Gastric Secretion in the Rat Following Histidine Loading

It is believed by some, that in the rat stomach, gastrin-induced gastric secretion and histamine (Hm) release, are coupled[1–3]. In addition, there is an increase in Hm formation through gastrin-induced activation of histidine decarboxylase (HdD), the rate limiting step in Hm synthesis[2–4]. Furthermore, in the rat oxyntic gland area (OGA), Hm is probably present in several pools[4,5], at least one of which is sensitive to gastrin-induced reduction. (However, it is not known whether Hm is partially reduced, or depleted from the pool following this secretagogue). We have previously shown[6], that we can increase Hm levels in the OGA area in the rat by giving either large doses of Hm, or its immediate precursor amino acid L-histidine (Hd). Presumably by these drugs, the gastrin-sensitive pool is replenished. We were interested in this study, to see whether gastrin-stimulated gastric secretion was normal or not after augmenting OGA Hm levels by giving a large Hd load.

Materials and methods. Male Sprague-Dawley rats[7] weighing 340.8 ± 3.6 g were used. They were housed as described previously[8] and fed commercial laboratory chow[9].

Gastric secretion was collected in 2 h pylorus-ligated rats[8] following a 40 h fast. Body weight prior to the 40 h fast period was used for the calculation of drug doses, and for calculation of gastric secretion/100 g. Drugs used were as follows: L-Histidine[10] 1000 mg/kg i.p.; pentagastrin (PG) butyloxycarbonyl-β-ala-try-met-asp-phe amide[11], 200 μg/ml/kg/h s.c.; 0.85 g/100 l w./v. NaCl, 1.0 ml/kg/h s.c. Techniques of collection and analysis of gastric secretion have been described previously[8]. Hm and HdD were measured as described previously[12–15].

Results and discussion. PG in control rats produced significant stimulation of gastric secretion (Table). In addition, PG depressed OGA Hm levels ($P < 0.01$) and increased OGA HdD activity ($P < 0.001$). Following the Hd load, PG-induced secretion remained unchanged; however, OGA Hm levels were depressed ($P < 0.005$) but HdD activity was not increased.

The results presented here show that a Hd load of 1000 mg/kg can effectively block PG-induced activation of HdD, but not PG-induced gastric secretion. In addition, basal gastric secretion was marginally higher, and PG-stimulated gastric secretion marginally lower, after the Hd load. These latter 2 trends, which occurred for all indices of gastric secretion were not statistically significant however, and the mechanisms of production are not clear.

We have previously shown that a large dose of Hd can block PG-induced activation of HdD[6]. It is well known that PG is a potent stimulant of gastric secretion[16], which may act, according to some, through release of histamine[1–3]. Release of rat OGA Hm by PG is impossible to completely quantitate, since even with repeated injections of PG, Hm levels never fall below a certain ceiling[4,17] even though gastric secretion continues to be stimulated. Present techniques cannot discriminate between the various tissue Hm pools[4,5], and thus, it is not clear after giving PG whether the PG-sensitive pool has been only partially or completely depleted by the hormone. Since the Hd load increased OGA Hm stores, which are released in turn by PG, the failure of PG to produce a greater gastric secretory response after the Hd load adds further evidence to the theory that gastrin-induced gastric

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Gastric secretion following pentagastrin (PG) with or without a prior histidine loading

	Histidine-control NaCl			Histidine, 1000 mg/kg		
	NaCl (12)	PG (12)	*P*	NaCl (12)	PG (12)	*P*
Gastric juice volume (ml)	1.0 ± 0.1	2.8 ± 0.3	< 0.005	1.2 ± 0.2	2.4 ± 0.3	< 0.005
Gastric juice volume (ml/100 g)	0.21 ± 0.04	0.81 ± 0.07	< 0.001	0.35 ± 0.05	0.70 ± 0.09	< 0.005
Acid concentration (mEq/l)	63.5 ± 3.9	112.3 ± 5.1	< 0.001	68.2 ± 5.8	106.5 ± 4.9	< 0.001
Acid output (μEq/2 h)	18.2 ± 2.1	83.6 ± 10.2	< 0.001	24.5 ± 4.9	74.3 ± 11.0	< 0.001
Pepsin concentration (mg/ml)	0.50 ± 0.05	0.43 ± 0.06	NS	0.52 ± 0.04	0.40 ± 0.04	NS
Pepsin output (mg/2 h)	0.48 ± 0.10	0.87 ± 0.07	NS	0.61 ± 0.08	0.90 ± 0.09	NS
Oxyntic gland area histamine	58.9 ± 5.2	43.0 ± 4.0	< 0.01	78.9 ± 4.8	55.4 ± 4.1	< 0.005
Oxyntic gland area histidine decarboxylase	< 0.02	3.6 ± 0.8	< 0.001	< 0.02	< 0.02	NS

Data reported as mean values ± SEM. Histidine or Histidine-control NaCl injected i.p. PG and NaCl injected s.c. 5 min after the Histidine or Histidine-control NaCl, and again 60 min prior to sacrifice (injections given at 5 min and 60 min during the 2 h of pylorus-ligation). Histamine expressed as μg/g mucosa and Histidine decarboxylase as pico moles CO_2/mg mucosa/h.

secretion is not primarily mediated through Hm release[18]. The possibility that the absence of PG-induction of HdD could be due to high tissue levels of Hd causing interference in the in vitro assay system has been ruled out. Similarly, we have shown that the transient ether anesthesia required for pylorus-ligation does not prevent PG-induced activation of HdD[19].

It is now well established in the rat, that exogenously supplied Hm can be taken up by the OGA, and PGA[19,20]. In addition, large loading doses of the immediate precursor amino acid Hd can produce a similar increase in Hm stores in the OGA[6], but not in the PGA, since this tissue is low in Hd decarboxylating activity[4]. It is not certain however into which pool this exogenously supplied histamine is going.

In the OGA uptake is at least partially into the PG-sensitive pool, since some reduction can be produced by this hormone (Table). Storage of Hm following the Hd load probably occurs in addition in the enterochromaffin-like cell system[21]; these cells have the capacity to take up precursor amino acids and decarboxylate them in situ to the corresponding amine. It is almost certain that, speculatively, the enterochromaffin-like cell Hm pool, and the PG-sensitive pool, are not within the same unit, since uptake of preformed amine by the enterochromaffin-like cells does not occur[21,22].

Zusammenfassung. Nachweis, dass Pentagastrin (ICI-50123) 200 µg/ml/kg/h die Magensaftsekretion in der Shay-Ratte 2 h nach Ligatur des Pylorus erhöht. Die Histaminspeicher des Rattenmagens wurden reduziert und Histidindecarboxylase-Aktivität gesteigert. Verabreichung von 1000 mg/kg Histidin erhöht den basalen Histaminspiegel, jedoch wurde die Histidindecarboxylase-Aktivität bei normaler Magensaftsekretion nicht durch Pentagastrin induziert.

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New Observations in the Metabolism of Morphine. The Formation of Codeine from Morphine in Man

During opiate screening of urines from heroin addicts, spots were frequently noted on the thin-layer chromatograms (TLC) corresponding in Rf and iodoplatinate color to codeine. It was believed that in man, codeine might be formed as the *O*-methylated metabolite of morphine, which had previously been observed in rats and dogs by ELISON and ELLIOTT[1].

In our study, urines from 75 heroin addicts applying for admittance into the Acute Detoxification Study Unit at San Francisco General Hospital were analyzed for opiates according to the method of PARKER et al.[2]. 85% of these urines which were found to be morphine positive also contained codeine. The presence of codeine was established by mass spectrometry[3]. The codeine concentration was determined in 5 samples by TLC[4] and gas-liquid chromatography (GLC)[5]. Codeine was found to be present in amounts between 12 and 15% relative to morphine.

To check whether the codeine in the addict urines was arising from codeine-contaminated street heroin, the street heroin currently available in the San Francisco Bay Area was analyzed by TLC, GLC, and mass spectrometry. The samples showed almost exclusively absence of codeine or acetylcodeine.

These findings make it reasonable to assume that codeine is formed as a metabolite of morphine via-*O*-methylation. To demonstrate whether the quantity of codeine formed is related to chronic morphine administration, the following study was performed on morphine tolerant and non-tolerant humans.

Group 1, non-tolerant subjects. Route of administration of morphine: oral-subjects A1, A2, 2 male volunteers, each receiving 50 mg morphine sulfate. intravenous-subjects B1, B2, 2 aortic-bypass patients, receiving as i.v. drip over 30 min., 180 and 195 mg morphine sulfate.

Group II, tolerant subjects. Route of administration of morphine: oral-subject C, male cancer patient, receiving for the sixth day, 220 mg morphine sulfate in Schlesinger solution as daily dose. Intravenous-subjects D1, D2, 2 known heroin addicts, who volunteered to inject heroin. Heroin is known to rapidly deacetylate to 6-monoacetylmorphine and morphine[6]. The dose injected was unknown. Urine samples supplied before injection and samples of the heroin injected served as control for absence of codeine.

The urines of all subjects were collected for 24 h after heroin and morphine administration. In Group 1 (non-tolerant subjects), urinalysis for opiates was performed according to the above procedures[2–5]. To recover the small amounts of codeine observed, the extraction method was modified[7] and the extract analyzed as before. Codeine was found to be present in all urines of Group 1 in amounts of 0.7–0.9% relative to morphine. No significant differences between oral and i.v. administration were noted.

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[5] Varian Aerograph, Model 204 B gas chromatograph was used, equipped with 6 foot $^1/_8$ inch diameter, 5% SE-30 column, and flame ion detector. Inlet temperature 260 °C, column temperature program 180–280 °C at 8 °C/min. 60 ml/min He carrier gas; 30 ml/min H_2 to flame ion detector. Analysis according to the one-column acetylation procedure of MULE (Analyt. Chem. *36*, 1907 (1964).
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