Effect of cysteamine on gastroduodenal mucosal histamine in rat

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SUMMARY Cysteamine administration to rats is followed by a high incidence of peptic ulceration. The aim of the present study was to investigate the effect of cysteamine on gastric and duodenal mucosal histamine and gastric mucosal histamine formation capacity. After a four hour fast, cysteamine in doses of 50, 100, 200, 300, 400, and 500 mg/kg bodyweight was injected subcutaneously to male Wistar rats; saline injection was used as control. After 24 hours the animals were killed; the stomach and duodenum were removed and examined for ulceration. Mucosal biopsies were taken for histamine studies. Gastric and duodenal ulceration tended to appear with increasing incidence with higher doses. A direct correlation was found between the dose of cysteamine and gastric mucosal histamine ($p < 0.02$), and duodenal mucosal histamine ($p < 0.05$). Further, a direct relationship was found between gastric mucosal histidine decarboxylase activity and the dose of cysteamine ($p < 0.05$). Gastric mucosal histamine and histidine decarboxylase activity showed a direct correlation ($p < 0.001$). Gastric and duodenal mucosal histamine and gastric mucosal histamine formation capacity were higher in rats with ulcers than in controls and rats without ulcers. In rat, cysteamine induces dose related changes in mucosal histamine and histidine decarboxylase activity. These changes are related to ulcer formation; histamine may be involved in the pathophysiology of cysteamine induced ulcer formation.

Cysteamine hydrochloride was first shown to produce duodenal ulcers in rats in 1973.¹ Since then several studies have been carried out concerning the mechanisms involved, and the model has been used to test anti-ulcer agents.²⁻⁹

Administration of cysteamine hydrochloride is followed by increased acid output and gastric acidity,^{2,3} increased serum gastrin,^{4,5} increased pepsin activity,⁶ decreased gastric emptying^{7,8} and changes in intraluminal⁹ and intracellular duodenal mucus.^{10,11}

So far, two studies have been published concerned with the possible role of histamine in the development of ulcers. In one study injection of cysteamine (100 mg/kg bodyweight) was not followed by any change in gastric mucosal histamine.² On the other hand, in another study histamine depletion before cysteamine administration was followed by a reduction in the intensity of ulcers.¹²

The amount of gastric mucosal histamine reflects

the acid secretory capacity and the acid secretory state. Further, the histamine formation capacity (histidine decarboxylase activity) seems to be the best measure of histamine metabolism in rats.¹³

The aim of the present study was to investigate the effect of cysteamine on gastric and duodenal mucosal histamine and gastric mucosal histamine formation capacity in rats.

Methods

MATERIALS

After four hours of fasting with access to water, cysteamine hydrochloride was injected subcutaneously into male Wistar rats with a bodyweight of 220 to 260 g. A single dose of 50, 100, 200 or 300 mg/kg bodyweight was injected. Doses of 400 and 500 mg/kg bodyweight were given as two injections separated by a four hour interval. Saline injection was used as control. The rats were then left in separate mesh bottom cages with free access to water. After 24 hours the rats were killed by an intraperitoneal injection of an overdose of barbiturate.

The stomach and duodenum were then removed

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Received for publication 27 January 1983

and examined for macroscopic ulceration. Only macroscopic ulcers were accepted as lesions. Mucosal biopsies from the stomach and duodenum were taken for histamine studies and further specimens were taken for histological evaluation of the macroscopic lesions, and also routinely from the stomach and duodenum. The histological assessment is not reported here.

HISTAMINE ASSAY

Histamine assay was based on the fluorimetric method of Rhode, Lorenz *et al.*¹⁴ and has been described in detail elsewhere.¹⁵

HISTAMINE FORMATION CAPACITY

The histidine decarboxylase activity was measured by a combination of two methods.^{16 17} Blank values were obtained by substituting L-histidine by D-histidine in the reaction mixture for each tissue sample.

STATISTICS

The Mann-Whitney U-test and Spearman's rank correlation test were used for statistical evaluation; *p* values less than 0.05 were accepted as significant.

Results

Gastric and duodenal ulcers tended to appear with increasing incidence with higher dose of cysteamine (Table).

A direct correlation was found between the dose of cysteamine and gastric mucosal histamine ($p < 0.02$) (Fig. 1). The dose of cysteamine also correlated with duodenal mucosal histamine ($p < 0.05$) (Fig. 2). A direct correlation was found between the dose of cysteamine and histamine formation capacity in the gastric mucosa ($p < 0.05$) (Fig. 3).

The results from control rats, rats which did not

Table Incidence of ulcers after different doses of cysteamine

	Group						
	I	II	III	IV	V	VI	VII
	Number of rats						
	10	9	9	9	10	9	10
	Dose (mg/kg)						
	0	50	100	200	300	400	500
No ulcer	10	8	5	4	6	4	2
Duodenal ulcer (DU)	0	1	3	1	0	2	2
Gastric ulcer (GU)	0	0	1	3	1	0	0
DU and GU	0	0	0	1	3	3	6

Fig. 1 Gastric mucosal histamine in rats 24 hours after injection of cysteamine in different doses.

Fig. 2 Duodenal mucosal histamine in rats 24 hours after injection of cysteamine in different doses.

develop ulcers and rats with ulcers were compared. In control rats and rats which did not develop ulcers no differences were found in gastric or duodenal mucosal histamine or gastric histamine formation capacity. Gastric mucosal histamine and histamine formation capacity were both higher in rats with

Fig. 3 Gastric mucosal histidine decarboxylase activity in rats 24 hours after injection of cysteamine in different doses.

Fig. 4 Gastric mucosal histamine in control rats, rats which did not develop ulcers, and rats which did develop ulcers 24 hours after injection of cysteamine in doses from 50 to 500 mg/kg bodyweight.

ulcers than in rats without ulcers and controls (Figs. 4 and 5). The same applied to duodenal mucosal histamine (Fig. 6). A direct correlation was found between mucosal histamine and histamine formation capacity (Fig. 7).

Discussion

A number of different mechanisms seem to be involved in the development of gastric and duodenal ulceration after administration of cysteamine. Increased acid and pepsin load^{2,3,6} combined with

Fig. 5 Histidine decarboxylase activity in control rats, rats which did not develop ulcers and rats which did develop ulcers 24 hours after injection of cysteamine in doses of 50 to 500 mg/kg bodyweight.

Fig. 6 Duodenal mucosal histamine in control rats, rats which did not develop ulcers and rats which did develop ulcers 24 hours after injection of cysteamine in doses of 50 to 500 mg/kg.

decreased mucosal resistance or at least changes in mucus substances and duodenal mucosal cell morphology are involved.⁹⁻¹¹ Gastric stasis follows cysteamine administration,⁸ and this alone may be ulcerogenic.

Histamine is a well recognised secretagogue in rats as well as in other animals and man,¹³ and histamine acts as an acid secretory mediator. Further, the gastric mucosal histamine content reflects the acid secretory state and capacity of the stomach. The histamine formation capacity seems to be correlated even more closely to the acid secretory

Fig. 7 The relationship between HFC and histamine in gastric mucosa.

capacity than histamine alone.¹³ A study has recently shown that serum gastrin increases within 30 minutes of the injection of cysteamine.⁵ Gastrin is also well known as an acid secretion stimulator. In rat, gastrin mediates the release of histamine and activates the gastric enzyme system for histamine formation.¹³

Development of ulcers after cysteamine is dose-dependent and is also associated with the increase in serum gastrin. In our study we found a dose-dependent increase in the mucosal histamine content as well as in the histamine formation capacity. Our present study did not include studies of gastrin and the changes we found apply only to the situation 24 hours after administration of cysteamine. The relationship between serum gastrin and the mobilisation of gastric histamine in response to cysteamine is the subject of another study. In that study, done in separate animals and to be reported elsewhere, cysteamine injection was followed by a transient rise in serum gastrin lasting less than four hours. This is consistent with the findings of Kirkegaard *et al.*⁵ More prolonged study of the gastrin response to cysteamine will be necessary to assess its importance in the aetiology of ulceration in a model such as ours; at present an important role would seem unlikely.

Gastric mucosal histamine and gastric mucosal histamine formation capacity were increased in rats with ulcers compared with both controls and rats which did not develop ulcers. The same changes in mucosal histamine were found in the duodenum. This might indicate that cysteamine acts by a general histamine release or production. Further studies into that problem are in progress. Our study confirmed a close relationship between histamine

formation capacity and histamine in the gastric mucosa.

The relationship between the development of peptic ulcers and the amounts of histamine in the gastric mucosa differs among species. In the present rat model, ulcers are accompanied by an increase of gastric mucosal histamine. In a rabbit model for the production of gastric lesions by intraperitoneal injection of adrenaline, severe mucosal changes are associated with a fall in gastric mucosal histamine concentration.^{18,19} Similarly, patients with peptic ulcer disease have less mucosal histamine than normal controls or patients with healed peptic ulcer.²⁰

In rats, histamine is actively synthesised by the enzyme, histidine decarboxylase. The mucosal histamine turns over at a rapid rate and is maintained at a constant concentration by rapid adjustment of histamine synthesis.²¹ In man and other mammalian species the significance of histamine synthesis cannot be readily shown. Thus depletion of histamine stores may be more readily detectable in man. In rats, cysteamine hydrochloride induces dose-related changes in gastric and duodenal mucosal histamine and in gastric mucosal histidine decarboxylase activity. These changes are related to ulcer formation, and suggest that histamine is involved in the pathophysiology of experimental ulcer formation after cysteamine administration.

We wish to thank M Blount, S K Li, A Brown, J Barr, and J Batten for skilled technical assistance, and Mrs Iris Fisher for typing the manuscript. Financial support was given by the Wellcome Trust.

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