the product was isolated by extraction with ether. The organic layer was washed with water, dried, and evaporated. The solid residue was washed with hot cyclohexane, the solvent was removed, and the product was chromatographed on silica gel ($CHCl_3$ as eluant) to give 2,6-dimethyl-4-hydroxythiophenol (15.5 g, 50%), mp 105–106 °C. Anal. ($C_8H_{10}OS$) C, H, N, S.

2,4-Dimethyl-6-hydroxythiophenol. 2,4-Dimethyl-6-hydroxyaniline was obtained [78%, mp 158–159 °C (lit.[16] mp 163 °C)] by catalytic hydrogenation (Raney Ni catalyst, 1 atm) of the corresponding nitro derivative,[17] and it was converted to 2,4-dimethyl-6-hydroxythiophenol by the procedure described for the preparation of 2,6-dimethyl-4-hydroxythiophenol: yield, 65%; bp 84 °C (1.5 mm); mp 39–40.5 °C [lit.[18] bp 126–127 °C (19 mm)].

4-Nitrophenyl 2-Methyl-4-methoxyphenyl Sulfide (14a) (Table II). To a stirred solution of sodium hydroxide (0.6 g, 15 mmol) in water (20 mL) were added 4-nitrophenyl 2-methyl-4-hydroxyphenyl sulfide[19] (2.6 g, 10 mmol) and dimethyl sulfate (1.9 g, 15 mmol). The mixture was heated at 100 °C for 24 h, diluted with water, and extracted with ether. The organic layer was washed with 5% aqueous NaOH and with water, dried, and evaporated. The residue was chromatographed on silica gel (benzene as eluant) and recrystallized from 2-propanol to give **14a** (2.4 g, 89%).

Synthesis of Aryl 4-Nitrophenyl Sulfide (17a, 20a, and 23a) (Table II). To a solution of the appropriate thiophenol (50 mmol) in dry acetone (80 mL) were added dry potassium carbonate (60 mmol) and a solution of 4-chloronitrobenzene (50 mmol) in dry acetone (80 mmol). The mixture was refluxed under a nitrogen atmosphere for 7 h and the solvent was removed. Ice and water were added and the crude product was collected by filtration, washed with water, and recrystallized.

4-Nitrophenyl 2,4-Dimethyl-6-methoxyphenyl Sulfide (19a) (Table II). A mixture of 4-nitrophenyl 2,4-dimethyl-6-hydroxyphenyl sulfide (5.5 g, 20 mmol), dimethyl sulfate (10 g, 79 mmol), and 45 mL of 8% aqueous NaOH was stirred at 100 °C for 24 h. After cooling, water was added and the mixture was extracted with benzene. The extracts were washed with 8% NaOH and water, dried, and evaporated to give a solid residue, which was recrystallized from 2-propanol to yield sulfide **19a** (4 g, 69%).

4-Nitrophenyl 2,6-Dimethyl-4-methoxyphenyl Sulfide (22a) (Table II). To a solution of sodium (2.5 g) in absolute ethanol (70 mL) was added 2,6-dimethyl-4-methoxythiophenol (16.8 g, 100 mmol) followed by 15.7 g (100 mmol) of 4-nitrochlorobenzene dissolved in 100 mL of absolute ethanol. The mixture was refluxed under a nitrogen atmosphere for 3 h and was allowed to stand overnight at room temperature. The precipitate obtained was collected by filtration, washed with cold ethanol and water, and recrystallized from ethanol to afford **22a** (24 g, 83%).

Table III. Aryl 4-Aminophenyl Sulfones ($4\text{-}NH_2C_6H_4SO_2C_6H_2\text{-}2'\text{-}X,4'\text{-}Y,6'\text{-}Z$)

no.	X	Y	Z	% yield[a]	mp, °C	recrystn solv[b]	formula[c]
14	CH_3	OCH_3	H	89	144–145	A	$C_{14}H_{15}NO_3S$
15	CH_3	OH	H	76	163–164	B	$C_{13}H_{13}NO_3S$
17	Cl	OH	H	95	205–207[d]	C–B	$C_{12}H_{10}ClNO_3S$
19	CH_3	CH_3	OCH_3	83	191–192	C	$C_{15}H_{17}NO_3S$
20	CH_3	CH_3	OH	74	170–171	D	$C_{14}H_{15}NO_3S$
22	CH_3	OCH_3	CH_3	77	202–203	A	$C_{15}H_{17}NO_3S$
23	CH_3	OH	CH_3	96	178–179	C–B	$C_{14}H_{15}NO_3S$

[a,c] See corresponding footnotes in Table II. [b] A = 2-propanol, B = water, C = ethanol, D = benzene. [d] Literature[20] mp 209–216 °C.

Synthesis of Aryl 4-Nitrophenyl Sulfones (Table II). The sulfones were prepared according to the following methods.

Method A. A solution of the sulfide (10 mmol) in acetic acid was heated at 100 °C and hydrogen peroxide (30% v/v, 25 mmol) was added dropwise. The solution was concentrated and diluted with ice–water. The crude product was collected by filtration, washed with water, and purified by crystallization or column chromatography on silica gel.

Method B. To a stirred solution of the sulfide (10 mmol) in chloroform was added slowly 3-chloroperbenzoic acid (85%, 25 mmol) in 70 mL of chloroform at 0 °C. The 3-chlorobenzoic acid and unchanged peroxy acid were removed by washing with dilute alkali and dilute aqueous sodium sulfite. Removal of the solvent gave the crude product, which was purified by crystallization or column chromatography on silica gel.

Synthesis of Aryl 4-Aminophenyl Sulfones (14, 17, 19, 20, 22, and 23) (Table III). The amino derivatives were prepared by catalytic hydrogenation (Raney Ni catalyst, 1 atm, room temperature) of the corresponding nitro compounds in methanol. When the calculated amount of H_2 had been absorbed, the catalyst was removed by filtration. Crude products obtained after removal of the solvent were purified by crystallization.

4-Aminophenyl 4-Hydroxy-2-methylphenyl Sulfone (15). A solution of sulfone 14 (2.5 g) in 22 mL of 48% hydrobromic acid was stirred at 130 °C for 36 h. Excess of hydrobromic acid was evaporated and the residue was dissolved in water (100 mL), basified with aqueous sodium hydroxide, and filtered with charcoal. The solution was neutralized with dilute hydrochloric acid, and the crude product was collected by filtration and recrystallized from water to afford sulfone **15** (1.8 g, 76%).

(17) Adams, R.; Stewart, H. W. *J. Am. Chem. Soc.* **1941**, *63*, 2859.
(18) Cabiddu, S.; Maccioni, A.; Secci, M. *Gazz. Chim. Ital.* **1969**, *99*, 1095.
(19) Foss, N. E.; Dunning, F.; Jenkins, G. L. *J. Am. Chem. Soc.* **1934**, *56*, 1978.
(20) Acharya, S. P.; Badiger, V. V.; Nargund, K. S. *J. Karnatak Univ.* **1958**, *3*, 25; *Chem. Abstr.* **1960**, *54*, 3289.

Dihydropyrimidines: Novel Calcium Antagonists with Potent and Long-Lasting Vasodilative and Antihypertensive Activity

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The novel calcium antagonists 3-*N*-substituted-3,4-dihydropyrimidines **1** and **9** and 3-*N*-substituted-dihydropyrimidin-2(1*H*)-ones **8** were regioselectively synthesized in good yields. Compounds **1** [especially **1s** [R^1 = $(CH_2)_2N$(benzyl)(2-naphthylmethyl), R^2 = *i*-Pr, X = *o*-NO_2] and **1t** [R^1 = $(CH_2)_2N$(benzyl)(3,4-dichlorobenzyl), R^2 = *i*-Pr, X = o-NO_2]] exhibited not only more potent and longer lasting vasodilative action but also a hypotensive activity with slow onset as compared with dihydropyridines. Moreover, some dihydropyrimidines [**1q** [R^1 = $(CH_2)_2N$(benzyl)(3-phenylpropyl), R^2 = CH_2(cyclopropyl), X = o-NO_2], **1s**, and **1t**] were weaker in blocking atrioventricular conduction in anesthetized open-chest dogs and less toxic than the dihydropyridines.

1,4-Dihydropyridine derivatives possessing calcium antagonistic action in the cardiovascular system have attracted much synthetic attention over the past 20 years. Calcium antagonists inhibit the influx of calcium ions

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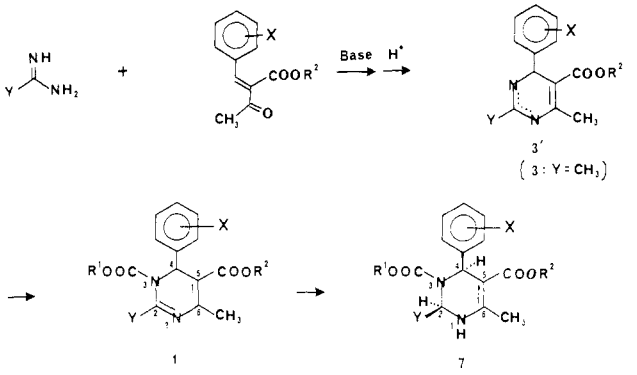
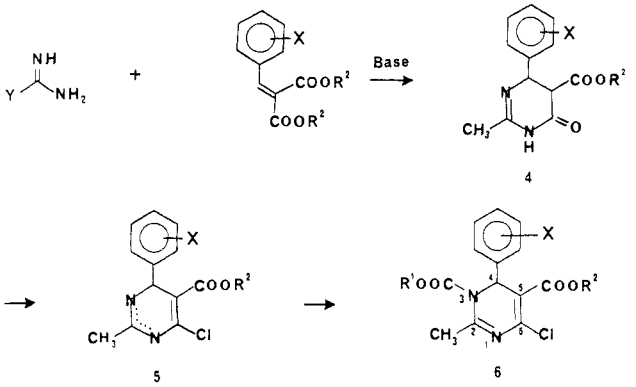
Scheme I



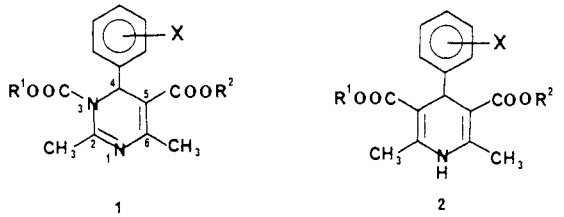
Scheme II



through plasma membrane channels and thus dilate vascular smooth muscle and alleviate the force of cardiac muscle contraction. Some calcium antagonists such as nifedipine and verapamil have been used as antihypertensive agents. However, nifedipine and verapamil have a serious disadvantage in the treatment of hypertension. Since their plasma half-lives are relatively short, these drugs must be administered repeatedly to achieve enough clinical efficacy, and the multiple dosages lower compliance.[1,2] Therefore, we approached the problems by synthesizing new types of compounds that have long-lasting hypotensive or antihypertensive activity.

Since 1967, a variety of dihydropyridine derivatives have been synthesized in the search for more potent vasodilating compounds with longer duration of action. Recently, Bayer A. G. synthesized analogues of the dihydropyridine skeleton itself, for instance, dihydropyran,[3a] dihydrothiopyran,[3b] dihydropyridazine,[3c] and dihydropyrazine.[3d]

Because of the structural similarity between dihydropyridine and dihydropyrimidine, we became interested in the synthesis and pharmacological activities of 3-*N*-substituted-3,4-dihydropyrimidines **1** or **6**. However, the chemistry of dihydropyrimidines had not been sufficiently investigated before our synthetic studies. This is because the chemical structure becomes ambiguous and complicated due to tautomerism and isomerization.[4,6,7] Moreover, in the case of synthesizing an *N*-substituted-dihydropyrimidine from a 1,4(3,4)-dihydropyrimidine, the selectivity of alkoxycarbonylation and alkylation toward the two nitrogen atoms of the skeleton cannot be clearly predicted.[5] In view of these problems, we carried out a novel dihydropyrimidine synthesis[5,8] (Scheme I).



Generally, in the case of dihydropyridine **2** with an *o*-nitro group on the phenyl ring [for example, nifedipine (**2**, X = *o*-NO_2, R^1 = R^2 = Me], the dihydropyridine skeleton is light-sensitive, because a nitroso radical generated from a nitro group by light irradiation removes an α-proton from the phenyl group causing aromatization.[9] The transposition of a nitro group from ortho substitution to meta substitution [for instance, nicardipine [**2**, X = *m*-NO_2, R^1 = $(CH_2)_2NMe(Bzl)$, R^2 = Me]] prevented the oxidation of the dihydropyridine ring, although the pharmacological activity of meta-substituted compounds is usually less potent than that of ortho-substituted compounds. However, in the case of dihydropyrimidines with *o*-nitro groups, the aza analogue of dihydropyridines, this problem could theoretically be overcome. They are stable against sunlight, because aromatization cannot occur due to the chemical structural characteristics of an *N*-substituted-3,4-dihydropyrimidine.

Usually, some of the dihydropyridines exhibit a potent and extremely rapid hypotensive effect, and sometimes this abrupt hypotension results in undesirable pharmacological side effects. Moreover, most potent dihydropyridines exhibit short durations of action (e.g., nifedipine, nilvadipine, and nitrendipine).[10]

Although compounds **1** do not have the N-1 hydrogen, which is thought to be prerequisite for good affinity for the dihydropyridine receptor, we found some 3,4-dihydropyrimidines (**1s** and **1t**) to displace competitively [^{3}H]nitrendipine from its binding site on the cell membrane prepared from the rat cerebral cortex (Table IV). Hence, we wish to report the details here.

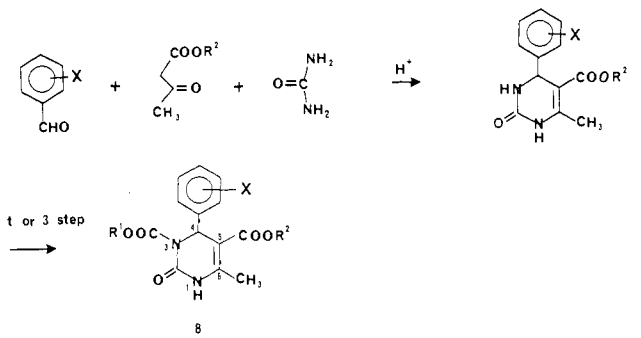
Chemistry

1,4(3,4)-Dihydropyrimidines **3′** were synthesized as illustrated in Scheme I (Y = Me, X = NO_2, CN, CF_3, Br, Cl, SMe). Novel N-alkoxycarbonylation of a series of compounds **3′** with alkyl chloroformate (e.g., ClCOOEt, $ClCOOC_7H_{15}$) in the presence of NaH or Et_3N was carried out by our reported procedure.[5] To introduce a nitrogen-containing alkyl ester group at position 3, the phosgene dimer procedure was developed to give regioselectively 3,4-dihydropyrimidines **1′** in good yields[8] (Scheme I) (Table I).

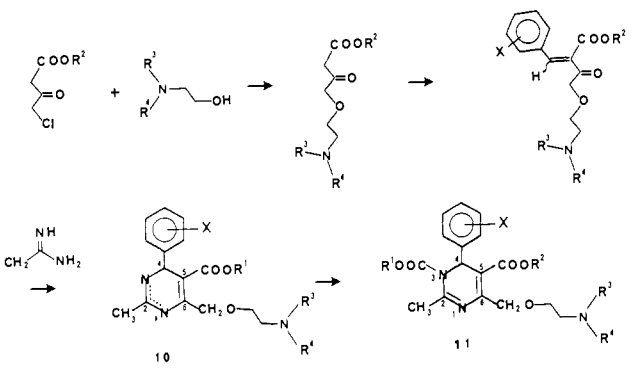
Subsequently, dihydropyrimidones **4** were prepared according to the previously reported procedure (Scheme II; Y = Me).[11]

(1) Buhler, F. R.; Hukthen, U. L.; Kiowski, W. *J. Cardiovasc. Pharmacol.* **1982**, *4*, 350.
(2) Belz, G. G.; Spies, G. *Excerpta Med.* **1986**, 177.
(3) (a) Bayer A. G. patent, DE 2235406.9, 1972. (b) Bayer A. G. patent, DE 3212737.5, 1982. (c) Bayer A. G. patent, DE 834624, 1978. (d) Bayer A. G. patent, DE 3400765, 1984.
(4) Silbersmith, E. F. *J. Org. Chem.* **1962**, *27*, 4090.
(5) Cho, H.; Shima, K.; Hayashimatsu, M.; Ohnaka, Y.; Mizuno, A.; Takeuchi, Y. *J. Org. Chem.* **1985**, *50*, 4227.
(6) Cho, H.; Iwashita, T.; Ueda, M.; Mizuno, A.; Mizukawa, K.; Hamaguchi, M. *J. Am. Chem. Soc.* **1988**, *110*, 4832.
(7) Cho, H.; Hamamoto, Y.; Shima, K.; Taira, Z. *Tetrahedron* **1986**, *42*, 6429.
(8) Cho, H.; Mizuno, A.; Shima, K.; Ueda, M.; Takeuchi, Y.; Hamaguchi, M.; Taniguchi, N. *Heterocycles* **1988**, *27*, 769.
(9) (a) Morad, N.; Goldman, Y. E.; Trentham, D. R. *Nature (London)* **1983**, *304*, 635. (b) Schlossmann, V. K. *Arzneim.-Forsch. (Drug Res.)* **1972**, *22*, 60.
(10) Pepine, C. J.; Conti, C. R. *Mod. Concepts Cardiovasc. Dis.* **1981**, *50*, 61.

Scheme III



Scheme IV



First, successful chlorination of **4** with $POCl_3$ gave dihydropyrimidines **5** whose alkoxycarbonylation yielded compounds 6.[12] Also, tetrahydropyrimidines **7** were prepared by stereoselective reduction of dihydropyrimidines **1** with $NaBH_4$.[5]

A variety of substituted dihydropyrimidines were synthesized in the search for more potent long-lasting antihypertensive and chemically stable compounds. Thus, compounds **8**, which have an alkoxycarbonyl group at position 2, were synthesized by the following reactions: (i) construction of dihydropyrimidin-2-one skeleton and (ii) regioselective alkoxycarbonylation[13] (Scheme III).

We intended to prepare dihydropyrimidine derivatives with various substituents at positions 2 and 6. Thus, 1,4(3,4)-dihydropyrimidine derivatives **3′** were constructed from various amidines and benzylidene derivatives as shown in Scheme I (Y = H, Et, *i*-Pr, SMe, CF_3, NMe_2). Subsequent reaction of **3′** with alkyl chloroformate or phosgene dimer yielded **9**.

In the case of synthesizing compound **11** with a long side chain, the benzylidene derivative was prepared as shown in Scheme III and then treated with an amidine to give **10**, which was reacted with alkyl chloroformate (Scheme IV).

Pharmacology

The pharmacological activities of the dihydropyrimidine derivatives were evaluated by the following tests.

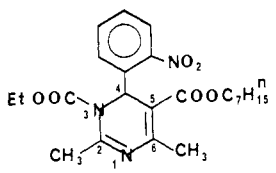
The potency (ED_{30}) and the duration ($t_{1/2}$) of the vasodilative action on the vertebral artery in anesthetized dogs were calculated according to the procedures described under Experimental Section (pharmacological method I).

The selected compounds in terms of the vasodilative potency were subjected to an antihypertensive test using conscious spontaneously hypertensive rats (SHR) (pharmacological method II), to a test of adverse effects on atrioventricular conduction system in anesthetized open-chest dogs, and to a [^{3}H]nitrendipine binding assay using the rat cerebral cortex (pharmacological tests IV and V; Tables III and IV).

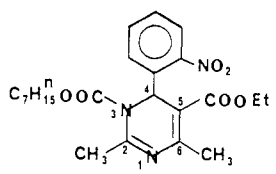
Results and Discussion

(1) 3-*N*-Substituted-3,4-dihydropyrimidines 1. Initially, compounds **3**, **4**, and **5** were subjected to pharmacological test I, but they were not potent vasodilators. However, a series of compounds **1** exhibited very potent vasodilative activity (ED_{30} = 1.0–3.0) comparable to that of nicardipine, but the duration of action was as short as nifedipine. Also, 3-*N*-alkylated-dihydropyrimidines showed weak activity. At this stage, the effect of the substituents on the phenyl ring was studied, and the vasodilative potency of compound **1** was found to decrease in the following order: X = *o*-NO_2 > *o*-Cl > *o*-Br > *m*-NO_2.

In order to obtain compounds that have a longer lasting vasodilative activity, the most desired length of an alkyl chain of an ester group at position 3 was changed from methyl to *n*-nonyl by keeping the alkyl chain of an ester group at position 5 fixed with ethyl. We found that the compounds with a shorter chain showed more potent vasodilative activity but of shorter duration, while the compounds with a longer chain exhibited less potent activity of longer duration. Interestingly, compound **1e** with a $COOC_7H_{15}$ group at position 3 was more potent than the corresponding compound **1f** with $COOC_7H_{15}$ at position 5. The generality of this phenomenon can be illustrated by other examples (**1h** vs **1i**, etc.) (Table I).



1f



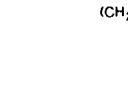
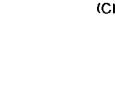
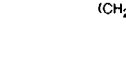
1e

As for duration of action, mostly oily compounds **1** with 5-(cyclopropylmethyl) groups were superior to those with smaller alkyl groups (e.g., Me, Et, *i*-Pr). However, the compounds **1** with an isopropyl ester group crystallized easily. For these reasons, the isopropyl ester group was chosen as the most suitable group to be substituted at position 5.

The potency and duration of vasodilator action of the dihydropyrimidines **1** were greatly improved by substituting an *N*-$(CH_2)_nC_6H_5$ (n = 0–3) or a 2-naphthylmethyl or a 3,4-dichlorobenzyl group for the *N*-methyl group of the alkyl chain at position 3 (Table I). The crystalline compounds **1l** and **1m** showed potent vasodilator action, but the duration of action was approximately equal to that of nicardipine. As n in the side chain *N*-$(CH_2)_nC_6H_5$ increases, the duration of action lengthens. Such promising results with the oily compounds **1q** and **1r** prompted us to search for crystalline compounds. Addition of a π electronic system (C=C) to the cinnamyl group of **1r** would furnish a 2-naphthylmethyl group. Thus, the crystalline compound **1s** was obtained, possessing the desired pharmacological profile. Compound **1t** with a 3,4-dichloro-substituted phenyl group was also synthesized, which showed promising activity similar to that of the naphthyl group. Both compounds **1s** and **1t** had the same R_f value (polarity) on TLC. Generally, the duration of the

(11) Cho, H.; Ueda, M.; Ohnaka, Y.; Hayashimatsu, M. *Heterocycles* **1984**, *22*, 1959.
(12) Cho, H.; Ohnaka, Y.; Hayashimatsu, M.; Ueda, M.; Shima, K. *Tetrahedron Lett.* **1986**, *27*, 6377.
(13) Cho, H.; Takeuchi, Y.; Ueda, M.; Mizuno, A. *Tetrahedron Lett.* **1988**, *42*, 5405.
(14) Meguro, K.; Aizawa, M.; Sohda, T.; Kawamatsu, Y.; Nagaoka, A. *Chem. Pharm. Bull.* **1985**, *33*, 3787.

Table I. Vasodilative Effects of Dihydropyrimidine Derivatives on Vertebral Vascular Beds in Anesthetized Dogs

compd[a]	R^1	R^2	X	ED_{30}[b]	T_P[c]	$T_{1/2}$[d]	N[e]	mp, °C	solvent of recrystn	molecular formula	anal.
1a	Et	Et	*o*-NO_2	3.0		0.4	3	130–131.5	acetone–*n*-hexane	$C_{18}H_{21}N_3O_6$	C, H, N
1b	Et	Et	*m*-NO_2	10.0		0.4	2	69		$C_{18}H_{21}N_3O_6$	C, H, N
1c	Et	Et	*o*-Cl	5.0			3	78–79	*n*-hexane	$C_{18}H_{21}ClN_2O_4$	C, H, N
1d	Et	Et	*o*-Br	5.0			3	74	*n*-hexane	$C_{18}H_{21}BrN_2O_4$	C, H, N
1e	C_7H_{15}	Et	*o*-NO_2	3.2		1.4	3	oil		$C_{23}H_{31}N_3O_6$	C, H, N
1f	Et	C_7H_{15}	*o*-NO_2	23.0			2	oil		$C_{23}H_{31}N_3O_6$	C, H, N
1g	C_7H_{15}	C_7H_{15}	*o*-NO_2	180			2	oil		$C_{28}H_{41}N_3O_6$	C, H, N
1h	$(CH_2)_2N(Me)(Bzl)$	Et	*o*-NO_2	0.75		4.0	3	oil		$C_{26}H_{30}N_4O_6$	C, H, N
1i	Et	$(CH_2)_2N(Me)(Bzl)$	*o*-NO_2	13.0			2	83–84	AcOEt–*n*-hexane	$C_{26}H_{29}N_4O_6$	C, H, N
1j	C_7H_{15}		*o*-NO_2	5.6		5.0	3	oil		$C_{25}H_{33}N_3O_6$	C, H, N
1k	$(CH_2)_2N(Me)(Bzl)$		*o*-NO_2	0.5		5.6	3	oil		$C_{28}H_{32}N_4O_6$	C, H, N
1l	$(CH_2)_2N(Bzl)(C_6H_5)$	*i*-Pr	*o*-NO_2	0.74	1.9	4.3	3	121–123	$CHCl_3$–*n*-hexane	$C_{32}H_{34}N_4O_6$	C, H, N
1m	$(CH_2)_2N(Bzl)_2$	*i*-Pr	*o*-NO_2	2.8	2.6	5.9	3	102–103	$CHCl_3$–*n*-hexane	$C_{33}H_{36}N_4O_6$	C, H, N
1n	$(CH_2)_2N(Bzl)_2$	*n*-C_4H_9	*o*-NO_2	6.8	4.7	13.0	2	oil		$C_{34}H_{38}N_4O_6$	C, H, N
1o	$(CH_2)_2N(Bzl)_2$	*n*-C_7H_{15}	*o*-NO_2	>30			1	oil		$C_{37}H_{44}N_4O_6$	C, H, N
1p	$(CH_2)_2N(Bzl)[(CH_2)_2C_6H_5]$	*i*-Pr	*o*-NO_2	1.2	2.7	6.8	3	oil		$C_{34}H_{38}N_4O_6$	C, H, N
1q	$(CH_2)_2N(Bzl)[(CH_2)_3C_6H_5]$		*o*-NO_2	4.3	4.8	13.5	3	oil		$C_{36}H_{40}N_4O_6$	C, H, N
1r			*o*-NO_2	3.9	5.6	9.6	3	oil		$C_{36}H_{38}N_4O_6$	C, H, N
1s		*i*-Pr	*o*-NO_2	1.8	7.9	15.6	3	85–89	*i*-Pr_2O–*n*-hexane	$C_{37}H_{38}N_4O_6$	C, H, N
1t		*i*-Pr	*o*-NO_2	2.1	10.8	>25.0	3	81–85	*i*-Pr_2O	$C_{33}H_{34}Cl_2N_4O_6$	C, H, N
6a		Et	*o*-NO_2	10.0	5.2	10	2	oil		$C_{35}H_{33}ClN_4O_6$	C, H, N
7a		*i*-Pr	*o*-NO_2	13.5	7.2	8.5	3	powder		$C_{37}H_{40}N_4O_6$	C, H, N
7b		*i*-Pr	*o*-NO_2	19.5	5.5	20	3	powder		$C_{33}H_{36}N_4O_6Cl_2$	C, H, N
8a	*n*-C_7H_{15}		*m*-NO_2	3.0		2.0	3	70–72	AcOEt–*n*-hexane	$C_{24}H_{31}N_3O_7$	C, H, N
8b	*n*-C_7H_{15}		*o*-NO_2	2.3	1.9	4.0	3	101–104	AcOEt–*n*-hexane	$C_{24}H_{31}N_3O_7$	C, H, N
8c	$(CH_2)_2N(Me)(Bzl)$		*m*-NO_2	13.0		2.0	3	oil		$C_{27}H_{30}N_4O_7$	C, H, N
8d	$(CH_2)_2N(Me)(Bzl)$	Et	*m*-Cl	43.0		1.0	2	oil		$C_{25}H_{28}ClN_3O_5$	C, H, N
8e		*i*-Pr	*o*-NO_2	10.0	7.8	10.9	2	oil		$C_{36}H_{36}N_4O_7$	C, H, N

8f	$CH_2CH(C_6H_5)_2$	Et	*m*-NO_2	10.6		2.8	2	oil	$C_{30}H_{29}N_3O_7$ C, H, N
nifedipine (**2a**)	Me	Me	o-NO_2	0.5	0.7	2.5	3		
nicardipine (**2b**)	$(CH_2)_2N(Me)(Bzl)$	Me	*m*-NO_2	3.0	1.2	5.6	3		
nitrendipine (**2c**)	Et	Me	*m*-NO_2	1.2	1.0	7.4	3		

[a] Each compound was administered intravenously. [b] ED_{30}: dose that lowered vascular resistance at 30% of control value (μg/kg). [c] T_p: time to peak response (min). [d] $T_{1/2}$: time to 50% recovery (min). [e] *N*: number of animals used. [f] Bzl: $CH_2C_6H_5$.

pharmacological activity seems to increase as the hydrophobicity increases.

Replacement of a methyl group with a chlorine atom at position 6 did not enhance pharmacological activity (compound **6**, Table I).

The dihydropyrimidines **1q**, **1s**, and **1t** were weaker than the dihydropyridines (nicardipine and nilvadipine) in blocking atrioventricular conduction in anesthetized open-chest dogs (Table III) and also less toxic in the acute toxicity test in mice.

Next, the vasodilative effects of tetrahydropyrimidines **7** were also examined. As shown in Table I, compounds **7a** and **7b** were less active than the corresponding dihydropyrimidines.

(2) *N*-Substituted-dihydropyrimidin-2(1*H*)-ones 8 or -2(3*H*)-ones. 3-*N*-Substituted-dihydropyrimidin-2-(1*H*)-ones **8** have a chemical similarity to dihydropyridines and 3-*N*-substituted-dihydropyrimidines **1**. Although compound **1** has an endocyclic π electronic system in a dihydropyrimidine ring, compound **8** has an exo π electronic system as a carbonyl group. We supposed that the difference in electronic density and location might influence pharmacological activity.

As reported previously, two different dihydropyrimidin-2-ones [1-*N*-substituted-dihydropyrimidin-2-(3*H*)-one and 3-*N*-substituted-dihydropyrimidine-2-(1*H*)-one] could be obtained according to the reaction conditions. The chemical structure of 1-*N*-substituted-dihydropyrimidin-2(3*H*)-ones is quite different from that of the ordinary dihydropyridines **2**. In fact, dihydropyrimidin-2(3*H*)-ones showed no vasodilative activity. However, 3-*N*-substituted-dihydropyrimidin-2(1*H*)-ones **8** showed fairly potent activity, especially **8a** and **8b** (Table I).

(3) Dihydropyrimidines 9 with Various Substituents at Position 2 and Position 6. Dihydropyrimidines with various substituents excluding methyl groups at position 2 were synthesized[5,6] (Table II). Replacement with a bulky isopropyl group instead of a methyl group resulted in weaker activity, although compounds **9c** and **9d** possessing an ethyl group remained active. Compounds **9e** and **9f** with a hetero atom at position 2 and **11** with a longer side chain at position 6 were inactive. However, a trifluoromethyl group proved to be a more effective substituent than the others. Thus, compound **9g** is comparable in activity to nicardipine.

9 (1′)

In conclusion, among a number of compounds tested in the primary screening, 3-*N*-substituted-3,4-dihydropyrimidines **1s** and **1t** exhibited very potent vasodilative activity in dilating the vertebral artery of the anesthetized dogs. Therefore, these dihydropyrimidines were subjected to the antihypertensive test using the conscious spontaneously hypertensive rats (SHR) (Figure 1).

Oral administration of compounds **1s** and **1t** (10 mg/kg) caused a decrease in mean blood pressure and an increase in heart rate similar to those caused by nicardipine and nitrendipine. When the peak response were compared, the activity of **1s** was almost the same as that of nicardipine and nitrendipine. The hypotensive action of nicardipine and nitrendipine reached the maximum immediately after

Table II. Vasodilative Effects of Dihydropyrimidine Derivatives on Vertebral Vascular Beds in Anesthetized Dogs

compd[a]	R^1	R^2	X	Y	Z	ED_{30}[b]	$T_{1/2}$[d]	N[e]	mp, °C	solvent of recrystn	molecular formula	anal.
9a	$(CH_2)_2N(Bzl)CH_2$	*i*-Pr	*o*-NO_2	*i*-Pr	Me	>100		2	oil		$C_{39}H_{42}N_4O_6$	C, H, N
9b	$(CH_2)_2N(Bzl)CH_2$	*i*-Pr	*o*-NO_2	H	Me	>100		2	oil		$C_{36}H_{36}N_4O_6$	C, H, N
9c	$(CH_2)_2N(Bzl)CH_2$	*i*-Pr	*o*-NO_2	Et	Me	27	19.5	2	oil		$C_{38}H_{40}N_4O_6$	C, H, N
9d	Et	*i*-Pr	*o*-NO_2	Et	Me	8.0	0.7	2	90–92	*i*-Pr_2O-*n*-hexane	$C_{20}H_{25}N_3O_6$	C, H, N
9e	$(CH_2)_2N(Bzl)CH_2$	*i*-Pr	*o*-NO_2	S(Me)	Me	>100		2	oil		$C_{37}H_{38}N_4O_6$	C, H, N
9f	$(CH_2)_2N(Bzl)CH_2$	*i*-Pr	*o*-NO_2	$N(Me)_2$	Me	>100		2	oil		$C_{38}H_{41}N_5O_6$	C, H, N
9g	$(CH_2)_2N(Bzl)_2$	*i*-Pr	*o*-NO_2	CF_3	Me	6.8	8.8	3	oil		$C_{33}H_{33}F_3N_4O_6$	C, H, N
9h	Et	*i*-Pr	*o*-NO_2	CF_3	Me	8.3	0.7	3	142–143	*i*-Pr_2O	$C_{19}H_{20}F_3N_3O_6$	C, H, N
11	Me	Et	*o*-NO_2	Me	$CH_2O(CH_2)_2N(Bzl)CH_2$ (2,3-dichlorophenyl)	>100		2	112–116 (HCl salt)	Et$_2$O-acetone	$C_{33}H_{34}Cl_2N_4O_6$	C, H, N

[a] Each compound was administered intravenously. [b] ED_{30}: dose that lowered vascular resistance at 30% of control value (μg/kg). [c] T_P: time to peak response (min). [d] $T_{1/2}$: time to 50% recovery (min). [e] N: number of animals used. [f] Bzl: $CH_2C_6H_5$.

Table III. Effects of Dihydropyrimidine Derivatives and Two Reference Compounds on Atrioventricular Conduction System in Anesthetized Open-Chest Dogs

compd	ED_{30}, μg/kg[a,c]	AV block[b]
1q	843 ± 375	1000–3000
1s	762 ± 317	1000–3000
1t	596 ± 246	1000–3000
nicardipine	175 ± 88	100–3000
nilvadipine	34 ± 13	100–300

[a] Dose at which they prolong AVCT by 30%. [b] Dose at which they induce AV block. [c] Each value represents mean ± SE (N = 4–5).

Table IV. Effects of Dihydropyrimidine Derivatives and Four Reference Compounds on [^{3}H]Nitrendipine Binding

compd	IC_{50}, nM[a]	compd	IC_{50}, nM[a]
1s	7.08 ± 1.95	nilvadipine	1.04 ± 0.26
1t	2.05 ± 0.51	nifedipine	2.53 ± 0.37
nicardipine	2.66 ± 0.49	CV-4093[b]	8.68 ± 2.42

[a] Each value represents mean ± SE (N = 3). [b] Reference 14.

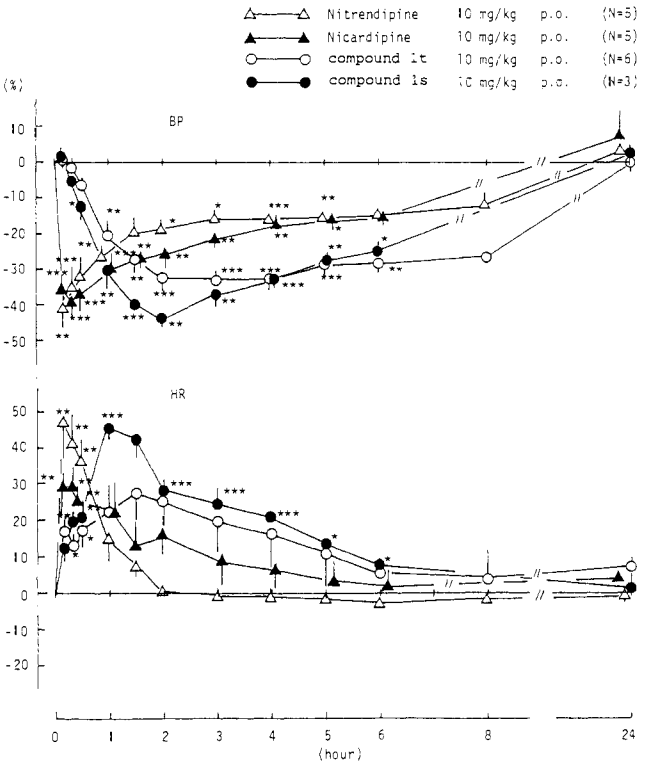


Figure 1. Effects of nitrendipine, nicardipine, and compounds **1s** and **1t** on mean blood pressure (BP) and heart rate (HR) in consciously hypertensive rats. (*) $p < 0.05$; (**) $p < 0.01$; (***) $p < 0.001$.

administration, while that of **1s** and **1t** did so in 2 h after administration. On the contrary, when the hypotensive responses were compared more than 2 h after administration, the hypotensive effects of **1s** and **1t** were more potent than those of nicardipine and nitrendipine. No animal died when compound **1s** at a dose of 1 g/kg or compound **1t** at a dose of 2 g/kg was administered orally, respectively (see pharmacological method III). These results suggest that the novel calcium antagonists **1s** and **1t** may have an antihypertensive property with favorable features.

Experimental Section

General Methods (Chemistry). Melting points were determined with a Yanagimoto micro-melting point apparatus and are uncorrected. ^{1}H NMR spectra were recorded with a JEOL GX-270 (270-MHz) spectrometer in $CDCl_3$ solution with tetramethylsilane (Me_4Si) as an internal standard. IR spectra were

taken on a Hitachi 260-10 infrared spectrometer in $CHCl_3$, unless otherwise noted. UV spectra were recorded on a Beckman DU-8 spectrophotometer. Thin-layer chromatography (TLC) was carried out on Merck silica gel plates 60F-254.

Column chromatography was performed on Merck silica gel (70–230 mesh). The dihydropyrimidines described here were synthesized according to our previous papers: compounds 1, 3 and 7 were prepared according to ref 5; compound 4 was prepared according to ref 11; compounds 5 and 6 were prepared according to ref 12; compound 8 was prepared according to ref 13; compound 9 was prepared according to refs 5 and 6.

Typical Procedure for the Preparation of Dihydropyrimidines. Synthesis of 3-[[2-[*N*-Benzyl-*N*-(2-naphthylmethyl)amino]ethoxy]carbonyl]-5-(isopropoxycarbonyl)-2,6-dimethyl-4-(2-nitrophenyl)-3,4-dihydropyrimidine (1s). To a stirred solution of 479 μL (4.00 mmol) of phosgene dimer in 10 mL of tetrahydrofuran (THF) was added a solution of 2.3 g (7.26 mmol) of 5-(isopropoxycarbonyl)-2,6-dimethyl-4-(2-nitrophenyl)-1,4(3,4)-dihydropyrimidine[5,6] and 6 mL of triethylamine in 50 mL of THF at −23 °C. After 1 h, a solution of 2.1 g (7.26 mmol) of 2-[*N*-benzyl-*N*-(2-naphthylmethyl)amino]ethanol[15] in 15 mL of THF was added at 0 °C. The mixture was stirred at 0 °C for 21 h, diluted with 10% NaOH solution, and extracted with *i*-Pr_2O. The organic layer was dried over $MgSO_4$ and evaporated to leave the residue, which was purified by SiO_2 column chromatography (Et_2O–*n*-hexane, 3:7) to give 2.38 g (52%) of compound 1s. Trituration with *i*-Pr_2O–*n*-hexane afforded pale yellow crystals: mp 85–89 °C; IR (cm^{-1}) ($CHCl_3$) 1725, 1695 NMR ($CDCl_3$) δ 1.04 (3 H, d, J = 6 Hz), 1.23 (3 H, d, J = 6 Hz), 2.23 (3 H, s), 2.50 (3 H, s), 2.85 (2 H, t, J = 6 Hz), 3.68 (2 H, s), 3.79 (2 H, s), 4.18–4.42 (2 H, m), 5.01 (1 H, m), 6.85 (1 H, s), 7.18–8.88 (16 H, m). Anal. ($C_{37}H_{38}N_4O_6$) C, H, N.

Synthesis of 3-[[2-[*N*-Benzyl-*N*-(3,4-dichlorobenzyl)amino]ethoxy]carbonyl]-5-(isopropoxycarbonyl)-2,6-dimethyl-4-(2-nitrophenyl)-3,4-dihydropyrimidine (1t). To a stirred solution of 317 mg (1 mmol) of 5-(isopropoxycarbonyl)-2,6-dimethyl-4-(2-nitrophenyl)-1,4(3,4)-dihydropyrimidine[5,6] and 0.83 mL (6 mmol) of triethylamine in 30 mL of tetrahydrofuran (THF) was added 60 μL (0.5 mmol) of phosgene dimer at −23 °C. After 1 h, a solution of 311 mg (1 mmol) of 2-[*N*-benzyl-*N*-(3,4-dichlorobenzyl)amino]ethanol in 10 mL of THF was added at 0 °C. The reaction mixture was stirred at room temperature for 20 h, quenched with H_2O, and extracted with $CHCl_3$.

The organic layer was dried and evaporated to leave 0.7 g of a residue, which was chromatographed on SiO_2 (benzene–acetone) to give 0.33 g (50%) of compound 1t.

Trituration with *i*-Pr_2O yielded colorless crystals: mp 81–85 °C; IR (cm^{-1}) ($CHCl_3$) 1735, 1705; NMR ($CDCl_3$) δ 1.04 (3 H, d, J = 6 Hz), 1.25 (3 H, d, J = 6 Hz), 2.27 (3 H, s), 2.47 (3 H, s), 2.80 (2 H, t, J = 6 Hz), 3.57 (2 H, s), 3.61 (2 H, s), 4.12–4.39 (2 H, m), 4.93–5.08 (1 H, m), 6.82 (1 H, s), 7.05–7.80 (12 H, m). Anal. ($C_{33}H_{34}Cl_2N_4O_6$) C, H, N.

Synthesis of 5-[(Cyclopropylmethoxy)carbonyl]-3-[(heptyloxy)carbonyl]-6-methyl-4-(2-nitrophenyl)-3,4-dihydropyrimidin-2(1*H*)-one (8b). To a stirred slurry of 16 mg (0.33 mmol) of 50% NaH–mineral oil in 1.5 mL of hexamethylphosphoramide (HMPA) was added a solution of 100 mg (0.30 mmol) of 5-[(cyclopropylmethoxy)carbonyl]-6-methyl-4-(2-nitrophenyl)-3,4-dihydropyrimidin-2(1*H*)-one[5] in 1.5 mL of HMPA at 0 °C, and stirring was continued at room temperature for 40 min. To the mixture was added 37 μL (0.49 mmol) of chloromethyl methyl ether at 0 °C, and stirring was continued at room temperature for 19h. The reaction mixture was quenched with aqueous $NaHCO_3$ solution and extracted with ether. The organic layer was washed with water, dried over $MgSO_4$, and evaporated to leave a residue, which was purified by SiO_2 chromatography to afford 60 mg (52%) of 5-[(cyclopropylmethoxy)carbonyl]-1-*N*-(methoxymethyl)-6-methyl-4-(2-nitrophenyl)dihydropyrimidin-2-one: mp 120–122 °C (pale yellow needles, AcOEt–*n*-hexane); IR (cm^{-1}) ($CHCl_3$) 3420, 1710, 1690, 1630; NMR ($CDCl_3$) δ −0.20–0.03 (2 H, m), 0.22–0.40 (2 H, m), 0.67–0.84 (1 H, m), 2.13 (3 H, s), 3.37 (3 H, s), 3.70 (2 H, d, J = 7 Hz), 5.11–5.37 (2 H, m), 5.80 (1 H, d, J = 3 Hz), 6.04 (1 H, br s), 7.38–8.00 (4 H, m); UV λ_{max}^{MeOH} 276 nm (ε 7500). To a stirred slurry of 9 mg (0.18 mmol) of 50% NaH–mineral oil in 8 mL of THF was added a solution of 58 mg (0.15 mmol) of the dihydropyrimidin-2(3*H*)-one at 0 °C. After stirring for 20 min at room temperature, 27 μL (0.15 mmol) of *n*-heptyl chloroformate was added at room temperature. After 20 min, the reaction mixture was quenched with brine and extracted with $CHCl_3$. The organic layer was dried over $MgSO_4$ and evaporated to leave a residue, which was chromatographed on SiO_2 (benzene–acetone, 20:1) to give 61.5 mg (77%) of 5-[(cyclopropylmethoxy)carbonyl]-3-[(heptyloxy)carbonyl]-1-*N*-(methoxymethyl)-6-methyl-4-(2-nitrophenyl)dihydropyrimidin-2-one.

To a stirred solution of 10 mg (0.0188 mmol) of the dihydropyrimidin-2(3*H*)-one in 0.2 mL of $CHCl_3$ was added 0.2 mL of concentrated HCl. The solution was heated at reflux for 2 h, quenched with saturated aqueous K_2CO_3, and extracted with $CHCl_3$. The organic layer was dried over $MgSO_4$ and evaporated to give 8.5 mg (100%) of **8b**: mp 101–104 °C (colorless needles, AcOEt–*n*-hexane); IR (cm^{-1}) ($CHCl_3$) 3400, 1770, 1730, 1645; NMR ($CDCl_3$) δ 0.10–0.28 (2 H, m), 0.39–0.56 (2 H, m), 0.86 (3 H, t, J = 7 Hz), 1.00–1.41 (9 H, m), 1.55–1.75 (2 H, m), 2.42 (3 H, s), 3.78–4.03 (2 H, m), 4.13–4.30 (2 H, m), 7.04 (1 H, s), 7.40–8.00 (5 H, m); UV λ_{max}^{MeOH} 274 nm (ε 9800). Anal. ($C_{24}H_{31}N_3O_7$) C, H, N.

Synthesis of 3-[[(*N*,*N*-Dibenzylamino)ethoxy]carbonyl]-2-(trifluoromethyl)-5-(isopropoxycarbonyl)-6-methyl-4-(2-nitrophenyl)-3,4-dihydropyrimidine (9g). To a stirred solution of 113 μL (0.94 mmol) of phosgene dimer in 9 mL of THF was added a solution of 289 mg (0.78 mmol) of 2-(trifluoromethyl)-5-(isopropoxycarbonyl)-6-methyl-4-(2-nitrophenyl)-1,4(3,4)-dihydropyrimidine[5,6] and 0.655 mL (4.68 mmol) of triethylamine in 10 mL of THF at −23 °C. After 1.5 h, a solution of 1.13 g (4.98 mmol) of 2-(dibenzylamino)ethanol[15] in 11 mL of THF was added at 0 °C. The reaction mixture was stirred at 0 °C for 1 h and at room temperature for 16 h, quenched with H_2O, and extracted with CH_2Cl_2. The organic layer was dried over $MgSO_4$ and evaporated to leave a residue, which was chromatographed on SiO_2 (AcOEt–*n*-hexane, 2:8–3:7) to afford 54 mg (11%) of compound **9g**: IR (cm^{-1}) ($CHCl_3$) 1740, 1700; NMR ($CDCl_3$) δ 1.06 (3 H, d, J = 6 Hz), 1.27 (3 H, d, J = 6 Hz), 2.55 (3 H, br s), 2.75–2.85 (2 H, m), 3.55 (4 H, m), 4.15–4.40 (2 H, m), 4.98–5.10 (1 H, m), 6.98 (1 H, s), 7.15–7.83 (14 H, m). Anal. ($C_{33}H_{33}F_3N_4O_6$) C, H, N.

Synthesis of 6-[[2-[*N*-Benzyl-*N*-(3,4-dichlorobenzyl)amino]ethoxy]methyl]-5-(ethoxycarbonyl)-3-(methoxycarbonyl)-2-methyl-4-(2-nitrophenyl)-3,4-dihydropyrimidine (11). To a stirred slurry of 583 mg (12.15 mmol) of 50% NaH–mineral oil in 6 mL of THF was added slowly a solution of 1.89 g (6.08 mmol) of 2-[*N*-benzyl-*N*-(3,4-dichlorobenzyl)amino]-ethanol[15] in 6 mL of THF at room temperature. Stirring was continued at 40 °C for 30 min. A solution of 1.0 g (6.08 mmol) of ethyl 4-chloroacetoacetate in 12 mL of THF was added in small portions over a 2-h period at room temperature. The mixture was stirred at room temperature for 15 h, quenched with 10% HCl solution, and extracted with AcOEt. The organic layer was washed with 10% NaOH solution, dried over $MgSO_4$, and evaporated to leave 2.1 g of the β-keto ester. Piperidine (45 μL, 0.45 mmol) was added to a solution of 2.0 g (4.56 mmol) of the β-keto ester and 680 mg (4.5 mmol) of 2-nitrobenzaldehyde in 20 mL of benzene.

The solution was refluxed for 14 h with the aid of a Dean–Stark aparatus. After evaporation of the solvent, the residue was chromatographed on SiO_2 (AcOEt–*n*-hexane, 3:7) to give 1.50 g of the benzylidene derivative. A mixture of 172 mg (1.82 mmol) of acetamidine hydrochloride and 204 mg (1.82 mmol) of *t*-BuOK in 2 mL of *N*,*N*-dimethylformamide (DMF) was stirred at 0 °C for 10 min. A solution of 840 mg (1.47 mmol) of the benzylidene derivative in 3 mL of DMF was added. Stirring was continued at 0 °C for 1 h and at room temperature for 3 h.

p-Toluenesulfonic acid monohydrate (559 mg, 2.94 mmol) and 5 mL of DMF were added to the reaction mixture at room temperature. The mixture was heated at 110 °C for 3 h, diluted with 10% NaOH solution, and extracted with *i*-Pr_2O. The organic layer was washed with H_2O, dried over $MgSO_4$, and evaporated to leave

(15) Cho, H.; Ueda, M. Suntory patent, JP 38345, 1987; EP 0280227, 1988.

564 mg of a residue, which was purified by SiO_2 column chromatography to yield 150 mg of compound **10**. A solution of 73 mg (0.12 mmol) of **10** in 3 mL of THF was added to a stirred slurry of 7 mg (0.14 mmol) of 50% NaH–mineral oil in 3 mL of THF at 0 °C. Methyl chloroformate (11 μL, 0.14 mmol) was added at 0 °C. The mixture was stirred at room temperature for 1 h, quenched with H_2O, and extracted with $CHCl_3$. The organic layer was dried over $MgSO_4$ and evaporated to leave 67 mg of a residue, which was purified by SiO_2 column chromatography (AcOEt–*n*-hexane, 4:1) to afford 25 mg (39% based on consumed starting material) of compound **11**: mp 112–116 °C (HCl salt; from Et_2O–acetone); IR (cm^{-1}) ($CHCl_3$) 1740, 1705; NMR ($CDCl_3$) δ 1.22 (3 H, d, J = 7 Hz), 2.34 (3 H, s), 2.75 (2 H, t, J = 6 Hz), 3.58–3.70 (6 H, m), 4.05–4.20 (2 H, m), 4.64 (1 H, br s), 6.83 (1 H, s), 7.15–7.80 (12 H, m). Anal. ($C_{33}H_{34}Cl_2N_4O_5$) C, H, N.

Pharmacological Methods. (I) Vasodilative Effects on the Vertebral Artery in Anesthetized Dogs. Mongrel dogs, weighing 7–15 kg, of either sex were anesthetized with an intravenous (iv) or an intraperitoneal (ip) injection of 30 mg/kg of sodium thiopental followed by a combination of urethane, 400 mg/kg (iv), and α-chloralose, 60 mg/kg (iv). The anesthetized animal was artificially ventilated with a positive pressure respirator after tracheal intubation. After left thoracotomy, the left vertebral artery was exposed and blood flow was measured with an implantable flow probe connected to an electromagnetic flow meter. Mean arterial blood pressure was measured from the right femoral artery. Vascular resistance of the vertebral artery was computed by dividing the mean arterial blood pressure by mean vertebral blood flow and was recorded continuously on a polygraph. The test compounds were dissolved in ethanol, and the solution was administered intravenously into the cannulated femoral vein at a volume of 0.05 mL/kg. The dose required to decrease vertebral vascular resistance by 30% (ED30) and half-recovery time (T1/2) of the vascular resistance decrease at ED30 were determined. The ED_{30} was determined by cumulative rising doses (N = 3).

(II) Hypotensive Effect in Conscious Spontaneously Hypertensive Rats. More than 1 day before the experiment, an indwelling catheter was inserted into the distal aorta via the left femoral artery for blood pressure measurement under petroleum ether anesthesia. The catheters were filled up with heparin sodium solution (300 units/mL) to prevent blood coagulation. About 1 h before the experiment, each animal was transferred into an individual translucent cyrindrical box. Systematic blood pressure was measured from the previously implanted catheter with the aid of a pressure transducer and heart rate, with a cardiotachometer triggered with the blood pressure pulse under conscious and unrestrained conditions. The test compounds were dissolved in 10% ethanol and administered orally.

(III) Acute Toxicity. Five fasted male ddY mice were used for each compound. Each compound was administered orally, and the survival of animals was observed for a week.

(IV) Effects on Atrioventricular Conduction System in Anesthetized Open-Chest Dogs. Thirteen dogs of either sex, weighing 6–14 kg, were used under anesthesia with sodium pentobarbital (30 mg/kg, iv). The animals were intubated with a cuffed endotracheal tube and ventilated with room air using a respirator in a tidal volume of 20 mL/kg at a rate of 20 strokes/min. The chest was opened by midsternal thoracotomy, and the heart was exposed and suspended in a pericardial cradle.

A stimulating bipolar electrode was sutured onto the epicardial surface of the right atrium. A bipolar recording electrode was also sutured onto the right atrium 1 cm apart from the stimulating one and another recording electrode onto a free wall of the right ventricle. The pacemaker activity was reduced by an injection of 0.1–0.2 mL of 37% formaldehyde into the wall of the right atrium near the sinus node. The atrium was driven by means of a cardiac stimulator (Nihon Kohden, Tokyo Japan, AB-621G) with square wave pulses of 1.5–2.5 V and of 2 ms in duration at a frequency of 2.5 Hz. The atrial and ventricular electrograms were amplified by ECG amplifiers (Nihon Kohden, AB-621G) and recorded on a polygraph recorder (Nihon Kohden, RM-6000) at a paper speed of 200 mm/s. The interval between the atrial (electrogram) and the ventricular electrogram was measured as an atrioventricular conduction time (AVCT).

Compounds were dissolved in 100% dimethyl sulfoxide (DMSO), and the solutions were injected into the femoral vein in a volume of 0.05 mL/kg.

(V) [^{3}H]Nitrendipine Binding Assay. The rat cerebral cortex was homogenized in 50 volumes of ice-cold 50 mM Tris-HCl buffer (pH 7.7) and centrifuged at 40000*g* for 10 min. The pellet was resuspended in the same volume of buffer and recentrifuged. This procedure was repeated twice. The pellets were finally resuspended in 70 volumes of ice-cold buffer. Binding assays were carried out in a total volume of 1 mL containing 0.2 nM [^{3}H]-nitrendipine (85.9 Ci/mmol; Du Pont New England Nuclear), drug, and tissue homogenate. Following 1-h incubation at 25 °C, the reaction was terminated by rapid vacuum filtration over Whatman GF/B glass fiber filters. The filters were rapidly washed three times with 5 mL of the same buffer. Nonspecific binding was defined as that which occurred in the presence of 1 μM nonradioactive nitrendipine and was subtracted from total binding to give the specific binding. Radioactivity was measured by liquid scintillation spectroscopy in 5 mL of scintillation cocktail (ACSII).

(VI) Statistical Methods. The data of Figure 1 were statistically analyzed by Student's t test.

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