Potent and specific blockade by AH 22216 of histamine-H2-receptor-mediated acid secretion in isolated rabbit gastric cells

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AH 22216 is a new histamine-H2-receptor antagonist which possesses a triazole ring. When compared to cimetidine, AH 22216 is about 100 times more potent (Ki = 0.21 x 10^-8 M) in inhibiting histamine-stimulated acid secretion in isolated rabbit gastric cells. These two antihistamines have no effect on carbachol-stimulated acid secretion in the system. The data indicate that AH 22216 interacts directly and specifically on the gastric H2-receptor of the parietal cell and are consistent with the reported pharmacological potencies of AH 22216 and cimetidine on histamine-induced gastric-acid secretion in vivo. AH 22216 could thus be a useful therapeutic agent in patients with peptic ulcers.

AH 22216 is a new H2-receptor antagonist (1) which differs in structure from cimetidine, the conventional H2-receptor antagonist (Fig. 1). Pharmacological studies in the dog have shown that AH 22216 is a long-acting inhibitor of histamine-stimulated gastric-acid secretion in vivo, with an oral potency 20-30 times that of cimetidine (1). The purpose of this study was to compare the anti-secretory activities of AH 22216 and cimetidine on gastric-acid secretion stimulated in vitro by histamine. The receptor specificity of the inhibitory action of AH 22216 (and cimetidine) is tested by combinations of the drugs with carbachol, a cholinergic agonist of gastric-acid secretion (2). The accumulation of the weak base aminopyrine in isolated rabbit fundic cells was measured as an indirect measure of hydrogen-ion production (3).

Materials and Methods

Chemicals

Collagenase (Type 1A), bovine serum albumin (fraction V), histamine dihydrochloride, and carbachol were from Sigma Chemical

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Fig. 1. Chemical structure of the H2-receptor antagonists AH 22216 and cimetidine.

Co. (St. Louis, MO). AH 22216 was a generous gift from Dr. Brittain (Glaxo Group Research Ltd., Ware, Hertfordshire, England), and cimetidine was kindly donated by Dr. Brimblecombe of Smith Kline and French Laboratories Ltd. (Welwyn Garden City, Hertfordshire, England). All other chemicals used were of the highest purity available.

Buffer solutions

The collagenase buffer solution for disaggregation of cells (buffer A) contained 132 mM NaCl, 5.4 mM KCl, 5 mM Na2HPO4, 1 mM NaH2PO4, 25 mM Hepes, 1.2 mM MgSO4, 1 mM CaCl2, 10 mM glucose, 0.2% bovine serum albumin, 0.02% phenol red, neutralized with HCl to pH 7.4. Buffer B was the Ca2+,Mg2+-free medium A, containing 2 mM EDTA and 0.4% bovine serum albumin, pH 7.4. The medium used for aminopyrine uptake (buffer C) contained 116 mM NaCl, 5.4 mM KCl, 1 mM NaH2PO4, 0.8 mM MgSO4, 1.8 mM CaCl2, 5 mM glucose, 0.2% bovine serum albumin, 0.2% phenol red, pH 7.4.

Gastric-cell preparation

Male rabbits of the New Zealand White strain, weighing approx. 3 kg, were fed a balanced diet of laboratory chow and given water ad libitum. Animals were killed by cervical fracture-dislocation and the fundic part of the stomach was immediately excised and rinsed in tap water and buffer A. Isolated gastric cells were prepared by a modification (4) of the procedure described by Soll for the preparation of isolated canine gastric cells (5). Briefly, this involved stripping and mincing the gastric mucosa, sequential exposure of the fragments to enzymatic digestion by collagenase, and chelation of divalent cations by EDTA. The entire cell preparation was done at 37°C under gentle stirring of mucosal fragments in mediums gassed with 95% O2 / 5% CO2 (pH 7.4). The minced mucosa was incubated for 15 min in buffer A containing 0.75 mg/ml collagenase. Then the mucosal fragments were washed twice in buffer B and incubated for 10 min in the same buffer. The settled material was resuspended in 10 ml of collagenase solution and digested for an additional 15 min. After centrifugation (3 min at 200 g), the pellet was resuspended in 15 ml.