and gastric output under these conditions in rats. However, the inhibition of basal gastric acid secretion produced by somatostatin might be useful clinically in humans.

Somatostatin Vagal stimulation Gastric acidity Gastric ulcer Histamine

DIRECT vagal stimulation induced gastric ulceration has been shown to share a similar etiology with stress ulceration [1, 2, 3, 4]. Histamine release could be one of the main and common factors in the causation of these two types of ulceration in the stomach [1,2].

Somatostatin has been reported to prevent stress-induced gastric ulcer formation [7,11]. It is reasonable to believe that somatostatin could also prevent gastric ulceration and reduce histamine release from stomach during direct vagal stimulation. Thus, the present study was designed to determine the pharmacological effects of somatostatin on gastric histamine, acid secretion and ulcer formation produced by electrical vagal stimulation.

METHOD

General

Male Sprague-Dawley rats, weighing 180–220 g were starved for 24 hr before experimentation but allowed free access to water. The animals, anesthetized with IP injections of urethane (1.25 mg/kg), were kept warm with a heating lamp during the experiment. Tracheal cannulation was performed. All the experiments were carried out over a total period of 2 hr after which animals were sacrificed and their stomachs were examined for the severity of ulceration. Ulcers were measured by the greater diameter of the lesions. If the ulcers were less than 1 mm, two such ulcers were regarded as 1 mm of lesion.

Electrical Vagal Stimulation

Left cervical vagus was ligated to prevent any central effects of electrical stimulation. A platinum electrode was placed on the distal part of the ligature. An alternate electrical current of 5 V, 20 Hz, 2 msec was delivered by a stimulator (Grass model SD9) over a period of 2 hr.

Sham-operated controls had their vagus nerves similarly exposed and ligated, but no stimulation was given.

Intragastric Perfusion

The method was similar to the technique described by Cho *et al.* [4]. The esophagus was cannulated at the cervical level with a polythene tube (2 mm o.d.), inserted until its tip reached the cardiac end of the stomach. It was then secured by a ligature around the esophagus at the cannulation site. The pyloric end of the stomach was cannulated through a central abdominal incision. A polythene tube (4 mm o.d.) was inserted through a small incision in the duodenum until its tip just entered the stomach. The tube was gently tied in place with a ligature over the duodenum before exteriorizing the distal end through an incision in the flank.

Normal saline was perfused through the esophageal tube at a rate of 6 ml/15 min and the perfusate was collected from the exteriorizing end of the pyloric tube. Total acid in each sample was determined by titration to pH 7.4 with 0.01 N NaOH. One ml of the perfusates were used to determine the histamine content [6].

TABLE 1

EFFECTS OF VAGAL STIMULATION (5 V, 20 Hz, 2 MSEC) ON HISTAMINE LEVEL IN GASTRIC SECRETION AND MAST CELL COUNT IN GASTRIC MUCOSA OF RATS WITH PYLORUS-LIGATION FOR 2 HOURS

Treatment	No. of rats	Histamine level in gastric secretion (μg/2 hr)	Glandular mast cell count/40 o.i.f.	
			mucosa	submucosa
Non-stimulated sham-operated group	12	1.10 ± 0.14	100.1 ± 9.7	57.0 ± 5.5
Vagal stimulated group	12	1.54 ± 0.09*	54.7 ± 8.3†	32.9 ± 4.2†

The values shown are the means ± SE. o.i.f.=oil immersion fields.
*$p<0.02$, †$p<0.01$ when compared with the corresponding value in non-stimulated sham-operated group.

Pylorus-Ligation and Examination of Gastric Mast Cells

In the separate experiment, pylorus was ligated instead of perfused with normal saline. After 2 hours of vagal stimulation, gastric secretion was collected and assayed for its histamine content [6]. The glandular portion of the stomach where the ulcers were located was fixed in a freshly prepared aqueous solution of 4% lead acetate (E. Merck) w/v for 2 days. Sections (7 μ thick) were made by cutting the paraffin block vertical to the mucosal surface and staining with an aqueous solution of 0.5% toluidine blue (E. Gurr Ltd) w/v. Mast cell counts were expressed as the number of granulated metachromatically stained mast cells seen in 40 adjacent oil immersion fields (magnification 1000×) in the following areas (a) immediately below and parallel to the mucosal surface epithelium (mucosal count), (b) in the submucosa (submucosal count).

Drugs

Synthetic cyclic somatostatin (Serono), 10 μg/kg and cimetidine (SKF) 25 mg/kg were injected subcutaneously 15 min before vagal stimulation. Similar volumes of saline were given to the controls by the same route.

Statistical Analysis

The data were analysed by means of Student's t-test.

TABLE 2

EFFECTS OF SOMATOSTATIN AND CIMETIDINE PRETREATMENT ON GASTRIC ULCERATION INDUCED BY VAGAL STIMULATION (5 V, 20 Hz, 2 MSEC) IN GASTRIC PERFUSED RATS FOR 2 HOURS

Pretreatment (SC)	No. of rats	Ulcer index (mm)
A. Non-stimulated sham-operated groups		
Saline 1 ml/kg	6	0.8 ± 0.4
Somatostatin 10 μg/kg	6	0.5 ± 0.3
Cimetidine 25 mg/kg	6	0.4 ± 0.3
B. Vagal-stimulated groups		
Saline 1 ml/kg	9	5.7 ± 1.4†
Somatostatin 10 μg/kg	8	4.8 ± 1.6*
Cimetidine 25 mg/kg	7	1.2 ± 0.2‡

The values shown are the means ± SE.
*$p<0.05$, †$p<0.02$ when compared with the corresponding value in non-stimulated sham-operated group.
‡$p<0.02$ when compared with the corresponding value in saline-pretreated vagal-stimulated group.

RESULTS

Direct electrical vagal stimulation for 2 hours significantly increased histamine level in gastric secretion accompanied with decreasing in mast cells numbers in gastric glandular mucosa and submucosa of pylorus-ligated rats (Table 1). These changes indicate that mast cells could be the main storage cells for histamine in stomach. Hemorrhagic ulcers were also observed in the glandular portion but not in the ruminal part of the stomachs after 2 hours of continuous vagal stimulation (Table 2). The severity of ulceration was markedly prevented by cimetidine ($p<0.02$) but not by somatostatin pretreatment (Table 2). Cimetidine also inhibited the increase of gastric acid secretion induced by vagal stimulation, whereas gastric acid secretion in somatostatin treated animals was not different from the saline-pretreated controls during vagal stimulation (Fig. 1). Cimetidine and somatostatin significantly reduced the basal acid secretion of the stomachs during the 2nd, 3rd and the 4th collection periods ($p<0.05$). However, basal gastric acid secretion at the 5th collection period was only significantly decreased by cimetidine. Histamine in gastric perfusate was also measured in the 2-hour vagal stimulated rats. Cimetidine and somatostatin pretreatment did not affect the histamine changes in gastric perfusate during vagal stimulation (Fig. 2).

DISCUSSION

Direct vagal stimulation produces gastric ulceration and gastric acid secretion which have been postulated to be due to excessive histamine release from the stomach [5]. This suggestion has been substantiated by the decreased stomach mast cell count after vagal stimulation [1]. The present study not only confirms that mast cell counts in the gastric glandular mucosa and submucosa were decreased, but also shows that histamine content in gastric secretion in pylorus-ligated rats was increased. Pylorus ligation could be an additional stress exerted on the stomachs. The effects on histamine and mast cell count might depend on an interaction between pylorus ligation and vagal stimulation. However, the latter

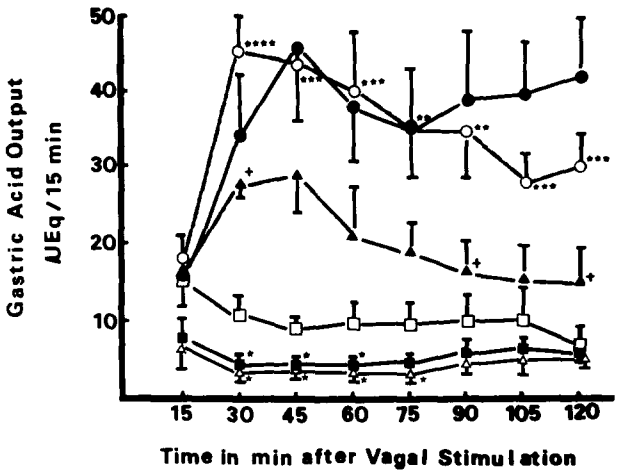

FIG. 1. Effects on gastric acid output during experiments. Various treatment groups are indicated as follows: saline pretreated-vagus stimulated=○; somatostatin pretreated-vagus stimulated=●; cimetidine pretreated-vagus stimulated=▲; saline pretreated-non-stimulated=□; somatostatin pretreated-non-stimulated=■ and cimetidine pretreated-non-stimulated=△. $^{*}p<0.05$, $^{**}p<0.02$, $^{***}p<0.01$, $^{****}p<0.001$ when compared with the corresponding value in saline pretreated-non-stimulated group. $^{+}p<0.05$ when compared with the corresponding value in saline pretreated-vagus stimulated group.

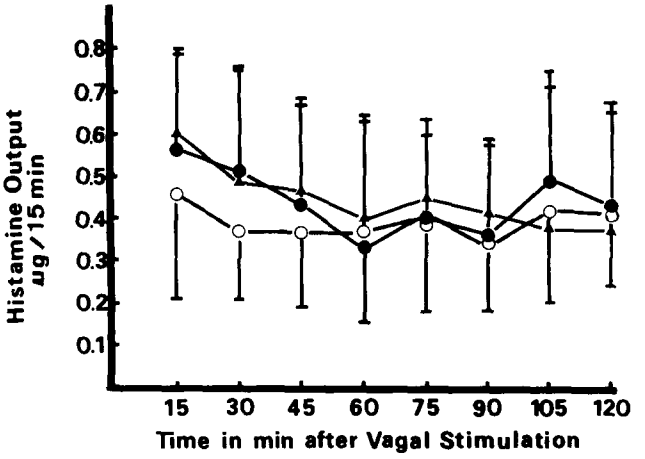

FIG. 2. Effects of vagus stimulation on histamine levels in gastric secretions for saline pretreated (○); somatostatin pretreated (●) and cimetidine pretreated (▲) animals.

could be largely responsible for these changes. Cimetidine pretreatment at the present dosage partially decreased acid output and ulcer formation but did not alter histamine levels in gastric secretion during vagal stimulation. Thus, one could conclude that histamine is partially involved in vagal stimulation-induced acid secretion and ulcer formation in the present study.

In the present experiment, somatostatin pretreatment using the dose and injection route shown to be effective in previous studies [7, 8, 10], did not affect histamine release, gastric acid secretion or ulcer formation induced by vagal stimulation. Thus, the protection of somatostatin against stress-induced gastric ulceration [7,11] is not histamine and acid dependent. The protective effect could be due to other mechanisms which remain to be clarified.

Although it has been shown that somatostatin is a strong inhibitor of gastrin-stimulated acid secretion, it is a weak inhibitor of histamine in dog [9]. This may partially explain why somatostatin did not modify the acid secretion which depended more than 40% on histamine release during vagal stimulation. Somatostatin also does not alter the hyperfunction of parietal cells induced by stress [7]. This hyperfunction is suggested to be due to cholinergic overactivity and also is true for direct vagal stimulation effects in the present study. This phenomenum also could explain why somatostatin does not inhibit acid secretion produced by vagal stimulation.

Somatostatin pretreatment significantly reduced basal acid secretion which cannot be explained at the present time. The secretory processes of vagal stimulation and basal gastric secretion might be different. However, somatostatin induced reductions in basal and gastrin-induced gastric acid secretion might be useful in the treatment of acid hypersecretion in duodenal ulcer patients.

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