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Effects of prostaglandin E_1 on acid secretion, mucosal histamine content and histidine decarboxylase activity in rat stomach

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Summary

1. Prostaglandin E_1 inhibits basal and pentagastrin-stimulated gastric acid secretion. The mechanism of this action is not clear. One possible explanation might be that prostaglandin E_1 interferes with the local release or synthesis of histamine which has been proposed as the mediator of the effects of gastrin on the parietal cell.

2. A single injection of prostaglandin E_1 did not affect mucosal histamine content or histidine decarboxylase activity in the rat stomach. Pentagastrin lowered the histamine content and activated the histidine decarboxylase to the same extent in prostaglandin E_1-pretreated and in control rats. We conclude therefore that the inhibitory effect of prostaglandin E_1 on basal and pentagastrin-stimulated acid secretion is not caused by inhibition of histamine release or histamine synthesis.

3. Repeated injections of prostaglandin E_1 resulted in a significant elevation of the gastric histidine decarboxylase activity in normal but not in antrectomized rats. Conceivably, this increase in enzyme activity is secondary to prostaglandin E_1-induced inhibition of acid secretion, which will stimulate release of gastrin due to the rise in intragastric pH.

Introduction

Prostaglandin E_1 and E_2 are known to inhibit both basal and pentagastrin-stimulated gastric acid secretion in the rat (Shaw & Ramwell, 1968; Robert, Nezamis & Phillips, 1968; Main, 1969). The mode of action is not known. The inhibitory effect may be due either to a direct action on the parietal cell or to interference with the synthesis or release of a mediator of the parietal cell response. Histamine, which in the rat stomach is produced and stored in endocrine cells in the basal part of the oxyntic mucosa (Håkanson, 1970), has been proposed as the final mediator in the stimulation of the parietal cell (Kahlson & Rosengren, 1968). The administration of gastrin (and pentagastrin) has been shown to mobilize histamine from its storage site in the gastric mucosa and to increase the activity of the histamine-forming enzyme, histidine decarboxylase (Kahlson, Rosengren, Svahn & Thunberg, 1964). Feeding and vagal stimulation (insulin hypoglycemia) induce identical changes in histamine turnover (Kahlson *et al.*, 1964; Kahlson, Rosengren & Thunberg, 1967). These effects are probably secondary to the release of endogenous gastrin from the antrum (Håkanson & Liedberg, 1970, 1971a, 1972). In an investigation of the

mechanism of action of prostaglandin on acid secretion it was decided to make a preliminary study of the effects of prostaglandin E_1 on the pentagastrin-induced release of gastric mucosal histamine and on the pentagastrin-induced activation of gastric histidine decarboxylase. In addition the effects of prostaglandin E_1 on basal acid secretion, histamine content and histidine decarboxylase activity were studied.

Methods

Animals. Ninety-six male Wistar rats (weight 200–350 g) were used. Fourteen rats were fitted with chronic gastric cannulae, using a slight modification of the technique described by Bel, Levrat, Nesmos & Girard (1966). Antrectomy was performed on 7 rats by resection of the pyloric gland area, including the entire lesser curvature and half the glandular stomach on the greater curvature and the duodenal bulb. Gastrointestinal continuity was re-established by a gastro-duodenostomy end-to-end. All operated animals were allowed to recover from the operation for a minimum of 3 weeks. Before experiments on gastric secretion, the fistula rats were deprived of food but not water for 24 hours. For determination of the gastric histamine content and the histidine decarboxylase activity the animals were fasted for 48 h before they were killed.

Drugs. Pentagastrin (Peptavlon, ICI) was given in a dose of 250 μg/kg subcutaneously 1 h before the rats were killed. Prostaglandin E_1 (a gift from Upjohn, Mich., USA) was given subcutaneously in various doses and at times as described under **Results.**

Determination of gastric histamine and histidine decarboxylase activity

The stomachs were removed, cut open along the greater curvature and the mucosa gently cleaned with ice-cold 0·9% w/v NaCl solution (saline). The mucosa of the oxyntic gland area was scraped off and homogenized in 0·1 M phosphate buffer pH 6·9, to a final concentration of 100 mg (wet weight) per ml. Aliquots (0·5 ml) were taken for histamine assay: proteins were precipitated with 5% TCA and spun down, the clear supernatant was extracted with an n-butanol-chloroform mixture (3:2), and histamine was assayed fluorometrically (Håkanson, Rönnberg & Sjölund, 1972). The remainder of the homogenates were centrifuged at $10{,}000 \times g$ for 15 min and aliquots of the supernatant were incubated with carboxyl-^{14}C-labelled L-histidine (4×10^{-4}M, 1·3 mCi/mM, New England Nuclear) in the presence of pyridoxal-5′-phosphate (10^{-5}M) and glutathione (5×10^{-4}M) at 37° for 1 h under nitrogen. The $^{14}CO_2$ produced during the reaction was released by acidification and trapped on a filter paper, immersed in Protosol (New England Nuclear, NEN Chemicals) for measurement by liquid scintillation counting. For details see Håkanson (1970).

Collection and determination of gastric acid secretion. The unanaesthetized fistula rats were immobilized in Bollman type cages, the cannulae opened and the stomachs rinsed with saline. Ten ml saline was given subcutaneously to replace fluid loss during the experiment. The cannulae were allowed to drain freely for one hour, and 2 one-hour samples of basal secretion were collected. Prostaglandin E_1 (250 μg/kg) or saline in a similar volume (0·5 ml/kg) were given s.c. after which 4 one-hour samples were collected. The volume was measured and the hourly acid output was determined by titration with 0·02 N NaOH, using phenolphthalein as indicator.

Results

Effect of prostaglandin E_1 on basal acid gastric secretion

With the 3 dose levels examined (50, 200 and 500 μg/kg) prostaglandin E_1 caused a significant and dose-dependent inhibition of the basal acid secretion (Fig. 1). The duration of inhibition was 2 h for the 50 and 200 μg/kg dose and 3 h for 500 μg/kg. No adverse effects were noted after the two lower doses; after the high dose some rats had slight diarrhoea.

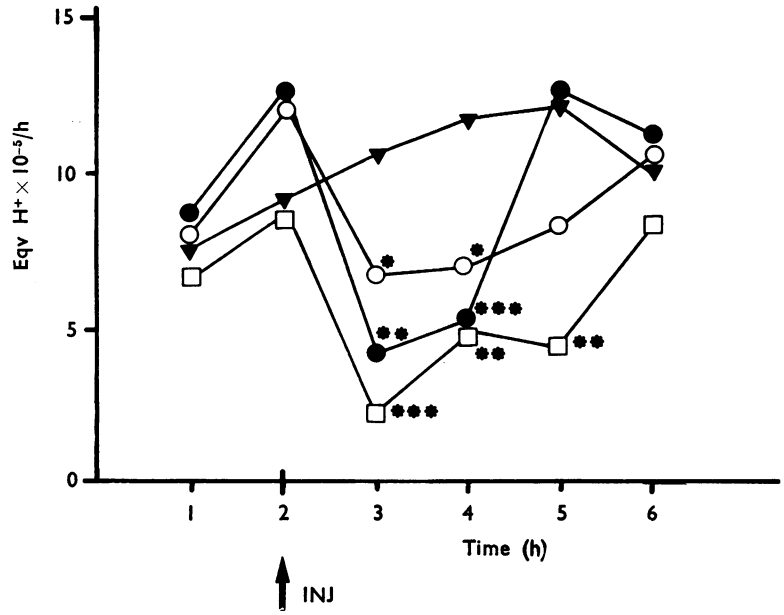

FIG. 1. Effect of 0·9% saline (▼——▼) and prostaglandin E_1 in doses of 50 (○——○), 200 (●——●) and 500 μg/kg (□——□) on acid secretion in gastric fistula rats. Mean values, at least 5 animals in each group. Significant differences between saline-treated and prostaglandin E_1-treated rats are shown by * for $0{\cdot}05>P>0{\cdot}01$, by ** for $0{\cdot}01>P>0{\cdot}001$ and by *** for $P<0{\cdot}001$, Student's t test.

TABLE 1. *Histamine content in rat gastric mucosa after various treatments*

Treatment	Histamine content μg/g, mean ± S.E.M. (*n*)
Saline	43·7±2·8 (10)
Prostaglandin E_1 250 μg/kg	42·2±3·1 (10)
Pentagastrin 250 μg/kg	32·2±3·2 (8)*
Prostaglandin E_1 250 μg/kg+pentagastrin 250 μg/kg	32·1±2·5 (12)**

Significant difference from saline-treated animals indicated by * for $0{\cdot}05>P>0{\cdot}01$ and ** for $0{\cdot}01>P>0{\cdot}001$, Student's t test.

TABLE 2. *Histidine decarboxylase activity in rat gastric mucosa after various treatments*

Treatment	Histidine decarboxylase activity, CO_2 (pmol/mg)/h, mean ± S.E.M. (*n*)
Saline	4·2±0·8 (12)
Pentagastrin 250 μg/kg	18·7±2·2 (11)***
Prostaglandin E_1 250 μg/kg	4·1±0·8 (15)
Prostaglandin E_1 250 μg/kg+pentagastrin 250 μg/kg	16·2±2·5 (20)***
Antrectomy+prostaglandin E_1 250 μg/kg	3·7±0·9 (4)

Significant difference from saline-treated rats indicated by *** for $P<0{\cdot}001$.

TABLE 3. *Effect of repeated injections of prostaglandin E_1 on histidine decarboxylase activity in normal and antrectomized rats*

Treatment	Histidine decarboxylase activity, CO_2 (pmol/mg)/h, mean ± S.E.M. (*n*)
Normal rats, saline × 6	5·8±1·2 (5)
Normal rats, prostaglandin E_1 250 μg/kg × 6	14·3±2·0 (12)**
Antrectomy, prostaglandin E_1 250 μg/kg × 6	6·9±0·9 (3)

Significant difference from normal rats is indicated by ** for $0{\cdot}01>P>0{\cdot}001$.

Effects of prostaglandin E_1 on gastric histamine content and histidine decarboxylase activity

Rats given a single dose of 250 μg/kg prostaglandin E_1 and killed 1 h after the injection had the same histamine content and histidine decarboxylase activity as control rats given saline (Tables 1 and 2). However, 6 hourly repeated injections of prostaglandin E_1 (250 μg/kg) caused a significant elevation of the histidine decarboxylase activity as compared to saline-injected controls (Table 3). This enzyme-activating effect was abolished by antrectomy (Table 3). The histamine content after repeated injections of prostaglandin E_1 (43·8 ± S.E.M. 5·9 μg/g; $n=5$) was not significantly different from that in the saline-treated controls (40·2 ± 1·9; $n=5$).

Effects of prostaglandin E_1 on pentagastrin-induced reduction of gastric histamine and activation of gastric histidine decarboxylase

Prostaglandin E_1 was given s.c. in doses of 250 μg/kg 5 min before the s.c. injection of pentagastrin (250 μg/kg) and the animals were killed 1 h after the injection of pentagastrin. This pretreatment did not prevent pentagastrin from lowering the mucosal histamine content and raising the histidine decarboxylase activity (Tables 1 and 2).

Discussion

The concept of histamine as the final common mediator for the stimulation of gastric acid secretion (MacIntosh, 1938; Kahlson & Rosengren, 1968) has recently been challenged by several authors (for references see Håkanson & Liedberg, 1971a; Johnson, 1971; Waton, 1971). However, it is an indisputable fact that stimulation of gastric acid secretion in the rat, by a number of stimuli, is accompanied by a reduction of mucosal histamine and an increase in the histidine decarboxylase activity (Kahlson *et al.*, 1964, 1967). The physiological significance of these changes in the histamine turnover is unknown. The present study does not lend support to the hypothesis that impaired synthesis or release of gastric histamine plays any role in the inhibition by prostaglandin E_1 of basal or pentagastrin-induced acid secretion, since pentagastrin still evoked histamine release and activation of histidine decarboxylase in the prostaglandin E_1-pretreated rats. The finding of an increased histidine decarboxylase activity after prolonged treatment with prostaglandin E_1 may be explained by the inhibition of acid output, resulting in an increase in intragastric pH and in a facilitated release of endogenous gastrin from the antrum, the released gastrin being responsible for the activation of the enzyme. This hypothesis is supported by the observation that the enzyme activation seen after repeated doses of prostaglandin E_1 is abolished by antrectomy. Inhibition of acid secretion by

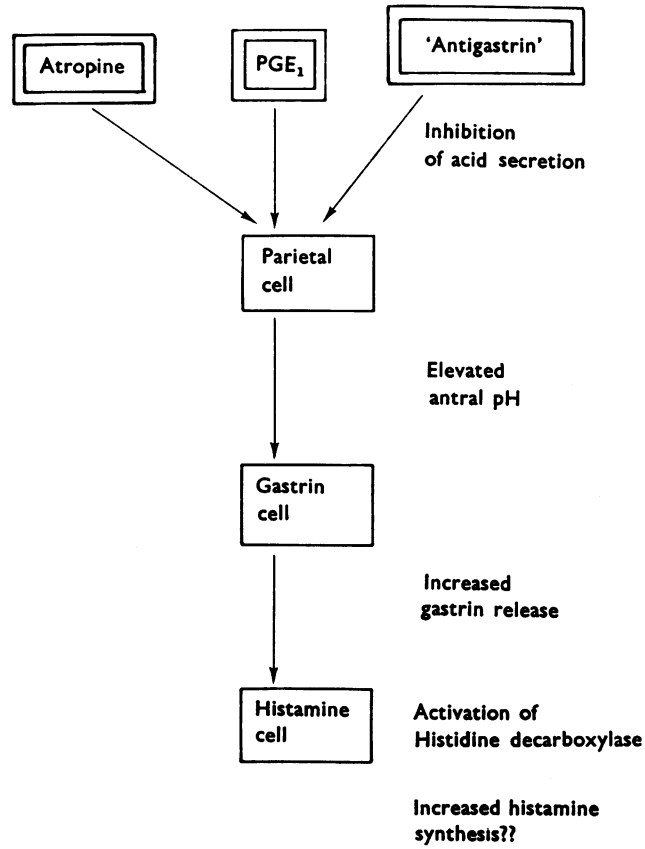

FIG. 2. Proposed chain of events following inhibition of acid secretion and leading to activation of rat gastric histidine decarboxylase.

other agents, such as atropine and SC 15396 (' Antigastrin '), also results in histidine decarboxylase activation in normal, fasted rats, but not in antrectomized rats (Håkanson & Liedberg, 1971b and c). Figure 2 gives a schematic presentation of our hypothesis.

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