2-[(2-Aminobenzyl)sulfinyl]-1-(2-pyridyl)-1,4,5,6-tetrahydrocyclopent[*d*]imidazoles as a Novel Class of Gastric H^+/K^+-ATPase Inhibitors

Masaki Yamada,*,† Takeshi Yura,† Masamichi Morimoto,† Tsunehiro Harada,† Koichiro Yamada,† Yasushi Honma,†,‖ Mine Kinoshita,‡ and Masaki Sugiura§

Organic Chemistry Research Laboratory, Biological Research Laboratory, and Research Laboratory of Applied Biochemistry, Tanabe Seiyaku Co., Ltd., 2-2-50 Kawagishi, Toda-shi, Saitama 335, Japan

Received August 15, 1995⊗

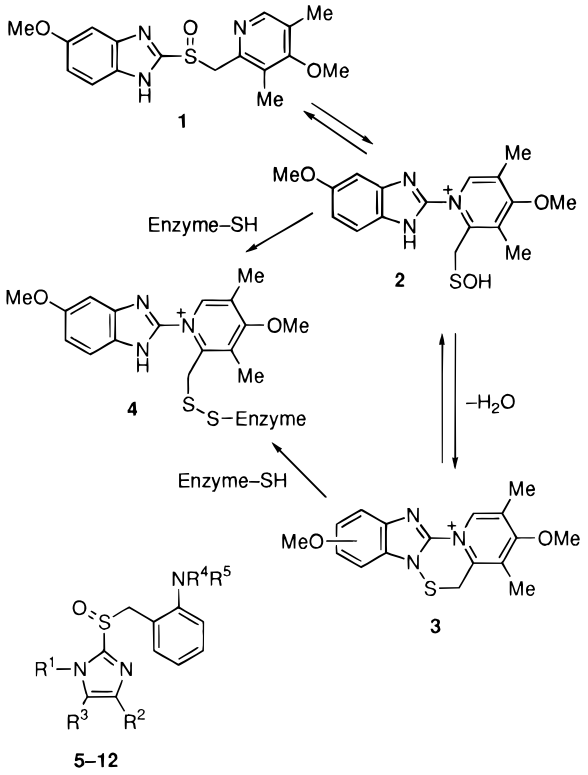
Substituted 2-sulfinylimidazoles were synthesized and investigated as potential inhibitors of gastric H^+/K^+-ATPase. The 4,5-unsubstituted imidazole series **6**−**11** and the 1,4,5,6-tetrahydrocyclopent[*d*]imidazole series **12** were found to be potent inhibitors of the acid secretory enzyme H^+/K^+-ATPase. Structure−activity relationships indicate that the substitution of 2-pyridyl groups at the 1-position of the imidazole moiety combined with (2-aminobenzyl)-sulfinyl groups at the 2-position leads to highly active compounds with a favorable chemical stability. Other substitution patterns in the imidazole moiety result in reducing biological activities. 2-[(2-Aminobenzyl)sulfinyl]-1-[2-(3-methylpyridyl)]-1,4,5,6-tetrahydrocyclopent[*d*]-imidazole (**12h**, T-776) was selected for further development as a potential clinical candidate. Extensive study on the acid degradation of **12h** indicates a mechanism of action different from that of omeprazole, the first H^+/K^+-ATPase inhibitor introduced to the market.

Introduction

The inhibition of gastric acid secretion has been proven to be a powerful therapeutic principle in the treatment of gastric and duodenal ulcer disease.[1] Different classes of drugs have been discovered from antimuscarinics, anticholinergics, and especially H2-receptor antagonists and are now available for antiulcer therapy. H^+/K^+-ATPase inhibitors, the so-called proton pump inhibitors like omeprazole (**1**), represent a new class of effective gastric acid secretion inhibitors.[2] The interest in this class of potent and long-lasting antisecretory agents is based on the highly specific and irreversible inhibitory action on gastric H^+/K^+-ATPase, which is responsible for the transport of gastric acid into the lumen of the stomach.[3]

Since omeprazole has been successfully introduced to the market in 1989, the side effects,[4] e.g., hypergastrinemia, have been emphasized resulting from the long-lasting inhibition of acid secretion. The mechanism of action of omeprazole on H^+/K^+-ATPase has been reported.[5] In the presence of acid, **1** is transformed into the highly thiophilic sulfenic acid **2** and cyclic sulfenamide **3**, both of which are able to react rapidly with thiol groups on the enzyme to form a complex (**4**) with a tightly bound S−S bond (Scheme 1). The long-lasting antisecretory effect of omeprazole results from the irreversible covalent binding of the inhibitor to the enzyme.[5] We focused, therefore, our efforts on H^+/K^+-ATPase inhibitors with an omeprazole-like mode of action but with pharmacodynamic properties different from omeprazole. We considered that a different kind of thiophile would afford an enzyme−inhibitor complex bound less tightly than **4** and lead to a reversible type of inhibition. This can be achieved by fine tuning the nucleophilic/electrophilic properties of the functional groups involved in the formation of the enzyme−inhibitor complex.[6] In modifying the structure of omeprazole, we were interested in evaluating substituted imidazoles[7] instead of benzimidazole and substitution of the nucleophilic pyridine moiety for the 2-aminobenzene moiety.[8] In this paper, we wish to report the synthesis, the pharmacological activities, and the mechanism of the inhibitory action of substituted 2-[(2-aminobenzyl)sulfinyl]-1-(2-pyridyl)imidazoles **5**−**12** as a new class of potent inhibitors of gastric H^+/K^+-ATPase.

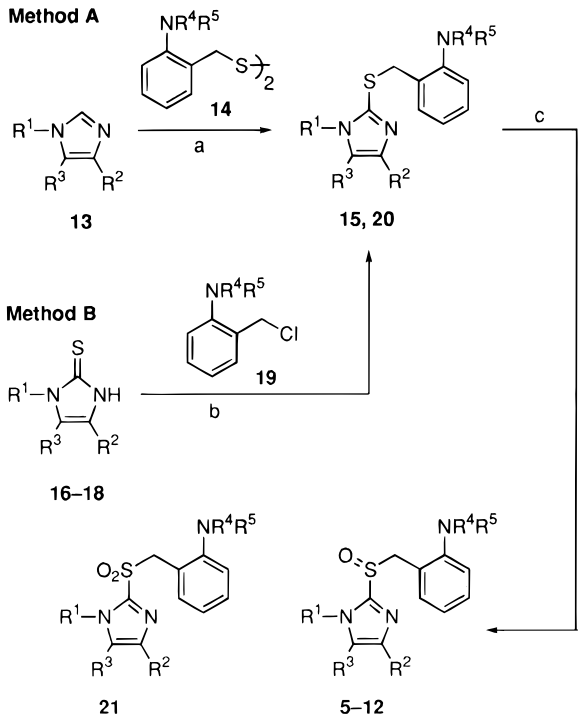
Scheme 1



* Author to whom correspondence should be addressed at Lead Generation Research Laboratory, Tanabe Seiyaku Co., Ltd., 16-89 Kashima-3-chome, Yodogawa-ku, Osaka 532, Japan.
† Organic Chemistry Research Laboratory.
‡ Biological Research Laboratory.
§ Research Laboratory of Applied Biochemistry.
‖ Present address: R&D Planning Division, Tanabe Seiyaku Co., Ltd., 16-89 Kashima-3-chome, Yodogawa-ku, Osaka 532, Japan.
⊗ Abstract published in *Advance ACS Abstracts,* December 15, 1995.

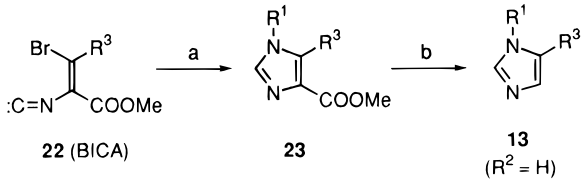
0022-2623/96/1839-0596$12.00/0

Scheme 2[a]



[a] (a) *n*-BuLi, −78 °C, THF; (b) NaOH or NaH, 0 °C, DMF; (c) mCPBA, room temperature, CH_2Cl_2, $CHCl_3$, or CCl_4.

Scheme 3[a]



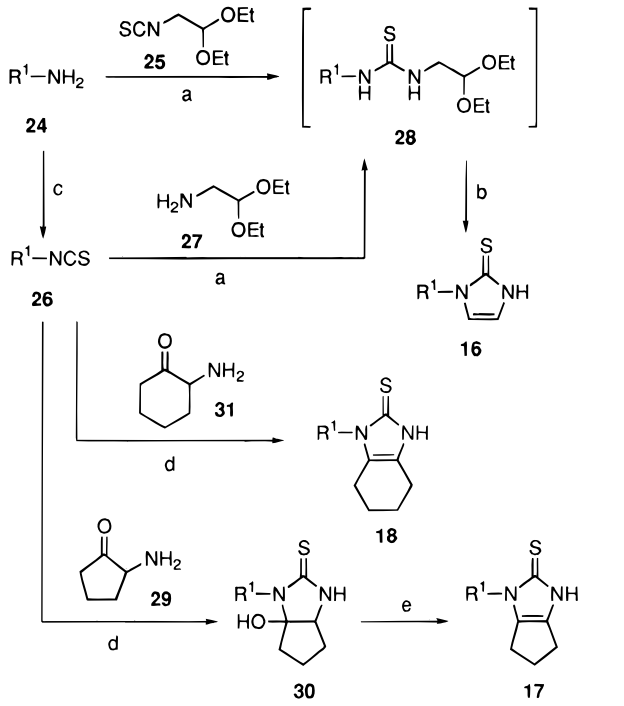
[a] (a) R^1NH_2, Et_3N, room temperature, DMF; (b) (1) NaOH, room temperature, H_2O−MeOH, (2) *N*,*N*-dimethylacetamide, reflux.

Chemistry

The new compounds **5**−**12** were synthesized as outlined in Scheme 2. The C2 anion of substituted imidazoles **13**, which was generated in dry THF at −60 °C by treatment with *n*-butyllithium,[9] reacted with the electrophilic disulfide **14** to provide the corresponding sulfides **15** (method A). Alkylation of substituted 2-mercaptoimidazoles **16**−**18** with *N*,*N*-disubstituted 2-aminobenzyl chlorides **19** under alkaline conditions yielded the desired sulfides **20** (method B). Oxidation of sulfides **15** and **20** was effected with *m*-chloroperbenzoic acid to give the desired sulfoxides **5**−**12**.

The 1,5-disubstituted imidazoles **13** were prepared by the reaction of primary amines with methyl 3-bromo-2-isocyanoacrylates (BICA) **22** followed by decarboxylation of the resulting methyl 1,5-disubstituted imidazole-4-carboxylates **23** according to our procedures previously reported[10] (Scheme 3). The other 1,4-disubstituted imidazoles **13** were prepared according to the literature.[11] The substituted 2-mercaptoimidazoles **16** were readily obtained by condensation of amines **24** with isothiocyanoacetaldehyde diethyl acetal (**25**) or isothiocyanates **26** with aminoacetaldehyde diethyl acetal (**27**) followed by cyclization of the resulting thioureas **28** under acidic conditions (Scheme 4). The condensation of isothiocyanates **26** with 2-aminocyclopentanone (**29**)[12] gave the hydroxy derivatives **30**, which were converted

Scheme 4[a]



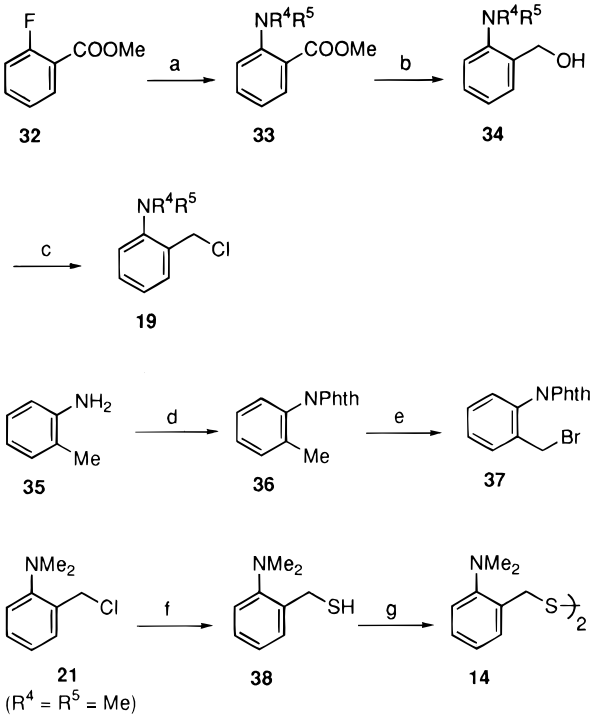
[a] (a) Toluene, reflux; (b) AcOH, catalytic concentrated HCl, reflux; (c) $CSCl_2$, $NaHCO_3$, $CHCl_3$−H_2O; (d) toluene, Et_3N, room temperature (e) PBr_3, pyridine, 0 °C to room temperature.

to the 1-substituted 2-mercapto-1,4,5,6-tetrahydrocyclopent[*d*]imidazoles **17**[13] by dehydration with PBr_3−pyridine. In contrast, the condensation of isothiocyanates **26** with 2-aminocyclohexanone (**31**)[12] gave the desired 4,5,6,7-tetrahydrobenzimidazoles **18** directly (Scheme 4). The substituted benzyl halides (**21**) were prepared from 2-substituted benzoates **33** either according to the literature or in analogy to known methods[8] (Scheme 5). 2-Phthalimidobenzyl bromide (**37**) was synthesized from *o*-toluidine (**35**), and the disulfide **14** was derived from **21** ($R^4 = R^5 =$ Me) via [2-(dimethylamino)phenyl]methyl mercaptan (**38**)[14] (Scheme 5). *N*-Unsubstituted aniline derivatives **9c**, **10d**, **11d**, and **12h** were derived from 2-phthalimidobenzyl derivatives **39** by oxidation and deprotection (Scheme 6), and *N*-monosubstituted aniline derivatives **9a**, **10a**,**c**, **11a**,**c**, and **12g** were synthesized by the alkylation of *N*-trifluoroacetyl derivative **41** followed by deacylation (Scheme 7).

Results and Discussion

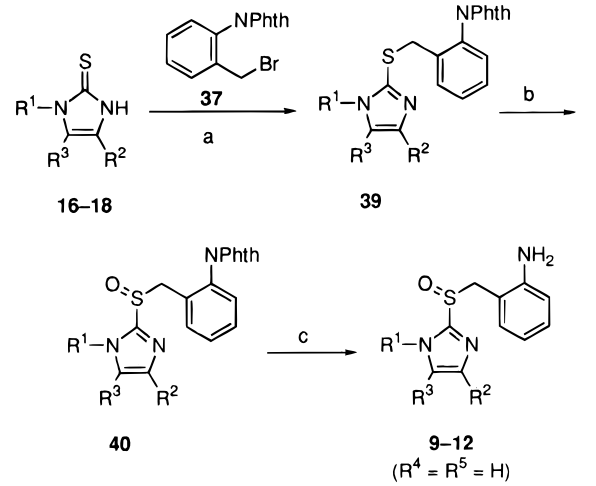
The effects of the substituted 2-sulfinylimidazoles **5a**−**o** on the inhibition of gastric acid secretion and H^+/K^+-ATPase were examined first (Table 1). The 4,5-unsubstituted series **5j**−**m** and the 1,4,5,6-tetrahydrocyclopent[*d*]imidazole series **5i**,**n** showed strong inhibition of gastric acid secretion both in vitro and in vivo, whereas the 4-phenyl-, 5-phenyl-, and 4,5-diphenylimidazoles **5a**,**d**−**h** and 4,5,6,7-tetrahydrobenzimidazole **5o** were found to be less effective in both biological assays. The substituent groups at the 1-position of imidazole were found to influence the inhibitory activities. Thus, 2-pyridyl (**5m**,**n**), methyl (**5k**), and benzyl (**5i**,**j**) derivatives showed strong activities in both assays, while the corresponding phenyl derivatives **5b**,**c** were only weakly inhibitory. Unfortunately, methyl (**5k**) and benzyl (**5i**,**j**) derivatives showed too short-lasting inhibitory activity in vivo.

Scheme 5[a]



[a] (a) R^4R^5NH, K_2CO_3 or NaH, room temperature, DMF; (b) $LiAlH_4$, THF; (c) $SOCl_2$, $CHCl_3$; (d) phthalic anhydride, AcOH, reflux; (e) NBS, CCl_4; (4) (1) thiourea, EtOH, (2) NaOH; (g) I_2, $NaHCO_3$.

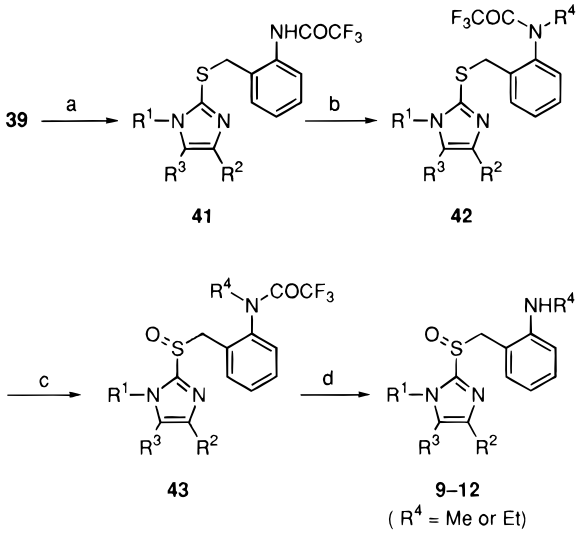
Scheme 6[a]



[a] (a) K_2CO_3, room temperature, DMF; (b) mCPBA, 0 °C, $CHCl_3$; (c) NH_2NH_2, MeOH.

Effects of the substituted pyridine group at the 1-position of 4,5-unsubstituted imidazole series **6**−**8** on activity were examined next in an attempt to maximize the potency (Table 2). 2-Pyridyl (**5m**), 4-pyridyl (**6b**), 2-pyridylmethyl (**6c**), and 2-pyridylethyl (**6d**) derivatives exhibited good activities in both assays, whereas 3-pyridyl **6a** showed diminished activities. The introduction of an electron-donating group such as the methyl (**7a**) or methoxy (**7b**,**h**,**k**) group onto the pyridine ring enhanced the activity, while the compounds bearing an electron-withdrawing group such as a halogen (**8b**,**c**), nitro (**8d**), or nitrile (**8e**) group showed decreased inhibitory activities. Substitution effects at the benzene ring of **9**−**11** are summarized in Table 3. The introduction of monomethylamine (**9a**, **10a**, **11a**), monoethylamine (**10c**, **11c**), diethylamine (**9b**, **10b**, **11b**), and acetamide (**9d**, **11e**) on the benzene ring showed strong activity in vivo, whereas the imidazole (**9h**) and pyrazole (**9i**) derivatives showed poor activity. The lack of correlation between the in vitro and in vivo tests has been cited as due to the chemical stability and bioavailability of the tested compounds.[15] The relative chemical stability of a series of the test compounds was examined in an acidic solution (pH 4.0, HCl) by monitoring their TLC. It has been found that the stability was remarkably affected by the substituents in the benzene ring, and the compounds with longer half-lives in this test showed poor in vitro activity. These results suggest that the preincubation conditions (pH 6.1, 37 °C, 30 min) of our in vitro assay are not acidic enough for the compounds with long half-lives to be activated to react with the enzyme. This may cause, at least in part, the discrepancy between in vitro and in vivo assays.

Scheme 7[a]

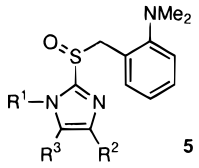


[a] (a) (1) NH_2NH_2, MeOH, (2) $(CF_3CO)_2O$, pyridine; (b) NaH, MeI or EtI, DMF; (c) mCPBA, 0 °C, $CHCl_3$; (d) $NaBH_4$, EtOH.

A variety of 1,4,5,6-tetrahydrocyclopent[*d*]imidazole derivatives (**12**) were further synthesized by modifying the pyridine and aniline groups (Table 4). The structure−activity relationship (SAR) was well in accordance with that observed for the 4,5-unsubstituted imidazole series. After pharmacological evaluation of the most active compounds, **12h** was selected for further study as a candidate for clinically useful proton pump inhibitors.

To obtain information about the mechanism of action at a molecular level, we undertook a detailed study on the chemical transformations of **12h** leading to its "active principle". An outline of the course of the reaction and the structures assigned to the isolated products are presented in Scheme 8. In order to identify the structure of the products, we carried out the reaction on a preparative scale under acidic conditions, which resemble the conditions at the site of action in the parietal cell. When **12h** was treated with 0.1 N HCl in the presence of 2-mercaptoethanol at 37 °C, **45**−**47** were obtained as isolable products. The formation of **45** could be well interpreted by the acid-catalyzed S(O)−C bond fission of **12h** and the subsequent nucleophilic displacement of the resulting sulfenic acid **44** with 2-mercaptoethanol. On acid treatment **12h** without 2-mercaptoethanol, the only isolable product was the symmetrical

Table 1. Physical Properties and Biological Activities of Substituted 2-[[2-(Dimethylamino)benzyl]sulfinyl]imidazoles **5** and Omeprazole (**1**)

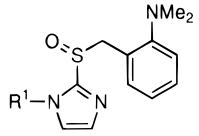


5

compd	R^1	R^2	R^3	yield, %	mp, °C	formula	inhibition of H^+/K^+-ATPase (IC_{50}, μM)[a]	antisecretory activity in rats at 30 mg/kg po (% inhibition[a])
5a	Ph	H	Ph	63	116−118	$C_{24}H_{23}N_3OS$	NA[b]	0
5b	Ph	H	H	54	97−99	$C_{18}H_{19}N_3OS$	68	9
5c	Ph	$-(CH_2)_3-$		66	116−117	$C_{21}H_{23}N_3OS$	68	20
5d	$PhCH_2$	Ph	H	60	syrup	$C_{25}H_{25}N_3OS$	NA	0
5e	Me	Ph	H	51	101−103	$C_{19}H_{21}N_3OS$	NA	0
5f	H	Ph	Ph	73	135−137	$C_{24}H_{23}N_3OS$	NA	0
5g	$PhCH_2$	H	Ph	58	syrup	$C_{25}H_{25}N_3OS$	NA	0
5h	Me	H	Ph	52	syrup	$C_{19}H_{21}N_3OS$	NA	0
5i	$PhCH_2$	$-(CH_2)_3-$		67	117−118	$C_{22}H_{25}N_3OS$	28	42
5j	$PhCH_2$	H	H	45	syrup	$C_{19}H_{21}N_3OS$	14	72
5k	Me	H	H	30	syrup	$C_{13}H_{17}N_3OS$	32	98
5l	H	H	H	49	121−123	$C_{12}H_{15}N_3OS$	32	80
5m	2-pyridyl	H	H	74	104−105	$C_{17}H_{18}N_4OS$	5.5	76
5n	2-pyridyl	$-(CH_2)_3-$		60	140−141	$C_{20}H_{22}N_4OS$	32	97
5o	2-pyridyl	$-(CH_2)_4-$		74	syrup	$C_{21}H_{24}N_4OS$	59	12
1	omeprazole						3.8	100

[a] See the Experimental Section. [b] NA = not active.

Table 2. Physical Properties and Biological Activities of 1-Substituted 2-[[2-(Dimethylamino)benzyl]sulfinyl]imidazoles **6**−**8**



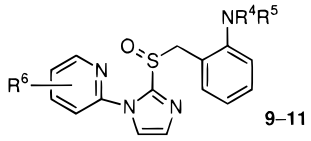
6–8

compd	R^1	yield, %	mp, °C	formula	inhibition of H^+/K^+-ATPase (IC_{50}, μM)[a]	antisecretory activity in rats at 30 mg/kg, po (% inhibition[a])
6a	3-pyridyl	26	syrup	$C_{17}H_{18}N_4OS$	NA[b]	*c*
6b	4-pyridyl	39	102−104	$C_{17}H_{18}N_4OS$	36	100
6c	2-pyridylmethyl	25	93−94	$C_{18}H_{20}N_4OS$	46	100
6d	2-pyridylethyl	27	syrup	$C_{19}H_{22}N_4OS$	44	87
6e	2-pyrimidinyl	44	153−155	$C_{16}H_{17}N_5OS$	NA	*c*
6f	2-pyrazinyl	16	109−111	$C_{16}H_{17}N_5OS$	NA	*c*
6g	1-isoquinolyl	33	149−150	$C_{21}H_{20}N_4OS$	43	65
7a	2-(3-Me)pyridyl	77	88−91	$C_{18}H_{20}N_4OS$	33	100
7b	2-(3-OMe)pyridyl	59	42−45	$C_{18}H_{20}N_4O_2S$	33	100
7c	2-(3-ONa)pyridyl	61	210−212	$C_{17}H_{17}N_4O_2SNa$	4.8	1
7d	2-(3-O-*i*-Pr)pyridyl	44	syrup	$C_{20}H_{24}N_4O_2S$	16	69
7e	2-(3-OBzl)pyridyl	35	96−97	$C_{24}H_{24}N_4O_2S$	16	34
7f	2-(3-OAc)pyridyl	40	syrup	$C_{19}H_{20}N_4O_3S$	12	2
7g	2-(4-Me)pyridyl	64	127−129	$C_{18}H_{20}N_4OS$	2.5	79
7h	2-(4-OMe)pyridyl	43	117−120	$C_{18}H_{20}N_4O_2S$	2.6	95
7i	2-(4-OCH_2CF_3)pyridyl	57	106−110	$C_{19}H_{19}F_3N_4O_2S$	6.8	11
7j	2-(5-Me)pyridyl	41	80−83	$C_{18}H_{20}N_4OS$	44	47
7k	2-(5-OMe)pyridyl	47	82−84	$C_{18}H_{20}N_4O_2S$	12	98
7l	2-(5-NH_2)pyridyl	34	140−143	$C_{17}H_{19}N_5OS$	5.7	48
8a	2-(5-NHAc)pyridyl	40	168−170	$C_{19}H_{21}N_5O_2S$	20	64
8b	2-(5-Cl)pyridyl	42	83−85	$C_{17}H_{17}ClN_4OS$	13	0
8c	2-(5-Br)pyridyl	26	73−75	$C_{17}H_{17}BrN_4OS$	ND[d]	0
8d	2-(5-NO_2)pyridyl	44	133−135	$C_{17}H_{17}N_5O_3S$	60	0
8e	2-(5-CN)pyridyl	40	137−139	$C_{18}H_{17}N_5OS$	35	0
8f	2-(5-CF_3)pyridyl	39	122−125	$C_{18}H_{17}F_3N_4OS$	7.4	36
8g	2-(6-Me)pyridyl	70	syrup	$C_{18}H_{20}N_4OS$	14	57
8h	2-(6-OMe)pyridyl	31	102−104	$C_{18}H_{20}N_4O_2S$	13	0
8i	2-(4,6-Me_2)pyridyl	67	syrup	$C_{19}H_{22}N_4OS$	11	0
8j	2-(4-OMe-6-Me)pyridyl	46	126−128	$C_{19}H_{22}N_4O_2S$	3.4	59
8k	2-(4,5-$(OMe)_2$)pyridyl	46	107−110	$C_{19}H_{22}N_4O_3S$	12	13
8l	2-(3-Me,5-Br)pyridyl	35	102−103	$C_{18}H_{19}BrN_4OS$	50	46

[a] See the Experimental Section. [b] NA = not active. [c] 20−30% inhibition at 30 mg/kg, sc. [d] ND = not determined.

disulfide **46**. This is well in accordance with the observation that a sulfenic acid is too reactive to be isolated, giving a disulfide by a disproportionation reaction.[5c,16] The sulfide **47** would be formed by reduc-

Table 3. Physical Properties and Biological Activities of 1-(2-Pyridyl)-2-[(2-substituted benzyl)sulfinyl]imidazoles **9**−**11**

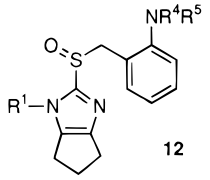


9–11

compd	R^6	NR^4R^5	yield, %	mp, °C	formula	inhibition of H^+/K^+-ATPase (IC_{50}, μM)[a]	antisecretory activity in rats at 30 mg/kg, po (% inhibition[a])
9a	H	NHMe	77	135−137	$C_{16}H_{16}N_4OS$	7.0	54
9b	H	NEt_2	81	115−116	$C_{19}H_{22}N_4OS$	31	96
9c	H	NH_2	34	145−146	$C_{15}H_{14}N_4OS$	27	29
9d	H	NHAc	87	172−173	$C_{17}H_{16}N_4O_2S$	NA[b]	99
9e	H	piperidino	78	94−96	$C_{20}H_{22}N_4OS$	NA	100
9f	H	morpholino	90	112−115	$C_{19}H_{20}N_4O_2S$	NA	90
9g	H	1-pyrrolyl	64	116−118	$C_{19}H_{16}N_4OS$	NA	88
9h	H	1-imidazolyl	49	188−190	$C_{18}H_{15}N_5OS$	NA	22
9i	H	1-pyrazolyl	75	134−137	$C_{18}H_{15}N_5OS$	NA	0
10a	3-Me	NHMe	74	syrup	$C_{17}H_{18}N_4OS$	7.5	ND[c]
10b	3-Me	NEt_2	60	syrup	$C_{20}H_{24}N_4OS$	100	100
10c	3-Me	NHEt	74	syrup	$C_{18}H_{20}N_4OS$	18	100
10d	3-Me	NH_2	56	113−115	$C_{16}H_{16}N_4OS$	98	98
10e	3-Me	piperidino	59	94−96	$C_{21}H_{24}N_4OS$	NA	82
10f	3-Me	morpholino	53	129−130	$C_{20}H_{22}N_4O_2S$	NA	100
10g	3-Me	1-pyrrolyl	48	113−115	$C_{20}H_{18}N_4OS$	NA	47
11a	3-OMe	NHMe	72	130−133	$C_{17}H_{18}N_4O_2S$	6.0	83
11b	3-OMe	NEt_2	79	syrup	$C_{20}H_{24}N_4O_2S$	NA	94
11c	3-OMe	NHEt	68	74−77	$C_{18}H_{20}N_4O_2S$	7.5	82
11d	3-OMe	NH_2	60	158−160	$C_{16}H_{16}N_4O_2S$	100	79
11e	3-OMe	NHAc	82	110−113	$C_{18}H_{18}N_4O_3S$	NA	90
11f	3-OMe	piperidino	79	syrup	$C_{21}H_{24}N_4O_2S$	NA	62
11g	3-OMe	morpholino	76	130−132	$C_{20}H_{22}N_4O_3S$	NA	85
11h	3-OMe	1-pyrrolyl	55	syrup	$C_{20}H_{18}N_4O_2S$	NA	39

[a] See the Experimental Section. [b] NA = not active. [c] ND = not determined.

Table 4. Physical Properties and Biological Activities of 1-Substituted 2-[(2-Substituted benzyl)sulfinyl]-1,4,5,6-tetrahydrocyclopent[*d*]imidazoles **12**



12

compd	R^1	NR^4R^5	yield, %	mp, °C	formula	inhibition of H^+/K^+-ATPase (IC_{50}, μM)[a]	antisecretory activity in rats at 30 mg/kg, po (% inhibition[a])
12a	2-pyridyl	NEt_2	43	107−109	$C_{22}H_{26}N_4OS$	11	98
12b	2-pyridyl	NHAc	94	188−191	$C_{20}H_{20}N_4OS$	NA[b]	75
12c	2-pyridyl	morpholino	63	135−136	$C_{22}H_{24}N_4O_2S$	NA	80
12d	2-pyridyl	1-pyrrolyl	67	147−148	$C_{22}H_{20}N_4OS$	NA	21
12e	2-(3-Me)pyridyl	NMe_2	46	119−120	$C_{21}H_{24}N_4OS$	>100	87
12f	2-(3-Me)pyridyl	NEt_2	81	syrup	$C_{23}H_{28}N_4OS$	NA	53
12g	2-(3-Me)pyridyl	NHMe	73	145−146	$C_{20}H_{22}N_4OS$	81	90
12h	2-(3-Me)pyridyl	NH_2	78	143−144	$C_{19}H_{20}N_4OS$	>100	95
12i	2-(3-OMe)pyridyl	NMe_2	63	syrup	$C_{21}H_{24}N_4O_2S$	95	73
12j	2-pyridylmethyl	NMe_2	51	syrup	$C_{21}H_{24}N_4OS$	55	12
12k	2-pyridylmethyl	NEt_2	46	syrup	$C_{23}H_{28}N_4OS$	NA	38
12l	3-pyridyl	NEt_2	65	syrup	$C_{22}H_{26}N_4OS$	NA	0
12m	4-pyridyl	NEt_2	65	syrup	$C_{22}H_{26}N_4OS$	NA	0

[a] See the Experimental Section. [b] NA = not active.

tion with 2-mercaptoethanol serving as a reducing agent.[5c] A sulfenic acid is known[3,4] to be involved in the inhibition of the H^+/K^+-ATPase enzyme, and therefore the transformations described in Scheme 8 can be considered as a model of the enzyme inhibition of **12h** in the acid compartments of the parietal cell. It is well known[17] that an aryl−S−S−R type disulfide is susceptible to thiol−disulfide exchange, and this tendency is apparent particularly for an arylthio group stabilized by resonance.[17a,18] Thus, the enzyme−inhibitor complex **48** would easily react with endogenous thiols to result in a rapid recovery of H^+/K^+-ATPase activity. In this regard, omeprazole blocks H^+/K^+-ATPase irreversibly by forming the stable disulfide **4**; therefore, recovery of the enzyme activity is owing to de novo synthesis of the enzyme.[19]

Table 5 shows the effect of cycloheximide, a protein synthesis inhibitor, on the **12h**- and omeprazole-induced

Scheme 8

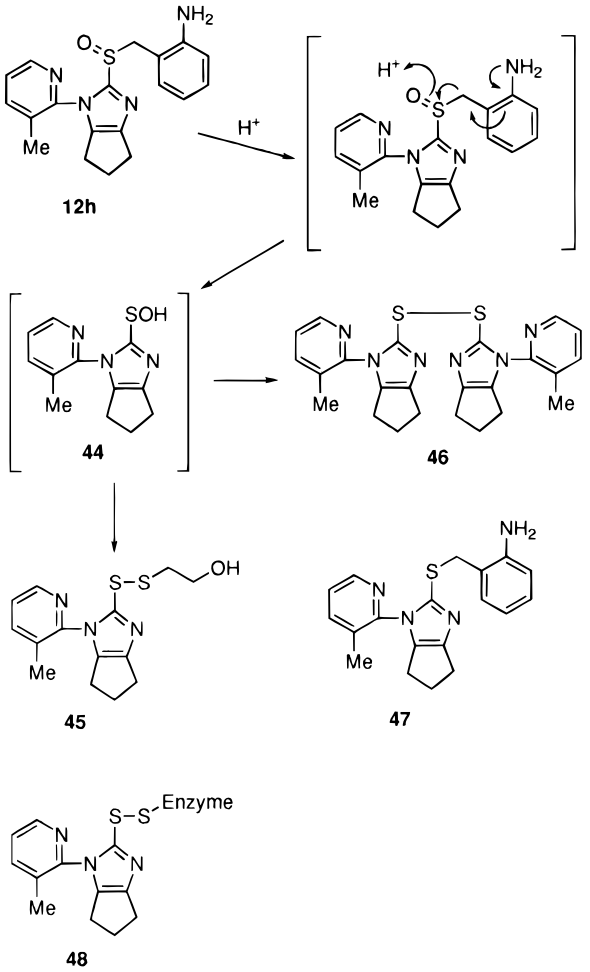


Table 5. Effect of Cycloheximide on the Recovery of Rat Gastric H^+/K^+-ATPase Activity Inhibited by **12h** and Omeprazole (OMZ) in Vivo

	H^+/K^+-ATPase activity (% of control)[a]					
	time after dosing of **12h** or OMZ (h)					
treatment	1.5	3	6	10	24	48
12h	48.8	46.0	69.2	79.6	88.9	
12h + cycloheximide				72.9	95.4	
OMZ	45.4	37.1	27.8		51.7	71.1
OMZ + cycloheximide					36.1	34.1

[a] Three, 13, 23, 33, and 43 h after intraduodenal administration of **12h** (40 mg/kg) or omeprazole (OMZ; 40 mg/kg), cycloheximide (1 mg/kg) was intraperitoneally injected. At the designated time after dosing of **12h** or OMZ, the stomach was excised and the crude gastric mucosal microsome was prepared. The H^+/K^+-ATPase activity of the microsome was assayed as described in the text. The data expressed are the means of 4−8 rats.

inhibition of H^+/K^+-ATPase activity in the rat gastric microsome. As shown in Table 5, the enzyme activities suppressed by the both inhibitors have been recovered time dependently. On addition of cycloheximide, omeprazole has sustained the inhibition[19] during the time course of measurements, while the enzyme activity with **12h** has not been affected. These observations clearly demonstrate that the restoration of the enzyme activity depressed by **12h** results from cleavage of the disulfide **48**, proving **12h** is a reversible inhibitor in nature.

Experimental Section

General. Melting points were determined on a 535 capillary melting point apparatus and are uncorrected. Elemental analyses were performed on a Perkin-Elmer 2400II analyzer. IR spectra were recorded on a Perkin-Elmer 1640 spectrophotometer. ^{1}H NMR spectra were obtained on a Bruker AC-200 (200 MHz) spectrometer with TMS as an internal standard. Mass spectra were recorded on a Hitachi M-2000A spectrometer. Column chromatography was performed on silica gel (E. Merck, no. 7734 or 9385 kieselgel 60) with the indicated solvent system. Radial chromatography was performed on a Chromatotron instrument (Harrison Research Model 7924T) using 1-, 2-, and 4-mm silica gel-coated (E. Merck kieselgel 60 PF_{254} plates. All reactions with air- and moisture-sensitive compounds were conducted in oven-dried glassware under an atmosphere of dry nitrogen.

Biology. H^+/K^+-ATPase Assay (in Vitro). Porcine gastric mucosal membrane vesicles containing H^+/K^+-ATPase were prepared according to the method of Lee and Forte.[20] The H^+/K^+-ATPase activity was assayed by the method of Keeling and co-workers.[21] The enzyme was preincubated with a test compound at 37 °C for 30 min in 2 mM Pipes−Tris buffer, pH 6.1. The enzyme reaction was started by addition of the mixture containing 2 mM $MgCl_2$, 2 mM ATP (pH 7.4), and valinomycin (5 mg/mL) in 80 mM Pipes−Tris buffer (pH 7.4) with or without 10 mM KCl. The reaction was terminated by addition of trichloroacetic acid (final 5%). After centrifugation, inorganic phosphate in the supernatant was determined by the method of Yoda and Hokin.[22] Protein was determined by the Bradford method.[23] Percent inhibition was calculated as follows: [(mean value of control activity − mean value of test activity)/mean value of control activity] × 100. The doses inhibiting H^+/K^+-ATPase by 50% (IC_{50}) were calculated by linear regression analysis.

H^+/K^+-ATPase Assay (in Vivo). Male Sprague−Dawley rats (Charles River Japan Inc.) weighing 200−250 g were used. Rats were deprived of food but allowed free access to water for 24 h prior to excision of the stomach. Via ether anesthesia, the abdomen of rats was incised and **12h** or omeprazole was intraduodenally administered. Then the abdomen was closed by suturing. To examine the effect of a protein synthesis inhibitor, we injected cycloheximide (1 mg/kg) intraperitoneally 3 h after the administration of the test drug and repeated this treatment every 10 or 12 h.[19] After designated times, the rats were given an overdose of ether and the stomach was excised. The stomach was immediately placed in an ice-cold buffer containing 250 mM sucrose and 2 mM Hepes−Tris (pH 7.4), and the crude microsomal preparation of the gastric mucosa (100000*g*, pellet) was prepared according to the previously published method.[24]

The microsomal preparations (10−20 μg of protein/mL) were incubated at 37 °C in 1 mL of a medium consisting of 180 mM sucrose, 40 mM Tris−acetate buffer (pH 7.4), 2 mM Na_2ATP, 2 mM $MgCl_2$, and nigericin (5 μg/mL) with or without 20 mM KCl. Ten minutes later, the reaction was terminated by adding 1 vol of 15% (w/v) trichloroacetic acid. Inorganic phosphate produced by hydrolysis of ATP was determined as described by Yoda and Hokin.[21] H^+/K^+-ATPase activity was measured by subtracting the ATPase activity in the absence of KCl from that in the presence of KCl.

Gastric Antisecretory Activity. Male Sprague−Dawley rats weighing 100−160 g (Charles River Japan Inc.) were deprived of food but allowed free access to water for 24 h prior to experiments. Each experiment was performed using 5 or 6 rats/group. Test drugs were dissolved or suspended in a 0.2% (v/v) poly(oxyethylene) sorbitan monooleate (Tween 80) solution in a volume of 10 mL/kg, body weight. One hour after oral administration of test drugs (30 mg/kg), pentagastrin (1 mg/kg) was subcutaneously injected. Thirty minutes later, the stomach was excised under overdose of ether anesthesia and the gastric contents were collected. After centrifugation (900*g*, 10 min), the volume of the supernatant was measured, the acid concentration was determined by automatic titration to pH 7.0 with 0.1 N NaOH, and the total acid output during 30 min was calculated. Antisecretory activity was expressed as the percentage of inhibition against the acid output of the control group given the vehicle only.

Chemistry. Typical Procedure for the Preparation of Sulfide Derivatives 15 by Method A: 2-[[2-(Dimethylami-

no)benzyl]thio]-1,5-diphenylimidazole (15a: $R^1 = R^3 =$ Ph, $R^2 =$ H, $R^4 = R^5 =$ Me). *n*-Butyllithium (1.6 M) in hexane (1.5 mL, 2.4 mmol) was added dropwise to a stirred solution of 1,5-diphenylimidazole (**13a**: $R^1 = R^3 =$ Ph, $R^2 =$ H) (440 mg, 2.0 mmol) in THF (5 mL) at −65 °C. After 1 h, 2-(dimethylamino)benzyl disulfide (**14a**: $R^4 = R^5 =$ Me) (666 mg, 2.0 mmol) in THF (5 mL) was added slowly while the temperature was kept between −65 and −40 °C. The reaction mixture was allowed to warm gradually to ambient temperature. Saturated aqueous ammonium chloride (15 mL) was added, and the reaction mixture was extracted with ether (10 mL × 2), whereupon the combined ether extracts were dried ($MgSO_4$), filtered, and concentrated in vacuo. The crude product was chromatographed on silica gel with AcOEt−hexane (4:1) as an eluent to give **15a** as a yellow syrup, which was crystallized from AcOEt−hexane to afford **15a** (391 mg, 63%) as colorless needles: mp 81−82 °C (AcOEt/hexane); ^{1}H NMR (δ in $CDCl_3$) 2.60 (s, 6H), 4.44 (s, 2H), 6.94−7.10 (m, 6H), 7.16−7.20 (m, 5H), 7.26−7.35 (m, 4H); IR (KBr) 2900, 1680, 1600, 1450, 1420, 1155, 1090 cm^{-1}; SIMS *m*/*z* 386 (M + H), 352, 134 (base). Anal. ($C_{24}H_{23}N_3S$) C, H, N, S.

In the same manner, other sulfides **15** were obtained.

Typical Procedure for the Preparation of Sulfide Derivatives 20 by Method B: 2-[[2-(Dimethylamino)-benzyl]thio]-1-[2-(3-methylpyridyl)]imidazole (20a: $R^1 =$ 2-(3-methylpyridyl), $R^3 = R^2 =$ H, $R^4 = R^5 =$ Me). To a solution of 2-mercapto-1-[2-(3-methylpyridyl)]imidazole (**16a**) (5.0 g, 26 mmol) and NaOH (1.2 g, 30 mmol) in DMF (30 mL) at 0 °C was added a solution of 2-(dimethylamino)benzyl chloride hydrochloride (**19a**: $R^4 = R^5 =$ Me) (5.1 g, 24.7 mmol) in DMF (20 mL), and the reaction mixture was stirred for 3 h. After the removal of DMF, the residue was extracted with $CHCl_3$, and the organic layer was washed with saturated aqueous $NaHCO_3$ and brine, dried ($MgSO_4$), filtered, and concentrated in vacuo. The residue was subjected to column chromatography on silica gel with $CHCl_3$−acetone (9:1) as an eluent to give **20a** (6.57 g, 78 %) as a yellow syrup: ^{1}H NMR (δ in $CDCl_3$) 2.08 (s, 3H), 2.61 (s, 6H), 4.45 (s, 2H), 7.21 and 7.47 (d, 1H each, $J = 1.5$ Hz), 6.94−7.76 (m, 6H), 8.39 (m, 1H); IR (film) 1595, 1575, 1495, 1450, 1360, 1300, 1195, 1150, 1095, 1050, 990 cm^{-1}; EIMS *m*/*z* 324 (M$^+$), 291, 134 (base).

In the same manner, other sulfides **20** were obtained.

Typical Procedure for the Preparation of Sulfoxide Derivatives 5−12: 2-[[2-(Dimethylamino)benzyl]sulfinyl]-1-[2-(3-methylpyridyl)]imidazole (7a). To a solution of 2-[[2-(dimethylamino)benzyl]thio]-1-[2-(3-methylpyridyl)]imidazole (**20a**) (2.49 g, 7.71 mmol) in $CHCl_3$ (30 mL) at 0 °C was added a solution of 80% *m*-chloroperbenzoic acid (1.83 g, 8.48 mmol) in $CHCl_3$ (30 mL) during a period of 10 min. After 30 min, the solution was washed with 5% NaOH and brine, dried ($MgSO_4$), filtered, and concentrated in vacuo. The residue was subjected to column chromatography on silica gel with $CHCl_3$−MeOH (95:5) as an eluent to give **7a** as a yellow syrup, which was crystallized from AcOEt−hexane to afford **7a** (2.02 g, 77%) as colorless prisms: mp 88−91 °C (AcOEt/*i*-Pr_2O); ^{1}H NMR (δ in $CDCl_3$) 2.11 (s, 3H), 2.59 (s, 6H), 4.63 and 4.96 (ABq, 1H each, $J = 12.3$ Hz), 6.94−7.83 (m, 8H), 8.35 (m, 1H); IR (KBr) 1590, 1475, 1440, 1325, 1305, 1155, 1080, 1045, 790 cm^{-1}; EIMS *m*/*z* 340 (M$^+$), 324, 134 (base). Anal. ($C_{18}H_{20}N_4OS$) C, H, N, S.

2-[[2-(Dimethylamino)benzyl]sulfonyl]-1-[2-(3-methylpyridyl)]imidazole (21a: $R^1 =$ 2-(3-methylpyridyl), $R^3 = R^2 =$ H, $R^4 = R^5 =$ Me): 137 mg, 5.0%; obtained from the subsequent eluate as colorless needles; mp 139−140 °C (AcOEt/*i*-Pr_2O); ^{1}H NMR (δ in $CDCl_3$) 2.13 (s, 3H), 2.58 (s, 6H), 4.97 (s, 2H), 6.94−7.83 (m, 8H), 8.33 (m, 1H); IR (KBr) 1590, 1475, 1440, 1325, 1305, 1155, 1080, 790 cm^{-1}; SIMS *m*/*z* 357 (M + H), 293, 160, 134 (base). Anal. ($C_{18}H_{20}N_4O_2S$) C, H, N, S.

In the same manner, other sulfoxides **5−12** were obtained.

Typical Procedure for the Preparation of 1,5-Disubstituted Imidazoles 13: 1,5-Diphenylimidazole (13a: $R^1 = R^3 =$ Ph, $R^2 =$ H). A mixture of methyl 1,5-diphenylimidazole-4-carboxylate[10] (**23a**: $R^1 = R^3 =$ Ph) (2.78 g, 10 mmol) and NaOH (1.2 g, 30 mmol) in MeOH−H_2O (1:1, 50 mL) was stirred for 18 h at room temperature. The mixture was neutralized with $KHSO_4$ and then extracted with AcOEt. The organic layer was washed with brine, dried over $MgSO_4$, and concentrated in vacuo. The resulting oil was crystallized from MeOH−H_2O to afford the corresponding carboxylic acid derivative. A suspension of the carboxylic acid in *N*,*N*-dimethylacetamide (20 mL) was refluxed for 3 h. The reaction mixture was poured into water and then extracted with AcOEt. The organic layer was washed with brine, dried over $MgSO_4$, and concentrated in vacuo. The resulting oil was chromatographed on silica gel with $CHCl_3$−MeOH (20:1) as an eluent to give **13a** as a yellow syrup, which was crystallized from AcOEt−hexane to afford **13a** (1.71 g, 78%) as colorless needles: mp 127−128 °C (AcOEt/hexane); ^{1}H NMR (δ in DMSO-d_6) 7.10−7.15 (m, 2H), 7.24−7.33 (m, 6H), 7.41−7.52 (m, 2H), 7.44 and 7.94 (s, 1H each); IR (KBr) 1600, 1500, 1465, 1270, 1120, 915 cm^{-1}; EIMS *m*/*z* 220 (M$^+$, base), 193, 165. Anal. ($C_{15}H_{12}N_2$) C, H, N.

In the same manner, other 1,5-disubstituted imidazoles **13** ($R^2 =$ H)[10] were obtained, and 1,4-disubstituted imidazoles **13** ($R^3 =$ H) were synthesized essentially by means of the procedure previously described.[11]

Typical Procedure for the Preparation of 1-Substituted 2-Mercaptoimidazoles 16: 2-Mercapto-1-[2-(3-methylpyridyl)]imidazole (16a: $R^1 =$ 2-(3-methylpyridyl), $R^2 = R^3 =$ H). A solution of 2-aminopicoline (**24a**: $R^1 =$ 2-(3-methylpyridyl)) (4.33 g, 40 mmol) and $(EtO)_2CHCH_2NCS$ (**25**) (7.0 g, 40 mmol) in toluene (100 mL) was refluxed for 18 h. The reaction mixture was concentrated in vacuo, and the resulting solid **28a** ($R^1 =$ 2-(3-methylpyridyl)) (7.55 g, 67%) was collected with hexane. The solid **28a** was dissolved in AcOH (50 mL), and a catalytic amount of concentrated HCl (0.5 mL) was added to the solution. The reaction mixture was refluxed for 18 h and then concentrated in vacuo. The resulting solid was dissolved in H_2O, and the solution was neutralized with $NaHCO_3$ to afford **16a** as a yellow solid, which was crystallized from AcOEt−hexane to afford **16a** (3.04 g, 63%) as colorless needles: mp 187−190 °C (AcOEt/hexane); ^{1}H NMR (δ in $CDCl_3$) 2.32 (s, 3H), 6.82 and 6.86 (d, 1H each, $J = 2.4$ Hz), 7.34 (dd, 1H, $J = 4.9$, 5.3 Hz), 7.70 (d, 1H, $J =$ 5.3 Hz), 8.48 (d, 1H, $J = 4.9$ Hz), 12.3 (br, 1H); IR (Nujol) 3100, 1570, 1450, 1310, 1290, 800 cm^{-1}; EIMS *m*/*z* 191 (M$^+$), 176, 158 (base). Anal. ($C_9H_9N_3S$) C, H, N, S.

In the same manner, other 1-substituted 2-mercaptoimidazoles **16** were obtained.

Typical Procedure for the Preparation of 1-Substituted 2-Mercapto-1,4,5,6-tetrahydrocyclopent[*d*]imidazoles 17: 2-Mercapto-1-[2-(3-methylpyridyl)]-1,4,5,6-tetrahydrocyclopent[*d*]imidazole (17a: $R^1 =$ 2-(3-methylpyridyl)). To a two-phase solution of 2-amino-3-picoline (**24a**) (7.57 g, 70 mmol) in $CHCl_3$ (80 mL) and $NaHCO_3$ (41.7 g, 496 mmol) in H_2O (150 mL) at 0 °C was added a solution of thiophosgen (8.4 g, 72.5 mmol) in $CHCl_3$ (40 mL) during a period of 1 h. After 1 h, the solution was washed with saturated aqueous $NaHCO_3$ and brine and dried over $MgSO_4$. 2-Aminocyclopentanone hydrochloride[12] (**29**) (9.5 g, 70 mmol) was dissolved in the $CHCl_3$ solution of **26a** ($R^1 =$ 2-(3-methylpyridyl)) described above. To the reaction mixture was added slowly a solution of Et_3N (7.5 g, 74 mmol) during a period of 6 h. After 18 h, the solution was washed with H_2O, dried ($MgSO_4$), and concentrated in vacuo to give a solid, which was crystallized from *i*-PrOH to afford 7-hydroxy-2-mercapto-1-[2-(3-methylpyridyl)]-1,4,5,6,7,8-hexahydrocyclopent[*d*]imidazole (**30a**: $R^1 =$ 2-(3-methylpyridyl)) (12.9 g, 74%) as colorless needles: mp 171−174 °C (*i*-PrOH); ^{1}H NMR (δ in DMSO-d_6) 1.64−1.84 (m, 6H), 2.34 (s, 3H), 3.94 (d-like, 1H), 6.58 (s, 1H, D_2O exchangeable), 7.31 (m, 1H), 7.70 (m, 1H), 8.33 (m, 1H), 8.70 (br s, 1H); IR (Nujol) 3300, 1580, 1510, 1460, 1450, 1420, 1380, 1270, 1250, 1080 cm^{-1}; EIMS *m*/*z* 249 (M$^+$), 231, 151 (base). Anal. ($C_{12}H_{15}N_3OS$) C, H, N, S.

To a solution of the alcohol derivative **30a** (12.9 g) in pyridine (130 mL) at 10 °C was added PBr_3 (7.8 mL, 82 mmol) during a period of 20 min. After 18 h, to the solution at 0 °C was added saturated aqueous $NaHCO_3$ (50 mL). After 3 h, the reaction mixture was extracted with AcOEt, and the organic layer was washed with brine, dried ($MgSO_4$), filtered, and concentrated in vacuo to give **17a** as a yellow solid, which

was crystallized from AcOEt−hexane to afford **17a** (4.7 g, 39%) as colorless prisms: mp 230−234 °C (AcOEt/*i*-Pr_2O); ^{1}H NMR (δ in DMSO-d_6) 2.23 (s, 3H), 2.23−2.72 (m, 6H), 7.42 (m, 1H), 7.85 (m, 1H), 8.39 (m, 1H), 12.2 (br s, 1H); IR (Nujol) 3160, 3080, 1660, 1575, 1460, 1450, 1420, 1080 cm^{-1}; EIMS *m/z* 231 (M^+), 216, 173 (base). Anal. ($C_{12}H_{13}N_3S$) C, H, N, S.

Typical Procedure for the Preparation of 1-Substituted 2-Mercapto-4,5,6,7-tetrahydrobenzimidazoles 18: 2-Mercapto-1-(2-pyridyl)-4,5,6,7-tetrahydrobenzimidazole (18a: R^1 = 2-pyridyl). To a solution of 2-isothiocyanopyridine (**26b**: R^1 = 2-pyridyl) (6.21 g, 45.6 mmol) and 2-aminocyclohexanone hydrochloride[12] (**31**) (6.82 g, 45.6 mmol) in toluene (100 mL) was added slowly a solution of Et_3N (4.76 g, 40.0 mmol). The reaction mixture was warmed to 50 °C and stirred for 3 h. The reaction mixture was washed with brine, dried ($MgSO_4$), filtered, and concentrated in vacuo to give **18a** as a yellow solid, which was crystallized from *i*-PrOH to afford **18a** (7.95 g, 86%) as colorless prisms: mp 203−204 °C (*i*-PrOH); ^{1}H NMR (δ in DMSO-d_6) 1.50−1.90 (m, 4H), 2.22−2.43 (m, 4H), 7.41 (m, 1H), 7.70−8.12 (m, 2H), 8.55 (m, 1H), 12.2 (br s, 1H); IR (Nujol) 3300, 1585, 1510, 1455, 1420, 1270, 1260, 1080 cm^{-1}; EIMS *m/z* 231 (M^+, base). Anal. ($C_{12}H_{13}N_3S$) C, H, N, S.

Typical Procedure for the Preparation of *N*-Unsubstituted Aniline Derivatives 9−12: 2-[(2-Aminobenzyl)sulfinyl]-1-[2-(3-methylpyridyl)]-1,4,5,6-tetrahydrocyclopent-[*d*]imidazole (12h). To a solution of 2-mercapto-1-[2-(3-methylpyridyl)]-1,4,5,6-tetrahydrocyclopent[*d*]imidazole (**17a**) (4.1 g, 17.7 mmol) and NaH (60% oil suspended) (850 mg, 21.2 mmol) in DMF (40 mL) at 0 °C was added a solution of 2-phthalimidobenzyl bromide (**37**) (5.6 g, 17.7 mmol) in DMF (20 mL), and the reaction mixture was stirred for 3 h. After the removal of DMF, the residue was extracted with $CHCl_3$, and the organic layer was washed with saturated aqueous $NaHCO_3$ and brine, dried ($MgSO_4$), filtered, and concentrated in vacuo. The residue was crystallized from AcOEt to afford 2-[(2-phthalimidobenzyl)thio]-1-[2-(3-methylpyridyl)]-1,4,5,6-tetrahydrocyclopent[*d*]imidazole (**39a**: R^1 = 2-(3-methylpyridyl), $R^2 = R^3 = -(CH_2)_3-$) (5.42 g, 66%) as pale yellow crystals: mp 140−142 °C (AcOEt). Anal. ($C_{27}H_{22}N_4O_2S$) C, H, N, S.

Oxidation of the sulfide **39a** (5.0 g, 10.7 mmol) was carried out with a procedure similar to that described for **7a** to give 2-[(2-phthalimidobenzyl)sulfinyl]-1-[2-(3-methylpyridyl)]-1,4,5,6-tetrahydrocyclopent[*d*]imidazole (**40a**: R^1 = 2-(3-methylpyridyl), $R^2 = R^3 = -(CH_2)_3-$) (4.6 g, 89%) as pale yellow crystals: mp 170−172 °C (AcOEt). Anal. ($C_{27}H_{22}N_4O_3S$) C, H, N, S.

To a suspension of **40a** (4.1 g, 8.5 mmol) in EtOH (65 mL) at 0 °C was added NH_2NH_2 hydrate (530 mg, 10.6 mmol). The reaction mixture was stirred at 50 °C for 18 h and then concentrated in vacuo. The resulting solid was dissolved in $CHCl_3$, and the organic layer was washed with saturated aqueous $NaHCO_3$ and brine, dried ($MgSO_4$), filtered, and concentrated in vacuo. The residue was crystallized from AcOEt−hexane to afford 2-[(2-aminobenzyl)sulfinyl]-1-[2-(3-methylpyridyl)]-1,4,5,6-tetrahydrocyclopent[*d*]imidazole (**12h**) (2.3 g, 77%) as colorless prisms: mp 143−144 °C (AcOEt/hexane); ^{1}H NMR (δ in $CDCl_3$) 2.13 (s, 3H), 2.46−2.90 (m, 6H), 4.3 (br, 2H, D_2O exchangeable), 4.47 and 4.85 (ABq, 1H each, J = 12.5 Hz), 6.6−7.4 (m, 5H), 7.70 (m, 1H), 8.42 (m, 1H); IR (Nujol) 3450, 3220, 1655, 1575, 1460, 1360, 1045, 750 cm^{-1}; EIMS *m/z* 352 (M^+), 247, 106 (base). Anal. ($C_{19}H_{20}N_4OS$) C, H, N, S.

Typical Procedure for the Preparation of *N*-Monosubstituted Aniline Derivatives 9−12: 2-[[2-(Methylamino)benzyl]sulfinyl]-1-[2-(3-methylpyridyl)]imidazole (10a). To a solution of 2-mercapto-1-[2-(3-methylpyridyl)]imidazole (**16a**) (4.23 g, 22.1 mmol) and NaH (60% oil suspended) (900 mg, 22.5 mmol) in DMF (40 mL) at 0 °C was added a solution of 2-phthalimidobenzyl bromide (**37**) (7.0 g, 22.1 mmol) in DMF (20 mL), and the reaction mixture was stirred for 3 h. After the removal of DMF, the residue was extracted with $CHCl_3$, and the organic layer was washed with saturated aqueous $NaHCO_3$ and brine, dried ($MgSO_4$), filtered, and concentrated in vacuo. The residue was crystallized from AcOEt−hexane to afford 2-[(2-phthalimidobenzyl)thio]-1-[2-(3-methylpyridyl)]-imidazole (**39b**: R^1 = 2-(3-methylpyridyl) $R^2 = R^3$ = H) (7.10 g, 75%) as colorless crystals: mp 121−122 °C (AcOEt/hexane). Anal. ($C_{24}H_{18}N_4O_2S$) C, H, N, S.

To a suspension of **39b** (7.05 g, 16.5 mmol) in EtOH (100 mL) at 0 °C was added NH_2NH_2 hydrate (1.0 g, 20.0 mmol). The reaction mixture was stirred at room temperature for 2 h and then concentrated in vacuo. The residue was dissolved in $CHCl_3$, and the organic layer was washed with saturated aqueous $NaHCO_3$ and brine, dried ($MgSO_4$), filtered, and concentrated in vacuo. The resulting oil (4.05 g, 83%) was dissolved in pyridine (50 mL), and trifluoroacetic acid anhydride (4.2 g, 20 mmol) was added to the solution at 0 °C. After the removal of pyridine, the residue was extracted with AcOEt, and the organic layer was washed with brine, dried ($MgSO_4$), filtered, and concentrated in vacuo. The residue was crystallized from ether to afford 2-[[2-(trifluoroacetoamido)benzyl]-thio]-1-[2-(3-methylpyridyl)]imidazole (**41a**: R^1 = 2-(3-methylpyridyl), $R^2 = R^3$ = H) (4.47 g, 86%) as colorless crystals: mp 146−147 °C (ether). Anal. ($C_{18}H_{15}F_3N_4OS$) C, H, N, F, S.

To a solution of the trifluoroacetyl derivative (2.3 g, 5.86 mmol) and NaH (60% oil suspension) (430 mg, 10.8 mmol) in DMF (30 mL) was added iodomethane (1.0 g, 7.04 mmol). After 3 h, the reaction mixture was poured over ice−water (100 mL) and extracted with AcOEt. The organic layer was washed with brine, dried ($MgSO_4$), filtered, and concentrated in vacuo. The resulting oil was chromatographed on silica gel with $CHCl_3$−MeOH (20:1) as an eluent to give **42a** (R^1 = 2-(3-methylpyridyl), $R^2 = R^3$ = H, R^4 = Me) (2.06 g, 87%) as a yellow syrup.

Oxidation of the sulfide **42a** (910 mg, 2.24 mmol) has been carried out with a procedure similar to that described for **7a** to give 2-[[2-[*N*-methyl-*N*-(trifluoroacetyl)amino]benzyl]sulfinyl]-1-[2-(3-methylpyridyl)]imidazole (**43a**: R^1 = 2-(3-methylpyridyl), $R^2 = R^3$ = H, R^4 = Me) (630 mg, 67%) as colorless crystals: mp 131−133 °C (ether). Anal. ($C_{19}H_{17}F_3N_4O_2S$) C, H, N, F, S.

To a solution of **43a** (250 mg, 0.62 mmol) in EtOH (5 mL) was added $NaBH_4$ (80 mg, 2.1 mmol). After 30 min, the reaction mixture was concentrated in vacuo, and the residue was dissolved in AcOEt. The organic layer was washed with brine, dried ($MgSO_4$), filtered, and concentrated in vacuo. The resulting oil was chromatographed on silica gel with $CHCl_3$−MeOH (20:1) as an eluent to give **10a** (158 mg, 78%) as a yellow syrup: ^{1}H NMR (δ in $CDCl_3$) 2.17 (s, 3H), 2.74 (s, 3H), 4.56 and 4.76 (ABq, 1H each, J = 13.0 Hz), 5.15 (br, 1H, D_2O exchangeable), 6.57−6.75 (m, 2H), 7.06−7.43 (m, 3H), 7.27 and 7.37 (d, 1H each, J = 1.0 Hz), 7.72 (m, 1H), 8.43 (m, 1H); IR (film) 3300, 1585, 1510, 1455, 1420, 1270, 1260, 1080, 1055 cm^{-1}; SIMS *m/z* 327 (M + H), 311, 138 (base).

Reaction of 12h (T-776) with 2-Mercaptoethanol under Acidic Conditions. Sulfoxide **12h** (352 mg, 1.0 mmol) was dissolved at room temperature in a stirred mixture of 0.1 N aqueous HCl (35 mL) and 1.2 mmol of 2-mercaptoethanol. After 30 min, the reaction was complete, and the pH of the reaction mixture was adjusted to 7.0 by means of a saturated $NaHCO_3$ solution in water. Extraction with ethyl acetate and evaporation under reduced pressure yielded a yellowish syrup, which was subjected to chromatography on silica gel with AcOEt as an eluent to give **45** (101 mg, 33%) as a yellow syrup, **46** (36 mg, 4%) as a yellow syrup, and **47** (81 mg, 24%) as a yellow syrup.

2-(4-Hydroxy-1,2-dithiabutyl)-1-[2-(3-methylpyridyl)]-1,4,5,6-tetrahydrocyclopent[*d*]imidazole (45): ^{1}H NMR (δ in $CDCl_3$) 2.19 (s, 3H), 2.39−2.62 (m, 5H), 2.71−2.78 (m, 2H), 2.97 (t, 2H, J = 5.0 Hz), 4.04 (t, 2H, J = 5.0 Hz), 7.35 (dd, 1H, J = 4.5, 7.5 Hz), 7.72 (dd, 1H, J = 1.0, 7.5 Hz), 8.44 (dd, 1H, J = 1.0, 4.5 Hz); IR (film) 3340, 3240, 2950, 2920, 2860, 1575, 1455, 1360, 1285, 1065, 1045, 1010, 800, 750, 665 cm^{-1}; EIMS *m/z* 307 (M^+), 231, 172 (base).

Bis[1-[2-(3-methylpyridyl)]-1,4,5,6-tetrahydrocyclopent-[*d*]imidazol-2-yl] disulfide (46): ^{1}H NMR (δ in $CDCl_3$) 2.14 (s, 6H), 2.28−2.62 (m, 8H), 2.70−2.77 (m, 4H), 7.27 (dd, 2H, J = 5.0, 7.5 Hz), 7.64 (dd, 2H, J = 1.5, 7.5 Hz), 8.36 (dd, 1H, J = 1.5, 5.0 Hz); IR (film) 2950, 2860, 1575, 1450, 1350, 750 cm^{-1}; FABMS *m/z* 461 (M + H), 232 (base); HRMS 461.1604 (calcd for $C_{24}H_{25}N_6S_2$ 461.1582).

2-[(2-Aminobenzyl)thio]-1-[2-(3-methylpyridyl)]-1,4,5,6-tetrahydrocyclopent[*d*]imidazole (47): ^{1}H NMR (δ in

$CDCl_3$) 2.09 (s, 3H), 2.36−2.63 (m, 4H), 2.71−2.79 (m, 2H), 3.90−4.40 (br, 2H), 4.19 (s, 2H), 6.57−6.66 (m, 2H), 6.95−7.08 (m, 2H), 7.33 (dd, 1H, J = 5.0, 7.0 Hz), 7.69 (dd, 1H, J = 1.0, 7.0 Hz), 8.44 (dd, 1H, J = 1.0, 5.0 Hz); IR (film) 3430, 3340, 3220, 2940, 2860, 1625, 1605, 1585, 1495, 1455, 1360, 1315, 750 cm^{-1}; EIMS m/z 336 (M^+), 231, 173, 106 (base).

Acknowledgment. We thank Drs. Mikio Takeda, Hajime Tamaki, Ikuo Iijima, and Tsutomu Takagi of our research laboratories for their useful comments and encouragement throughout this study and Ms. Yoko Yamamoto and Yuko Dainaka for their technical assistance.

References

(1) Prous, J. R. The year's new drugs. *Drug News Perspect*. **1988**, *1* (1), 38.

(2) (a) Fellenius, E.; Berglindh, T.; Sachs, G.; Olbe, L.; Elander, B.; Sjöstrand, S. E.; Wallmark, B. Substituted Benzimidazoles Inhibit Gastric Acid Secretion by Blocking (H^+,K^+)-ATPase. *Nature* **1981**, *290*, 159−161. (b) Olbe, L.; Berglindh, T.; Elander, B.; Helander, H.; Fellenius, E.; Sjöstrand, S. E.; Sundell, G.; Wallmark, B. Properties of a New Class of Gastric Acid Inhibitors. *Scand. J. Gastroenterol*. **1979**, *14* (Suppl. 55), 131−133.

(3) Lindberg, P.; Nordberg, P.; Alminger, T.; Brändström, A.; Wallmark, B. The Mechanism of Action of the Gastric Acid Secretion Inhibitor Omeprazole. *J. Med. Chem*. **1986**, *29*, 1327−1329.

(4) (a) Hakanson, R.; Sundler, F. Proposed Mechanism of Introduction of Gastric Carcinoids: The Gastric Hypothesis. *Eur. J. Clin. Invest*. **1990**, *20* (Suppl. 1), 65−71. (b) Creutzfeldt, W.; Stöckmann, F.; Conon, J. M.; Fölsch, U. R.; Bonatz, G.; Wülfrath, M. Effect of Short- and Long-Term Feeding of Omeprazole on Rat Gastric Endocrine Cells. *Digestion* **1986**, *35* (Suppl. 1), 84−97.

(5) (a) Figala, V.; Klemm, K.; Kohl, B.; Krüger, U.; Rainer, G.; Schaefer, H.; Senn-Bilfinger, J.; Sturm, E. Acid Activation of (H+,K+)-ATPase Inhibiting 2-[(2-Pyridylmethyl)sulfinyl]benzimidazoles: Isolation and Characterization of the Thiophilic 'Active Principle' and Its Reactions. *J. Chem. Soc., Chem. Commun*. **1986**, 125−127. Sturm, E.; Krüger, U.; Senn-Bilfinger, J.; Figala, V.; Klemm, K.; Kohl, B.; Rainer, G.; Schaefer, H. (H^+K^+)-ATPase Inhibiting 2-[(2-Pyridylmthyl)sulfinyl]benzimidazoles. 1. Their Reaction with Thiols under Acidic Conditions. Disulfide Containing 2-Pyridiniobenzimidazolides as Mimics for the Inhibited Enzyme. *J. Org. Chem*. **1987**, *52*, 4573−4581. (c) Senn-Bilfinger, J.; Krüger, U.; Sturm.; E.; Figala, V.; Klemm, K.; Kohl, B.; Rainer, G.; Schaefer, H.; Huttner, G.; Zsolnai, L. (H^+-K^+)-ATPase Inhibiting 2−[(2-Pyridylmthyl)sulfinyl]-benzimidazoles. 2. The Reaction Cascade Induced by Treatment with Acids. Formation of 5H-Pyrido[1′,2′:4,5][1,2,4]thiadiazino-[2,3-a]benzimidazol-13-ium Salts and Their Reactions with Thiols. *J. Org. Chem*. **1987**, *52*, 4582−4592. Krüger, U.; Senn-Bilfinger, J.; Sturm, E.; Figala, V.; Klemm, K.; Kohl, B.; Rainer, G.; Schaefer, H.; Blake, T. J.; Darkin, D. W.; Ife, R. J.; Leach, C. A.; Mitchell, R. C.; Pepper, E. S.; Salter, C. J.; Viney, N. J. (H^+-K^+)-ATPase Inhibiting 2-[(2-Pyridylmthyl)sulfinyl]-benzimidazoles. 3. Evidence for the Involvement of a Sulfenic Acid in Their Reactions. *J. Org. Chem*. **1990**, *55*, 4163−4168. (e) Kohl, B.; Sturm, E.; Senn-Bilfinger, J.; Alexander Simon, W.; Krüger, U.; Schaefer, H.; Rainer, G.; Figala, V.; Klemm, K.; Ife, R. J.; Leach, C. A. (H^+-K^+)-ATPase Inhibiting 2-[(2-Pyridylmthyl)sulfinyl]-benzimidazoles. 4. A Novel Series of Dimethoxypyridyl-Substituted Inhibitors with Enhanced Selectivity. The Selection of Pantoprazole as a Clinical Candidate. *J. Med. Chem*. **1992**, *35*, 1049−1057. (f) Herling, A. W.; Weidmann, K. Gastric H^+/K^+-ATPase Inhibitors. *Prog. Med. Chem*. **1994**, *31*, 233−264.

(6) The possibility of fine tuning the nucleophilic/electrophilic properties of the functional groups was raised by Kohl and coworkers[4e] prior to our studies. However, their modification was limited to the pyridine and benzimidazole moieties of [(pyridylmethyl)sulfinyl]benzimidazoles (PSBs).

(7) Independent of our work, a similar series of imidazole derivatives was described; see: Yamakawa, T.; Matsukura, H.; Nomura, Y.; Yoshioka, M.; Masaki, M.; Igata, H.; Okabe, S. Synthesis and Structure-Activity Relationships of *N*-Substituted 2-[(2-Imidazolylsulfinyl)methyl]anilines as a New Class of Gastric H^+/K^+-ATPase Inhibitors. *Chem. Pharm. Bull*. **1991**, *39*, 1746−1752.

(8) The syntheses of aniline derivatives were recently disclosed in literatures; see: (a) Okabe, S.; Satoh, M.; Yamakura, T.; Nomura, Y.; Hayashi, M. Belgian Patent No. BE 903128, 1986; *Chem. Abstr*. **1986**, *105*, 133881w. (b) Adelstein, G. W.; Yen, C. H.; Haack, R. A.; Yu, S.; Gullikson, G.; Price, D. V.; Anglin, C.; Decktor, D. L.; Tsai, H.; Keith, R. H. Substituted 2-[(2-Benzimidazolylsulfinyl)methyl]anilines as Potential Inhibitors of H^+/K^+ ATPase. *J. Med. Chem*. **1988**, *31*, 1215−1220. (c) Hirai, K.; Koike, H.; Ishiba, T.; Ueda, S.; Makino, I.; Yamada, H.; Ichihashi, T.; Mizushima, Y.; Ishikawa, M.; Ishihara, Y.; Hara, Y.; Hirose, H.; Shima, N.; Doteuchi, M. Amino Acid Amides of 2-[(2-Aminobenzyl)sulfinyl]benzimidazole as Acid-stable Prodrugs of Potential Inhibitors of H^+/K^+ ATPase. *Eur. J. Med. Chem*. **1991**, *26*, 143−158.

(9) (a) Manoharan, T. S.; Brown, R. S. 1-(1-Ethoxyethyl): An Effective Protecting Group for Imidazole Nitrogen. *J. Org. Chem*. **1988**, *53*, 1107−1110. (b) Dudfield, P. J.; Ekwuru, C. T.; Hamilton, K.; Osbowrn, C. E.; Simpson, D. J. Convenient Synthesis of 2-Cyanoimidazoles. *Synlett* **1990**, 277−278.

(10) Nunami, K.; Yamada, M.; Fukui, T.; Matsumoto, K. A Novel Synthesis of Methyl 1,5-Disubstituted Imidazole-4-carboxylates Using 3-Bromo-2-isocyanoacrylates. *J. Org. Chem*. **1994**, *59*, 7635−7642.

(11) Sorrell, T. N.; Allen, W. E. A Regiospecific Synthesis of 1,4-Disubstituted Imidazoles. *J. Org. Chem*. **1994**, *59*, 1589−1590.

(12) (a) Smith, H. E.; Hicks, A. A. Optically Active Amines. XII. Synthesis and Spectral Properties of Some Optically Active α-Oximino Ketones and α-Amino Ketone Hydrochlorides. Dimerization of α-Amino Ketones. *J. Org. Chem*. **1971**, *36*, 3659−3668. (b) *Organic Syntheses*; Wiley: New York, 1973; Collect. Vol. 5, p 909.

(13) Schubert, H.; Rühberg, B.; Fiedrich, G. Notiz zur Synthese des 4,5-Trimethylenimidazols. (Syntheses of 4,5-Trimethyleneimidazoles.) *J. Prakt. Chem*. **1966**, *32*, 249−253.

(14) Khan, R. H.; Rastogi, R. A. Simple Procedure for the Oxidation of Thiols to Disulphides. *Chem. Ind. (London)* **1989**, 282−283. (b) Fujihara, H.; Chiu, J.-J.; Furukawa, N. Synthesis, Conformation, and the Stereoselective Sulfur Extrusion Reaction of a New Intraannular Dibromo-Substituted Tetrathia[4.4]metacyclophane. *Chem. Lett*. **1991**, 141−144. McAllan, D. T.; Cullum, T. V.; Dean, R. A.; Fidler, F. A. The Preparation and Properties of Sulfur Compounds Related to Petroleum. I. The Dialkyl Sulfides and Disulfides. *J. Am. Chem. Soc*. **1951**, *73*, 3627−3631.

(15) The similar lack of correlation between the in vitro and in vivo tests was reported previously.[7,8b]

(16) (a) Ju, T.-L.; Kice, J. L. Mechanisms of Reactions of Thiolsulfinates (Sulfenic Anhydrides). 4. Acid- and Sulfide-Catalyzed Decomposition of *tert*-Butyl Benzenethiolsulfinate. *J. Org. Chem*. **1979**, *44*, 610−614. (b) Ostermayer, F.; Tarbell, D. S. Products of Acidic Hydrolysis of S-Methyl-L-cysteine Sulfoxide; the Isolation of Methyl Methanethiolsulfonate, and Mechanism of the Hydrolysis. *J. Am. Chem. Soc*. **1960**, *82*, 3752−3755.

(17) (a) Danehy, J. P.; Elia, V. J.; Lavelle, C. J. The Alkaline Decomposition of Organic Disulfides. IV. A Limitation 2,2′-Dinitro-5,5′-dithiodibenzoic Acid. *J. Org. Chem*. **1971**, *36*, 1003−1005. (b) Parker, A. J.; Kharasch, N. Derivatives of Sulfenic Acid. XXXVI. The Ionic Scission of the Sulfur-Sulfur Bond. Part 1. *J. Am. Chem. Soc*. **1960**, *82* 3071−3075. (c) Fava, A.; Iliceto, A.; Camera, E. Kinetics of the Thiol-Disulfide Exchange. *J. Am. Chem. Soc*. **1957**, *79*, 833−838. (d) Jayasuriya, N.; Regen, S. L. A. Convenient Procedure for Converting Organic Thiols into Ethyldithio Derivatives. *Tetrahedron Lett*. **1992**, *33*, 451−452. (e) Nagano, T.; Arakane, K.; Hirobe, M. Disulfide Interchange Reactions by Superoxide. *Tetrahedron Lett*. **1980**, *21*, 5021−5024. (f) Matsueda, R.; Higashida, S.; Ridge, R. J.; Matsueda, G. R. Activation of Conventional *S*-Protecting Groups of Cysteine by Conversion into the 3-Nitro-2-pyridinesulfenyl (NPYS) Group. *Chem. Lett*. **1982**, 921−924.

(18) (a) Attanasi, O. A.; Foresti, E.; Liao, Z.; Serra-Zanetti, F. Reaction of Heterocryl Thiols with Conjugated Azoalkenes: Regioselective Preparation of 4-(Heteroarylthio)-1*H*-pyrazol-5(2*H*)-ones. X-ray Crystal Structures of Methyl 2-((Pyrimid-2-yl)thio)acetoacetate (Aminocarbonyl)hydrazone and 1-(Aminocarbonyl)-3-methyl-4-((pyrimid-2-yl)thio)-1*H*-pyrazol-5(2*H*)-one. *J. Org. Chem*. **1995**, *60*, 149−155. (b) Matsueda, R.; Kimura, T.; Kaiser, E. T.; Matsueda, G. R. 3-Nitro-2-pyridinesulfenyl Group for Protection and Activation of the Thiol Function of Cysteine. *Chem. Lett*. **1981**, 737−740.

(19) Im, W. B.; Blakeman, D. P.; Davis, J. P. Irreversible Inactivation of Rat Gastric (H^+-K^+)-ATPase In Vivo by Omeprazole. *Biochem. Biophys. Res. Comm*. **1985**, *126*, 78−82.

(20) Lee, H. C.; Forte, J. G. A Study of H^+ Transport in Gastric Microsomal Vesicles Using Fluorescent Probes. *Biochem. Biophys. Acta* **1978**, *508*, 339−356.

(21) Keeling, D. J.; Falloowfield, C.; Milliner, K. J.; Tingley, S. K.; Ife, R. J.; Underwood, A. H. Study on the Mechanism of Action of Omeprazole. *Biochem. Pharmacol*. **1985**, *34*, 2967−2973.

(22) Yoda, A.; Hokin, L. E. On the Reversibility of Binding of Cardiotonic Steroids to a Partially Purified (Na + K)-Activated Adenosinetriphosphatase from Beef Brain. *Biochem. Biophys. Res. Commun*. **1970**, *40*, 880−886.

(23) Bradford, M. M. A Rapid and Sensitive Method for the Quantitation of Microgram Quantities of Protein Utilizing the Principle of Protein-Dye Binding. *Anal. Chem*. **1976**, *72*, 248−254.

(24) Wallmark, B.; Larsson, H.; Humble, L. The Relationship between Gastric Acid Secretion and Gastric H^+,K^+-ATPase Activity. *J. Biol. Chem*. **1985**, *260*, 13681−13684.

JM950610N