Second-Generation Histamine H_2-Receptor Antagonists with Gastric Mucosal Defensive Properties

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Abstract: Unlike the earlier agents in this class, certain of the newer histamine H_2-receptor antagonists (so-called second-generation H_2-receptor antagonists) have recently been reported to promote gastric mucosal defenses. We review herein the structure, specificity, and mechanisms of these agents with a special focus on their cytoprotective/gastroprotective actions.

Key Words: Calcitonin gene-related peptide (CGRP), capsaicin-sensitive nerves, gastroprotective action, histamine H_2-receptor antagonists, lafutidine, mucin, roxatidine, structure-activity relationship.

1. INTRODUCTION

Acid related diseases, including gastric and duodenal ulcer, have plagued human and animals throughout recorded history. Schwarz's dictum [1]:, "no acid, no ulcer", has been challenged but rarely [2]. The discovery of the histamine H_2-receptor [3] and the subsequent introduction of cimetidine [4] are regarded as milestones in the history of peptic ulcer disease. During the 1980s, these gastric antisecretory agents became the first-line therapy in peptic ulcer disease, and led to an improved quality of life for many patients. An immense amount of work has been done in this field of pharmacological research [5-7]. More than 10,000 compounds have been synthesized and tested for their H_2-receptor antagonistic activity. Many H_2-receptor antagonists are being tested in clinical or pre-clinical trials.

While acid, pepsin, and *Helicobacter pylori* are thought to be major factors in the pathophysiology of peptic ulcer diseases, the importance of the mucosal defense system has also been emphasized [8-16]. Based on the belief that ulcers occur as a result of an imbalance between aggressive and defensive factors, such as mucus and mucosal blood flow, related to mucosal resistance, in Japan it is often treated with a combination of an acid suppressant (e.g. H_2-receptor antagonist or proton-pump inhibitor) and a mucosal protectant [17-22]. Recent prospective randomized trials indicated that the addition of a mucosal protectant significantly augmented gastric ulcer healing and symptom relief by cimetidine [20, 21]. Compared with the aggressive factors, little attention has been paid to the mucosal defensive factors in ulcer therapy and the role of the H_2-receptor antagonists in gastric mucosal protection has not been well characterized.

Recently, some of the newer H_2-receptor antagonists (so-called second-generation histamine H_2-receptor antagonists) have been reported to promote the gastric mucosal defense mechanisms [18, 23-41]. We review herein the structure, specificity, and mechanism of the histamine H_2-receptor antagonists with a special focus on their cytoprotective/gastroprotective actions.

2. STRUCTURE OF H_2-RECEPTOR ANTAGONISTS

The chemical structures of some frequently used H_2-receptor antagonists are shown in Fig. (**1**). All the known H_2-receptor antagonists comprise an aromatic ring with a flexible chain joined to a polar group. Despite considerable diversity, these compounds can be grouped into two main series according to the nature of the aromatic rings, namely five-membered and six-membered aromatic ring series. Cimetidine, ranitidine, nizatidine, and famotidine belong to the first-generation group characterized by a five-membered aromatic ring. Second-generation histamine H_2-receptor antagonists contain a six-membered aromatic ring, instead of a five-membered heterocyclic ring. The polar groups of cimetidine and ranitidine are bioisosteric groups. This means that they can be exchanged without great loss of activity [7, 42].

In the search for a selective antagonist for H_2-receptors, the structure of histamine served as the chemical starting point. Studies of its chemistry suggest that tautomerism of the imidazole ring of histamine may be involved as a proton-transfer agent [43]; consequently, the first H_2-receptor antagonists developed were derivatives of imidazole, chemical modification of which led to cimetidine. It was originally thought that a basic imidazole ring was required for H_2-receptor affinity. It later became apparent that the imidazole ring could be successfully replaced by other five-membered rings. Ranitidine contains a dimethylaminomethylfuran ring [44]. If the furan ring of ranitidine is replaced by a thiazole ring, nizatidine is the result; the H_2-receptor antagonistic activity of this drug is in the same range as that of ranitidine [45]. The guanidinothiazole moiety of famotidine, however, is a group with exceptionally high H_2-receptor affinity [46]. The guanidinothiazole group has been incorporated into

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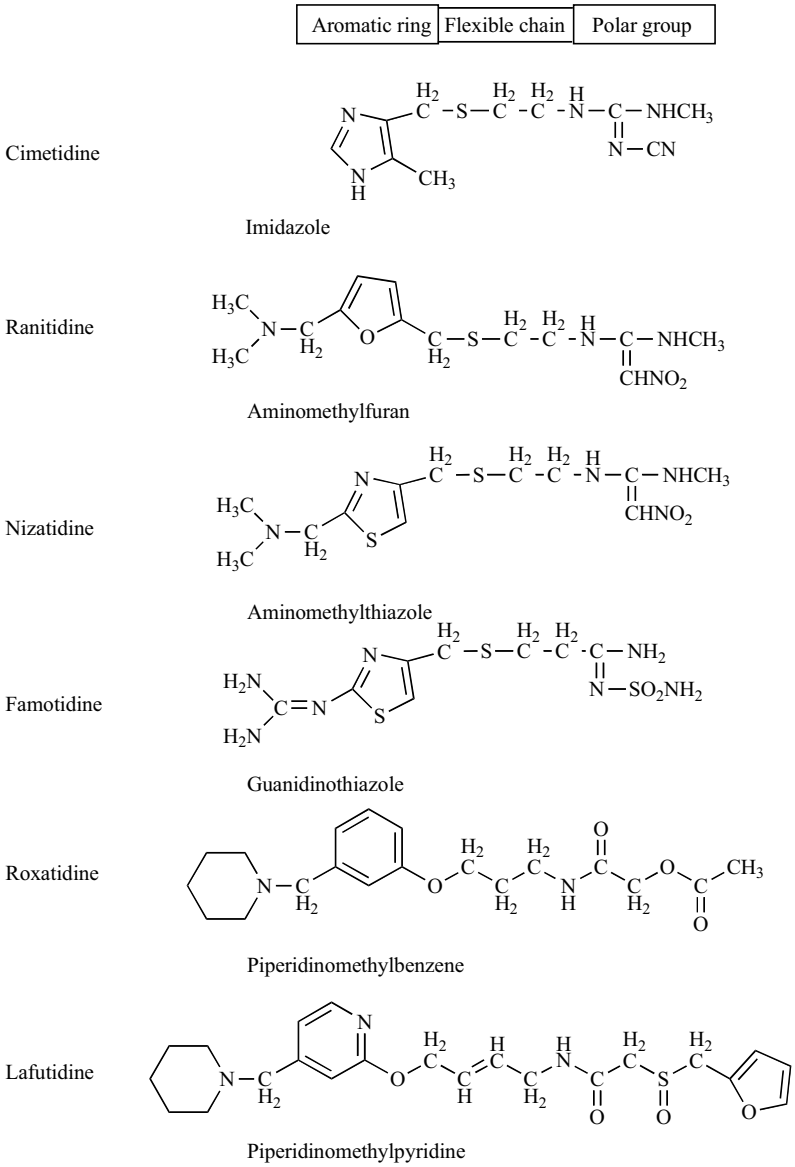


Fig. (1). Chemical structures of the histamine H_2-receptor antagonists.

many other compounds to produce potent H_2-receptor antagonists. This chemical group, therefore, is responsible for the potency and H_2 selectivity of famotidine. Moreover, molecular modelling studies by Schunack *et al.* have demonstrated that the best fit to the binding site H_2-receptor protein is seen with famotidine, but not cimetidine or ranitidine [42].

Roxatidine and lafutidine belong to the second-generation histamine H_2-receptor antagonist group characterized by a six-membered aromatic ring. These drugs contain piperidinomethylbenzene or piperidinomethylpyridine groups, instead of a five-membered aromatic ring, and also possess strong antisecretory properties. The negative logarithm of the molar concentration of antagonist in the presence of which the potency of the agonist is reduced 2-fold (pA_2), and antagonist dissociation constant (K_B) values of some H_2-receptor antagonists, derived from *in vitro* models are summarised in Table [45, 47-49]. Comparison of average K_B values obtained using guinea-pig atrium indicates that famotidine is about 50 times more potent than cimetidine and 10 times more potent than ranitidine. The K_B values of nizatidine and roxatidine acetate are in the same range as ranitidine. Inaba *et al.* demonstrated that the histamine dose-response curve could not show typical competitive antagonism in the guinea-pig right atrium in the absence or presence of increasing concentrations of lafutidine, indicating that it is an unsurmountable and selective histamine H_2-receptor antagonist [50]. In a [^{3}H]tiotidine binding study using a guinea-pig cerebral cortex preparation, the K_B values showed that the affinity of lafutidine was 2 and 80 times more potent than those of famotidine and cimetidine, respectively [50].

3. GASTROPROTECTIVE ACTIONS INDEPENDENT OF ANTI-ACID SECRETORY EFFECT

Gastric 'cytoprotection' refers to a reduction or prevention of chemically induced acute hemorrhagic erosions by

Table. Pharmacological Activities of Histamine H2-Receptor Antagonists

H_2-Receptor Antagonist	Species	Tissue	pA_2	K_B (¥10^{-6}mol/L)	Reference
Cimetidine	Guinea-pig	Gastric gland	6.41	0.389	[49]
		Right atrium	6.08	0.832	[47]
Ranitidine	Guinea-pig	Gastric gland	6.87	0.135	[49]
		Right atrium	6.75	0.178	[47]
Nizatidine	Rat	Uterus	7.10	0.079	[45]
Famotidine	Guinea-pig	Gastric gland	7.60	0.025	[49]
		Right atrium	7.74	0.018	[47]
			7.80	0.016	[48]
Roxatidine	Guinea-pig	Gastric gland	6.94	0.115	[49]
		Right atrium	7.00	0.100	[48]

compounds such as prostaglandin (PG) and SH derivatives without inhibiting acid secretion in rodents [51-53]. Since the concept of 'cytoprotection' was introduced, increasing attention has been paid to the effect of medications on the gastric mucosal defensive mechanisms. Although the exact mechanisms of the mucosal defense system are unknown, it involves one or more of the naturally occurring gastric mucosal defensive factors such as gastric blood flow, bicarbonate secretion, and mucin metabolism. For estimation of the gastroprotective function, many drugs have been investigated for their activity to protect the gastric mucosa from a variety of necrotizing agents such as ethanol and HCl. Of the six histamine H_2-receptor antagonists shown in Fig. (**1**), roxatidine and lafutidine have been reported to prevent the formation of gastric mucosal lesions induced by necrotizing agents in rats [24, 34, 39], and this effect may be due not only to the inhibition of aggressive factors such as acid, but also to the maintenance of defensive factors such as gastric blood flow, bicarbonate and mucus secretion. On the other hand, many reports have indicated that cimetidine and ranitidine lack a protective effect against necrotizing agent-induced gastric mucosal damage in the rat [39, 54].

Considerable information has accumulated about the gastroprotective function of the mucus that covers the mucosal surface of the stomach [55]. Mucin, a major component of gastric mucus, is a high-molecular-weight compound of unique structure and an important mucosal defensive factor [56]. Stimulation of mucin synthesis is shown to be closely related to mucosal protective activity [26]. Roxatidine and lafutidine, the second-generation histamine H_2-receptor antagonists characterized by a six-membered aromatic ring, have a stimulant effect on mucin biosynthesis in the rat gastric mucosa. In contrast, first-generation H_2-receptor antagonists such as cimetidine, ranitidine and famotidine, failed to stimulate mucin biosynthesis [18, 26].

The above findings have clarified that the second-generation H_2-receptor antagonists have a unique structure, and not only inhibit acid secretion but also enhance the protective mechanisms of the gastric mucosa. This should stimulate new interest in the chemical analysis of these drugs to determine the structural requirements for their gastroprotective actions.

4. STRUCTURE-ACTIVITY RELATIONSHIP FOR GASTROPROTECTIVE ACTIONS

Compared with the structural requirements of the acid-inhibitory mechanisms of the H_2-receptor antagonists, only a few detailed analyses have been reported of the structural aspects of their gastroprotective actions [27, 28, 37, 57] because of the complicated mechanisms of mucosal protection. However, the cardinal chemical features of lafutidine that determine its mucin biosynthetic activity, as a quantitative index of its gastroprotective action, were identified by considering the structural analogs (Fig. **2**) of this drug using an rat stomach organ culture system [28]. As shown in Fig. (**2**), compounds A, B and C bear the pyridine ring and compounds D and E bear the furan ring, which are commonly present in the structure of lafutidine. Mucin biosynthetic activity was increased by the addition of two pyridine derivatives, lafutidine and compound A. In contrast, compounds D and E, lacking a pyridine ring, failed to stimulate mucin biosynthesis. Similar results were obtained for compounds B and C, which have a pyridine ring but lack an amide structure. These results indicate that pyridine-based compounds containing an amide structure may be essential for activating the gastroprotective function. Furthermore, comparison with the histamine H_2-receptor antagonistic activities of these compounds suggests that H_2-receptor antagonism is not directly correlated with lafutidine-induced stimulation of mucin biosynthesis.

A more detailed analysis has been performed using roxatidine and its structural analogs to reveal the structural requirements of second-generation H_2-receptor antagonists for the stimulant effect on rat gastric mucin biosynthesis, particularly with regard to whether the cardinal features of roxatidine are only the six-membered aromatic ring and amide structure, and its relation to histamine H_2-receptor antagonism [27]. Of six compounds containing both a benzene ring and an amide structure, analogs A and B, but not C, stimulated mucin biosynthesis in a manner similar to that of roxatidine. These three compounds contain a piperidine ring (in-

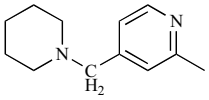
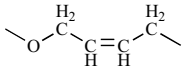
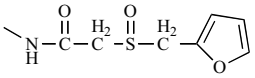
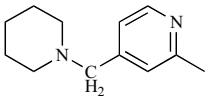
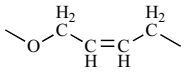
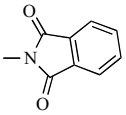
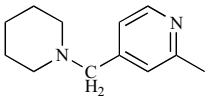
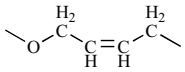
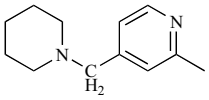
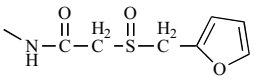
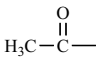
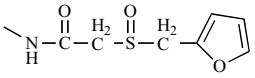
	A—B—C			Mucin biosynthetic activity	Histamine H_2-receptor antagonistic activity
	A	B	C		
Lafutidine				+++	+++
Compound A				++	-
Compound B			—OH	-	+
Compound C		Cl	-	-	-
Compound D	-	H		-	-
Compound E	-			-	-

Fig. (2). Structures and pharmacological activities of lafutidine and its analogs. Mucin biosynthetic activity was evaluated in an organ culture system of the rat stomach. Score was divided into the following 4 groups: -, no effect at 1 x 10^{-6} M; +, under 20% increase from the baseline at dose of 1 x 10^{-6} M; ++, significant 20-30% increase of biosynthetic activity ($p < 0.05$) at 1 x 10^{-6} M; +++, significant over 30% increase of mucin biosynthesis ($p < 0.01$) at 1 x 10^{-6} M. Histamine H_2-receptor antagonistic activity was investigated on the histamine-induced positive chronotropic responses in the isolated guinea-pig right atria. Score was divided into the following 4 groups: -, no effect at 1 x 10^{-5} M; +, under 70% inhibition at 1 x 10^{-6} M; ++, 70-90% inhibition at 1 x 10^{-6} M; +++, over 90% inhibition at 1 x 10^{-6} M. Data are taken from reference [28].

dicated by R_1 in Fig. **3**) attached to the benzene ring *via* a methylene bridge, but the length of the flexible chain (indicated by R_2 in Fig. **3**) of analog C differs from that of roxatidine. This means that the length of the flexible chain between the benzene ring and the amide structure is essential for this stimulation of mucin biosynthesis. Analogs D, E and F, having different ring structures or no ring structure at R_1 of the roxatidine molecule, failed to activate mucin biosynthesis. Analogs D, E and F contain the same flexible chain as roxatidine. Thus, the piperidine ring is also important for their activity. These results indicate that the structural requirements for the stimulant effect of roxatidine on mucin biosynthesis are not only the six-membered aromatic ring and amide structure, but the attachment of the piperidinomethyl group and the appropriate length of the flexible chain are also important for this function. With regard to their histamine H_2-receptor antagonistic properties, the six analogs were investigated using competition with the binding of the radiolabeled H_2-receptor antagonist [^{125}I]iodoaminopotentidine to membranes of the guinea pig striatum [58, 59]. All compounds except analog F in Fig. (**3**), displaced the specific [^{125}I]iodoaminopotentidine binding to histamine H_2-receptor sites. The relative potencies of these antagonists were: analog B > A > roxatidine > D > C > E. Compared with the IC_{50} value (concentration required to inhibit 50% of specific binding) for cimetidine obtained under similar experimental conditions, roxatidine and analogs A, B, C and D were 4.6, 9.5, 13.7, 1.6 and 2.7 times more potent than cimetidine, respectively [27]. These results suggest that H_2-receptor antagonism does not directly correlate with roxatidine-induced stimulation of mucin biosynthesis.

Structural differences between roxatidine and lafutidine have been examined with regard to the strength of their gastroprotective ability [37, 57]. Structures and pharmacological activities of *N*-[3-[3-(piperidinomethyl)phenoxy]propyl]acetamide derivatives are summarized in Fig. (**4**). Gastroprotective activity was evaluated in terms of the degree of inhibition of lesion formation by necrotizing agent in the rat stomach [37]. As shown in Fig. (**4**), *N*-phenoxypropylacetamide derivatives with thioether function expressed more potent gastroprotective activity than roxatidine. As a thioether moiety, the furfurylsulfinyl part was found to provide the optimal gastroprotective activity. In addition, oxidation of sulfur atoms tended to strengthen gastroprotective action (Fig. (**4**): compound 6 *vs*. 11, 12 and 20 *vs*. 21, 22).

Taken together, these data indicate that the structural requirements for mucosal protective activity in the second-generation H_2-receptor antagonists are their amide structure and six-membered aromatic ring, such as benzene and pyri-

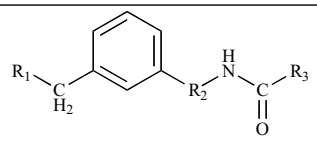
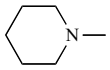
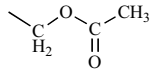
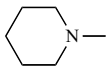
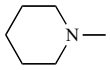
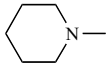
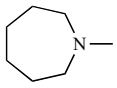
				Mucin biosynthetic activity	Histamine H_2-receptor antagonistic activity
	R_1	R_2	R_3		
Roxatidine		—$OCH_2CH_2CH_2$—		+++	+++
Analog A		—$OCH_2CH_2CH_2$—		+++	+++
Analog B		—$OCH_2CH_2CH_2$—	—CH_3	++	++++
Analog C		—$OCH_2CH_2CH_2CH_2$—	—CH_3	-	++
Analog D		—$OCH_2CH_2CH_2$—	—CH_3	-	++
Analog E		—$OCH_2CH_2CH_2$—	—CH_3	-	+
Analog F		—$OCH_2CH_2CH_2$—	—CH_3	-	-

Fig. (3). Structures and pharmacological activities of roxatidine and its analogs. Mucin biosynthetic activity was evaluated in an organ culture system of the rat stomach. Score was divided into the following 4 groups: -, no effect at 1 x 10^{-6} M; +, under 20% increase from the baseline at dose of 1 x 10^{-6} M; ++, significant 20-30% increase of biosynthetic activity ($p < 0.05$) at 1 x 10^{-6} M; +++, significant over 30% increase of mucin biosynthesis ($p < 0.01$) at 1 x 10^{-6} M. Histamine H_2-receptor antagonistic activity was investigated on the competition studies with [^{125}I]iodoaminopotentidine binding to membranes of the guinea-pig striatum. IC_{50} values (concentration required to inhibit 50% of specific binding) were determined and divided into the following 5 groups: -, IC_{50} > 4000 nM; +, 800 > IC_{50} > 500 nM (similar to cimetidine in the antagonism); ++, 500 > IC_{50} > 200 nM; +++, 200 > IC_{50} > 50 nM; ++++, 50 nM > IC_{50}. Data are taken from reference [27].

dine derivatives. The cardinal chemical features of roxatidine for the activation of mucin biosynthesis are the appropriate length of the flexible chain between the amide structure and the aromatic ring system bearing the methylpiperidinyl group at the meta position. The thioether function can confer increased gastroprotective activity on lafutidine.

5. MECHANISMS OF GASTROPROTECTIVE ACTION

Although the exact mechanisms that underlie the gastroprotective activity of the second-generation H_2-receptor antagonists are not well understood, recent findings suggest that the activation of capsaicin-sensitive sensory neurons is associated with their maintenance of gastric mucosal integrity [24, 25, 31, 32, 40, 60]. The gastrointestinal tract is known to possess a rich neural network, among which afferent neurons of extrinsic origin are reported to operate as the emergency protective system [61, 62]. The discovery of these sensory neuron functions was made possible by capsaicin, a pharmacological tool with which the activity of certain primary afferent neurons can be manipulated selectively [63]. Capsaicin is an excitotoxin that acutely stimulates a group of afferent neurons with unmyelinated (C) or thinly myelinated (Aδ) nerve fibers. This excitotoxic action is restricted to neurons with C- and Aδ-fibers because only these cells express receptor-binding sites (transient receptor potential vanilloid type 1: TRPV1) for capsaicin and structurally related ligands. The mammalian stomach, particularly in the lamina propria mucosa and submucosa, is densely innervated with capsaicin-sensitive afferent neurons [64, 65]. These neurons not only serve a sensory and afferent role, but also display a local effector function initiated by the release of neuropeptide mediators, such as calcitonin gene-related peptide (CGRP) and substance P [66], from their peripheral nerve endings. CGRP is reported to exhibit significant mucosal protective roles in the gastrointestinal tract [67-73]. The action of CGRP is in part mediated by endogenous nitric oxide (NO) [61, 70].

The gastroprotective action of lafutidine has been reduced or abolished by treatment with tetrodotoxin, $CGRP_{8-37}$, or chemical defunctionalisation of afferent nerves [31, 34, 74], indicating that capsaicin-sensitive nerves contribute significantly to the mechanisms underlying the actions of lafutidine [60]. Moreover, lafutidine has been shown to sig-

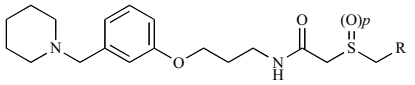
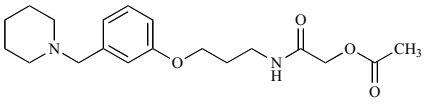
			Gastroprotective activity	Histamine H_2-receptor antagonistic activity
	P	R		
Compound 6	0		++	+++
Compound 11	1		+++	++
Compound 12	2		+++	+++
Compound 20	0		++	+++
Compound 21	1		+++	+++
Compound 22	2		+++	+++
Roxatidine			-	+++

Fig. (4). Structures and pharmacological activities of *N*-[3-[3-(piperidinomethyl)phenoxy]propyl]acetamide derivatives. Gastroprotective activity was evaluated in terms of the degree of inhibition of acidified ethanol-induced lesion formation of rat stomach. Score was divided into the following 4 groups: -, no effect at an orally administration dose of 30 mg/kg; +, inhibition of lesion formation at doses of 10-30 mg/kg; ++, significant inhibition of lesion formation ($p < 0.05$) at a dose of 10 mg/kg; +++, significant inhibition of lesion formation ($p < 0.01$) at a dose of 10 mg/kg. Histamine H_2-receptor antagonistic activity was investigated on the histamine-induced positive chronotropic responses in the isolated guinea-pig right atria. Score was divided into the following 4 groups: -, no effect at 1 x 10^{-5} M; +, under 70% inhibition at 1 x 10^{-6} M; ++, 70-90% inhibition at 1 x 10^{-6} M; +++, over 90% inhibition at 1 x 10^{-6} M. Data are taken from reference [37].

nificantly increase CGRP release in both experimental animal models and humans [25, 60, 75, 76]. Several reports indicate that the TRPV1 of capsaicin-sensitive afferent nerves may not contribute the CGRP release by lafutidine, suggesting the existence of yet unidentified sites for lafutidine other than TRPV1 on these nerves [24, 60]. The gastroprotective effects of lafutidine are decreased by treatment with NO synthase inhibitors or NO scavenger [60, 77], indicating the involvement of NO generation in lafutidine function. Similar results have been obtained with another second-generation H_2-receptor antagonist, roxatidine [27, 78].

Lafutidine has been shown to enhance the healing of gastrointestinal mucosal lesions in a manner independent of its antacid secretory action [34, 38, 79, 80]. However, lafutidine by itself does not have any direct effects on cell migration or proliferation. An earlier study demonstrated that lafutidine does not influence the impaired healing of epithelial wounds in RGM1 cells under *in vitro* conditions without neuronal innervations [32], again confirming the importance of sensory neurons in the healing-promoting action of this agent. Several studies show that luminal lafutidine stimulates capsaicin-sensitive afferent nerves *via* presumably direct diffusion rather than after its absorption from intestine followed by *via* circulation, suggesting the rapid local diffusion reaching to the afferents before H_2-receptor blockade from the circulation [81, 82]. Second-generation H_2-receptor antagonists such as lafutidine are thought to facilitate capsaicin-sensitive sensory afferent nerves and exert gastroprotective effects through CGRP and in part *via* NO release in the stomach.

6. SUMMARY AND PERSPECTIVES

The findings reviewed here put a new perspective on the ability of second-generation H_2-receptor antagonists to strengthen gastric mucosal defense in a manner independent of their histamine H_2-receptor antagonistic activity. The structural requirements for mucosal protective activity in these antagonists were shown to be the amide structure and six-membered aromatic ring, such as benzene and pyridine derivatives. The cardinal chemical features of roxatidine for the activation of mucin biosynthesis are the appropriate length of the flexible chain between the amide structure and the aromatic ring system bearing the methylpiperidinyl group at the meta position. Although the exact mechanism underlying the gastroprotective action associated with these agents is unknown, capsaicin-sensitive nerves and CGRP/NO pathway are considered responsible for their anti-ulcer effects in experimental animal models of various gastric mucosal injuries. These mechanisms are also involved in the cytoprotective properties of gastrin, which is a physiologically important bioactive peptide [70, 83]. Taken together, these find-

ings suggest the gastroprotective effects of second-generation histamine H_2-receptor antagonists may be of physiological relevance.

Second-generation histamine H_2-receptor antagonists offer the possibility of more effective prevention of gastrointestinal mucositis through the activation of mucosal defense mechanisms. Recently, lafutidine has been demonstrated to be an impressively effective therapeutic agent for acid-unrelated bowel diseases [35, 79, 84-86]. Its mechanism of action has been suggested to be related to capsaicin-sensitive sensory afferent neurons in the intestine. These afferent neurons are widely distributed throughout the entire body in mammals [63]. Furthermore, certain components of the immune system have been shown to be closely associated with CGRP-containing nerve fibers in the skin [87, 88], suggesting that some functions of the immune system are regulated by sensory neurons. It will be worth examining whether these functional aspects of sensory neurons are relevant to the restitution and healing of damaged gastrointestinal mucosa. Second-generation H_2-receptor antagonists will be useful tools for investigating this hypothesis.

REFERENCES

[1] Schwarz, K. Über penetrierende Magen und Jejunalgeschwüre. *Beitr. Klin. Chir.,* **1910,** *67*, 96-128.

[2] Isenberg, J.I.; Spector, H.; Hootkin, L.A.; Pitcher, J.L. An apparent exception to Schwarz's dictum, "no acid--no ulcer". *N. Engl. J. Med.,* **1971,** *285*, 620.

[3] Black, J.W.; Duncan, W.A.; Durant, C.J.; Ganellin, C.R.; Parsons, E.M. Definition and antagonism of histamine H_2–receptors. *Nature,* **1972,** *236*, 385-90.

[4] Brimblecombe, R.W.; Duncan, W.A.; Durant, G.J.; Ganellin, C.R.; Parsons, M.E.; Black, J.W. The pharmacology of cimetidine, a new histamine H_2-receptor antagonist. *Br. J. Pharmacol.,* **1975,** *53*, 435-6.

[5] Buyniski, R.L.; Cavanagh, A.W.; Pircio, A.A.; Crenshaw, R.R., In *Highlights in Receptor Chemistry*; Melchiorre, C.; Gianella, M., Eds.; Elsevier Science Publishers: Amsterdam, **1984**; pp. 195-215.

[6] Ganellin, R. 1980 Award in Medicinal Chemistry: Medicinal chemistry and dynamic structure-activity analysis in the discovery of drugs acting at histamine H_2 receptors. *J. Med. Chem.,* **1981,** *24*, 913-20.

[7] Schunack, W. What are the differences between the H_2-receptor antagonists? *Aliment. Pharmacol. Ther.,* **1987,** *1*(Suppl 1), 493S-503S.

[8] Kinoshita, M.; Kume, E.; Tamaki, H. *J. Pharmacol. Exp. Ther.,* **1995,** *275*, 494-501.

[9] Kinoshita, M.; Tamaki, H. Roles of prostaglandins, nitric oxide and the capsaicin-sensitive sensory nerves in gastroprotection produced by ecabet sodium. *Dig. Dis. Sci.,* **1997,** *42*, 83-90.

[10] Kishimoto, M.; Yanai, H.; Okazaki, Y.; Matsui, H.; Yoshida, T.; Okita, K. Characteristics of gastric mucus in elderly patients with gastric ulcers. *Hepatogastroenterology,* **2001,** *48*, 1594-8.

[11] Lam, S.K. Why do ulcers heal with sucralfate? *Scand. J. Gastroenterol. Suppl.,* **1990,** *173*, 6-16.

[12] Marshall, B.J.; Warren, J.R. Unidentified curved bacilli in the stomach of patients with gastritis and peptic ulceration. *Lancet,* **1984,** *1*, 1311-5.

[13] Peterson, W.L. Helicobacter pylori and peptic ulcer disease. *N. Engl. J. Med.,* **1991,** *324*, 1043-8.

[14] Sato, N.; Kawano, S.; Tsuji, S.; Ogihara, T.; Yamada, S. Gastric blood flow in ulcer diseases. *Scand. J. Gastroenterol. Suppl.,* **1995,** *208*, 14-20.

[15] Tsuji, S.; Kawano, S.; Hayashi, N.; Tsujii, M.; Takei, Y.; Nagano, K.; Sasayama, Y.; Kobayashi, I.; Fusamoto, H.; Kamada, T. Simultaneous analysis of mucosal and submucosal hemodynamics using infrared electronic endoscopy: effects of intraluminal acid on ulcer scars. *Endoscopy,* **1995,** *27*, 679-82.

[16] Wolfe, M.M.; Lichtenstein, D.R.; Singh, G. Gastrointestinal toxicity of nonsteroidal antiinflammatory drugs. *N. Engl. J. Med.,* **1999,** *340*, 1888-99.

[17] Hagiwara, T.; Mukaisho, K.; Ling, Z. Q.; Sakano, T.; Sugihara, H.; Hattori, T. Rebamipide contributes to reducing adverse effects of long-term administration of omeprazole in rats. *Dig. Dis. Sci.,* **2007,** *52*, 988-94.

[18] Ichikawa, T.; Ito, Y.; Saegusa, Y.; Iwai, T.; Goso, Y.; Ikezawa, T.; Ishihara, K. Effects of combination treatment with famotidine and methylmethionine sulfonium chloride on the mucus barrier of rat gastric mucosa. *J. Gastroenterol. Hepatol.,* **2009**, in press.

[19] Kinoshita, M.; Noto, T.; Tamaki, H. Effect of a combination of ecabet sodium and cimetidine on experimentally induced gastric lesions and gastric mucosal resistance to ulcerogenic agents in rats. *Biol. Pharm. Bull.,* **1995,** *18*, 223-6.

[20] Murata, H.; Kawano, S.; Tsuji, S.; Kamada, T.; Matsuzawa, Y.; Katsu, K.; Inoue, K.; Kobayashi, K.; Mitsufuji, S.; Bamba, T.; Kawasaki, H.; Kajiyama, G.; Umegaki, E.; Inoue, M.; Saito, I. Combination therapy of ecabet sodium and cimetidine compared with cimetidine alone for gastric ulcer: prospective randomized multicenter study. *J. Gastroenterol. Hepatol.,* **2003,** *18*, 1029-33.

[21] Murata, H.; Kawano, S.; Tsuji, S.; Tsujii, M.; Hori, M.; Kamada, T.; Matsuzawa, Y.; Katsu, K.; Inoue, K.; Kobayashi, K.; Mitsufuji, S.; Bamba, T.; Kawasaki, H.; Kajiyama, G.; Umegaki, E.; Inoue, M.; Saito, I. Combination of enprostil and cimetidine is more effective than cimetidine alone in treating gastric ulcer: prospective multicenter randomized controlled trial. *Hepatogastroenterology,* **2005,** *52*, 1925-9.

[22] Terano, A.; Arakawa, T.; Sugiyama, T.; Yoshikawa, T.; Haruma, K.; Asaka, M.; Shimosegawa, T.; Sakaki, N.; Ishii, H.; Sakamoto, C.; Takahashi, S.; Kinoshita, Y.; Fujioka, T.; Kobayashi, K. A pilot study to evaluate a new combination therapy for gastric ulcer: Helicobacter pylori eradication therapy followed by gastroprotective treatment with rebamipide. *J. Gastroenterol. Hepatol.,* **2006,** *21*, 103-9.

[23] Ajioka, H.; Miyake, H.; Matsuura, N. Effect of FRG-8813, a new-type histamine H_2-receptor antagonist, on the recurrence of gastric ulcer after healing by drug treatment in rats. *Pharmacology,* **2000,** *61*, 83-90.

[24] Fukushima, K.; Aoi, Y.; Kato, S.; Takeuchi, K. Gastro-protective action of lafutidine mediated by capsaicin-sensitive afferent neurons without interaction with TRPV1 and involvement of endogenous prostaglandins. *World J. Gastroenterol.,* **2006,** *12*, 3031-7.

[25] Harada, N.; Okajima, K. Inhibition of neutrophil activation by lafutidine, an H_2-receptor antagonist, through enhancement of sensory neuron activation contributes to the reduction of stress-induced gastric mucosal injury in rats. *Dig. Dis. Sci.,* **2007,** *52*, 469-77.

[26] Ichikawa, T.; Ishihara, K.; Saigenji, K.; Hotta, K. Effects of acid-inhibitory antiulcer drugs on mucin biosynthesis in the rat stomach. *Eur. J. Pharmacol.,* **1994,** *251*, 107-11.

[27] Ichikawa, T.; Ishihara, K.; Saigenji, K.; Hotta, K. Structural requirements for roxatidine in the stimulant effect of rat gastric mucin synthesis and the participation of nitric oxide in this mechanism. *Br. J. Pharmacol.,* **1997,** *122*, 1230-6.

[28] Ichikawa, T.; Ishihara, K.; Shibata, M.; Yamaura, T.; Saigenji, K.; Hotta, K. Stimulation of mucin biosynthesis in rat gastric mucosa by FRG-8813 and its structural analogs. *Eur. J. Pharmacol.,* **1996,** *297*, 87-92.

[29] Ichikawa, T.; Ota, H.; Sugiyama, A.; Maruta, F.; Ikezawa, T.; Hotta, K.; Ishihara, K. Effects of a novel histamine H_2-receptor antagonist, lafutidine, on the mucus barrier of human gastric mucosa. *J. Gastroenterol. Hepatol.,* **2007,** *22*, 1800-5.

[30] Marazova, K.; Klouchek, E.; Popov, A.; Ivanov, C.; Krushkov, I.; Ichikawa, T.; Ishihara, K.; Hotta, K. Effect of roxatidine bismuth citrate (MX1) against acetylsalicylic acid- and indomethacin-induced gastric mucosal damage in rats. *Methods Find. Exp. Clin. Pharmacol.,* **1998,** *20*, 667-72.

[31] Mimaki, H.; Kagawa, S.; Aoi, M.; Kato, S.; Satoshi, T.; Kohama, K.; Takeuchi, K. Effect of lafutidine, a histamine H_2-receptor antagonist, on gastric mucosal blood flow and duodenal HCO_3^- secretion in rats: relation to capsaicin-sensitive afferent neurons. *Dig. Dis. Sci.,* **2002,** *47*, 2696-703.

[32] Murashima, Y.; Kotani, T.; Hayashi, S.; Komatsu, Y.; Nakagiri, A.; Amagase, K.; Takeuchi, K. Impairment by 5-fluorouracil of the

healing of gastric lesions in rats: effect of lafutidine, a histamine H_2 receptor antagonist, mediated by capsaicin-sensitive afferent neurons. *Dig. Dis. Sci.,* **2009,** *54,* 36-45.

[33] Okabe, S.; Takagi, K.; Igata, H.; Kato, S.; Shimosako, K.; Yamaji, Y.; Seiki, M. Effects of a new histamine H_2-receptor antagonist, Z-300, on gastric secretion and gastroduodenal lesions in rats: comparison with roxatidine. *Jpn. J. Pharmacol.,* **1992,** *59*, 275-89.

[34] Onodera, S.; Shibata, M.; Tanaka, M.; Inaba, N.; Yamaura, T.; Ohnishi, H. Gastroprotective activity of FRG-8813, a novel histamine H_2-receptor antagonist, in rats. *Jpn. J. Pharmacol.,* **1995,** *68*, 161-73.

[35] Saegusa, Y.; Ichikawa, T.; Iwai, T.; Goso, Y.; Ikezawa, T.; Nakano, M.; Shikama, N.; Saigenji, K.; Ishihara, K. Effects of acid antisecretory drugs on mucus barrier of the rat against 5-fluorouracil-induced gastrointestinal mucositis. *Scand. J. Gastroenterol.,* **2008,** *43*, 531-7.

[36] Sato, H.; Kawashima, K.; Yuki, M.; Kazumori, H.; Rumi, M. A.; Ortega-Cava, C. F.; Ishihara, S.; Kinoshita, Y. Lafutidine, a novel histamine H_2-receptor antagonist, increases serum calcitonin gene-related peptide in rats after water immersion-restraint stress. *J. Lab. Clin. Med.,* **2003,** *141*, 102-5.

[37] Sekine, Y.; Hirakawa, N.; Kashiwaba, N.; Matsumoto, H.; Kutsuma, T.; Yamaura, T.; Sekine, A. A novel histamine 2(H_2) receptor antagonist with gastroprotective activity. I. Synthesis and pharmacological evaluation of N-phenoxypropylacetamide derivatives with thioether function. *Chem. Pharm. Bull. (Tokyo),* **1998,** *46*, 610-5.

[38] Shibata, M.; Yamaura, T.; Inaba, N.; Onodera, S.; Chida, Y.; Ohnishi, H. Gastric antisecretory effect of FRG-8813, a new histamine H_2 receptor antagonist, in rats and dogs. *Eur. J. Pharmacol.,* **1993,** *235*, 245-53.

[39] Shiratsuchi, K.; Fuse, H.; Hagiwara, M.; Mikami, T.; Miyasaka, K.; Sakuma, H. Cytoprotective action of roxatidine acetate HCl. *Arch. Int. Pharmacodyn. Ther.,* **1988,** *294*, 295-304.

[40] Sugiyama, T.; Hatanaka, Y.; Iwatani, Y.; Jin, X.; Kawasaki, H. Lafutidine facilitates calcitonin gene-related peptide (CGRP) nerve-mediated vasodilation *via* vanilloid-1 receptors in rat mesenteric resistance arteries. *J. Pharmacol. Sci.,* **2008,** *106*, 505-11.

[41] Takahashi, S.; Okabe, S. A histamine H_2 receptor antagonist, roxatidine, stimulates mucus secretion and synthesis by cultured rabbit gastric mucosal cells. *J. Physiol. Pharmacol.,* **1995,** *46*, 503-11.

[42] Schunack, W. Pharmacology of H_2-receptor antagonists: an overview. *J. Int. Med. Res.,* **1989,** *17*(Suppl 1), 9A-16A.

[43] Ganellin, C. R. In *Molecular and Quantum Pharmacology*, Bergmann, E.; Pullman, B., Eds.; Kluwer Academic Publishers: Dordrecht, **1974**; pp. 45-53.

[44] Brittain, R.T.; Daly, M.J. Pharmacology of H_2-receptor antagonists: an overview. *Scand. J. Gastroenterol. Suppl.,* **1981,** *69*, 1-9.

[45] Lin, J.H. Pharmacokinetic and pharmacodynamic properties of histamine H_2-receptor antagonists. Relationship between intrinsic potency and effective plasma concentrations. *Clin. Pharmacokinet.,* **1991,** *20*, 218-36.

[46] Campoli-Richards, D.M.; Clissold, S.P. Famotidine. Pharmacodynamic and pharmacokinetic properties and a preliminary review of its therapeutic use in peptic ulcer disease and Zollinger-Ellison syndrome. *Drugs,* **1986,** *32*, 197-221.

[47] Black, J.W.; Leff, P.; Shankley, N.P. Further analysis of anomalous pK_B values for histamine H_2-receptor antagonists on the mouse isolated stomach assay. *Br. J. Pharmacol.,* **1985,** *86*, 581-7.

[48] Ganellin, C.R., In *Frontiers in Histamine Research*, Ganellin, C.R., Ed.; Pergamon Press: Oxford, **1985**; pp. 47-59.

[49] Tanaka, A.; Nishihara, S.; Misawa, T.; Ibayashi, H. Effects of H_2-receptor antagonists on ^{3}H-cimetidine binding and histamine-stimulation of cellular cAMP in isolated guinea pig gastric glands. *Jpn. J. Pharmacol.,* **1987,** *45*, 97-105.

[50] Inaba, N.; Shibata, M.; Onodera, S.; Tanaka, M.; Suzuki, T.; Yamaura, T.; Ohnishi, H. Studies on histamine H_2-receptor antagonistic property of FRG-8813, a novel anti-ulcer drug. *Nippon Yakurigaku Zasshi,* **1995,** *105*, 231-41.

[51] Chaudhury, T.K.; Jacobson, E.D. Prostaglandin cytoprotection of gastric mucosa. *Gastroenterology,* **1978,** *74*, 58-63.

[52] Robert, A. Cytoprotection by prostaglandins. *Gastroenterology,* **1979,** *77*, 761-7.

[53] Szabo, S.; Trier, J.S.; Frankel, P.W. Sulfhydryl compounds may mediate gastric cytoprotection. *Science,* **1981,** *214*, 200-2.

[54] Tarnawski, A.; Hollander, D.; Gergely, H.; Stachura, J. Comparison of antacid, sucralfate, cimetidine, and ranitidine in protection of the gastric mucosa against ethanol injury. *Am. J. Med.,* **1985,** *79*, 19-23.

[55] Rees, W.D.W.; Brown, C.M., In *Bockus Gastroenterology, 5th edn*, Haubrich, W.S.; Schaffner, F.; Berk, J.E., Eds.; Saunders Company: Philadelphia, **1995**; pp. 582-614.

[56] Allen, A.; Hutton, D.A.; Leonard, A.J.; Pearson, J.P.; Sellers, L.A. The role of mucus in the protection of the gastroduodenal mucosa. *Scand. J. Gastroenterol. Suppl.,* **1986,** *125*, 71-8.

[57] Hirakawa, N.; Matsumoto, H.; Hosoda, A.; Sekine, A.; Yamaura, T.; Sekine, Y. A novel histamine 2(H_2) receptor antagonist with gastroprotective activity. II. Synthesis and pharmacological evaluation of 2-furfuryl-thio and 2-furfurylsulfinyl acetamide derivatives with heteroaromatic rings. *Chem. Pharm. Bull. (Tokyo),* **1998,** *46*, 616-22.

[58] Leurs, R.; Smit, M.J.; Menge, W.M.; Timmerman, H. Pharmacological characterization of the human histamine H_2 receptor stably expressed in Chinese hamster ovary cells. *Br. J. Pharmacol.,* **1994,** *112*, 847-54.

[59] Ruat, M.; Traiffort, E.; Bouthenet, M.L.; Schwartz, J.C.; Hirschfeld, J.; Buschauer, A.; Schunack, W. Reversible and irreversible labeling and autoradiographic localization of the cerebral histamine H_2 receptor using [^{125}I]iodinated probes. *Proc. Natl. Acad. Sci. USA,* **1990,** *87*, 1658-62.

[60] Nishihara, K.; Nozawa, Y.; Nakano, M.; Ajioka, H.; Matsuura, N. Sensitizing effects of lafutidine on CGRP-containing afferent nerves in the rat stomach. *Br. J. Pharmacol.,* **2002,** *135*, 1487-94.

[61] Holzer, P. Neural emergency system in the stomach. *Gastroenterology,* **1998,** *114*, 823-39.

[62] Sobue, M.; Joh, T.; Oshima, T.; Suzuki, H.; Seno, K.; Kasugai, K.; Nomura, T.; Ohara, H.; Yokoyama, Y.; Itoh, M. Contribution of capsaicin-sensitive afferent nerves to rapid recovery from ethanol-induced gastric epithelial damage in rats. *J. Gastroenterol. Hepatol.,* **2003,** *18*, 1188-95.

[63] Holzer, P. Capsaicin: cellular targets, mechanisms of action, and selectivity for thin sensory neurons. *Pharmacol. Rev.,* **1991,** *43*, 143-201.

[64] Green, T.; Dockray, G.J. Characterization of the peptidergic afferent innervation of the stomach in the rat, mouse and guinea-pig. *Neuroscience,* **1988,** *25*, 181-93.

[65] Sternini, C.; Reeve, J.R.Jr; Brecha, N. Distribution and characterization of calcitonin gene-related peptide immunoreactivity in the digestive system of normal and capsaicin-treated rats. *Gastroenterology,* **1987,** *93*, 852-62.

[66] Green, T.; Dockray, G.J. Calcitonin gene-related peptide and substance P in afferents to the upper gastrointestinal tract in the rat. *Neurosci. Lett.,* **1987,** *76*, 151-6.

[67] Arai, K.; Ohno, T.; Saeki, T.; Mizuguchi, S.; Kamata, K.; Hayashi, I.; Saigenji, K.; Murata, T.; Narumiya, S.; Majima, M. Endogenous prostaglandin I_2 regulates the neural emergency system through release of calcitonin gene related peptide. *Gut,* **2003,** *52*, 1242-9.

[68] Boku, K.; Ohno, T.; Saeki, T.; Hayashi, H.; Hayashi, I.; Katori, M.; Murata, T.; Narumiya, S.; Saigenji, K.; Majima, M. Adaptive cytoprotection mediated by prostaglandin I_2 is attributable to sensitization of CRGP-containing sensory nerves. *Gastroenterology,* **2001,** *120*, 134-43.

[69] Hayashi, H.; Ohno, T.; Nishiyama, K.; Boku, K.; Katori, M.; Majima, M. Transient prevention of ethanol-induced gastric lesion by capsaicin due to release of endogenous calcitonin gene-related peptide in rats. *Jpn. J. Pharmacol.,* **2001,** *86*, 351-4.

[70] Ichikawa, T.; Ishihara, K.; Kusakabe, T.; Hiruma, H.; Kawakami, T.; Hotta, K. CGRP modulates mucin synthesis in surface mucus cells of rat gastric oxyntic mucosa. *Am. J. Physiol.,* **2000,** *279*, G82-9.

[71] Mizuguchi, S.; Ohno, T.; Hattori, Y.; Kamata, K.; Arai, K.; Saeki, T.; Saigenji, K.; Hayashi, I.; Kuribayashi, Y.; Majima, M. Calcitonin gene-related peptide released by capsaicin suppresses myoelectrical activity of gastric smooth muscle. *J. Gastroenterol. Hepatol.,* **2005,** *20*, 611-8.

[72] Ohno, T.; Hattori, Y.; Komine, R.; Ae, T.; Mizuguchi, S.; Arai, K.; Saeki, T.; Suzuki, T.; Hosono, K.; Hayashi, I.; Oh-Hashi, Y.; Kurihara, Y.; Kurihara, H.; Amagase, K.; Okabe, S.; Saigenji, K.; Majima, M. Roles of calcitonin gene-related peptide in maintenance of

gastric mucosal integrity and in enhancement of ulcer healing and angiogenesis. *Gastroenterology,* **2008,** *134*, 215-25.

[73] Saeki, T.; Ohno, T.; Kamata, K.; Arai, K.; Mizuguchi, S.; Katori, M.; Saigenji, K.; Majima, M. Mild irritant prevents ethanol-induced gastric mucosal microcirculatory disturbances through actions of calcitonin gene-related peptide and PGI_2 in rats. *Am. J. Physiol.,* **2004,** *286*, G68-75.

[74] Onodera, S.; Tanaka, M.; Aoyama, M.; Arai, Y.; Inaba, N.; Suzuki, T.; Nishizawa, A.; Shibata, M.; Sekine, Y. Antiulcer effect of lafutidine on indomethacin-induced gastric antral ulcers in refed rats. *Jpn. J. Pharmacol.,* **1999,** *80*, 229-35.

[75] Ikawa, K.; Shimatani, T.; Azuma, Y.; Inoue, M.; Morikawa, N. Calcitonin gene-related peptide and somatostatin releases correlated with the area under the lafutidine concentration-time curve in human plasma. *J. Clin. Pharm. Ther.,* **2006,** *31*, 351-6.

[76] Shimatani, T.; Inoue, M.; Kuroiwa, T.; Xu, J.; Nakamura, M.; Tazuma, S.; Ikawa, K.; Morikawa, N. Lafutidine, a newly developed antiulcer drug, elevates postprandial intragastric pH and increases plasma calcitonin gene-related peptide and somatostatin concentrations in humans: comparisons with famotidine. *Dig. Dis. Sci.,* **2006,** *51*, 114-20.

[77] Ichikawa, T.; Ishihara, K.; Saigenji, K.; Hotta, K. Lafutidine-induced stimulation of mucin biosynthesis mediated by nitric oxide is limited to the surface mucous cells of rat gastric oxyntic mucosa. *Life Sci.,* **1998,** *62*, PL259-64.

[78] Ichikawa, T.; Ishihara, K.; Kusakabe, T.; Kawakami, T.; Hotta, K. Stimulant effect of nitric oxide generator and roxatidine on mucin biosynthesis of rat gastric oxyntic mucosa. *Life Sci.,* **1999,** *65*, PL41-6.

[79] Kato, S.; Tanaka, A.; Kunikata, T.; Umeda, M.; Takeuchi, K. Protective effect of lafutidine against indomethacin-induced intestinal ulceration in rats: relation to capsaicin-sensitive sensory neurons. *Digestion,* **2000,** *61*, 39-46.

[80] Onodera, S.; Nishida, K.; Takeuchi, K. Unique profile of lafutidine, a novel histamine H_2-receptor antagonist: mucosal protection throughout gastrointestinal tract mediated by capsaicin-sensitive afferent neurons. *Curr. Pharm. Des.,* **2004,** *1*, 133-44.

[81] Onodera, S.; Shibata, M.; Tanaka, M.; Inaba, N.; Arai, Y.; Aoyama, M.; Lee, B.; Yamaura, T. Antiulcer effect of lafutidine on indomethacin-induced gastric antral ulcers in refed rats. *Arzneimittelforschung,* **1999,** *49*, 519-26.

[82] Nagahama, K.; Yamato, M.; Kato, S.; Takeuchi, K. Protective effect of lafutidine, a novel H_2-receptor antagonist, on reflux esophagitis in rats through capsaicin-sensitive afferent neurons. *J. Pharmacol. Sci.,* **2003,** *93*, 55-61.

[83] Ichikawa, T.; Ishihara, K.; Kusakabe, T.; Kurihara, M.; Kawakami, T.; Takenaka, T.; Saigenji, K.; Hotta, K. Distinct effects of tetragastrin, histamine, and CCh on rat gastric mucin synthesis and contribution of NO. *Am. J. Physiol.,* **1998,** *274*, G138-46.

[84] Okayama, M.; Tsubouchi, R.; Kato, S.; Takeuchi, K. Protective effect of lafutidine, a novel histamine H_2-receptor antagonist, on dextran sulfate sodium-induced colonic inflammation through capsaicin-sensitive afferent neurons in rats. *Dig. Dis. Sci.,* **2004,** *49*, 1696-704.

[85] Someya, A.; Horie, S.; Yamamoto, H.; Murayama, T. Modifications of capsaicin-sensitive neurons in isolated guinea pig ileum by [6]-gingerol and lafutidine. *J. Pharmacol. Sci.,* **2003,** *92*, 359-66.

[86] Tanaka, A.; Mizoguchi, H.; Hase, S.; Miyazawa, T.; Takeuchi, K. Intestinal protection by lafutidine, a histamine H_2-receptor antagonist, against indomethacin-induced damage in rats--role of endogenous nitric oxide. *Med. Sci. Monit.,* **2001,** *7*, 869-77.

[87] Asahina, A.; Moro, O.; Hosoi, J.; Lerner, E.A.; Xu, S.; Takashima, A.; Granstein, R.D. Specific induction of cAMP in Langerhans cells by calcitonin gene-related peptide: relevance to functional effects. *Proc. Natl. Acad. Sci. U S A,* **1995,** *92*, 8323-7.

[88] Beresford, L.; Orange, O.; Bell, E.B.; Miyan, J.A. Nerve fibres are required to evoke a contact sensitivity response in mice. *Immunology,* **2004,** *111*, 118-25.

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